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Poster

P012. Postnatal Neurogenesis: Temporal and Spatial Patterns

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P012.01

Topic: A.02. Postnatal Neurogenesis

Support: Research Grants Council of Hong Kong C4055-19GF

Title: Investigation of brain cortical development in moderate-late preterm infants

Authors: *X. GENG, P. H. CHAN, C. LAI, W. C. W. CHU, H. S. LAM, P. C. M. WONG; Chinese Univ. of Hong Kong, Shatin NT, Hong Kong, Hong Kong

Abstract: Preterm birth is among the risk factors for children's developmental delay and language impairment. The majority of preterm babies (up to 84%) are moderate to late preterm (MLPT) infants, and relatively less attention has been paid to their potential neurodevelopmental issues. One recent study have reported abnormal total CSF volume in MLPT infants. Another study showed that rain volumes were associated with cognitive and language development in MLPT children at 2 years of age. However, it is not clear whether MLPT infants show differences in cortical measures. Cortical measures have been shown to relate to later behavioral outcomes in full term infants.

A total of 31 newborn infants were included in the study: 17 MLPT (32-36 weeks' gestational age (GA)) vs 14 term babies (>37 weeks' GA) with an age range of 6-89 days (corrected chronological age) at MRI scan. Age and sex between the two groups were matched (MLPT: 47.5 days, 9 girls; control: 52.3 days, 8 girls). MRI data were acquired during natural sleep. An MRI-compatible incubator was used for individuals with weight < 5KG. T1-weighted structural images were acquired to extract neural cortical features, with spatial resolution=0.8x0.8x0.8 mm3, TR=1.8s, TE=2.82ms, flip angle=8o. The raw data was sent to infantFS, a FreeSurfer based pipeline that provides infants templates for template-guided segmentation. The major processing steps include: intensity uniformization, skull stripping, spatial normalization, tissue segmentation, surface reconstruction of the outer and inner surfaces of the cortex, calculating cortical thickness and surface area. Whole brain vertex-based statistical analysis was conducted using a general linear model: imaging measures=age+Group (MLPT and control). The effect of Group was examined on cortical thickness and surface area.

Compared with controls, decreased cortical thickness in right fusiform gyrus and right primary occipital cortex were found in the MLPT group. Increased cortical thickness in right supramarginal gyrus, and enlarged surface area in left precentral gyrus were also observed in the MLPT group. The cluster of decreased cortical thickness in right primary occipital cortex, and that of increased surface area in left precentral gyrus remained significant after multiple comparison correction. These findings suggest that moderate to late preterm birth may affect the

development of primary visual and sensori-motor cortices as early as perinatal period of life. These abnormal sensory processing areas may later affect higher-order cognitive processing, which may eventually contribute to neurodevelopmental problems later in life.

Disclosures: X. Geng: None. **P.H. Chan:** None. **C. Lai:** None. **W.C.W. Chu:** None. **H.S. Lam:** None. **P.C.M. Wong:** None.

Poster

P012. Postnatal Neurogenesis: Temporal and Spatial Patterns

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P012.02

Topic: A.02. Postnatal Neurogenesis

Support: MH091844 (AD)
MH115215 (AD)
MH091427 (EDL)
MH123153 (EDL)
NS085502 (LMS)

Title: Adult-born neurons maintain hippocampal cholinergic inputs and support working memory during aging

Authors: *M. BOMPOLAKI[1,2], G. KIRSHENBAUM[1,2], C.-Y. CHANG[3], R. SHANSKY[4], A. HARRIS[1,2], L. M. SAVAGE, Ph.D.[5], E. D. LEONARDO[1,2], A. DRANOVSKY[1,2];
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Abstract: Adult neurogenesis contributes to the maintenance of normal hippocampal functions and its impairment has been implicated in disorders of stress, memory and cognition. However, a systems level understanding of how low levels of ongoing neurogenesis contribute to major brain functions over time is lacking. We have performed a connectivity analysis of inputs into the dentate gyrus in mice living without neurogenesis. Animals that do not have neurogenesis in adulthood developed a slow-emerging deficit in the spontaneous alternation task indicating working memory impairments. This deficit was accompanied by selective reorganization of the cholinergic projection to the dentate gyrus (DG). Somatotopic organization of medial septal cholinergic hippocampal inputs was lost after 5 months without neurogenesis as neurons that normally project to the ventral hippocampus also innervated the dorsal hippocampus. Thus, more cholinergic neurons projected to the dorsal DG overall. Despite increased cholinergic connectivity, the levels of acetylcholine (ACh) in the dorsal DG were significantly lower in

animals without neurogenesis than controls. Behavioral pharmacology studies revealed that functional deficits in acetylcholine signaling precede the delayed changes in cholinergic innervation and ACh levels. We hence used fluorescent acetylcholine sensor fiber photometry to study the dynamics of ACh release in the DG before behavioral, anatomic, and biochemical changes emerged. During these early stages we observed slower and attenuated ACh responses upon task exposure when cognitive demand is highest. This result suggests that adult neurogenesis is important for maintaining normal cholinergic signaling. Hence, slow structural reorganization of cholinergic innervation, decline in baseline ACh, and accompanying working memory deficits are likely a failed compensatory response to functional disruption of cholinergic signaling. Importantly, chemogenetic increase in cholinergic signaling rescued behavioral deficits in animals long after anatomic and biochemical changes took place. Our findings support the notion that ongoing neurogenesis throughout life is necessary to maintain normal septohippocampal circuit function and optimal working memory performance. These results have important implications for understanding age and stress related cognitive decline and for treatment development.

Disclosures: M. Bompolaki: None. **G. Kirshenbaum:** None. **C. Chang:** None. **R. Shansky:** None. **A. Harris:** None. **L.M. Savage:** None. **E.D. Leonardo:** None. **A. Dranovsky:** None.

Poster

P012. Postnatal Neurogenesis: Temporal and Spatial Patterns

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P012.03

Topic: A.02. Postnatal Neurogenesis

Support: ANR FINEST (ANR-11-BSV2-0029)

Title: Ultrastructure of the teleost optic tectum : electron microscopic analysis

Authors: J.-M. HERMEL[1], ***K. YAMAMOTO**[1], J.-S. JOLY[2];
[1]Univ. Paris-Saclay, CNRS UMR9197, Gif-sur-Yvette, France; [2]TEFOR Paris-Saclay, CNRS UMS2010, INRA UMS1451, Gif-sur-Yvette, France

Abstract: Unlike the mammalian brain, the optic tectum (OT) is particularly well-developed in the teleost brain. It has been known that the teleost OT contains stem cell niche that contributes post-embryonic growth of the structure, nevertheless, very little is known about the ultrastructure of this area. Here, we acquired, stitched and combined images of low to meso resolution (500X-10000X) in order to get both a wide (200µmx200µm) portion of the structure with a good picture of the ultrastructure of the cells present in the OT stem cell niche. Based on this technique we observed several genuine anatomical data. We observed the presence of a basal lamina with several branch in the area of stem cell niche. The cells of the same putative age i.e. being at the

same distance from the cell niche dividing area have an inter-digitized membrane but not with their older or younger neighbor. In the ventricle between some ventricle contacting cells, there is a network of extracellular space that might play a role in the cell movement thus in the physical growth of the OT. The OT ventricle contains many closed membrane structure that appear to be actively produced by cells bordering the ventricle. Contrary to the adult, at 7 dpf, there is no blood capillaries in the posterior part of the OT. At 43hpf, there are many small bundles of axon in the process of being ensheeted. Most often, a single oligocyte is enwrapping up to 15 axons at the same time. At early stages, very few cells are ciliated with only one cilium but there is also few multiciliated cells. We observed also few peculiar cases of nuclear mitochondria. Finally, we describe the presence of stretched cells organized in bundles. These cells, most probably tanycytes have often a single cilia in the ventricle and their long process extend most of the time toward blood capillaries where it is possible to observe fenestration. Altogether, we believe that the technic of stitching images obtained at several resolution and these anatomical observations will contribute to a better understanding of the brain stem cell niches.

Disclosures: J. Hermel: None. **K. Yamamoto:** None. **J. Joly:** None.

Poster

P012. Postnatal Neurogenesis: Temporal and Spatial Patterns

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P012.04

Topic: A.02. Postnatal Neurogenesis

Support: Wesleyan University

Title: Singing-related activation of darpp-32+ adult-born neurons in zebra finch hvc

Authors: *J. V. ARONOWITZ[1], B. FILIO, III[1], Y. ZHAO[1], K. NELSON-MAROIS[1], C. L. PYTTE[2], G. B. AARON[1];
[1]Wesleyan Univ., Middletown, CT; [2]Psychology, Queens Col., Flushing, NY

Abstract: In many songbirds including zebra finches (*T. guttata*), new neurons are continually incorporated into brain regions that subserve song learning, perception, and production. HVC (proper name), a brain region necessary for song production, receives and integrates new neurons (HVC NNs) throughout the life of the animal. Approximately half of these HVC NNs can be retrogradely labeled from the robust nucleus of the arcopallium, the first synaptic relay in the vocal motor pathway underlying learned song production. Interestingly, both backfilled and non-backfilled HVC NNs express the activity-dependent immediate early genes ZENK and c-Fos during song production, suggesting that they are active during song production once they are integrated into existing HVC circuitry (Tokarev, et al., 2016). Our lab has recently shown that approximately 50% of HVC NNs express DARPP-32 during their first month post-mitosis.

DARPP-32 is a phosphoprotein related to the expression of dopamine receptors, although its activity is modulated by many different neurotransmitter systems including glutamate and norepinephrine. Furthermore, these DARPP-32+ HVC NNs receive putative tyrosine hydroxylase synaptic contacts, have transiently larger nuclei than DARPP-32-neg HVC NNs, and cannot be retrogradely labeled from RA at 28 days post mitosis, suggesting that they may be represent a unique subtype of adult-born neuron in HVC. Here, we show that DARPP-32+ HVC NNs express immediate early genes following song production, and that singing-related immediate early gene expression is related to neuronal age and may be related to the social context in which the song is sung.

Disclosures: J.V. Aronowitz: None. **B. Filio:** None. **Y. Zhao:** None. **K. Nelson-Marois:** None. **C.L. Pytte:** None. **G.B. Aaron:** None.

Poster

P012. Postnatal Neurogenesis: Temporal and Spatial Patterns

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P012.05

Topic: A.02. Postnatal Neurogenesis

Support: NRF-2020R1C1C1011857
NRF-2020M3C1B8081320

Title: Age-dependent electroencephalographic spectral patterns during midazolam-induced general anesthesia for congenital heart surgery in children

Authors: I.-K. SONG[1], Y.-S. KIM[2], B. A. LEE[1], W.-J. SHIN[1], *S.-E. KIM[2];
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Abstract: Background: Age-dependent electroencephalographic (EEG) characteristics may guide brain monitoring during general anesthesia (GA) in children, considering the neurodevelopmental state of the brain [1,2]. However, the dynamics of EEG in children receiving GA for congenital heart surgery (CHS), which can be affected by hypothermia, fluctuations in cerebral metabolism, and anesthetics [3], has not been elucidated. This study investigated the changes in EEG spectral patterns according to age and to body temperature variation during CHS under midazolam-induced GA in children. **Methods:** Four-channel frontal EEG was recorded continuously from 55 children (aged 0-48 months) undergoing CHS with midazolam-induced GA. The effect of midazolam-induced GA on brain was compared across four age-groups in EEG spectral power: 0-2 months (n=6), 3-4 months (n=18), 5-7 months (n=6), and 8-42 months (n=25). In addition, EEG oscillatory patterns were analyzed according to the five stages of CHS determined by the core body temperature: pre-hypothermia, cooling,

hypothermia, rewarming, and recovery. We computed group-median spectrograms by taking the median power across all subjects at each time and frequency of each stage. Peak frequencies within the alpha (8-13 Hz) and beta (20-30 Hz) range were assessed by a peak detection algorithm and manually adjusted according to the stages. **Results:** During maintenance of midazolam-induced GA, the frontal spectrogram demonstrated distinct distributions of slow-delta, alpha, and beta oscillations according to age group. Alpha oscillations were prominent in subjects aged 3-4 months, whereas beta oscillations were prominent in subjects aged five months and older. In the cooling period, the peak frequency of beta oscillations slowed down, shifting significantly toward the alpha band boundaries. The rewarming period showed a gradual acceleration of a slowed beta band that returns to physiological levels as core body temperature rises. **Conclusions:** A significant age-dependent EEG spectral patterns were observed during midazolam-induced GA. Anesthesia-induced age-dependent EEG characteristics changed in different ways following body temperature alterations during CHS, likely due to differences in brain development. These results indicate that EEG monitoring is essential during GA and can provide useful information on the appropriate depth of anesthesia in relation to the neurodevelopmental status of children.
References[1] Cornelissen et al (2015*) eLife*. 4:e06513. [2] Lee et al (2017) *Anesthesiology* 127(2):293-306.[3] Kaiser et al (2020) *Br. J Anaesth* 25(3):291-297.

Disclosures: I. Song: None. **Y. Kim:** None. **B.A. Lee:** None. **W. Shin:** None. **S. Kim:** None.

Poster

P012. Postnatal Neurogenesis: Temporal and Spatial Patterns

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P012.06

Topic: A.02. Postnatal Neurogenesis

Support: FUSF grant

Title: Prolonged Integration of Adult-born Axin2 Cell Lineage into Granule Neurons of the Dentate Gyrus

Authors: *K. SHARIFI[1], F. FARZAD[1], S. SOLDOZY[1], M. KALANI[2], P. TVRDIK[1];
[1]Univ. of Virginia, Charlottesville, VA; [2]St. John's Neurosci. Inst., Tulsa, OK

Abstract: The Wnt pathway plays multiple roles during adult neurogenesis. The expression of Axin2 is induced by Wnt/β-catenin signaling, lending this gene a sensitive indicator of canonical Wnt activity. We have employed pulse-chase genetic lineage tracing with the Axin2-CreER allele to follow the fate of Axin2-positive cells in the hippocampus. Simultaneously with tamoxifen induction of Axin2 fate mapping, the dividing cells were marked with 5-ethynyl-2´-deoxyuridine (EdU). Tamoxifen induction resulted in significant increase of dentate gyrus

granule cells three months later; however, none of these neurons contained EdU signal. Conversely, six months after the tamoxifen/EdU pulse-chase labeling, EdU-positive granule neurons were identified in each animal. Our data implies that Axin2 is expressed in several different stages of adult granule neuron differentiation, including the dividing neural stem cells. However, our data also indicates that the process of integration of the adult-born neurons from certain cell lineages may take longer than previously thought.

Disclosures: K. Sharifi: None. **F. Farzad:** None. **S. Soldozy:** None. **M. Kalani:** None. **P. Tvrdik:** None.

Poster

P012. Postnatal Neurogenesis: Temporal and Spatial Patterns

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P012.07

Topic: A.02. Postnatal Neurogenesis

Support: Junta de Comunidades de Castilla la Mancha

Title: Rostral migratory stream description along the olfactory tract of Macaca fascicularis

Authors: *J. RASPEÑO[1], J. AGUILAR-GOMEZ[1], S. GONZALEZ-GRANERO[4], F. SANCHO-BIELSA[2], A. COZAR-CUESTA[3], E. ARTACHO-PERULA[1], R. INSAUSTI[1], J. GARCIA-VERDUGO[4], C. DE LA ROSA-PRIETO[1];
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Abstract: Most mammals generate new neurons during adulthood that are incorporated into a pre-existing olfactory circuit, presumably to keep their functionality intact. This process is known as adult neurogenesis, and involve different types of cells: stem cells, amplifying cells and migrating cells. This migrating neuroblasts, depart from the walls of the lateral ventricles (SVZ), and travel through the olfactory peduncle (OP) conforming the rostral migratory stream (RMS), until they reach the olfactory bulbs (BO) where they differentiate into interneurons. This migratory process has been widely described in rodents but not as well in primates. The potential of this process as a model for developing neuronal replacement strategies for brain repair in humans, requires better characterization in animal models more similar to own. In this work we present 6 cases of young adult primates (*Macaca fascicularis*) in which the olfactory tract was longitudinally dissected into the main olfactory bulb, and an anterior and a posterior region of OP. Immunofluorescence (IF) and Electron microscopy (EM) techniques were performed to identify the cytoarchitecture and cell populations, such as glia (GFAP-positive cells) and

migrating neuroblasts (DCX-positive cells) along the OP. In addition, stereological estimation of the number and occupied area of DCX-positive cells present in the OP were performed.Our findings show that, unlike rodents, where there is an internal, single, and continuous RMS, macaques show a different organization with neuroblasts conforming several chains at level of the caudal regions (closer to the SVZ), which tend to converge reaching the OB, and even groups of migrating cells over the periphery of the OP. Furthermore, all these migrating cells travel in waves forming clusters, not in a continuous way. We also observed some examples of morphologies compatible with radial migrations along the OP. These cells seem to be capable to leave the glial scaffold that guides the RMS and join into the surrounding tissue, reaching even the outermost layers for unknown reason. Further studies in humans and close phylogenetically species are necessary to elucidate the underlying process of adult human neurogenesis and its clinical applications. **KEYWORDS: Adult neurogenesis, doublecortin, macaque, migration, olfactory peduncle**

Disclosures: J. Raspeño: None. **J. Aguilar-Gomez:** None. **S. Gonzalez-Granero:** None. **F. Sancho-Bielsa:** None. **A. Cozar-Cuesta:** None. **E. Artacho-Perula:** None. **R. Insausti:** None. **J. Garcia-Verdugo:** None. **C. de la Rosa-Prieto:** None.

Poster

P167. Oligodendrocytes: Disease Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P167.01

Topic: B.11. Glial Mechanisms

Support: Grant #5010, NSB
NIH Grant R25 NS 80686

Title: Activity-based anorexia in mice downregulates oligodendrocyte lineage cells

Authors: *Z. WANG[1], C. V. MELENDEZ-VASQUEZ[2], N. S. BURGHARDT[3], A. ALMAKTAR[1];
[1]Hunter Col. CUNY, New York, NY; [2]Hunter Col., New York, NY; [3]Dept. of Psychology, Hunter College, CUNY, New York, NY

Abstract: Anorexia nervosa (AN) is a life-threatening eating disorder that occurs predominately in women, usually begins during adolescence, and frequently involves excessive exercise, which is thought contribute importantly to the development and maintenance of the disorder. Previous studies indicate that AN is associated with deficits in white matter, but the effects of AN on central nervous system myelination are poorly understood. We used the activity-based anorexia model (ABA) in mice to study how the combination of food restriction and exercise affects oligodendrocytes, the glial cells responsible for forming myelin. Adolescent (postnatal day 42)

female C57Bl/6N mice were distributed into 4 groups: ABA (restricted food access; unlimited wheel access), wheel control (WH; unlimited food and wheel access), food-restricted control (FR; restricted food access, unlimited access to a locked wheel), and homecage control (HC; unlimited access to food and a locked wheel). For the ABA and FR groups, access to food was limited to the first 2 hours of the dark cycle, during which time mice were free to eat as much as they wanted. Mice that lost 25% of their baseline weight were immediately removed from the experiment while the remaining mice were tested for 11 days. Tissue was collected after one day of recovery and immunocytochemistry was used to stain coronal brain slices (n=4 mice/group) for CC1, a marker of mature oligodendrocytes, and NG2, a marker of immature oligodendrocyte progenitor cells. Fiji was used to quantify stained cells in the corpus callosum and cortex. As expected, ABA and FR groups lost substantial weight compared to the non-food restricted groups, with the FR group surviving in the model longer than the ABA group ($p<0.001$). Weight loss in the ABA group correlated with levels of wheel running ($r=0.38$), which were significantly higher during the light cycle than the WH control group ($p<0.001$). In the corpus callosum, mice in the ABA condition had 74-81% fewer cells stained for CC1 (10.25±1.1 vs 38.75 ±8.9) and NG2 (16.25±3.3 vs. 88±31.7) than the HC control group. Although the FR group had 30-38% fewer oligodendrocyte lineage cells than the HC controls, this change was not statistically significant. Of note, exercise alone did not affect cell counts in the WH controls. Interestingly, no group differences in CC1+ or NG2+ cells were detected in the cortex. Together, these findings indicate that ABA drives downregulation of oligodendrocyte lineage cells in the corpus callosum. We continue to study the underlying cause of these changes, consequent effects on myelination, and implications for patients with AN.

Disclosures: Z. Wang: None. **C.V. Melendez-Vasquez:** None. **N.S. Burghardt:** None. **A. Almaktar:** None.

Poster

P167. Oligodendrocytes: Disease Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P167.02

Topic: B.11. Glial Mechanisms

Support: Research Grant Council: GRF16124916
Food and Health Bureau: HMRF05163736
PolyU Start-up Fund: P0030307

Title: Apoe4 compromises white matter integrity by harnessing oligodendrocyte differentiation - a radio-pathological investigation

Authors: *G. W. Y. CHENG[1], J. HUANG[2], K. K. S. MOK[1], S. H. S. YEUNG[1], Z. CHEN[2], K. HERRUP[3], H. K. F. MAK[4], K. W. Y. CHAN[2,5], K.-H. TSE[1];

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Abstract: Apolipoprotein E *(APOE)* ε4 allele is not only the strongest genetic risk factor for sporadic Alzheimer's disease (AD) but is also associated with poor white matter (WM) integrity in the healthy population. We previously reported a significant reduction of oligodendrocyte (OL) population in the frontal cortex of AD subjects carrying *APOE4* alleles, but the cellular mechanism remains unknown. Here, we aim to test if *APOE4* disrupts OL plasticity and alters WM integrity. To investigate, the relationship between neuroimaging and OL lineage was examined in *Apoe*-knockout (*Apoe*$^{-/-}$), humanized APOE3 and APOE4 (*hAPOE*)-knockin mice at 1, 3, 6, & 12 months ($n \geq 4$/group) using preclinical 3T MRI, histology analysis, Y-maze behavioral test as well as gene and protein expression assays. In the neocortex and corpus callosum, unexpectedly, minimal differences in myelin expression (MBP, PLP, MAG, MOG) were found between all mouse strains across all age groups by immunohistochemistry, immunoblotting and real-time PCR. Despite the constant myelin level, diffusion kurtosis imaging consistently identified significant alterations of WM integrity in the anterior cingulate among the transgenic mice, where abnormally increased radial diffusivity ($P = 0.0275$) and mean diffusivity ($P = 0.044$) were found in *Apoe*$^{-/-}$ and hAPOE4 genotype mice reflecting disrupted myelination vs control. Intriguingly, stereotactic radiological-histological analysis of gray matter (*Anterior cingulate, motor, somatosensory cortex*) and WM (*Genu, external capsule, cingulum and anterior commissure*) at 10 months (Bregma 0.8) revealed significant and region-specific disruption of OL lineage by APOE4 genotype. In the anterior cingulate of hAPOE4 mouse, altered WM integrity was accompanied by 23% reduction in the overall Olig2^{+} OL population and 60% loss of CC1^{+} mature OLs, despite a 3.2 fold increase of the committed Nkx2.2^{+} OL precursor cells (OPC, relative to wildtype). Such accumulation of OPCs was also found in the genu, body and splenium of the corpus callosum along the midline of hAPOE4, but not hAPOE3 mice. No cognitive impairment nor neuronal loss was identified. These in vivo findings were further validated by primary OL cultures where we found that postnatal OL derived from *Apoe*$^{-/-}$, hAPOE3 and hAPOE4 animals developed normally. However, lipidated recombinant human ApoE4, but not ApoE3 protein, suppressed the differentiation and halved the formation of MBP^{+} myelin sheet formation in *Apoe*$^{-/-}$ OPCs (-52.3%, $P = 0.026$). Together, *APOE4* is likely to hamper OPC differentiation and their myelination is sustained only by a disproportionally reduced mature OLs that may ultimately jeopardize WM integrity.

Disclosures: G.W.Y. Cheng: None. **J. Huang:** None. **K.K.S. Mok:** None. **S.H.S. Yeung:** None. **Z. Chen:** None. **K. Herrup:** None. **H.K.F. Mak:** None. **K.W.Y. Chan:** None. **K. Tse:** None.

Poster

P167. Oligodendrocytes: Disease Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P167.03

Topic: B.11. Glial Mechanisms

Support: Research Grant Council ECS25104220
Food and Health Bureau HMRF06173836
PolyU Start-up Fund P0030307

Title: Divergent effects of ATM mutations on oligodendrocytes - the cellular basis of myelin pathology in Ataxia Telangiectasia

Authors: *S. H. S. YEUNG[1], A. CHENG[2], G. W. Y. CHENG[1], K. K. S. MOK[1], K. HERRUP[3], K.-H. TSE[1];
[1]Dept. of Hlth. Technol. and Informatics, Hong Kong Polytechnic Univ., Hong Kong, Hong Kong; [2]Dept. of Ocean Sci., Hong Kong Univ. of Sci. and Technol., Hong Kong, Hong Kong; [3]Dept. of Neurobiology, Sch. of Med., Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Ataxia Telangiectasia (A-T) Mutated protein (ATM) is a major kinase that responds to DNA double strand breaks. A spectrum of pathogenic mutations of *ATM* is known to cause the rare genetic disease, A-T with cerebellar symptoms of different severity. Myelin pathology emerges as one of the neurological deficits in young A-T patients, but the mechanism is unknown. We proposed that oligodendrocytes (OL) are lost in the A-T cerebellum and here, we aim to delineate the divergent effects on OL lineage mediated by different *ATM* mutations. To test, we characterized the OL population in post-mortem cerebella of ten A-T cases where the ATM mutations were profiled by Illumina-based targeted genomic sequencing. First, 13 distinct pathogenic mutations were identified among A-T cases with four cases were compound heterozygous. In A-T cases with frameshift mutations, the overall cerebellar $Olig2^+$ OL population and mature OLs (mOL, $CC1^+$) were reduced by 53.6 and 87.8% (vs controls), respectively. Importantly, the demise of OL was not correlated with the neuronal atrophy. These histological results were validated by *Atm*-deficient mice, $Atm^{-/-}$ (B6;129S4-Atm^{tm1Bal}/J), where the *Atm* exons was disrupted with no residual ATM activity. In $Atm^{-/-}$ cerebellum, the overall OL, mOL, and their progenitor (OPC) density and myelin proteins were reduced, but OL reduction in the neocortex was accompanied by $NG2^+$ OPC accumulation. We hypothesize that ATM is required for OL differentiation and investigated the putative interactions between ATM and myelin-related proteins in silico. ATM showed strong putative interactions with Myelin regulatory factor (MyRF, $E_{balanced}$ = -1225 KJ mol^{-1}, 113 contacts) and Myelin basic protein (MBP, $E_{balanced}$ = -1123.6 KJ mol^{-1}, 206 contacts). Intriguingly, the ATM-MBP interaction was validated by in co-immunoprecipitation (IP) and IP-based liquid chromatography-mass spectrometry analysis, and the site (MBP^{Ser99}) is a known phosphorylation site for myelin compaction. Importantly, the strength of the putative ATM-MBP was significantly reduced among the A-T cases carrying mutations terminating ATM at the spiral/pincer region (≤1238 a.a, $p < 0.003$), and the cases carrying early ATM disruption showed a significantly reduced density

of Olig2^{+} and CC1^{+} OLs without affecting neurons. The impact of partial ATM mutation is being validated in a model harbouring truncated but active ATM residue (*Atm*^{Awb};B6.129S6-Atm^{tm1Awb}/J). Together, ATM is intrinsically required for OL differentiation. The diverse effects of mutation variants to hinder interactions between ATM and myelin-related proteins may be the molecular mechanisms of the diverse myelin pathology in A-T.

Disclosures: S.H.S. Yeung: None. **A. Cheng:** None. **G.W.Y. Cheng:** None. **K.K.S. Mok:** None. **K. Herrup:** None. **K. Tse:** None.

Poster

P167. Oligodendrocytes: Disease Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P167.04

Topic: B.11. Glial Mechanisms

Support: NIH R01-MH098742
NIH R01-MH126773
NIH T32-NS007413

Title: The HIV-1 antiretroviral cocktail Triumeq disrupts oligodendrocyte maturation through dolutegravir-induced integrated stress response dysfunction.

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Abstract: Despite the development of antiretroviral therapies (ART), approximately half of all HIV-1 infected individuals develop a spectrum of cognitive and psychiatric sequelae, collectively known as HIV-1 associated neurocognitive disorders (HAND). While ART decreased the clinical severity of HAND, notable histopathological features, such as white matter abnormalities and myelin loss, have persisted, lending credence to the possibility that ART drugs themselves may be modulating CNS dysregulation. Our laboratory has implicated five front-line ART drugs in hampering oligodendrocyte maturation and myelin protein production, raising concern about the use of ART drugs during CNS development. Therefore, we proposed to examine the front-line ART drug combination given to HIV-1 positive pregnant women: Dolutegravir, Abacavir, and Lamivudine (also known as Triumeq). We hypothesized that this combination would inhibit oligodendrocyte maturation. Treating primary rat oligodendrocyte precursor cells (OPCs) with the Triumeq drug combination for 24 hours, followed by co-treatment with this combination of drugs after induction of differentiation decreased the number of mature oligodendrocytes and myelin protein production after 3 days in a concentration-

dependent manner. As the number of OPCs and apoptotic cells remained similar to controls, these data suggest that this drug combination attenuates OPC differentiation. Subsequent analyses demonstrated that Dolutegravir was primarily responsible for this differentiation deficit. Finally, treatment of OPCs with trans-ISRIB, a drug that inhibits the integrated stress response (ISR) after Dolutegravir treatment prevented low, but not mid-dose differentiation deficits. Together, these data demonstrate a potential role for the ISR in Dolutegravir-induced oligodendrocyte differentiation deficits.

Disclosures: R. Putatunda: None. **L.M. Roth:** None. **B. Di Sanza:** None. **M.A. Romer:** None. **K.L. Jordan-Sciutto:** None. **J.B. Grinspan:** None.

Poster

P167. Oligodendrocytes: Disease Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P167.05

Topic: B.11. Glial Mechanisms

Support: NHMRC 1100626

Title: Loss of very long chain sphingolipid synthesis in oligodendrocytes causes rapid microglial activation, followed by myelin degeneration and early lethality.

Authors: *J. D. TEO[1,2], O. C. MARIAN[1,2], H. P. MCEWEN[1], H. SONG[1,2], J. C. BRÜNING[3], A. S. DON[2,1];
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Abstract: Myelin is comprised 70-80% of lipids. The sphingolipids galactosylceramide (GalCer), sulfatide (ST), and sphingomyelin (SM) are essential for myelin stability and function. Whether the specific acyl chain composition of these lipids affects myelin stability is less clear. GalCer and ST are synthesized from C22-C24 ceramides (very long chain ceramides) by ceramide synthase 2 (CerS2), which is very highly expressed in oligodendrocytes. Prior studies have shown than myelin is destabilised and deteriorates in mice with universal CerS2 deletion, which was attributed to a reduction in total GalCer content. To clarify the requirement for very long chain sphingolipids in myelin biosynthesis and stability, we generated C57BL6 mice lacking CerS2 specifically in myelinating cells ($CerS2^{\Delta O}$), by crossing mice homozygous for a floxed CerS2 allele to those expressing cre recombinase from the CNP gene locus (CNP-cre). Littermates not expressing CNP-cre were used as controls. Motor function was assessed by rotarod, balance beam and grip strength tests. Mice (n=5/6 per group) were culled at 6, 10 and 16 weeks of age (6W, 10W, 16W). Myelin content was assessed by lipidomic mass spectrometry, luxol fast blue (LFB) staining, and western blotting for myelin proteins. Immunofluoresent

staining for mature oligodendrocytes, astrocytes, and microglia was quantified blinded to the experimental groups. This project was approved by the University of Sydney animal ethics committee. At 6W, total levels of GalCer, ST, and SM were not reduced in CerS2$^{\Delta O}$ mice, as loss of C22-C24 sphingolipids was compensated by increased levels of shorter-chain C18 lipids. Myelin content and glial response were unaffected at 6W. At 10W, reactive astrocytes and microglia accumulated in white matter of CerS2$^{\Delta O}$ mice, even though myelin content, mature oligodendrocyte density, and motor function remained comparable to controls. Pronounced myelin atrophy, motor deficits and severe gliosis were observed in 16W CerS2$^{\Delta O}$ mice. 50% of CerS2$^{\Delta O}$ mice died by 5 months of age. In conclusion, synthesis of very long chain sphingolipids by CerS2 is not essential for myelin formation during development. However, the loss of CerS2 in oligodendrocytes results in myelin degeneration, motor deficits and lethality prior to 6 months of age. Astrogliosis and morphological transformation of microglia precedes loss of oligodendrocytes and myelin, suggesting that the shorter acyl chain composition in CerS2$^{\Delta O}$ myelin lipids destabilises myelin and elicits phagocytic microglia. Understanding the mechanisms by which these events occur could lead to the development of new therapeutics for demyelinating diseases.

Disclosures: J.D. Teo: None. **O.C. Marian:** None. **H.P. McEwen:** None. **H. Song:** None. **J.C. Brüning:** None. **A.S. Don:** None.

Poster

P167. Oligodendrocytes: Disease Mechanisms

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Program #/Poster #: P167.06

Topic: B.11. Glial Mechanisms

Support: 1R01NS109227

Title: DYT6 dystonia mutation causes hypomyelination and the loss of YY1 binding

Authors: *A. E. ROGERS[1], A. J. KIM[3], S. KIM[2], S. S. PAPPAS[3,4], W. T. DAUER[3,4,5], D. YELLAJOSHYULA[6];
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Abstract: Dystonia is a disabling neurological disorder that manifests as prolonged involuntary twisting movements. DYT6 is an inherited form of isolated dystonia caused by mutations in *THAP1* encoding the transcription factor THAP1. In previous studies, we demonstrated that THAP1 is essential for CNS myelination due to its cell-autonomous role in the oligodendrocyte

lineage using mouse models with conditional deletion of *Thap1*. However, it is not known if the many DYT6 disease mutations reported have any effect on CNS myelination. The phe81leu (F81L) missense mutation is representative of a category of poorly understood mutations that do not occur on residues critical for DNA binding. Using a knock-in mouse line that we generated containing the *Thap1*^{F81L} allele, we demonstrate that the F81L mutation (THAP1^{F81L}) impairs THAP1 transcriptional activity and results in CNS hypomyelination. Strikingly, THAP1^{F81L} showed normal DNA binding at target loci, but disrupted the binding of YY1 - a cofactor of THAP1 with a role in the development of oligodendrocyte lineage. Our studies suggest a model of molecular pathogenesis in which THAP1^{F81L} normally binds DNA but cannot form a functional transcription complex.

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Poster

P167. Oligodendrocytes: Disease Mechanisms

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Program #/Poster #: P167.07

Topic: B.11. Glial Mechanisms

Support: Research Grant Council: GRF16124916
Food and Health Bureau: HMRF05163736
PolyU Start-up Fund: P0030307

Title: Does demyelination accelerate the neuropathology progression of Alzheimer's disease? - a radio-pathological investigation

Authors: *K.-H. TSE[1], G. W. Y. CHENG[1], J. HUANG[2], K. K. S. MOK[1], S. H. S. YEUNG[1], Z. CHEN[2], H. K. F. MAK[3], K. W. Y. CHAN[2,4], K. HERRUP[5];
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Abstract: Myelin abnormalities such as white matter (WM) volume reduction, microstructural changes and WM hyperintensity are common in the aging brain. Such neuroimaging detections are often found in cognitive decline and early in late-onset Alzheimer's disease (AD). We recently reported the loss of oligodendrocytes (OLs), the only myelinating glia, in human brain before amyloid plaques formation in dementia. Here, we hypothesize that such early demise of

myelin and OL population may exacerbate AD pathogenesis. To test, we asked if induced myelin loss can accelerate classic AD pathology in common transgenic mouse models. The APP/PS1 (*B6.Cg-Tg(APPswe,PSEN1dE9)85Dbo/Mmjax*) carrying human *APP*$^{K595N/M596L}$ plus Presenilin 1(PS1^{dE9}) and the R1.40 (*B6.129-Tg(APPSw)40Btla/Mmjax*) harboring just human *APP*$^{K670N/M671L}$ normally exhibit amyloid deposition by 6 and 14 months, respectively. To induce demyelination, both strains and wildtype controls between 9-12 weeks (male, n = 4-6 / group) were subjected control or cuprizone diet (0.2% w/w) for four weeks before behavioral tests and stereotactic radiological-histological analysis using 3T preclinical MRI and immunohistochemistry. While no behavioral changes were observed, diffusion kurtosis imaging revealed the region-specific WM microstructural alternations (fractional anisotropy, radial diffusivity, and mean diffusivity) after demyelination in all animals. In the anterior cingulate and the genu of the corpus callosum, the overall Olig2^{+} OL population and ASPA^{+} and CC1^{+} mature OLs were reduced by more than 50%, as expected. To investigate the classic AD pathology, we examined if the demyelination accelerated the formation of amyloid plaque (6E10) and the DNA double strand breaks (DSBs) the brain. In APP/PS1, but not R1.40, demyelination increased 6E10 signals within cortical neurons and across neuropil at four months old. In both AD strains and the wildtype control, demyelination increased the number of neurons exhibiting 53BP1^{+} DSB nuclei, suggestive of the loss of neuronal genomic integrity. This observation is consistent with our previous report of human multiple sclerosis and AD where myelin and OL degeneration in frontal cortex were associated with neuronal DSBs and ectopic cell cycle re-entry. These DNA damage inductions were also found in the OL in the genu and is likely correlated with the demise of their population amid myelin loss. Together, our data suggested that myelin loss may drive AD pathology in a premature stage. We are currently examining the radiological-histological analysis between neuronal cell cycle re-entry, OL pathology, and the magnetization transfer signals upon demyelination.

Disclosures: K. Tse: None. **G.W.Y. Cheng:** None. **J. Huang:** None. **K.K.S. Mok:** None. **S.H.S. Yeung:** None. **Z. Chen:** None. **H.K.F. Mak:** None. **K.W.Y. Chan:** None. **K. Herrup:** None.

Poster

P167. Oligodendrocytes: Disease Mechanisms

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Program #/Poster #: P167.08

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NS103433
NS108529

Title: Post-spinal cord injury oligodendrocyte translatome analysis reveals paracrine contributions to white matter repair

Authors: *M. D. FORSTON[1,2], G. Z. WEI[1,3], J. H. CHARIKER[4,5], R. M. HOWARD[1,6], K. R. ANDRES[1,6], S. A. MYERS[1,6], D. A. BURKE[1,6], T. L. STEPHENSON[1], B. A. STOLL[1], E. C. ROUCHKA[4,5], S. R. WHITTEMORE[1,2,6], M. HETMAN[1,2,3,6];
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Abstract: Oligodendrocyte (OL) death and white matter loss contribute to functional deficits after spinal cord injury (SCI). OL protection and remyelination by oligodendrocyte precursor cells (OPCs) is critical for functional recovery after contusive SCI. However, SCI-induced, OL-specific changes in gene expression have not yet been reported at the transcriptomic/translatomic level. RiboTag technology allows analysis of mRNAs that are bound to translating ribosomes from specific cell populations marked genetically with a hemagglutinin (HA) tag on a core ribosomal subunit, RPL22. Using *Plp1-Cre^{ERT2}:Rpl22^{HA}* female mice to induce the RiboTag in mature OLs, we isolated OL-specific mRNAs from the injury epicenter following moderate T10 contusion. We initially confirmed reliable isolation of OL-specific mRNAs as evidenced by enrichment of myelin-associated genes only found in OLs when comparing the OL translatome to total RNA samples. Using uninjured cord from naïve mice as a control, extensive changes in OL translatome mRNAs were documented at acute (2 days post injury, dpi), subacute (10 dpi) and chronic (42 dpi) timepoints. These genes included upregulation of the OL injury response markers Klk8 and C4b. Among 77 mRNAs that were highly upregulated selectively in OLs at all timepoints post-SCI, enriched gene ontology (GO) terms included organismal development, cell differentiation, nervous system development and cell-cell signaling. OLs upregulated secreted inhibitors of BMP (*Sostdc1*), WNT (*Sostdc1*, *Sfrp5*) and Notch (*Egfl8*, *Dlk2*) signaling, pathways which are implicated in inhibiting OPC to OL differentiation. Therefore, after SCI, surviving OLs may secrete paracrine factors to enhance OPC/OL differentiation and promote white matter repair. Such a notion is consistent with a documented occurrence of OPC-mediated remyelination in areas where OLs are still present.

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Poster

P167. Oligodendrocytes: Disease Mechanisms

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Program #/Poster #: P167.09

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NS073584
GM103507

Title: Differential effects of the integrated stress response kinases PERK, PKR, GCN2 and HRI on oligodendrocyte survival and locomotor recovery after spinal cord injury

Authors: *S. SARASWAT OHRI[1], Y. LIU[1], K. ANDRES[1], R. HOWARD[1], S. MYERS[1], D. CAVENER[2], M. HETMAN[1], S. WHITTEMORE[1];
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Abstract: The integrated stress response (ISR) pathway is integral to the proteostasis network that is crucial to maintain cellular homeostasis under physiological and pathological conditions. The mammalian ISR is mediated by 4 independent kinases: protein kinase RNA (PKR)-like kinase (PERK), general control non-de-repressible 2 (GCN2), double-stranded RNA-activated protein kinase (PKR) and heme-regulated inhibitor (HRI). Activation of all ISR kinases phosphorylates eIF2α (eukaryotic initiation factor 2α) which results in general protein synthesis inhibition and in selective translation of stress response mediator proteins such as activating transcription factor 4 (ATF4) and CCAAT/enhancer binding homologous protein (CHOP). However, the spectrum of their upstream activating stimuli differs, albeit with some overlap. Using genetic and pharmacological inhibition of the unfolded protein response (UPR), we previously demonstrated improvements in hindlimb locomotion after thoracic SCI and implicated oligodendrocyte (OL) survival as a potential mechanism. Here, we explore the functional contribution of the individual kinases of the ISR pathway to the pathogenesis of SCI. Thoracic contusive SCI resulted in induction of all ISR kinases 24 hours post-injury. Global deletion of *Pkr* or *Gcn2* failed to improve locomotor recovery after SCI. Targeted loss of *Perk* in OL lineage cells resulted in enhanced pro-apoptotic *Atf4* and *Chop* mRNA levels in mice acutely (72 hours) after thoracic contusive SCI. Consistent with this, post-injury recovery of hindlimb locomotion and white matter sparing were impaired in OL-*Perk*$^{-/-}$ mice and correlated with greater loss of oligodendrocyte precursor cells (OPCs)/OLs at the injury epicenter six weeks post-SCI. In contrast, after SCI, constitutive *Hri*$^{-/-}$ mice showed reduced *Atf4* and *Chop* mRNA levels acutely and demonstrated improvement in hindlimb locomotor recovery at six weeks post-SCI that correlated with enhanced white matter sparing and reduced loss of OPCs/OLs at the injury epicenter. Collectively, data show that the ISR kinases differentially contribute to OPC/OL loss and implicate PERK and HRI as critical pro- and anti-survival mediators after SCI, respectively. The differential effects of *Perk, Pkr, Gcn2* and *Hri* deletion on functional recovery after SCI potentially reflects fundamental differences in outcome of ISR activation in specific signaling and/or cellular contexts.. Supported by NS073584 and GM103507.

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Poster

P175. Human and Animal Studies and Therapeutics: Brain Injury

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P175.01

Topic: B.12. Demyelinating Disorders

Support: NRF-2020R1I1A1A01072653
NRF-2020R1A2C2008480

Title: A Role for Zinc in Axon Formation via mTORC1 Pathway.

Authors: *S. CHOI[1], D. KANG[1], J.-E. KANG[2], D. HONG[1], B. KANG[1], A. KHO[1], D. YOO[2], S.-O. HUH[2], S. SUH[1];
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Abstract: Zinc is an essential micronutrient required for proper function during neuronal development, due to its ability to modulate neuronal function and structure. A fully functional description of zinc on axonal processing in the central nervous system remains elusive. Here we defined the role of intracellular zinc in axon formation and elongation, involving mammalian target of rapamycin complex 1 (mTORC1). To investigate the involvement of zinc on axon growth, we performed an ex vivo culture of mouse hippocampal neurons and administrated $ZnCl_2$ as a media supplement. At 2 days *in vitro*, administration of zinc induced the formation of multiple and elongated axons in the ex vivo culture system. A similar outcome was witnessed in callosal projection neurons in developing mouse brain. Treatment with extracellular zinc activated the mTORC1 signaling pathway in mouse hippocampal neuronal cultures. The zinc dependent enhancement of neuronal processing was moreover inhibited either by deactivation of mTORC1 with RAPTOR shRNA or by mTOR-insensitive 4EBP1 mutants. Additionally, zinc-dependent mTORC1 activation enhanced axonal translation of TC10 and Par3 may responsible for axonal growth. Collectively, we have identified a promising role of zinc in controlling axonogenesis in the developing brain, which in turn, may possess a novel role for the treatment of neurological disorders such as callosal agenesis.

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Poster

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Program #/Poster #: P175.02

Topic: B.12. Demyelinating Disorders

Support: NRF-2017M3C7A1028937

Title: The effects of pyruvate kinase M2 (PKM2) gene deletion in astrocytes on global cerebral ischemia-induced neuronal death

Authors: *B. KANG, S. SUH, B. CHOI, A. KHO, S. LEE, D. HONG, S. LEE;
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Abstract: Ischemic stroke is one of the most severe brain diseases and is caused by insufficient supply of nutrients and oxygen when there is decreasing blood flow to the brain. The astrocyte has an important role in bidirectionally converting pyruvate generated via glycolysis into lactate, and supplying lactate to neurons through the astrocyte-neuron lactate shuttle (ANLS). Pyruvate kinase M2 (PKM2) is an enzyme that catalyzes dephosphorylation of phosphoenolpyruvate to pyruvate in astrocyte-specific glycolysis. To test whether astrocyte-specific PKM2 gene deletion influences energy production, and also exacerbates brain damage, we used PKM2 knock out (KO) mice. PKM2 gene was deleted by administrating tamoxifen to Aldh1l1-Cre^{ERT2}; PKM2$^{f/f}$ mouse. The global cerebral ischemia (GCI) performed blood flow restoration and resuscitation after occlusion of the bilateral common carotid artery (BCCA) for 20 min in mice. So, we hypothesized that the decrease in lactate supply over the ANLS in the astrocyte-specific PKM2 KO will lead to more widespread neuronal death compared to PKM2 wild-type mice after GCI. To confirm whether lactate administration in the astrocyte-specific PKM2 KO reduces neuronal death after GCI, sodium L-lactate (250mg/kg, i.p.) was injected immediately after GCI and then once daily for 3 days before sacrifice. We demonstrated that PKM2 KO exacerbates GCI-induced neuronal death and reactive oxygen species (ROS) production, and that these aberrant processes are attenuated by lactate administration. In conclusion, we suggest that lactate supply via the ANLS in astrocytes plays a crucial role in maintaining sufficient energy production in neurons, therefore lactate administration has a potential therapeutic effect on neuronal death after GCI.
Key Words: global cerebral ischemia, astrocyte-neuron lactate shuttle, pyruvate kinase M2, sodium L-lactate, neuronal death

Disclosures: B. Kang: None. **S. Suh:** None. **B. Choi:** None. **A. Kho:** None. **S. Lee:** None. **D. Hong:** None. **S. Lee:** None.

Poster

P175. Human and Animal Studies and Therapeutics: Brain Injury

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Program #/Poster #: P175.03

Topic: B.12. Demyelinating Disorders

Support: NRF-2017M3C7A1028937

Title: An acid sphingomyelinase inhibitor, imipramine, reduces hippocampal neuronal death after traumatic brain injury

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Abstract: Traumatic brain injury (TBI), broadly, is a decline in the normal function of the brain after experiencing a bump, blow or jolt to the head. It leads to aggravation of pre- existing brain dysfunction and promotes neurotoxic cascades that involve such processes as oxidative stress, dendritic loss, and zinc accumulation. Acid sphingomyelinase (ASMase) is an enzyme which hydrolyzes sphingomyelin to ceramide in the cells. Ceramide is involved in various physiological functions such as cell migration, differentiation and apoptosis. However, under pathological settings such as cerebral ischemia and Alzheimer's disease, excessively ceramide production causes activation of cell death mechanisms to occur. So, we hypothesized that inhibition of ASMase activity by the ASMase inhibitor, imipramine, could reduce ceramide formation and thus prevent TBI-induced neuronal death. To test our hypothesis, imipramine was injected (10mg/kg, i.p., once-daily for 7 days) after TBI. The experimental animals were sacrificed at 24 hour and 7 days following TBI. As the result, we confirmed that neuronal death, dendritic loss and oxidative stress were significantly reduced in the TBI-imipramine group compared to TBI-vehicle group at 24 hours. Additionally, we validated that the administration of imipramine for 7 days has neuroprotective effects on TBI, and that neurological behavior testing (mNSS test) confirmed that imipramine prevents cognitive dysfunction after TBI. Taken together, we suggest that imipramine, an ASMase inhibitor, decreases ceramide levels and thus prevents TBI-induced neuronal death. Consequently, ASMase inhibition may be therapeutic target for TBI-induced hippocampal neuronal death.
Key Words: Traumatic brain injury, Acid sphingomyelinase, Ceramide, Imipramine, Neuronal death

Disclosures: S. Lee: None. **B. Choi:** None. **A. Kho:** None. **S. Lee:** None. **D. Hong:** None. **B. Kang:** None. **M. Park:** None. **S. Suh:** None.

Poster

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Topic: B.12. Demyelinating Disorders

Support: NRF-2021R1C1C2012889

Title: Novel zinc chelators ameliorate experimental autoimmune encephalomyelitis by modulating zinc toxicity and AMPK activation

Authors: *B. CHOI[1], J. JEONG[2], J.-W. EOM[3], J.-Y. KOH[4], Y.-H. KIM[3], S. SUH[1];
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Abstract: Previous studies in our lab revealed that chemical zinc chelation or zinc transporter 3 (ZnT3) gene deletion suppresses the clinical features and neuropathological changes associated with experimental autoimmune encephalomyelitis (EAE). In addition, although protective functions are well documented for AMP-activated protein kinase (AMPK), paradoxically, disease-promoting effects have also been demonstrated for this enzyme. Recent studies have demonstrated that AMPK contributes to zinc-induced neurotoxicity and that compound A or B, an inhibitor of AMPK, reduces zinc-induced neuronal death and protects against oxidative stress, excitotoxicity, and apoptosis. Here, we sought to evaluate the therapeutic efficacy of compound A or B against myelin oligodendrocyte glycoprotein 35-55-induced EAE. Compound A (5 ug/kg) or B (20 ug/kg) was intraperitoneally injected once per day for the entire experimental course. Histological evaluation was performed three weeks after the initial immunization. We found that Compound A or B profoundly reduced the severity of the induced EAE and that there was a remarkable suppression of demyelination, microglial activation, and immune cell infiltration. Compound A or B also remarkably inhibited EAE-associated BBB disruption, MMP-9 activation and aberrant synaptic zinc patch formation. Furthermore, the present study showed that long term-treatment with compound A or B also reduced the clinical course of EAE. Therefore, the present study suggests that zinc chelation and AMPK inhibition with compound A or B may have great therapeutic potential for the treatment of multiple sclerosis.

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Poster

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Topic: B.12. Demyelinating Disorders

Support: NRF-2020R1A2C2008480
NRF-2020R1A6A3A01096433

Title: Administration of an acidic sphingomyelinase (ASMase) inhibitor, imipramine, reduces hypoglycemia-induced hippocampal neuronal death

Authors: *A. KHO, B. CHOI, S. LEE, D. HONG, B. KANG, S. LEE, S. SUH;
Dept. of Physiol., Hallym University, Col. of Med., Chuncheon, Korea, Republic of

Abstract: Severe hypoglycemia (below 35 mg/dL) appears most often in diabetes patients who continuously inject insulin. To rapidly cease the hypoglycemic state, glucose reperfusion is conducted to restore blood glucose levels, which can induce a secondary neuronal death cascade following hypoglycemia. Acid sphingomyelinase (ASMase) hydrolyzes sphingomyelin into ceramide and phosphoryl choline. ASMase activity can be influenced by cations, pH, redox, lipids, and other proteins in the cells. In particular, severe hypoglycemia can lead to acidosis; oxidative stress; and excessive cation release—for example, the release of Ca^{2+} or Zn^{2+}. Therefore, we expect that ASMase is activated excessively after hypoglycemia. Ceramide is known to cause free radical production, excessive inflammation, calcium dysregulation, and lysosomal injury, resulting in apoptosis and the necrosis of neurons. Imipramine is mainly used in the treatment of depression and certain anxiety disorders. Additionally, it is known as ASMase inhibitor. In the present study, we hypothesized that imipramine can decrease hippocampal neuronal death by reducing ceramide via the inhibition of ASMase after hypoglycemia. In the present study, we found that the administration of imipramine significantly reduced hypoglycemia-induced neuronal death. Therefore, the present study suggests that imipramine may be a promising therapeutic tool for preventing hypoglycemia-induced neuronal death.
Keywords: Hypoglycemia; Sphingolipid; Acidic sphingomyelinase; Ceramide; imipramine; Neuron death; Cognitive dysfunction

Disclosures: A. Kho: None. **B. Choi:** None. **S. Lee:** None. **D. Hong:** None. **B. Kang:** None. **S. Lee:** None. **S. Suh:** None.

Poster

P175. Human and Animal Studies and Therapeutics: Brain Injury

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P175.06

Topic: B.12. Demyelinating Disorders

Support: NRF-2017M3C7A1028937
NRF-HU20C0206

Title: Acid sphingomyelinase inhibitor, imipramine, increases the survival of newborn neurons after pilocarpine-induced seizure

Authors: *S. LEE[1], B. CHOI[1], A. KHO[1], D. HONG[1], B. KANG[1], S. LEE[1], H. SONG[2], H. CHOI[2], S. SUH[1];
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Abstract: Acid sphingomyelinase (ASM) is an enzyme that hydrolyzes sphingomyelinase into ceramides, and the ceramides produced at this time can cause viral and bacterial diseases as well as a wide range of diseases such as cancer, cystic fibrosis, diabetes, Alzheimer's and depression. Ceramide metabolized from sphingomyelin by ASM are the simplest sphingolipids in the brain. Ceramide is known to cause reactive oxygen species (ROS) production, excessive inflammation and lysosomal damage. After epilepsy, excessive zinc release occurs in neurons, and ASM is activated to promote ceramide formation, which plays a pivotal role in inflammation, cell signaling and cell death, leading to apoptosis. ASM inhibitor, imipramine, can reduces the activity of ASM caused by epilepsy, thereby reducing ceramide production. In this study, we tested the hypothesis that imipramine can suppress excessive oxidative stress and improve the survival of newly generated neurons under pathological states such as epilepsy. This study used Sprague-Dawley rats (250-300g) and seizure was induced by pilocarpine (25 mg/kg, i.p.). Imipramine (10 mg/kg, i.p.) was continuously injected once per day for 4 weeks after seizures. Histological analysis and cognitive function (barnes maze) test was performed at 4 weeks after seizure. We found that long-term treatment of imipramine increased the number of $BrdU^+$ and $BrdU^+NeuN^+$ cells in the dentate gyrus of hippocampus after pilocarpine-induced seizure. In the barnes maze test, imipramine treatment improved seizure-induced cognitive impairment. The present study demonstrates that imipramine increases the survival of newborn neurons in the dentate gyrus of hippocampus and improved cognitive function after seizure. Therefore, this study suggests that imipramine may be a therapeutic tool that reduces neuronal death after seizure and increases the survival of newborn neurons.
Keywords: epilepsy, imipramine, ceramide, Acid sphingomyelinase, neuron death

Disclosures: S. Lee: None. **B. Choi:** None. **A. Kho:** None. **D. Hong:** None. **B. Kang:** None. **S. Lee:** None. **H. Song:** None. **H. Choi:** None. **S. Suh:** None.

Poster

P175. Human and Animal Studies and Therapeutics: Brain Injury

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P175.07

Topic: B.12. Demyelinating Disorders

Support: NRF-2019R1A6A3A13093671
NRF-2017M3C7A1028937

Title: Targeting the AMP-activated protein kinase and zinc: a therapeutic target for global cerebral ischemia

Authors: *D. HONG[1], B. CHOI[1], J.-W. EOM[2], S. CHOI[1], A. KHO[1], S. LEE[1], B. KANG[1], S. LEE[1], Y.-H. KIM[2], S. SUH[1];
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Abstract: Ischemic stroke causes severe brain damage and it is a leading threat to the global population at large. Insufficient supply of glucose, oxygen and blood by blood clot-induced blockade of vessels or considerably reduced blood flow due to heart attack are two common examples of situations that can result in cerebral ischemia. These deleterious situations lead to activation of cell death cascades that involve production of reactive oxygen species (ROS), excitotoxicity and ionic imbalance. AMP-activated protein kinase (AMPK) has three subunits: catalytic unit alpha, regulatory unit beta and regulatory unit gamma. Under the physiological situations, AMPK is necessary for maintaining energy balance and processes such as glycolysis, gene transcription, glucose uptake and several other biological functions. However, central nervous system (CNS) injury-induced energy and metabolic stress, such as cerebral ischemia, increases AMPK phosphorylation. Highly phosphorylated AMPK contributes to excitotoxicity, oxidative and metabolic problems. As well, CNS injuries-induced excessively released chelatable zinc contributes to brain damage, including ROS production, apoptosis, and elevation of intracellular calcium levels. For this reason, we assert that regulation of abnormal AMPK activity and zinc release is necessary for prevention against brain damage under ischemic conditions. In compound C (a well-known AMPK inhibitor) structure-based screening, novel agent 2G11 significantly decreased zinc toxicity and AMPK activity. In this study, we verified whether novel AMPK inhibitor, 2G11 administration has neuroprotective effects via regulation of AMPK activity on the global cerebral ischemia (GCI). In order to demonstrate 2G11 effects, we used a GCI animal disease model (2G11: 20μg/kg) and oxygen glucose deprivation/reoxygenation (OGD/R, 2G11: 20μM) in primary cultured neurons. As a result, 2G11 reduced AMPK phosphorylation, the number of degenerating neurons, zinc accumulation, lipid peroxidation and astrocyte & microglia activation. These collective findings suggested that the novel AMPK inhibitor, 2G11 is a therapeutic candidate for cerebral ischemia-induced cell death cascades.

Disclosures: D. Hong: None. **B. Choi:** None. **J. Eom:** None. **S. Choi:** None. **A. Kho:** None. **S. Lee:** None. **B. Kang:** None. **S. Lee:** None. **Y. Kim:** None. **S. Suh:** None.

Poster

P185. Metabolism, Oxidative Stress, and Cell Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P185.01

Topic: C.01. Brain Wellness and Aging

Title: Altered expression of peroxisomal markers in a mouse model of amyotrophic lateral sclerosis

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Abstract: Amyotrophic lateral sclerosis (ALS) is a degenerative disorder affecting both upper motor neurons in the neocortex, and lower motor neurons in the brainstem and spinal cord, and leading to skeletal muscle atrophy and weakness. As a multisystemic disease, alterations in one or more cellular pathways in extra-neural tissues have been considered fostering ALS onset and progression. Some of these include oxidative stress, mitochondrial dysfunction, axonal and vesicular transport defects, excitotoxicity, autophagic dysfunction, neuroinflammation and altered RNA metabolism. Increased energy expenditure and lipid metabolism alterations related to mitochondrial dysfunction manifest prior to motor neuron loss. Being a key determinant of whole-body metabolic rate, skeletal muscle plays a pathological role in ALS, contributing to defective energy metabolism and determining a derangement of basal metabolic rate, switching its metabolism towards an oxidative phenotype at the expense of a glycolytic one, and to the preferential use of fatty acids as fuel. Over the years, light has been shed on the tightly connected functions of mitochondria and peroxisomes, mainly in controlling lipid and reactive oxygen species (ROS) metabolism, so that (dys)-functions in either of the two determine altered metabolism of the other. Importantly, peroxisomes respond to physiological and pathological changes and extracellular stimuli, by altering their enzyme content, morphology, size, and abundance. These dynamic features are modulated by the activation of a subfamily of transcription factors, namely peroxisome proliferator activated receptors (PPARs), upon binding to their ligands, which in turn exert neuroprotective effects in ALS. The aim of this study is to address the role played by peroxisomes in ALS onset and progression, by analyzing the expression of peroxisomal markers in the spinal cord, muscle and liver of SOD1-G93A mouse model. Immunoblotting analysis of peroxisomal proteins and antioxidant enzymes shows an increase concomitant with pathology progression, suggesting an enhancement in the number of peroxisomes and in β-oxidation of very long chain fatty acids. This correlates with the increase of PGC1-α that, stimulating both peroxisomal and mitochondrial biogenesis, elicits an antioxidant response to cope with redox imbalance through the progression of the disease. This data is consistent with what observed by qRT-PCR analysis of the same markers. Overall, our data strongly suggests an involvement of both neural and non-neural peroxisomes in ALS onset and progression, possibly counteracting mitochondrial dysfunction.

Disclosures: R. Price: None. **S. Scaricamazza:** None. **M. Terricola:** None. **I. Salvatori:** None. **G. Taglialatela:** None. **R. Kayed:** None. **M. Cervelli:** None. **F. Berardinelli:** None. **A. Ferri:** None. **C. Valle:** None. **S. Moreno:** None.

Poster

P185. Metabolism, Oxidative Stress, and Cell Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P185.02

Topic: C.01. Brain Wellness and Aging

Support: DFG Excellence cluster 2145 Munich Cluster for Systems Neurology (SyNergy)
DFG Individual Research Grant 323061152
Institute of Advanced Study (TUM-IAS), Technical University Munich
PhD Program Life Science and Technology, Technical University Munich

Title: A retrograde transit filter in distal axons mediates mitochondrial quality control in synapses

Authors: *N. A. MARAHORI[1,2], B. PLOMER[1], A. IATROUDI[1], T. KLEELE[4,1], M. SCHIFFERER[5,6], P. AVRAMOPOULOS[3,7], S. ENGELHARDT[3,7], M. LAKADAMYALI[8,2], M. BRILL[1,6], T. MISGELD[1,2,5,6];
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Abstract: Organelle homeostasis is an intricate process, requiring axonal transport to relocate organelles between sites of high demand and their sites of biogenesis and eventual degradation. For example, mitochondria are transported anterogradely toward axonal branch tips and their synapses, where they provide local energy to drive neurotransmission - damaging mitochondria over time via reactive oxygen species (ROS) production. While some presynaptic mitochondria are eventually transported back toward the soma, a substantial residue is left behind, raising the question of how mitochondrial homeostasis - ‘mitostasis’ - is ultimately achieved. Although principal pathways for mitochondria degradation, such as bulk autophagy or selective mitophagy, are known, current models of mitostasis do not explain how these processes are compartmentalized and regulated in synapses *in vivo*. By measuring mitochondrial volume flux in mouse motor axons, we identified major sites of degradation near presynaptic compartments. Using pulse-chase tracking of photo-tagged synaptic mitochondria in *Thy1*-mitoDendra mice, we discovered that ~60% of mitochondria that attempted to leave neuromuscular synapses (NMJs) stopped and vanished close to the most distal node of Ranvier. Nodes located further proximally captured retrogradely, but not anterogradely moving mitochondria with decreasing frequency, indicating a selective mechanism acting on specific mitochondria only. In line with local

degradation, we found a high density of lysosomal markers at capture sites, and colocalization with captured mitochondria over time. Locally depolarizing presynaptic mitochondria using a photo-caged uncoupler increased capture rates, indicating that damaged mitochondria were preferentially targeted for capture. Accordingly, in mouse models of Amyotrophic lateral sclerosis (ALS), where synaptic mitochondria are prone to damage, we observed increased capture (SOD and dipeptide-repeat-protein based models). Our ongoing genetic analysis shows that ablating the disease-associated protein optineurin (*Optn*), but not the *Pink1-Parkin* pathway, led to reduced capture and higher retrograde transport of mitochondria in terminal branches, implying that the mitophagy receptor optineurin is involved. In summary, our data support a new model, where mitochondrial quality control occurs in distal axonal compartments. Perinodal sites serve as a cascade of checkpoints that filter out damaged mitochondria as they transit towards the soma for local degradation - a mitostatic system that establishes mitochondrial mass balance in healthy axons and is adjusted in degenerative axonopathies.

Disclosures: N.A. Marahori: None. **B. Plomer:** None. **A. Iatroudi:** None. **T. Kleele:** None. **M. Schifferer:** None. **P. Avramopoulos:** None. **S. Engelhardt:** None. **M. Lakadamyali:** None. **M. Brill:** None. **T. Misgeld:** None.

Poster

P185. Metabolism, Oxidative Stress, and Cell Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P185.03

Topic: C.01. Brain Wellness and Aging

Support: US Public Health Service Grant DA000266

Title: Inositol hexakisphosphate kinase-2 regulates mitochondrial functions through creatine kinase-B

Authors: *L. NAGPAL, M. D. KORNBERG, S. H. SNYDER;
Johns Hopkins Univ. Sch. of Med., Baltimore, MD

Abstract: Inositol hexakisphosphate kinases (IP6Ks) regulate various biological processes. IP6Ks convert IP6 to pyrophosphates such as diphosphoinositol pentakisphosphate (IP7) and bis-diphosphoinositol tetrakisphosphate (IP8). IP7 is produced in mammals by a family of inositol hexakisphosphate kinases, IP6K1, IP6K2, and IP6K3, which have distinct biological functions. We explored the role of IP6K2 by elucidating its protein interactome. IP6K2 was found to robustly bind to creatine kinase-B (CK-B), a key enzyme in energy homeostasis. Cerebella of IP6K2-deleted mice (IP6K2-KO) produced less phosphocreatine and ATP and generated higher levels of reactive oxygen species and protein oxidative damage. In IP6K2-KO mice, mitochondrial dysfunction was associated with impaired expression of the cytochrome-c1

subunit of complex III of the electron transport chain. Overactive mitochondrial fission was observed in IP6K2 knocked down cells. We also observed upregulation of mitochondrial fission protein, Drp1, as well as mitochondrial biogenesis markers (PGC1α and Nrf-1). We reversed some of the effects of mitochondrial dysfunction by combined treatment with N-acetylcysteine and phosphocreatine. These findings establish a role for IP6K2-CK-B interaction in energy homeostasis associated with neuroprotection.

Disclosures: L. Nagpal: None. **M.D. Kornberg:** None. **S.H. Snyder:** None.

Poster

P185. Metabolism, Oxidative Stress, and Cell Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P185.04

Topic: C.01. Brain Wellness and Aging

Title: Hydrogen peroxide-induced neurite degeneration via mitochondrial dysfunction

Authors: *K. FUKUI, M. WAKUZAWA, N. YOSHIDA, Y. KATO;
Shibaura Inst. Technol., Saitama, Japan

Abstract: Reactive oxygen species (ROS) induce neuronal cell death and deeply relate to the development of many severe neurodegenerative disorders such as Alzheimer's disease (AD), Parkinson's disease and dementia. These diseases show the same neuronal changes in the early stages, and neuronal axons cause alteration such as aggregation, shrinking and accumulation. If we find to the early sign of the neuronal change such as axon/neurite degeneration, it has a chance to improve neuronal functions. We tried to elucidate the mechanism of neurite degeneration in hydrogen peroxide-treated neuroblastoma cells. To clarify the mechanism of neurite degeneration in hydrogen peroxide-treated N1E-115 cells, we established neurite-isolation methods and analyzed proteins using LC/MS/MS and determined some specific proteins. HSP 60 which is one mitochondrial chaperon protein was increased in the isolated neurite of a low concentration hydrogen peroxide-treated cells. However, it expression was gradually decreased in AD-transgenic mouse brain in an age-dependent manner. It was confirmed that mitochondria accumulated to the neurite degenerated region and released superoxide in cultured cells. The fluorescent intensity of Mitosox in hydrogen peroxide-treated cells was significantly increased compared to the normal cells. 3-nitrotyrosin and nNOS expressions in the isolated mitochondria on AD-transgenic mouse brain tended to increase compared to the age-matched controls. SOD-2 and NMNAT-3 expressions tended to decrease in the isolated mitochondria on AD-transgenic mouse brain tended to increase compared to the age-matched controls. We also found that endoplasmic reticulum (ER) was also accumulated at neurite degenerated regions. Mitochondria and ER are famous calcium stores in the cells, and we

are focusing on the relation between changes in calcium homeostasis and ROS-induced mitochondrial dysfunction.

Disclosures: K. Fukui: None. **M. Wakuzawa:** None. **N. Yoshida:** None. **Y. Kato:** None.

Poster

P185. Metabolism, Oxidative Stress, and Cell Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P185.05

Topic: C.01. Brain Wellness and Aging

Title: Standardization of a low-cost western diet as a model for increased weight gain during pregnancy and overweight in offspring at birth in CD-1 mice

Authors: *R. PEDRAZA-MEDINA, S. ROSALES-HERRERA, M. ORTIZ-VALLADARES, J. GUZMÁN-MUÑIZ, N. MOY-LÓPEZ;
Univ. De Colima, Colima, Mexico

Abstract: Western diet (WD) is a set of processed foods, which are high in calories, fat, and sugar, low in fiber, but pleasant in taste and texture. In recent years, studies of WD have increased, however, in developing countries is hard to have access to a standardized model of WD. They are expensive and manufactured in other countries. We aimed to develop a low-cost western diet model capable to induce increased weight gain during pregnancy and overweight in the neonatal offspring of CD-1 mice. To develop our diet, we analyzed the composition of three commercial WD. Using the standard diet (SD) "Nutricubos" from Purina Mexico as a basis we defined the composition of the diet to be developed. Then we analyzed the diet for the next parameters: crude protein, fat, crude fiber, carbohydrates, and ash, following the official methods of analysis from the Association of Official Analytical Chemists. Finally, we realized two experiments in CD-1 mice, in the first one; we analyzed bodyweight on PND 5 and spatial learning and memory on Morris Water Maze on PND 90 in 27 mice from mothers exposed to our WD during pregnancy and lactation, and 27 from animals exposed to the SD (Nutricubos). In the second experiment, we analyzed bodyweight on PND 5 and brain weight on PND 26 in 20 mice from mothers exposed to the WD during pregnancy and lactation, and 10 from mothers with an SD. We registered maternal weight gain and food intake during pregnancy in both protocols. Analysis of WD shows that our diet has 4.30 Kcal/g^2, of which 41.39% were from fats, 15.28% from proteins, and 43.43% from carbohydrates. Mice exposed to WD during pregnancy show an increased intake of food, calories and have a greater weight gain. On the PND 5, offspring from a WD were heavier than control. We did not found differences in brain weight; however, WD decreased memory skills during adulthood. In conclusion, our diet proved to have the composition of a characteristic WD, in addition to being useful to increase weight gain during pregnancy and induce overweight in neonatal offspring of CD-1 mice.

Disclosures: R. Pedraza-Medina: None. **S. Rosales-Herrera:** None. **M. Ortiz-Valladares:** None. **J. Guzmán-Muñiz:** None. **N. Moy-López:** None.

Poster

P185. Metabolism, Oxidative Stress, and Cell Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P185.06

Topic: C.01. Brain Wellness and Aging

Support: Larry L Hillblom Foundation Postdoctoral Fellowship
NIH P01 grant: P01AG036695

Title: Disrupting glucose uptake rejuvenates neural stem cells in the aged brain.

Authors: *T. J. RUETZ, C. M. KASHIWAGI, B. MORTIN, R. W. YEO, D. S. LEEMAN, M. C. BASSIK, A. BRUNET;
Stanford Univ., Stanford, CA

Abstract: Neural stem cells in the aging mammalian brain are impaired in their ability to activate to produce new neurons and regenerate damaged tissue. Here we use a genome-wide CRISPR/Cas9 knockout screen to identify gene networks to improve neural stem cell activation in old mice. We establish a high throughput system to screen primary cultures of neural stem cells. We screened 600 million neural stem cells from 3 cohorts of young and old mice to identify gene knockouts that specifically boost old neural stem cell activation. We identify 300 gene knockouts that rescue old neural stem cell function. The top gene knockouts that boost old neural stem cell activation are involved in cilium organization, cytoplasmic granule structures, and glucose import, and include the glucose transporters GLUT4 and GLUT12. To identify rejuvenating gene knockouts most relevant for the aging brain, we developed an *in vivo* screening platform. We find that knockout of GLUT4 in the sub ventricular zone stem cell niche rejuvenates neural stem cell activity in old mice. The GLUT4 protein increases in expression in the stem cell niche during aging, and indeed old neural stem cells uptake ~2-fold more glucose than their young counterparts. Importantly, a brief glucose starvation increases the ability of old neural stem cells to activate, which is not improved with knockout of GLUT4. These results show that a shift in GLUT4 activity and increase in glucose uptake drives the decline in neural stem cell activity with age, thereby providing a robust target for rejuvenation strategies. Our work also identifies a large set of gene knockout interventions to boost neural stem cell activity in old brains, which should help counter cognitive decline during aging.

Disclosures: T.J. Ruetz: None. **C.M. Kashiwagi:** None. **B. Mortin:** None. **R.W. Yeo:** None. **D.S. Leeman:** None. **M.C. Bassik:** None. **A. Brunet:** None.

Poster

P185. Metabolism, Oxidative Stress, and Cell Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P185.07

Topic: C.01. Brain Wellness and Aging

Title: A novel role for BRIP1/ FANCJ in neuronal cell health and in resolving oxidative and metabolic stress induced DNA lesions

Authors: *C. MANI[1], P. H. REDDY[2], K. PALLE[1];
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Abstract: A novel role for BRIP1/ FANCJ in neuronal cell health and in resolving oxidative and metabolic stress induced DNA lesions

Chinnadurai Mani[1], P. Hemachandra Reddy[2] and Komaraiah Palle[1]. 1. Cell Biology and Biochemistry, 2. Internal Medicine, Texas Tech University Health Sciences Center, Lubbock, TX, 79430 USA.**Presenting Author: Chinnadurai Mani[1]**

Abstract: The purpose of our study is to understand the role of BRIP1/ FANCJ in neuronal cell health and oxidative/metabolic stress. DNA damage accumulation and mitochondrial abnormalities are elevated in neurons of AD patients, and contribute to neuronal dysfunction and AD pathogenesis/progression. BRCA1 interacting protein 1 or BRIP1 is a 5' to 3' DNA helicases that catalyzes abnormal DNA structures during replication, gene transcription, recombination, and contributes to genomic integrity. BRIP1 functions are well studied in DNA repair; however, there is limited data on its role and regulation during aging and neurodegenerative diseases.

Methods: We used immunohistochemistry, western blot and RT-PCR assays to analyze BRIP1 expression in mice and human brains. Immunofluorescence assay was used to study R-loop, ROS formation and mitochondrial morphology. Flow cytometry and Electron microscopy was used to evaluate mitochondrial ROS and mitochondrial structures, respectively. Seahorse assay was performed to study the OCR, and Prestoblue assay was used to evaluate cell viability.

Results: Our results show BRIP1 expression in the brain and in neuronal cells, and its levels were elevated in hippocampal region of brain, specifically in dentate gyrus. BRIP1 downregulation in human and mouse neuronal cells caused increased R-loop formation basally and in response to H_2O_2 treatment. Additionally, knocking down BRIP1 in neuronal cells caused downregulation of BRCA1, increased DNA damage and cell death both in cycling and terminally differentiated cells. Furthermore, in BRIP1 deficient cells, elevated levels of excitotoxicity was observed in response to L-Glutamic acid exposure as evidenced by mitochondrial ROS levels, deteriorated mitochondrial health, and cell death.

Conclusions: Overall, our results indicate that BRIP1 plays important role in maintaining neuronal cell health and homeostasis by suppressing cellular oxidative stress, resolving abnormal

DNA structures, and protects mitochondria. As these metabolic stress conditions are common during aging and neurodegenerative conditions, our data suggests BRIP1 contributes to healthy aging and may have protective role in neurodegenerative diseases.
Keywords: BRIP1; R-loop; DNA damage; Mitochondrial Stress; L-Glutamic acid.

Disclosures: C. Mani: None. **P.H. Reddy:** None. **K. Palle:** None.

Poster

P185. Metabolism, Oxidative Stress, and Cell Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P185.08

Topic: C.01. Brain Wellness and Aging

Support: NIMH R00MH109626

Title: Molecularly distinct mitochondria display synapse-specific localization

Authors: *K. PANNONI[1], D. GIL[1], L. CAMPBELL[1], S. FARRIS[1,2];
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Abstract: CA2 is an understudied subregion of the hippocampus that is critical for encoding social memories. To uncover which genes play a role in CA2 function, we transcriptionally profiled subregions of the mouse hippocampus and found that genes encoding mitochondrial proteins involved in energy generation and calcium uptake have higher expression in CA2 compared to CA1. In particular, multiple components of the mitochondrial calcium uniporter (MCU) complex are selectively enriched in CA2. The MCU complex regulates calcium entry into mitochondria, which in turn can regulate bioenergetics, mitochondrial transport and localization to active synapses. We hypothesize that mitochondria in CA2 are molecularly and functionally distinct from mitochondria in CA1, which may confer unique synaptic and circuit properties underlying CA2 function. It is well-known that CA2 synapses in stratum radiatum (SR) that receive input from CA3 are resistant to long-term potentiation (LTP). Conversely, CA2 synapses in stratum lacunosum moleculare (SLM) that receive input from entorhinal cortex layer II readily express LTP. To determine if there are circuit-specific differences in mitochondria across dendritic layers, we immunostained for MCU and quantified the size, fluorescence, number, and percent area of mitochondria. We found that MCU fluorescence is strikingly enriched in CA2 SLM compared to SR. Furthermore, the MCU-enriched mitochondria in CA2 SLM are larger compared to MCU-labeled mitochondria in SR. This suggests that larger mitochondria preferentially localize to distal and/or more plastic synapses, perhaps due to enriched MCU expression. To determine if MCU expression regulates mitochondrial size and/or localization, we used adeno-associated viral vectors to overexpress MCU or GFP in CA1, which

normally has 4-fold less MCU than CA2. MCU overexpression led to larger MCU-labeled mitochondria in CA1 SR and SLM compared to GFP controls, with a preferential enrichment in SR compared to SLM. Our findings demonstrate that mitochondria are molecularly diverse across hippocampal cell types and circuits and that MCU is a key regulator of mitochondrial size and localization at synapses. Ongoing studies are using a CA2-specific MCU conditional knockdown mouse line to investigate whether there is bi-directional effect on mitochondrial size and localization in CA2 that could impact circuit-specific function.

Disclosures: K. Pannoni: None. **D. Gil:** None. **L. Campbell:** None. **S. Farris:** None.

Poster

P185. Metabolism, Oxidative Stress, and Cell Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P185.09

Topic: C.01. Brain Wellness and Aging

Title: The relationship between obesity and cognitive dysfunction; Prevention by tocotrienols

Authors: *Y. KATO, J. BEN, K. FUKUI;
Shibaura Inst. of Technol., Saitama, Japan

Abstract: Recently, it is social problem to increasing of the people with cognitive dysfunction such as Alzheimer's (AD) and Parkinson's disease. Obesity is also increasing all over the world. Obesity raises a risk of secondary diseases such as type-2 diabetes and cardiovascular disease. It has been reported that obesity induces cognitive dysfunction. However, detailed mechanisms of it have been yet unclear. The diseases induced by obesity strongly related to accumulation of oxidative damages by reactive oxygen species. In fact, the amount of lipid peroxide in AD model mice cortex and hippocampus are higher than that of control mice. We expected that obesity may induce cognitive dysfunction via elevation of brain oxidation.
Vitamin E is one of lipophilic vitamins and have chroman ring structure with side chain. Tocotrienols (T3s) are the vitamin E homologs with unsaturated side chain and have strong antioxidant function. It was reported that T3s but not the saturated vitamin E (tocopherols) have beneficial functions such as anti-cancer and neuroprotective effects. In addition, antioxidant activity of T3s is higher than that of tocopherols. Additionally, we demonstrated that T3s have anti-obesity effects such as inhibition of body weight gain and decrease of serum cholesterols. So, it is possible that T3s can prevent obesity-induced cognitive impairment via antioxidant and/or anti-obesity effects.
We fed a high-fat diet or high-fat high-sucrose diet in the presence or absence of T3s. We prepared some experimental conditions (diets, treatment duration, age of mice, treatment volume of T3s, etc.). We measured body weight at once a week. After duration, we assessed the effects of obesity and T3s on mice cognition by the Morris water maze, rota rod, Y-maze and Open filed

test. Furthermore, we measured oxidative stress makers and the expressions of antioxidative enzymes, neurotrophic factors and acetylated histone H3 in the high-fat diet and high-fat high-sucrose diet-treated mice brain. We would like to present the detailed-results and comparison between each experimental condition.

References
1. Fukui K, et al., *Nutrients*, 2019.
2. Kato Y, et al., *Int J Mol Sci*., 2020.
3. Kato Y, et al., *J Clin Biochem Nutr*., 2021.

Disclosures: Y. Kato: None. **J. Ben:** None. **K. Fukui:** None.

Poster

P185. Metabolism, Oxidative Stress, and Cell Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P185.10

Topic: C.01. Brain Wellness and Aging

Support: Wellcome Trust

Title: The role of oxidative stress in microtubule regulation and neuronal ageing

Authors: *S. SHIELDS, O. R. WILKES, N. SANCHEZ-SORIANO;
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Abstract: In healthy axons, microtubules are arranged into parallel bundles providing the structural backbones and highways for life-sustaining transport from and to the cell body. During ageing and across different species, neurons experience changes at the subcellular level including decay of the microtubule (MT) cytoskeleton and a rise in reactive oxygen species (ROS) leading to oxidative stress (OS). We intend to understand the nexus between oxidative stress and microtubule damage and the contribution to axonal and synaptic decay during neuronal ageing. Using primary neuronal cultures derived from *Drosophila*, we show that OS (induced by loss-of-function of superoxide dismutases, Sod; Gss1 and Gss2 knockdown, the enzymes catalysing the antioxidant glutathione; or treatment with ROS-inducing compounds) disrupts microtubule organisation within axons in vitro and is rescued via administration with the antioxidant vitamin E analogue trolox. At the mechanistic level, we find that the association of Eb1 (end binding protein) to microtubules is reduced in conditions of OS. Since Eb1 is a master regulator of microtubule polymerisation, dysregulation of Eb1 could explain MT defects observed in conditions of OS in vitro. Confirming the latter, we find that the neuroprotective peptide NAP, which enhances Eb1 binding to MTs, fully rescues ROS-induced MT disorganisation. To assess the importance of our findings for neuronal ageing, we established a novel in vivo model within the fly optic lobe, to study the cell biology of ageing. Within neurons, we observed the

accumulation of MT defects within axons and synaptic terminals during ageing and such changes preceded the thinning of axons and the appearance of membrane swellings. All these ageing phenotypes were exacerbated in both paraquat and diethyl maleate-fed flies. Conversely, the deterioration of axons and synapses could be improved upon knockdown of the NRF2 inhibitor Keap1 and by the overexpression of Eb1. We propose that age-related OS alters the functions of key MT regulators, such as Eb1, leading to the formation of MT aberrations and, in turn, axon and synapse decay. From this, enhancing the function or stability of Eb1 provides an effective strategy for combatting OS-induced neuronal decay during ageing.

Disclosures: S. Shields: None. **O.R. Wilkes:** None. **N. Sanchez-Soriano:** None.

Poster

P200. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vitro Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P200.01

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: An efficient iPSC-derived “brain-in-a-dish” tri-culture model identifies a role for TREM2 in modulating a neuroinflammatory cascade implicated in neurodegeneration

Authors: M. OHSHIMA, T. WAKUI, **M. GOEDLAND**, M. LIVINGSTON, S. BURTON, C. B. CARLSON, *W. W. POON, S. HILCOVE, D. RAJESH;
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Abstract: Neuron-glia interactions play a pivotal role in normal brain homeostasis in which perturbations to disrupt this delicate balance are implicated in neurodegenerative disease pathogenesis. Although mouse models have uncovered many aspects related to the role of brain neuroinflammation in disease etiology, salient features are not captured due to species differences. Hence, there is a critical need to develop human disease models that are more translatable to the clinic. Here, we describe a defined, scalable *in vitro* tri-culture model combining human induced pluripotent stem cell (iPSC)-derived GABAergic neurons, astrocytes, and microglia that can be used to study cellular interactions and cell type-specific genetic contributions to model neurodegeneration for drug screening applications. This “brain-in-a-dish” system was used to investigate the role of TREM2 in neuroinflammation, as TREM2 mutations are implicated in Alzheimer’s Disease (AD) risk. By examining these tri-cultures that contain iPSC-derived microglia differentiated from either an apparently healthy donor (AHN), an engineered heterozygous (HZ) TREM2 knockout (KO) iPSC line, or a homozygous (HO) TREM2 KO line, we found intrinsic differences between the AHN and TREM2 engineered lines and that LPS treatment led to neurotoxicity in tri-cultures, as analyzed by an image-based quantification system coupled to immunocytochemistry. Further analysis of tri-cultures with either TREM2 HZ or TREM2 HO KO microglia revealed a several fold increase in

neuroinflammatory cytokines and chemokines (TNF-alpha, IL-6, IL-10, IL-12, CCL2, CCL3, CCL4 and CX3CL1), and augmented the generation of A1 astrocytes, implicating a role for TREM2 in restraining the A1 astrocyte phenotype. Importantly, we identified intrinsic differences in complement component production (C3 and C1q) due to TREM2 loss of function in microglia in mono-culture compared to when in tri-culture. Thus, iPSC-based tri-cultures can serve as a powerful tool to screen for therapeutics to rescue neuroinflammation-mediated neurotoxicity, as well as to study autocrine and paracrine signaling pathways involved in neuronal dysfunction, synaptic pruning, and provide mechanistic insights into pathways disrupted by disease associated microglia (inherited genetic mutations) in the context of a more relevant brain environment.

Disclosures: M. Ohshima: None. **T. Wakui:** A. Employment/Salary (full or part-time):; FUJIFILM CDI. **M. Goedland:** A. Employment/Salary (full or part-time):; FUJIFILM CDI. **M. Livingston:** A. Employment/Salary (full or part-time):; FUJIFILM CDI. **S. Burton:** A. Employment/Salary (full or part-time):; FUJIFILM CDI. **C.B. Carlson:** A. Employment/Salary (full or part-time):; FUJIFILM CDI. **W.W. Poon:** A. Employment/Salary (full or part-time):; FUJIFILM CDI. **S. Hilcove:** A. Employment/Salary (full or part-time):; FUJIFILM CDI. **D. Rajesh:** A. Employment/Salary (full or part-time):; FUJIFILM CDI.

Poster

P200. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vitro Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P200.02

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Innate immunity protein IFITM3 is involved in Alzheimer’s disease-associated microglial response

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Abstract: Alzheimer’s disease (AD) is the most common neurodegenerative disorder. It has complex neuropathology which includes an accumulation of amyloid beta (Aβ) plaques, hyper-phosphorylated tau tangles, and neuroinflammation. Our lab has identified that expression of the interferon response protein IFITM3 is upregulated in human AD brain as well as in 5xFAD mouse brain, which expresses human APP and PSEN1 transgenes. We have shown that IFITM3 regulates γ-secretase activity in neurons and astrocytes, and a deficit in IFITM3 results in decreased γ-secretase cleavage of APP. Further, IFITM3 is highly expressed and inducible in

microglia. Research has indicated that microglia play a critical role in AD progression. Our current focus is on investigating the function of IFITM3 in the phagocytic clearance of Aβ by microglia and its interplay with TREM2 and downstream signaling in the context of neuroinflammation. For this, we have generated 5xFAD ; IFITM3$^{-/-}$; TREM2$^{-/-}$ mice and have established stem cell-derived IFITM-deficient microglial cells. With our studies, we hope to determine the role of IFITM3 in microglia, which has not been previously described before, using cellular assays and mouse models.

Disclosures: T. Jain: None. **G. Frost:** None. **J. Hur:** None. **L. Studer:** None. **Y. Li:** None.

Poster

P200. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vitro Models

Location: Virtual Only

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Program #/Poster #: P200.03

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH/NIA 1R01AG059639
3R01AG05963
AARGD-591887

Title: Activated Endothelial Cells Induce a Distinct Type of Astrocytic Reactivity Associated to Cerebral Amyloid Angiopathy

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Abstract: **Background:** Recently, transcriptomic profiling has helped identify the diverse heterogeneity and distinct molecular states of astrocytes in different disease models. Mouse astrocytes adopt an ‘A1’ neurotoxic phenotype, characterized by the increase expression of complement 3 (C3), after exposures to specific cytokines secreted by microglia, whereas they acquire an ‘A2’ neuroprotective phenotype in a model of ischemic stroke. Although, now it is unclear if these binary divisions are fixed and if A1 and A2 phenotypes are all encompassing, suggesting a much more complex disease specific interplay. Interestingly, we and others have reported in response to the cerebrovascular deposition of amyloid in CAA, severe astrogliosis with an C3^{+}-astrocytic phenotype without reactive microgliosis, suggesting that other cell types

may be responsible for A1 astrocytic induction independent of microglia activation. **Methods:** Here, we use LPS-mediated inflammation of microglia and endothelial cells to identify distinctive reactive astrocytic profiles in primary culture assays and in mouse models and AD/CAA patients. **Results:** We show that LPS-mediated inflammation of endothelial cells induces the formation of $C3^+$-reactive astrocyte with a strong extracellular matrix remodeling profile, distinct from astrocytes classically activated by neuroinflammatory microglia. We show that activated endothelial cells strongly induce C3 positive astrocytes enriched in A1-astroctyic genes and exhibit a gain of neurotoxic function while retaining homeostatic phagocytic capacity. Additionally, the transcriptomes of astrocytes differ between endothelial and microglial inflammatory stimulus and GBP2 and extracellular matrix remodeling gene Decorin stratify to molecular subtypes of $C3^+$-astrocytes. Finally, we demonstrate $Decorin^+$ $C3^+$ astrocytes are associated to vascular amyloid deposits but not amyloid plaques in mouse models and AD/CAA patients showing the existence of potentially extensive and subtle functional diversity of $C3^+$-reactive astrocytes and the multidimensional roles of reactive astrocytes after CNS injury and neurodegenerative diseases. **Conclusion:** The presence of $C3^+Decorin^+$ astrocytes associated to vascular amyloid deposits suggests vascular damage induces reactive astrocytes with an extracellular matrix remodeling profile in mouse models and AD/CAA patients that are distinct from astrocytes classically activated by neuroinflammatory microglia demonstrating the existence of potentially extensive and subtle functional diversity of neurotoxic A1- astrocytes.

Disclosures: X. Taylor: None. **P. Cisternas:** None. **N. Jury:** None. **X. Huang:** None. **Y. You:** None. **J. Redding:** None. **R. Vidal:** None. **J. Zhang:** None. **J.C. Troncoso:** None. **C.A. Lasagna-Reeves:** None.

Poster

P200. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vitro Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P200.04

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: LKCMedicine Strategic Academic Initiative

Title: Tnf-mediated neuroinflammation linked to neuronal necroptosis signaling in alzheimer's disease hippocampus

Authors: *A. JAYARAMAN[1], T. T. HTIKE[1], R. JAMES[2], C. PICON[2], R. REYNOLDS[1,2]; [1]Lee Kong Chian Sch. of Med. - NTU, Singapore, Singapore; [2]Div. of Neuroscience, Dept. of Brain Sciences, Fac. of Medicine, Imperial Col. London, London, United Kingdom

Abstract: Neuroinflammation has been implicated in the pathogenetic mechanisms underlying neuronal death and dysfunction in Alzheimer's disease (AD). In chronic inflammatory

environments, TNF-TNFR1 can lead to cell death via necroptosis. Although necroptosis has recently been demonstrated in AD, its significance in neuronal death and the role of TNF signaling is unclear. Using post-mortem human brain tissues and human iPSC-derived glutamatergic neurons, we demonstrate that TNF signaling is linked to neuronal necroptosis. We show evidence of altered expression of ESCRT III components, which antagonize necroptosis and provide a possible mechanism for cell survival after necroptosis. Snap-frozen post-mortem hippocampal tissue blocks from AD (n=30) and non-neurological control subjects (n=11), were compared. For RNA/protein work, 5-10 serial 30µm cryosections of grey matter tissue were pooled for each sample. The differential spatial distribution and cellular source were examined by multiple immunolabelling on 10µm cryosections. All imaging data were correlated with pathological and clinical outcomes. Human iPSC-derived glutamatergic neurons were treated with TNF+Smac-mimetic+zVAD, with or without pretreatment with RIPK1, RIPK3 or MLKL inhibitors. LDH assays were used to determine the extent of neuronal death. qRTPCR analysis showed 2–3-fold upregulation of TNF and necroptosis pathway genes in AD. Immunolabelling showed increases in TNFR1, RIPK1, pRIPK3 and pMLKL expression in the AD hippocampus, predominantly in CA1-2 pyramidal neurons. Western blot analysis showed significant increases in pMLKL, pRIPK3, TNFα, FADD, and TNFR1 expressions in AD. Colocalization of pMLKL with pRIPK3/RIPK1, and co-immunoprecipitation of pRIPK3-MLKL in the AD hippocampus further provided evidence for neuronal necroptosis and necrosome formation. We showed chronic inflammatory factors (CD8+ T-cells and activated microglia) around pMLKL+ neurons. ESCRT-III components showed a significant overlap at the subcellular level in pMLKL+ neurons. TNF+Smac-mimetic+zVAD exposure induced necroptosis in human iPSC-derived glutamatergic neurons with ~67% neuronal death at 24 h, and significant increase in ROS production by 6 h, both of which were effectively decreased by inhibitors. Our results suggest that increased TNF/TNFR1-mediated inflammatory signaling leads to necroptotic cell death in AD brains. Demonstration of TNF-mediated necroptosis in human iPSC derived neurons and its reduction with small molecule inhibitors targeting RIPK1, RIPK3 or MLKL suggests that this pathway could potentially be therapeutically manipulated in AD.

Disclosures: A. Jayaraman: None. **T.T. Htike:** None. **R. James:** None. **C. Picon:** None. **R. Reynolds:** A. Employment/Salary (full or part-time):; 2Division of Neuroscience, Department of Brain Sciences, Faculty of Medicine, Imperial College London, London, UK..

Poster

P200. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vitro Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P200.05

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant T32NS096050

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NIH Grant RF1AG071587
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Title: Identification of state-specific proteomic characteristics of microglia-derived exosomes

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Abstract: Alzheimer's disease (AD) is the most common neurodegenerative disorder defined by progressive pathological protein aggregation (amyloid-beta and tau) and deterioration of cognitive function. Microglia-mediated neuroinflammation is a key pathological component of AD; however, there are critical gaps in our understanding of how microglia perpetuate AD pathology. One proposed mechanism of microglia-mediated neuroinflammation and neurodegeneration is exosome release because of their critical role in the transport of macromolecules between cells to facilitate intercellular communication. Thus, it is possible that microglia-derived exosomes transfer pathogenic cargo which could perpetuate AD pathology. The proteomic profiles and influence of different microglia-derived exosomal populations on AD pathology remain unknown. We hypothesize that different microglia states determine the molecular composition of exosomes. To test this, we treated a murine microglia cell line, BV2 cells, with various cytokines to polarize them and collect their exosomes for downstream mass spectrometry (MS) analyses. Three groups of BV2 cells (n=4/group) were treated with either lipopolysaccharide (LPS) to polarize to a pro-inflammatory state, interleukin 10 (IL-10) to polarize to an anti-inflammatory state, or transforming growth factor beta (TGF-β) to polarize to a homeostatic state. Untreated BV2 cells served as a control group. Following 72 hours of treatment, BV2 cells were lysed and cell culture media was collected for exosome isolation. Transmission electron microscopy images and western blotting for exosomal marker, CD9, confirmed exosome purification by our isolation method. In MS studies, we identified 533 proteins in exosome fractions and 1,866 proteins in BV2 cell proteomes. We found that known exosome related proteins, Sdcbp and Igsf8, were significantly increased in the exosomal proteome compared to the whole cell proteome. We identified proteins that are differentially expressed across polarization with unique proteins increased in exosomes derived from LPS, IL-10, and TGF-β treated BV2 cells. Validation studies of these top hits are ongoing, and studies are underway to replicate these findings in primary microglia. Our results indicate that exosomes derived from microglia adopt distinct state-associated protein profiles which may have differential effects on other cell types. This work will guide future studies concerning the role of exosomal cargo in perpetuating AD pathology.

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Poster

P200. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vitro Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P200.06

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Exploring cell signaling pathways engaged by two novel small molecule drug candidates (MW151 and MW189) that selectively modulate stressor-induced neuroinflammation

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Abstract: Neuroinflammation plays an important role in many different brain pathologies and injuries and therefore may represent a beneficial therapeutic target across neurological conditions. Two brain-penetrant, anti-inflammatory small molecule drug candidates, MW151 and MW189, are in or have completed Phase 1 clinical trials for safety (clinicaltrials.gov ID NCT04120233 and NCT02942771, respectively). MW151 and MW189 are structural analogs, and both ameliorate neuroinflammatory effects across a broad range of preclinical animal models, such as reducing glial activation and pro-inflammatory cytokine production. However, their molecular target(s) are not defined. To maximize the likelihood of eventual clinical success for one or both drugs (or similar compounds), we have started exploring potential target pathways *in vitro*. For these studies, we are utilizing the BV-2 mouse microglial-type cell line. It is already known that these drugs do not inhibit the p38 MAPK pathway, so other major inflammatory pathways such as NFκB, STAT3, JNK, and ERK are currently being explored. We found that MW151 and MW189 inhibit lipopolysaccharide (LPS)-induced iNOS stimulation and LPS-induced cytokine secretion, which may occur in part via inhibition of NFκB signaling. Both drugs also block extended IL-6 induced STAT3 phosphorylation (1 to 4 hours post-stimulation), with no effect on the early phase (30 minutes or less). While these findings offer intriguing clues, an unbiased proteomic exploration of BV-2 cells exposed to MW151 and MW189 is being undertaken to more clearly identify the molecular mechanism(s) and target pathway(s) of these drugs. Successful identification of the signaling pathways and proteins modulated by MW151 and MW189 *in vitro* will provide the biological rationale for design of future clinical trials, and possibly open new avenues of exploration in future drug development programs.

Disclosures: E.D. Roepke: None. **D.J. Braun:** None. **L.J. Van Eldik:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Dr. Linda J. Van Eldik is an inventor on the patents covering MW151 and MW189., Dr. Linda J. Van Eldik is a scientific founder of ImmunoChem Therapeutics, LLC, a Northwestern University spinout formed to commercialize the compounds..

Poster

P200. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vitro Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P200.07

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Assessing and resolving the enigmatic role Complement Receptor 1 (CR1) plays in Alzheimer's Disease

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Abstract: Complement receptor 1 (*CR1*) was implicated as a top Alzheimer's Disease (AD) risk factor in 2009, yet its role in AD remains an enigma. *CR1* encodes a transmembrane, regulatory protein with an extracellular binding domain that binds complement cascade proteins (C3b, C4b, and iC3b), and is thought to improve Aβ clearance. Oddly, while *CR1* RNA and proteins are highly expressed in various blood cells, *CR1* RNA is not expressed in brain tissue at a meaningful level, making it difficult to understand *CR1*'s involvement in AD. A study by Crehan et al. suggests CR1-homologue proteins (Cr2) are present on rat microglia, and that blocking Cr2 significantly decreases microglial phagocytosis of $A\beta_{42}$. A major challenge here, however, is whether murine *Cr2* in fact encodes a valid human *CR1* homologue.Adding to the peculiarity of *CR1* involvement in AD, the gene has been genetically linked as a potential loss-of-function risk gene, yet certain apparent gain-of-function variants are also associated with AD risk. Specifically, CR1 variable C3b/C4b binding domains in the population, where more binding domains are associated with increased AD risk. Furthermore, we recently demonstrated the *CR1* binding domains are "camouflaged," meaning they are entirely overlooked by standard short-read sequencing methods and likely have undiscovered DNA variants. In fact, by rescuing mutations from these regions in *CR1* in the Alzheimer's Disease Sequencing Project (ADSP), we identified a rare 10-nucletotide loss-of-function frameshift deletion that could be involved in disease. In short, significant evidence suggests *CR1* plays a major role in AD pathogenesis, yet exactly how *CR1* is involved remains an enigma, demonstrating the importance of research into *CR1*'s involvement. We propose a future study to clarify *CR1*'s apparent enigmatic role in AD. Using a combination of human tissue, cell lines, and murine models, our primary goals are to: (1) determine whether CR1 proteins are present in human brain tissue, and assess function in the brain; and (2) experimentally modify the binding domain by increasing, decreasing, and mutating the region, including the 10 bp frameshift to test its effects on AD phenotypes. We hope these experiments clarify the enigmatic nature of CR1's involvement in AD.

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Poster

P200. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vitro Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P200.08

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIA Grant K01AG066747

Title: Altering microglia phenotype in Alzheimer's disease by blockade of L-type calcium channels in 5XFAD mouse model with isradipine

Authors: *J. WICKLINE[1], S. SMITH[1], K. ODFALK[1], R. SHIN[2], J. SANCHEZ[1], M. JAVORS[1], B. GINSBURG[1], S. C. HOPP[3];
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Abstract: Alzheimer’s disease (AD) is the sixth leading cause of death in the United States and is pathologically characterized by tau tangles and amyloid beta (aβ) plaques. In AD, microglia take on a neurodegenerative phenotype characterized by toxic gain-of function (e.g., proinflammatory cytokine production) and loss-of-function (e.g., reduced ability to remove aβ). Intracellular calcium (Ca^{2+}) dysregulation is a key factor that may drive the microglia neurodegenerative phenotype in AD. Microglia display dysregulated intracellular Ca^{2+} near plaques. Epidemiological studies show that use of Ca^{2+} channel blockers targeting L-type Ca^{2+}channels (LTCCs) reduce the incidence of neurodegenerative diseases, but the mechanism of this observation is unknown. We hypothesize that LTCCs on microglia mediate their Ca^{2+} dysregulation and neurodegenerative phenotype near aβ plaques. To test this hypothesis, we treated male and female 5XFAD plaque pathology model mice with the LTCCs antagonist isradipine (3 mg/kg/day) using three different drug delivery methods: osmotic minipumps, mouse chow and subcutaneous slow-release pellet. Following three weeks of treatment, we tested whether isradipine treatment could improve learning and memory behavior measured using novel object recognition and Morris water maze (MWM). We also measured markers of neurodegenerative microglia using qPCR, immunohistochemistry (IHC) and protein biochemistry. qPCR was used to measure phenotypic markers of neurodegenerative microglia such as IL1β, iNOS, TNFα, TREM2 and CD68, and the LTCCs Cav1.2 and 1.3. IHC was used to examine the relationship between microglia and aβ plaques and the presence of dystrophic neurites. Finally, protein biochemistry was used to quantify the ratio of aβ 42/40. All data was analyzed with a two- or three-way ANOVA using post hoc Tukey HSD and G-power was used

to calculate the correct sample size for behavior assays at a power of 0.80. We observed a genotypic difference in 5XFAD and wild-type (WT) littermates in the MWM but observed no effect of isradipine treatment. However, mass spectrometry revealed subtherapeutic levels of drug in the plasma and brain for 2 out of 3 delivery methods. Our studies showed that previously published literature using isradipine in osmotic minipumps was not reproducible and that a novel chow delivery of isradipine is not effective. We show the most reliable drug delivery method for isradipine is the subcutaneous pellet; future studies will focus on understanding the gene expression, histological, and biochemical changes following this treatment paradigm in the 5XFADs.

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Poster

P200. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vitro Models

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Program #/Poster #: P200.09

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: OSU Carlson College of Veterinary Medicine and OSU Foundation
Oregon Tax Check-off Alzheimer's Research Fund

Title: Early changes in nmda receptors in 5xfad mice are enhanced through the treatment of ibuprofen in females but not males

Authors: *E. SACKINGER[1], J. RAUENHORST[1], T. VU[1], A. RADKE[1], F. NIGUSSIE[2], K. R. MAGNUSSON[3];
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Abstract: Changes in amyloid occur early in Alzheimer's disease (AD), before behavioral signs. Based on this, and early changes seen in N-methyl-D-aspartate receptors (NMDARs) in presenilin models, we aimed to study the development of changes in α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid receptors (AMPARs) and NMDAR prior to behavioral signs of amyloid-related degeneration in 5xFAD mice. We performed 2 studies to examine glutamate receptor responses, with and without ibuprofen (IBU) intervention. In order to assess early changes in 5xFAD mice at 15-days, 1, 2 or 4 months old, we examined the field excitatory postsynaptic potentials (fEPSPs) of CA1 synapses in hippocampal slices using a MED64 multielectrode array system. Antagonists of AMPAR, GluN2A and GluN2B NMDAR subunits, and full NMDARs were applied. Hippocampal slices began in regular artificial cerebrospinal fluid (aCSF), followed by 1) 30μM DNQX (AMPAR antagonist), 2) 0.5mM $MgSO_4$ aCSF,

30μM DNQX & 10μM picrotoxin (γ-aminobutyric acid (GABA) receptor antagonist) solution; 3) 4μM Ro-25-6981 (GluN2B antagonist), 4) 500nM PEAQX (GluN2A antagonist), and 5) 50μM AP5 (NMDAR antagonist). A stimulus response was performed after each step and input (fiber volley amplitude) / output (fEPSP amplitude) was analyzed. At 4 months old, both male and female hemizygous (5xFAD) mice showed increases in I/O responses for GluN2A, GluN2B and all NMDARs (p = .014-<.0001) but decreases at 15 days in AMPARs (p =.015, <.0001), and at 2 months for AMPARs and GluN2A (p <.0001). 5xFAD females showed increased I/O responses for AMPARs (1 and 4 months, p <.0001), GluN2B and NMDAR (1 month, p=.02), but decreases at 2 months for GluN2B (p <.0001). 5xFAD males had increased I/O responses for GluN2B (p=.0003) at 15 days, but decreased responses for NMDAR (p. <.0001) at 1 month and GluN2B and NMDAR (p =.001- <.0001) at 2 months. There appeared to be early increases in glutamate transmission at .5-1 month, followed by a reduction and subsequent return to hyperresponsive NMDARs by 4 months in 5xFAD mice. Previously, our lab determined IBU can decrease NMDAR expression in male mice. We treated 2 month old 5xFAD mice with IBU (375 ppm in chow) for 2 months, in order to reduce the increased synaptic NMDAR responses that we saw at 4 months in 5xFAD mice. Both male and female 5xFAD showed increased I/O responses for NMDAR and GluN2A (p=.007-<.0001). IBU increased responses for NMDAR, GluN2A and AMPAR in both WT (p<.0001) and 5xFAD (p=.02-.0009) females, but decreased responses to GluN2B and AMPAR in 5xFAD males (p=.045-.0002). IBU had small reducing effects on glutamate receptor responses in males, but enhanced NMDAR responses in females.

Disclosures: E. Sackinger: None. **J. Rauenhorst:** None. **T. Vu:** None. **A. Radke:** None. **F. Nigussie:** None. **K.R. Magnusson:** None.

Poster

P200. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vitro Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P200.10

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Modeling the Impact of AD Genetic Risk on Microglia States and Function

Authors: *M. THERRIEN[1], M. J. DOLAN[2], S. JEREB[2], T. KAMATH[2], N. LOJEK[2], T. ATKESON[2], S. E. MARSH[4], M. B. JOHNSON[5], K. EGGAN[6], E. MACOSKO[3], B. A. STEVENS[7];
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Abstract: Microglia, the brain's resident immune cells, are highly dynamic and reactive to environment and genetic challenges. More than 40 genomic loci have been linked to late onset Alzheimer's disease (AD) and many risk genes are highly expressed in microglia. Our goal is to connect insights from genetic association studies to new ways of functionally modeling the cellular and molecular causes of disease to enable predictive tracking and targeting of detrimental immune cell states in patients in the early stages of disease. Single cell transcriptomic studies reveal diverse microglial states in human and mouse brains, however we currently lack specific approaches to track and manipulate specific populations of microglia in Alzheimer's and other diseases. To answer this question, we turned toward human iPSC-induced microglia (iMGL) and single cell transcriptomics.Single cell transcriptomics revealed the presence of multiple microglia states when iMGLs are stimulated with different brain-relevant challenges, including apoptotic neurons, synaptosomes, myelin and amyloid. Moreover, alignment of these data using LIGER (Welch et al. Cell 2019) shows these states are similar to the ones observed in human and mouse in vivo, revealing several disease associated states, including disease-associated microglia (DAM). We also observed changes in microglia states depending on the challenge and genetic background.Together, our data identified key elements causing the formation of DAM and how AD risk genes affect disease-associated states and functions. This work will open the door to the identification of modulators of DAM and highlight new therapeutics avenues of AD.

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Poster

P200. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vitro Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P200.11

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH T32 NS082145
NIH K01 AG066747

Title: Microglia internalize tau monomers and preformed fibrils via macropinocytosis and clathrin-mediated endocytosis

Authors: *K. F. ODFALK[1], C. M. BYRNE[1], D. LIM[2], S. C. HOPP[1];
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Abstract: Aggregation of misfolded tau is a hallmark of many neurodegenerative diseases including Alzheimer's disease (AD) and is correlated with cognitive decline. Neurons release tau which enters nearby cells and serves as a template for tau aggregation, thus propagating the progression of tau pathology through the brain. Microglia do not express the tau gene. Yet, microglia containing tau are observed in brains of AD patients and mouse models of tauopathy. This suggests that microglia take up neuronal tau, but how this occurs is unclear. Microglia undergo constitutive macropinocytosis and thus efficiently internalize liquid-phase particles in their extracellular milieu. We hypothesized that microglia employ macropinocytosis to internalize free extracellular tau. To test this hypothesis, we treated microglial cells (BV2) in culture with different endocytic pathways inhibitors and probed their uptake of fluorescently conjugated monomer and preformed fibril (PFF) tau. These compounds include: cytochalasin D (CTD), dynasore, chlorpromazine (CPZ), and methyl-β-cyclodextrin (MβCD), inhibitors of actin polymerization, dynamin, AP2, and lipid rafts, respectively. As predicted CTD robustly inhibited tau uptake in BV2 microglia, suggesting an actin-dependent process is involved. Interestingly, CPZ and dynasore also inhibited monomer and PFF uptake but neither as robustly as did CTD. This suggests that macropinocytosis, an actin-dependent but dynamin-independent process, and clathrin-mediated endocytosis (CME), an AP2- and dynamin-dependent process, constitute the two potential endocytic pathways of tau uptake in BV2 microglia. We next asked whether microglia tau uptake was a saturable, receptor-mediated process. To test this, examined whether increasing concentrations of unlabeled tau altered the internalization of a constant concentration of labeled tau. Here we observed that labeled tau monomer uptake decreased with the addition of unlabeled tau, but tau PFFs uptake did not. This suggests that tau monomer uptake in BV2 microglia is receptor mediated. In future experiments we will co-administer tau with 70 kDa dextran or transferrin, positive controls for macropinocytosis and CME, respectively, and measure co-localization within endosomal compartments. The literature suggests that microglia play a dual-role role in tau pathology, as they have been implicated in both tau clearance and spread. By determining the tau uptake mechanisms employed by microglia, we hope to advance our understanding of microglia-tau interaction and aid in the development of therapies aimed at preventing pathological tau progression and cognitive decline.

Disclosures: K.F. Odfalk: None. **C.M. Byrne:** None. **D. Lim:** None. **S.C. Hopp:** None.

Poster

P200. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vitro Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P200.12

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: ISHIZUE 2021 of Kyoto University Research Development Program.

Title: The roles of oligodendrocytes in the pathogenesis of Alzheimer's disease

Authors: *K. YASUDA[1], T. MAKI[1], M. TOYOKAWA[2], S. TODA[1], Y. KUWATA[1], R. TAKAHASHI[1];
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Abstract: [Aim] Neuron-centric view cannot explain entire aspects of Alzheimer's disease (AD). Among non-neuronal cells, disruption of oligodendrocytes (OLs) and myelin has been reported as an early sign in the AD. Given that oligodendrocyte precursor cells (OPCs) regulate brain homeostasis by interacting with neuronal, vascular, glial, and immune system, pathological OPCs/OLGs could be associated with the onset and progression of AD. However, the mechanisms by which OLs exert harmful effects in AD remain elusive. Therefore, the purpose of present study is to examine the roles of OLs on the pathogenesis of AD. [Methods]For *in vitro* experiments, we examined the effect of Aβ oligomers on OLs as well as the expression levels of Aβ and its related factors in primary cultured OLs. Quantitative RT-PCR was performed to confirm the effects of Aβ oligomers on the phenotype of OPCs. For *in vivo* studies, we evaluated the behaviour of OLs in AD model mice (Tg-SwDI) and postmortem human brains with AD. [Results] *In vitro* studies showed that Aβ oligomers induced cytotoxicity in OPCs in a dose-dependent manner. In addition, OLs themselves produced Aβ40/42, which are pathogenic for AD, via amyloidogenic pathway, and secreted sAPPα, which has neuroprotective and neurogenic functions, via non-amyloidogenic pathway. RT-PCR revealed that Aβ42 oligomers increase the multiple gene expression levels of inflammation-related factors and metalloproteinases in OPCs. In AD model mice and patients, OPCs were located in close proximity with Aβ depositions. [Conclusions] The present study indicated the bi-directional relationship between APP processing and OLs in AD. Further studies are warranted to elucidate the pathological shift in OLs from beneficial non-amyloidogenic process to detrimental amyloidogenic process during AD progression.[Grant/Other Support] ISHIZUE 2021 of Kyoto University Research Development Program.

Disclosures: K. Yasuda: None. **T. Maki:** None. **M. Toyokawa:** None. **S. Toda:** None. **Y. Kuwata:** None. **R. Takahashi:** None.

Poster

P200. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vitro Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P200.13

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Nu Rho Psi Undrgraduate Research Grant

Title: Cxcl14 as a Potential Modulator for Amyloid Beta Clearance and Cell Death in Alzheimer's Disease

Authors: J. GALSKE, J. ZHOU, R. YAN;
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Abstract: The abnormal accumulation of beta-amyloid (Aβ) plaques is viewed to initiate events that eventually cause Alzheimer's Disease (AD) neuronal cell death. In AD, both cytokines and chemokines are responsible for recruiting astrocytes and microglia towards Aβ plaque sites for uptake and degradation. Activated microglia and reactive astrocytes are known to regulate the expression and/or secretion of certain cytokines and chemokines that may play beneficial or detrimental effects on the development of AD pathology. We have now found higher levels of CXCL14 localized around Aβ plaques in the 5XFAD mouse model. CXCL14 is a chemokine possessing a C-X-C domain and has been identified as an inflammatory modulator in the periphery. However, its' role in neurodegeneration and AD is unknown. In this study, we explored whether CXCL14 is a chemoattractant to microglia migration in AD mouse models. We showed that in vitro administration of Aβ increased CXCL14 levels in primary astrocytes. A transwell migration assay revealed that CXCL14 recruited microglia in a dose-dependent manner, but not astrocytes. CXCL14 also enhanced Aβ uptake in primary microglia with both acute and recurrent Aβ treatments. Furthermore, CXCL14 protects both microglia and neurons from recurrent Aβ insults. The interaction between CXCL14 and one of its putative receptors CXCR4 mediates neuronal protection. In summary, our study suggests that CXCL14 may serve as a potent chemokine by recruiting microglia to Aβ plaques to enhance Aβclearance and to prevent cell death in AD.

Disclosures: J. Galske: None. **J. Zhou:** None. **R. Yan:** None.

Poster

P218. Novel Models and Approaches to Study Alzheimer's

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P218.01

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: 2019-466-236 PMI-AD
European Regional Development Funds-European Union (ERDF-EU)
EULACH16/T010131
International Brain Research Organization (IBRO)

Title: Translational potential of synaptic dysfunction in Alzheimer`s disease patients and App knock-in mouse models

Authors: *D. MEDINA-VERA[1,2], D. ENACHE[3], E. ABUHASHISH[3], S. TAMBARO[3], C. ROSELL-VALLE[1], B. WINBLAD[3], F. RODRÍGUEZ DE FONSECA[1], E. BERECZKI[3], P. NILSSON[3];
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Abstract: Functional synapses are crucial for memory formation and learning processes. Synaptic dysfunction is an early biological mechanism in neurodegenerative diseases such as Alzheimer's Disease (AD). Previous post-mortem studies suggest that changes in synaptic proteins such as zinc transporter protein 3 (ZnT3), dynamin1 (Dyn1) and AMPA glutamate receptor 3 (GluA3) are associated with cognitive decline in AD. We aimed to evaluate the diagnostic value of ZnT3, Dyn1 and GluA3 in cerebrospinal fluid (CSF) of Mild cognitive impairment (MCI) and AD patients. In addition, we aimed to assess the translational potential of these synaptic proteins in two established App knock-in AD animal models by assessing the CSF, hippocampal and cortical synaptic protein concentrations within the same animal. ELISA immunoassay was used to measure the concentration of ZnT3, Dyn1 and GluA3 in CSF from a cohort of 54 MCI, AD and control patients, as well as in CSF and/or brain of 12 and 24 months old App^{NL-F}, App^{NL-G-F} and wild-type mice. Furthermore, age and brain regional- distribution (12 months and 24 months) and expression changes of these proteins were further explored in App knock-in models by immunofluorescence. We found a significant increase in ZnT3 and GluA3 concentrations both in MCI and AD patients as compared to control patients. Dyn1 concentration was significantly higher in AD patients only. MCI patients converting to AD exhibited more elevated CSF ZnT3 concentrations than stable MCI patients. CSF GluA3 concentration was significantly higher in App^{NL-G-F} knock-in mice as compared to wild-type controls. Furthermore, we have detected age and brain regional specific changes of the synaptic proteins in both App^{NL-F} and App^{NL-G-F} knock-in mice. Notably, the concentrations of all the three synaptic proteins correlated negatively with their concentrations in brain hippocampal lysates. We have also observed age dependent neuroinflammation and distinct accumulation of these synaptic proteins around amyloid-beta plaques. The pronounced synaptic dysfunction in MCI and AD patients reflected by increased CSF concentrations of synaptic proteins was a discerned characteristic of the App knock-in mouse models highlighting the translational potential of these AD mouse models. These translatable synaptic biomarkers could potentially help reducing the current disparities between human and animal models-based studies aiding the translation of preclinical discoveries of pathophysiological changes into clinical research.

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Poster

P218. Novel Models and Approaches to Study Alzheimer's

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P218.02

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Innovation and Technology Commission of the Hong Kong Special Administrative Region Government (ITS/168/19FP).

Title: Alternative treatment strategy and validation of a novel brain target for deep brain stimulation in neurodegenerative disease

Authors: *S. C. L. CHAN[1], G. KUMAR[1], C. H. E. MA[1,2];
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Abstract: Alzheimer's disease (AD) is the major cause of dementia worldwide. AD progression significantly affects mental and physical health of patients as well as families and resulting in a heavy financial burden to Public Health Services. AD patients with disease progression fails to respond conventional drug therapy after long-term medication, and therefore new therapeutic strategies has been explored. Deep brain stimulation (DBS) has been emerging as a potential alternative treatment for AD. Applying DBS to fornix (fornix-DBS) was found therapeutically effective in improvement of cognitive symptoms. However, significant cognitive impairments were reported in patients below 65 years old during phase II clinical trial. Moreover, no tangible evidence supported the long-term efficacy and delay of disease progression. Therefore, we proposed a novel brain target of DBS (novel target-DBS) in brain stem for AD treatment due to its strategic anatomical location and indirect projection to hippocampus. In our exploratory study, we used streptozotocin (STZ) induced and transgenic APP/PS1 (2xTg) mice model of AD to evaluate the therapeutic potential of DBS at novel area in the brain stem for amelioration of learning and memory. To assess the spatial and recognition memory, we conducted neurobehavioral tests, including Y-maze test, object recognition (NOR) test and Barnes maze test in the STZ and transgenic mice model of AD. All the neurobehavioral experiments were performed in a quiet room and observer was blinded to the mouse genotypes, surgery and treatment group. Our results showed that acute treatment (1 week) in STZ model and chronic treatment (3 weeks) in transgenic mice model significantly improved spatial and recognition memory as assessed by Y-maze, NOR test and Barnes maze test from day 7 or day 21 of the experimental paradigm of STZ or transgenic mice respectively. We also performed immunohistology of Nissl and BrdU staining for quantification of newly proliferated cell to evaluate the activity of neurogenesis and delineate the mechanism of DBS efficacy. Neurogenesis is one of the potential mechanisms of DBS for memory formation. We observed that the novel target-DBS significantly increased the number of Nissl positive and BrdU positive cells in the hippocampus. Interestingly, a wide migration of BrdU positive cells towards the vital nuclei of memory formation, suggesting the widespread therapeutic effect on neuronal circuit for enhancement of memory function. In conclusion, our novel target-DBS showed better

therapeutic efficacy than conventional DBS (fornix-DBS) to improve learning and memory in mice model of AD.

Disclosures: S.C.L. Chan: None. **G. Kumar:** None. **C.H.E. Ma:** None.

Poster

P218. Novel Models and Approaches to Study Alzheimer's

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P218.03

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Mechanisms of Mitochondrial Disruption by Amyloid Beta 42 Identified by Proteomics and Pathway Mapping

Authors: *P. L. SINCLAIR[1], T. SEPAHPOUR[2], A. BARANOVA[3], N. KABBANI[4]; [1]Interdisciplinary Program in Neurosci., George Mason Univ., Annandale, VA; [2]Biosci., [3]Sch. of Systems Biol., George Mason Univ., Fairfax, VA; [4]Mol. Neurosci, Krasnow Inst., Fairfax, VA

Abstract: Abstract
Alzheimer's Disease (AD) is marked by chronic neurodegeneration of the human brain that correlates with the occurrence of the pathogenic amyloid beta $A\beta_{42}$ peptide, the primary component of plaques. With limited symptom management and new drugs that target amyloid peptides a fundamental understanding of early cellular changes that occur as a result of amyloids. This problem is not unique to AD, but pertinent to other human neurodegenerative disorders such as Parkinson's and Lewy body dementia, which are also marked by amyloid protein pathology. Here we examined the effect of 3-day $A\beta_{42}$ exposure on differentiated PC12 cells using liquid-chromatography electrospray ionization mass spectrometry (LC-ESI MS/MS) coupled to protein identification and precursor ion peptide quantification. Gene set enrichment analysis (GSEA) of the resulting $A\beta_{42}$ proteome was conducted using Pathway Studio to identify differentially represented gene ontology (GO) pathways. Findings indicate a critical role for mitochondrial signaling, bioenergetics, and morphology in $A\beta_{42}$ cellular responses and provide potential new biomarkers for AD. Mitochondrial adaptation to $A\beta;_{42}$ mediated high calcium load within the matrix compartment may be achieved by stemming ER calcium transfer with an aid of FK-506 binding protein 1a (FKBP1a).

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Poster

P218. Novel Models and Approaches to Study Alzheimer's

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Program #/Poster #: P218.04

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Frontier Research unit (6401005490)
KMUTT Research Fund2020
KMUTT Financial support

Title: The simple and cost-efficient workflow of small molecule-induced amyloid-beta production in neuronal cell culture encapsulated in a three-dimensional agarose-laminin hydrogel scaffold

Authors: *P. NAMCHAIW[1], P. BUNREANGSRI[2];
[1]Biol. Engin. Program, Fac. of Engin., [2]Neurosci. Ctr. for Res. and Innovation, Learning Inst., King Mongkut's Univ. of Technol. Thonburi, Thung Kru, Thailand

Abstract: A 3D culture is now considered a relevant in vitro culture model for studying neurological diseases such as Alzheimer's disease (AD). In the past several years, hiPSC derived from familial AD (FAD-hiPSC) was used to generate an in vitro AD model. However, 90% of all AD cases are sporadic AD (SAD), which is not inherited genetic mutations. While the cause of the disease is still not fully understood, the amyloid-β accumulation has been thought to be the key event explaining the impairment of neurons and cognitive functions in both AD. Herein, we utilize Aftin-4, a small molecule β- and γ-secretase modulator to induce the amyloidogenic pathway in the neuronal-like cell encapsulated in agarose-laminin hydrogel scaffolds. We present here a small molecule-induced amyloidogenic condition in 3D neuronal culture workflow and provide the methods to isolate cells, RNAs, and proteins from a hydrogel scaffold. The isolated RNA and protein are suitable for real-time PCR and ELISA to study the expression profile in 3D neuronal culture. Besides the isolated cells from a hydrogel scaffold is suitable for flow cytometry analysis. Our results show that at 7 days in vitro, neuroblastoma cell lines become neuronal-like cells expressing TUJ1+. The real-time PCR analysis revealed the amyloid beta-linked genes expression *APP, BACE1, PS1,* and *PS2* in 3D culture. The secreted amyloid beta-40 and -42 (Aβ40, Aβ42) diffuse into an agarose-laminin hydrogel part and culture medium, which Aftin-4 specifically increases the level of Aβ42 but not Aβ40 in both parts. This increases Aβ42/Aβ40 ratios to 0.24-fold and 0.27-fold upon treatment when compared with vehicle control of 0.19-folds and 0.23-folds in cell culture medium and agarose-laminin hydrogel, respectively. The alteration of the Aβ42/Aβ40 ratio tightly correlates with a pathological hallmark of AD. The results presented here demonstrate the feasible and cost-efficient method to generate amyloidogenic induction in a 3D neuronal-like culture. Besides the isolated RNAs, proteins, and cells from the 3D scaffold would facilitate the drug screening and drug development to slow the progression or delay the onset of AD.

Disclosures: P. Namchaiw: None. **P. Bunreangsri:** None.

Poster

P218. Novel Models and Approaches to Study Alzheimer's

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Program #/Poster #: P218.05

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: 1RF1AG071587-01
R01AG061800

Title: Cellular proteomic profiling using proximity labeling by TurboID in microglial and neuronal cell lines

Authors: *S. SUNNA[1], S. RAYAPROLU[1], C. BOWEN[1], P. BAGCHI[1], N. T. SEYFRIED[2], S. RANGARAJU[1];
[1]Emory Univ., Atlanta, GA; [2]Biochem., Emory Sch. Med., Atlanta, GA

Abstract: Proximity labeling of proteins by promiscuous biotin ligases such as TurboID, coupled with mass spectrometry (MS) analyses of biotinylated proteins, can potentially resolve cell-type specific proteomes from complex tissues such as brain without cell type enrichment. Before *in vivo* applications, it is important to characterize the types of proteins preferentially labeled by TurboID in mammalian cells, and directly compare biotinylated proteomes with whole cell proteomes. We determined whether TurboID-labeled proteomes in microglial (BV2) and neuronal (N2A) cell lines are representative of total cellular proteomes under homeostatic and activated conditions. We generated N2A and BV2 cell lines that stably express TurboID fused to a nuclear export sequence, and treated cells with 200μM biotin and 1μg/mL lipopolysaccharide (LPS) for 48hrs to mimic inflammatory stress. Whole cell lysates (inputs) and streptavidin bead-enriched biotinylated proteins were used for western blot and label free quantitation MS (LFQ-MS). After confirming robust proteomic biotinylation in TurboID N2A and BV2 cell lines by western blot, LFQ-MS quantified >3,000 proteins in inputs and >2,400 proteins in biotinylated proteomes from both cell types. In BV2 and N2A inputs, TurboID expression and biotinylation did not impact the proteome, evidenced by very few differentially-expressed proteins and absence of cellular respiration measures in mitochondrial stress tests, comparing TurboID and non-TurboID cells. Of all the proteins quantified in BV2 and N2A TurboID inputs, over 50% were also quantified in the biotinylated proteomes in both cell types. As compared to input proteomes of both cell types, the biotinylated proteome was enriched in vesicular transport, ribosomal, mitochondrial, cytoplasmic and cytoskeletal proteins. The biotinylated proteome successfully differentiated BV2 and N2A cells, similar to the input proteomes. LPS stimulation altered levels of 488 proteins in BV2 cells in the input proteome, of which the top increased (e.g. Irg1, Oasl1, Cyb5a) and decreased (e.g. Mrc1, Mgl2, Arg1) proteins showed concordant changes in the biotinylated BV2 proteome. Our analyses show that

cellular proteomic labeling by TurboID in microglial and neuronal cell lines does not disrupt cellular function, and the resulting biotinylated proteomes are reasonably representative of the whole cell proteome and is able to capture the most robust alterations induced by immune stimuli in microglia. These represent foundational data to guide future applications of proximity-labeling for cell type specific proteomics *in vivo*.

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Poster

P218. Novel Models and Approaches to Study Alzheimer's

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Program #/Poster #: P218.06

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Brookhaven National Laboratory general user grant for National Synchrotron Light Source beamline access

Title: Relationships between potassium, copper, iron, and zinc in plaques in a mouse model for Alzheimer's disease

Authors: *P. KAKALEC[1], K. BOGGS[1], C. M. GROEBER TRAVIS[3], A. LANZIROTTI[4], J. M. FLINN[2];
[2]Psychology, [1]George Mason Univ., Fairfax, VA; [3]BTNN, Walter Reed Army Inst. of Res., Silver Spring, MD; [4]Univ. of Chicago, Chicago, IL

Abstract: One of the hallmarks of Alzheimer’s disease is the presence of amyloid plaques in the brain. While these plaques are primarily composed of amyloid, the presence of several trace metals has been detected within these plaques. Given the presence of these trace metals, it is probable that metal dyshomeostasis plays a role in the disease process and could indicate a mechanism by which the dietary intake of trace metals could affect disease progression. To better understand the role of dietary metal intake in Alzheimer’s disease pathology we administered waters supplemented with copper, iron, and copper + iron to CRND8 animals expressing the Indiana and Swedish mutation of the human APP gene, with transgenic animals administered lab water serving as a control. Following 5 months of supplementation, unfixed brain tissue was analyzed at Brookhaven National Laboratory to quantify the metal distribution in the tissue using synchrotron X-ray fluorescent microscopy. Fixing tissue results in the washing of potassium, thus our use of unfixed tissue allowed us to visualize the potassium content of plaques. These data were then used to model the relationships between potassium, iron, zinc, and copper, as well as the changes in plaque metal content observed in response to dietary metal supplementation. Results indicated that potassium content correlated positively with both iron

and zinc, but a relationship was not observed between iron and zinc. We also observed that the administration of copper- and iron-supplemented waters reduced the presence of potassium in the plaques relative to plaques in animals administered lab water ($p < 0.05$), and that the administration of copper water increased the zinc content of plaques relative to lab water ($p < 0.05$).

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Poster

P218. Novel Models and Approaches to Study Alzheimer's

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Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant RO1NS094201

Title: Brain regional proteomics reveals mechanistic insight to the progression of distinct pathologies in the rTg-DI rat model of cerebral amyloid angiopathy.

Authors: *J. M. SCHRADER, F. XU, W. E. VAN NOSTRAND;
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Abstract: Cerebral amyloid angiopathy (CAA), characterized by cerebral vascular amyloid accumulation, intracerebral hemorrhages and microbleeds, and cerebral infarction, is a prevalent comorbidity in Alzheimer’s disease (AD) and prominent contributor to vascular cognitive impairment and dementia (VCID). rTg-DI rats, a preclinical model of CAA, develop varying degrees of microvascular amyloid deposition and neuroinflammation in several brain regions, while thrombotic events of occluded vessels and microbleeds are mainly restricted to the thalamus. We employed liquid chromatography coupled with tandem mass spectrometry (LC-MS/MS) using a sequential window acquisition of all theoretical mass spectra (SWATH) method to report numerous brain regional specific proteomic changes in the cortex, hippocampus and thalamus of rTg-DI rats, followed by Ingenuity Pathway Analysis (IPA) to connect these changes with specific pathways and disease functions. IPA suggested TNFα activation, abnormal nervous system morphology, and neutrophil degranulation in all three regions, TGFβ activation in the hippocampus and thalamus, and thrombin activation in the thalamus. Proteomics analysis and immunostaining also revealed the presence of neutrophil extracellular traps (NETs) specifically

in the thalamus, a potentially important mechanism in the progression of the more severe vasculopathies observed in that region. Thus, we report new and significant insight linking specific proteomic changes to differential CAA-pathologies, and present potential mechanisms for the regionally distinct disease progression.

Disclosures: J.M. Schrader: None. **F. Xu:** None. **W.E. Van Nostrand:** None.

Poster

P218. Novel Models and Approaches to Study Alzheimer's

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P218.08

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Purdue University PIIN/PIDD Alzheimer's Award.

Title: Validation of PAL test in 3xTG and APP/PS1 transgenic AD mouse model

Authors: *T. SYEDA[1,2], S. ENKH-AMGALAN[3], M. HADDAD CORREA[4], J. M. BROWN[1,2], J. R. CANNON[1,2], W. A. KOSS[2];
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Abstract: Alzheimer's disease (AD) is characterized by the accumulation of amyloid-beta plaques, intracellular neurofibrillary tangles, and progressive cognitive decline. Despite the significant public health crisis that it poses, there is no clinically available disease-modifying therapy. The behavioral tasks used currently, such as the morris water maze, novel object recognition, and battery of spatial mazes in rodents, do not detect early mild cognitive deficits and do not detect the nuances of deficits seen in AD patients; this major translational weakness limits preclinical AD modeling, particularly evaluating early diagnostic markers of disease and efficacy testing of novel therapeutics. The Cambridge neuropsychological test automated battery (CANTAB) is one of the most popular tests to assess cognition in AD patients. Paired-associates learning (PAL) task, part of the CANTAB, requires the subject to learn to associate a given object with a particular location. A touchscreen automated object-in-place PAL task has been recently adapted for mice and rats. PAL touchscreen task has not been tested and validated in the most commonly used 3xTG and APP/PS1 transgenic AD mouse model. In the present study, we hypothesized that the PAL test effectively detects cognitive deficits in this model earlier than the traditional cognitive test. If the PAL test is found to be more effective than other traditional cognitive tests, we will further validate rodent cognitive tests with higher translational value to aid the AD studies and drug discovery. In the present study, we will validate the PAL test in Bussey-Saksida touchscreen chambers using two transgenic 3xTG (4 months old) and APP/PS1

(5.5 months old) AD mouse models. We will validate this test by comparing it with traditional cognitive tests (water maze, Y-maze, and object recognition test). Since cognitive deficits are more significant in females than males in both of these mouse strains, we will use female mice for the study. Preliminary data show that the behavioral performance of 3xTG performed poorer in the training; the average number of trials in the PAL test completed was significantly lower compared to C57BL6 mice. Other test will be done to analyze whether the non-performance in the PAL test is associated with motivation. Collectively, our preliminary data suggests that PAL can detect cognitive deficits sooner in 3xTG mice compared to the traditional cognitive test.

Disclosures: T. Syeda: None. **S. Enkh-Amgalan:** None. **M. Haddad Correa:** None. **J.M. Brown:** None. **J.R. Cannon:** None. **W.A. Koss:** None.

Poster

P218. Novel Models and Approaches to Study Alzheimer's

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P218.09

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: AMED Grant JP21dm0207065

Title: Generation of PSEN1 gene-manipulated common marmoset using the base-editor system

Authors: *W. KUMITA[1], H. SASAGURI[2], K. SATO[1], N. OURA[1], T. C. SAIDO[2], E. SASAKI[1];
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Abstract: Animal models are beneficial in elucidating the pathogenic mechanism of neurodegenerative diseases and in developing treatment methods and effective drugs. Rodent models have contributed substantially to advancements in neuroscience research. However, the brain structure differs significantly between rodents and humans, limiting the use of rodents as models of neurological disorders. The use of non-human primates that are neurologically similar to humans could be valuable to study psychiatric and neurological disorders. The common marmoset (*Callithrix jacchus*) is a small non-human primate for which a genetic modification method has been established. In 2016, we developed a marmoset model of X-linked severe combined immunodeficiency model generated by gene editing. The presenilin 1 gene (*PSEN1*) is associated with Alzheimer's disease (AD), and many *PSEN1* mutations have been reported in AD patients. Especially the P117L amino acid mutation of the *PSEN1* (*PSEN1*-P117L mutant) is found in early-onset familial AD patients. In this study, the *PSEN1*-P117L mutant was targeted for gene editing to develop a marmoset model of AD using the base-editor system, which can generate single-nucleotide changes in DNA or RNA without double-strand breaks. First, *PSEN1*-

P117 sequence-targeted guideRNA mRNA and base-editor mRNA were injected into 896 marmoset embryos with nine injection conditions. Injected embryos were analyzed, and efficient conditions were established to cause *PSEN1*-P117L modification. Next, 69 of the injected embryos were transferred into the uterus of surrogate female marmosets resulting in 4 neonates (4.7% of the transferred embryos). The sub-clones of the PCR products derived from the genome of hair root cells from each neonate were analyzed. The *PSEN1*-P117L modification was detected in all 4 neonates (100%). The *PSEN1*-P117L mutant marmosets are expected to be early-onset of the AD, and could be valuable in elucidating the peculiar mechanism of AD in primates.

Disclosures: W. Kumita: None. **H. Sasaguri:** None. **K. Sato:** None. **N. Oura:** None. **T.C. Saido:** None. **E. Sasaki:** None.

Poster

P233. Therapeutic Strategies: Preclinical Animal Models and Small Molecule Therapeutics III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P233.01

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Grant from National Research Foundation, Ministry of Science, ICT and future planning (2012R1A5A2A44671346)
Grant from Korea Healthcare Technology R&D Project, Ministry of Health and Welfare (A062260)
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Grant for Shaperon from Ministry of Health and Welfare (HI19C0429)

Title: Hy209 regulates p2x7r-mediated nlrp3 inflammasomal activation of microglia in mice with alzheimer’s diseases

Authors: *J. ISLAM[1,2,3], J.-A. CHO[1], J.-Y. KIM[1], K.-S. PARK[1], Y.-J. KOH[3], C. CHUNG[3], S.-Y. SEONG[1,4,2,3];
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Abstract: NLRP3 inflammasome (N3I) activation by P2X7R of microglia is crucial in neuroinflammation of Alzheimer’s disease (AD). Damage-associated molecular patterns such as Amyloid β (Aβ) and/or ATP activates N3I thru P2X7R pathway. Inhibitors for N3I have been

tested to alleviate progression of AD in animal models. Due to polymorphisms, subtypes, and ubiquitous expression of P2X7R and N3I components (NLRP3, ASC or pro-caspases etc.), clinical benefits have been controversial. G-protein coupled receptor 19 (GPCR19) is preferentially expressed on myeloid immune cells including microglia. For the first time, we found that GPCR19 co-localized with P2X7R on the membrane of microglia and regulates P2X7R-mediated N3I activation. HY209, a GPCR19 ligand suppressed IL-1β production in WT mice but not in GPCR19KO and P2X7RKO microglia. Ca++ mobilization of microglial in response to ATP or BzATP was diminished in GPCR19KO and P2X7RKO cells when compared with WT cells, suggesting GPCR19 is crucial for P2X7R mediated N3I activation which is essential for production of IL-1β/IL-18. HY209 also inhibited the priming phase of N3I activation via GPCR19-cAMP-PKA-NF-kB pathway. Further, HY209 suppressed expression of P2X7R, enhanced phagocytosis of Aβ, and decreased number of Aβ plaque in the brain of 5xFAD mice. HY209 also reduced microgliosis, prevented neuronal loss, and improved memory function of 5xFAD mice. Aβ and/or ATP enables the open state of the P2X7R on the cell membrane, thereby increases exchange of cytosolic and extracellular cations. The crystal structure of the P2X7 receptor displays clear differences between the open and closed pore state. Taken together with colocalization of GPCR19-P2X7R, HY209 might inhibit N3I activation by changing configuration of GPCR19 which hampers opening of P2X7R necessary for cation exchange. In summary, GPCR19 ligands might be a novel option for ameliorating neuroinflammation as Alzheimer’s disease to prevent neuronal loss which is crucial to preserve cognition and memory.

Disclosures: J. Islam: None. **J. Cho:** None. **J. Kim:** None. **K. Park:** None. **Y. Koh:** None. **C. Chung:** None. **S. Seong:** None.

Poster

P233. Therapeutic Strategies: Preclinical Animal Models and Small Molecule Therapeutics III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P233.02

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: University Grants Commission

Title: Apocynin a nootropic, neurotrophic and neuroprotective treatment modality for Alzheimer's disease

Authors: *N. CHIMTHANAWALA, S. SATHAYE, S. BAGLE;
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Abstract: Learning from daily life experiences, and their reconstruction in the form of memories, remain a crucial facet of our personality. This ability gets eroded in Alzheimer's disease (AD). A multifactorial disease with a complex etiology, AD presents extracellular amyloid beta (Aβ) deposits and intracellular hyperphosphorylated tau as hallmark features. Pathologically, immense oxidative stress, mitochondrial dysfunction, impaired mitochondrial biogenesis, altered oxidative phosphorylation system, perturbed calcium homeostasis, hampered adult neurogenesis, inflammation, etc. are observed. Progressive degenerative nature of the disease demands exploration of a molecule possessing multifaceted activity to offer a better quality of life. We evaluated the role of apocynin (phytoactive), an NADPH oxidase inhibitor, obtained from Picrorhiza kurroa, possessing anti-inflammatory and antioxidant activity, in the AD pathology in an $A\beta_{1-42}$ peptide-induced AD rodent model. Male Wistar rats were subjected to intrahippocampal $A\beta_{1-42}$ administration in N=12 per group using stereotaxy in the study. Further, per-oral treatment with 3 graded doses of apocynin for 28 days was done. Neurobehavioral activity was evaluated using Barnes maze test (BMT), Open field test (OFT). Biomarkers like anti-oxidants [Superoxide dismutase (SOD), reduced glutathione (GSH), Catalase], membrane peroxidation [Lipid peroxidation (LPO)], inflammatory parameter (TNF-α), immunohistochemistry for $A\beta_{1-42}$ peptide were measured. RT-PCR analysis for caspase-3, and RAGE (receptor for advanced glycation end-products) gene expression was performed. Histopathological staining using Congo red, Nissl and H&E was also carried out. Our data revealed that apocynin had a multidimensional role showing nootropic, neuroprotective, anti-inflammatory, and anti-oxidant effects. Our aim was to stop the overall progression of disease and explore the synaptogenic and neurogenic potential. We assessed the activity of apocynin on adult hippocampal neurogenesis [using BDNF, Ki-67, DCX (doublecortin), NeuN, synaptophysin, and ChAT (choline acetyltransferase) as markers] in the same AD model. Apocynin enhanced the proliferation and differentiation of the neuronal stem cells, instigating adult hippocampal neurogenesis. Lastly, apocynin restored the impaired mitochondrial biogenesis in the same model, modulating levels of PGC-1α (PPAR-$_\gamma$ coactivator) and Mt-CO1 (cytochrome c oxidase I) as the parameters expressed providing neuroprotective effect. We hence propose a novel treatment modality for AD that can be further upscaled for clinical studies as a disease modifying agent.

Disclosures: N. Chimthanawala: None. **S. Sathaye:** None. **S. Bagle:** None.

Poster

P233. Therapeutic Strategies: Preclinical Animal Models and Small Molecule Therapeutics III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P233.03

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: R01AG061114
RO1AG061114-01S1
R21AG06175
R21AG053876

Title: Evaluating the Effect of Angiotensin Receptor Blocker Treatment in Mice that Express Human APOE4

Authors: *S. B. SCHEINMAN, L. M. TAI;
Univ. of Illinois at Chicago Dept. of Anat. and Cell Biol., Chicago, IL

Abstract: Human *APOE4* is the greatest genetic risk factor for the development of Alzheimer's disease (AD), the most common form of dementia. In addition to earlier AD onset and more rapid cognitive decline in human *APOE4* carriers, *in vivo* data support that *APOE4* is associated with memory-relevant behavioral deficits, higher Aβ levels, and greater neuroinflammation as compared to *APOE3*. These same dysfunctions have also been linked to activation of the Angiotensin II Type 1 (AT1) receptor in the brain. As such, antagonism of the AT1 receptor could represent a potential therapeutic strategy for *APOE4*-associated AD risk and progression. In fact, Angiotensin receptor blockers (ARBs), a class of pharmaceuticals commonly used to treat hypertension, have been shown to have beneficial effects in both human AD patients and AD-relevant rodent models, although results are mixed. However, the effect of ARBs in the context of *APOE* genotype remains unknown and understanding this interaction could inform the effective development of ARBs for AD therapy. Therefore, we treated both male and female mice that express the human *APOE4* gene, either with (E4FAD+) or without (E4FAD-) high Aβ levels, with the ARB candesartan for a period of 4 months. We found that in female, but not male, E4FAD- and E4FAD+ mice, ARB treatment resulted in greater performance on the novel object recognition (NOR) task of memory-relevant behavior as well as higher levels of synaptophysin, SV2A, and SNAP25 in the hippocampus. Further, female E4FAD- and E4FAD+ mice treated with ARBs also had lower hippocampal GFAP levels (16-43%) and lower Iba1 levels (30-43%), the latter of which correlated with enhanced NOR performance. Overall, these results suggest that ARB treatment modulates behavior, synaptic protein levels, and hippocampal gliosis in female mice, but not male mice, that express human *APOE4*, most likely through an Aβ independent pathway as similar results were observed in both E4FAD- and E4FAD+ mice. Findings support the therapeutic potential of ARBs for the targeting of neuroinflammation in AD, in particular for female *APOE4* carriers.

Disclosures: S.B. Scheinman: None. **L.M. Tai:** None.

Poster

P233. Therapeutic Strategies: Preclinical Animal Models and Small Molecule Therapeutics III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P233.04

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: ADDF-TPEP-18-579974C
NIH NIA-R01 AG059799

Title: Inhibition of EV biogenesis reduces tau propagation in seeded tau model of Alzheimer's Disease

Authors: *B. BELL[1], C. TALLON[1], M. M. MALVANKAR[1], K. COLEMAN[1], T. JOE[1], A. SHARMA[1], E. EREN[2], K. RANGARAMANUJAM[1], D. KAPOGIANNIS[2], B. S. SLUSHER[1]; [1]Johns Hopkins Med. Institutions, Baltimore, MD; [2]NIH Natl. Inst. on Aging, Baltimore, MD

Abstract: Alzheimer's Disease (AD) is the most common form of dementia worldwide, yet no effective therapeutics are available despite extensive research. AD is characterized by progressive neurodegeneration and cognitive decline, as well as the accumulation of putatively pathological Amyloid-β (Aβ) and hyperphosphorylated Tau. Numerous clinical trials which reduced levels of aggregated Aβ plaques in patient brains were unsuccessful in alleviating cognitive symptoms. These results renewed focus on tau, which spreads throughout the brain along characteristic pathways and correlates strongly with the severity of disease progression. Recent evidence highlights the importance of extracellular vesicles (EVs) in enabling the transcellular transmission of pathological tau in the brain in a 'prion-like' manner and identified the inhibition of EV biogenesis via small-molecule inhibitors of nSMase2 as a potential avenue for clinical intervention. However, the presently available tool inhibitors are unsuitable for clinical development. To assess this, our lab performed a >350K high-throughput screen against human nSMase2 and identified two new inhibitor chemotypes. After substantial medicinal chemistry efforts on these hit clusters, two lead nSMase2 inhibitors were identified - PDDC (IC_{50}=300 nM) with excellent oral bioavailability (F%=87) and brain penetration (AUC_{brain}/AUC_{plasma}=0.6) and DPTIP (IC_{50}=30 nM) with poor oral availability and limited brain exposure. To circumvent the pharmacokinetic limitations of DPTIP, we conjugated it to a brain-penetrable hydroxyl dendrimer system and subsequently demonstrated CNS target engagement following oral dosing. To characterize the potential of these two inhibitors as AD therapeutics *in vivo*, we optimized a rapid seeded tau mouse model in which we stereotaxically injected an AAV vector containing mutant human tau (P301L) into the CA1 region of the left hippocampus in young WT mice (male and female), and quantified the degree of tau and phosphorylated tau species in the contralateral dentate gyrus (DG) region after 6 weeks. Mice were immediately dosed following injection with PDDC-containing chow or oral dendrimer-DPTIP (100mg/kg each), as well as matched vehicle controls. Both treatment paradigms significantly reduced pThr181 phosphorylated tau fluorescence intensity in the contralateral DG. Similarly, drug treatments suppressed tau-containing neuronally-derived EVs circulating in plasma compared to vehicle-treated controls. These results confirm the efficacy of nSMase2 inhibition at retarding tau propagation through EVs, and highlight these promising therapeutics as treatments for AD.

Disclosures: B. Bell: None. **C. Tallon:** None. **M.M. Malvankar:** None. **K. Coleman:** None. **T. Joe:** None. **A. Sharma:** None. **E. Eren:** None. **K. Rangaramanujam:** None. **D. Kapogiannis:** None. **B.S. Slusher:** None.

Poster

P233. Therapeutic Strategies: Preclinical Animal Models and Small Molecule Therapeutics III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P233.05

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: AG062817

Title: Small-molecule Shc Blockers as Novel Therapeutics for Alzheimer's Disease

Authors: *G. CORTOPASSI[1], A. TOMILOV[1], C. GARCIA[1], C. MONTGOMERY[1], D. KOVALSKYY[2], I. MAEZAWA[3], L.-W. JIN[3];
[1]Univ. of California, Davis, Davis, CA; [2]Resistomix, San Antonio, TX; [3]M.I.N.D. Inst. UC Davis, Davis, CA

Abstract: Background. The human Shc locus has recently been identified as extremely important in the conversion from Mild Cognitive Impairment (MCI) to full Alzheimer's Disease (AD) [PMID: 33172501], and Shc is ranked as the 12th most important locus in AD, ahead of BACE, to which substantial drug development effort has been expended [PMID: 30944912]. Shc is more highly expressed in Hippocampus of human AD cases vs. Controls [PMID: 12084708] and activated A-beta binds to and increases Shc activity and gene expression in cells, and Shc knockdown reduces A-beta toxicity [PMID: 30459314]. PSAPP mice with genetically reduced Shc reduction resist cognitive deficits, memory loss and age-related cerebrovascular dysfunction [PMID: 25012499] in an interesting way**.** These Shc-deficient PSAPP mice have improved memory and cognition and survival relative to PSAPP mice, *with the same amyloid burden* [PMID: 27431297]. Thus Shc reduction is neuroprotective, *downstream* of the toxic A-beta signal. Under MTA we have received small molecule Shc Blockers from Buto Corp, to identify their relative efficacy in cell and animal models of disease.**Method.** We tested the neuroprotective potency of Shc blockers to rescue N2A cells challenged with A-beta in 384-well plates. We also tested the anti-inflammatory potency of Shc blockers in human and mouse microglial cells challenged with pro-inflammatory A-beta or LPS and observe decreased inflammatory gene expression (IL-6, IL-10, and Nos). The impact of Shc Blockers on memory loss by Barnes Maze and Novel Object Recognition was also tested in the ApoE4 and PSAPP mouse models of AD is ongoing. **Results.** We observe significant protection of several Shc blockers from A-beta-dependent neurotoxicity to N2a cells. We observe significant protection of multiple Shc blockers from A-beta or LPS mediated microglial inflammation. We observe

protection from memory loss in the ApoE4 mouse model of AD in vivo by a molecule that protects N2a cells from A-beta toxicity and inflammation in vitro. **Conclusion.** These preclinical data suggest that small-molecule Shc blockers are a novel therapeutic strategy of potential utility for AD.

Disclosures: G. Cortopassi: None. **A. Tomilov:** None. **C. Garcia:** None. **C. Montgomery:** None. **D. Kovalskyy:** None. **I. Maezawa:** None. **L. Jin:** None.

Poster

P233. Therapeutic Strategies: Preclinical Animal Models and Small Molecule Therapeutics III

Location: Virtual Only

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Program #/Poster #: P233.06

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R01AG057767
NIH Grant R01AG061937
Dale and Deborah Smith Center for Alzheimer's Research and trEatment
Kenneth Stark Endowment
IDPH Grant 03282005H
Illinois Health Improvement Association

Title: Sex-based early intervention may be key to improving long-term cognitive outlook: evidence from mouse models of Alzheimer's disease

Authors: *E. R. HASCUP[1,2], S. A. MCFADDEN[1], M. F. COX[1], C. A. FINDLEY[1,2], S. PANDEY[1,2], M. R. PECK[1], L. A. ALEWELT[1], K. QUINN[1], D. MEDINA[3], R. STOCKWELL[3], Y. FANG[1], A. BARTKE[1,4,3], K. N. HASCUP[1,2,4];
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Abstract: Age is the predominate risk factor for developing neurodegenerative diseases, including Alzheimer's disease (AD). We have recently shown in mouse models of AD that changes in glutamatergic neurotransmission are altered as a function of age and disease progression, and that the glutamatergic system can be targeted prior to cognitive decline to positively influence cognitive outcome. However, the underlying mechanisms responsible for these changes are currently unknown, and may be influenced from additional factors both internal and external to the brain, such as glucose metabolism, body composition, cell senescence, and environmental factors. Their impact on disease progression and interventional strategies was assessed in AβPP/PS1 (transgenic) and $APP^{NL\text{-}F/NL\text{-}F}$ (knock-in) mouse models of

AD, and their shared genetic background controls, C57BL/6 mice. Separate groups of mice underwent a single early intervention strategy beginning at 2-6 months of age, including oral riluzole treatment (356 µM or vehicle control), visceral fat removal (VFR; or sham surgery control), oral senolytic treatment (100 mg/kg bw fisetin, 5 mg/kg bw dasatinib + 50 mg/kg bw quercertin (D+Q; or vehicle control), or altered environmental temperature (chronic housing at 30 or 23°C control). Sex was also probed as a biological variable in all experiments. At approximately 12 or 18 months of age, all mice underwent insulin and glucose tolerance tests (ITT and GTT, respectively) to assess glucose metabolism, body composition measures, and cognitive testing using the Morris water maze. Brain (hippocampus) and peripheral (white and brown adipose and hepatic) tissues were assed for markers of glutamatergic neurotransmission, cell senescence, and glucose metabolism. Data support sex differences in disease progression and response to intervention across all groups. Riluzole treatment resulted in improved long-term memory compared to control mice in males, but not females. VFR intervention altered insulin sensitivity in female AD mouse models, but not males; however, preliminary data do not support cognitive improvement in either sex. Senolytic treatment with D+Q reduced abdominal white adipose depots and improved long-term memory in female mouse models of AD, but not males. Chronically housing mice at 30°C improved insulin sensitivity and glucose tolerance in both sexes, but procognitive effects were only observed in male AD models. Observed sexual dimorphic effects may indicate differences in disease progression profiles, which supports a need for sex-based, disease-stage specific therapeutic strategies.

Disclosures: E.R. Hascup: None. **S.A. McFadden:** None. **M.F. Cox:** None. **C.A. Findley:** None. **S. Pandey:** None. **M.R. Peck:** None. **L.A. Alewelt:** None. **K. Quinn:** None. **D. Medina:** None. **R. Stockwell:** None. **Y. Fang:** None. **A. Bartke:** None. **K.N. Hascup:** None.

Poster

P233. Therapeutic Strategies: Preclinical Animal Models and Small Molecule Therapeutics III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P233.07

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Sigma-1 receptor is critical for UPR response and mitochondrial activity in larval zebrafish

Authors: *B. DELPRAT, L. CROUZIER, M. DENUS, E. M. RICHARD, A. TAVERNIER, C. DIEZ, N. CUBEDO, T. MAURICE;
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Abstract: The sigma-1 receptor (S1R) is a highly conserved transmembrane protein highly enriched in mitochondria-associated endoplasmic reticulum (ER) membranes (MAMs), where it

interacts with several partners involved in ER-mitochondria Ca^{2+} transfer and/or activation of the ER stress pathways. By stabilizing the conformation of inositol 1,2,4-trisphosphate receptor type 1, S1R enhances the efflux of Ca^{2+} from the ER into the mitochondria, thus improving mitochondria function. Very interestingly, S1R can be activated/inactivated by small molecules and accumulating preclinical data suggest that S1R agonists are protectants in neurodegenerative diseases. In addition, mutations in S1R led to juveniles ALS and distal Hereditary Motor Neuropathies (dHMN), underlying the critical role of S1R in motor neurons function and survival. Even if deficient S1R mice model shed light on its role on motor deficits and muscle weakness associated with denervation at the neuromuscular junction and loss of motor neurons in the spinal cord, the precise function of the protein is still not fully understood. In order to determine the physiological role of S1R, we characterized a new S1R deficient zebrafish line at 5 days post fertilization (dpf). We analyzed the impact of S1R deficiency on the visual, auditory and locomotor functions using the technology developed by Viewpoint. The *sigmar1*$^{+25/+25}$ deficient line showed impairments in its visual and/or locomotor functions compared to *sigmar1*^{WT}. Indeed, the locomotion of the larvae is increased in the light and dark phases of the visual motor response in *sigmar1*$^{+25/+25}$. In contrast, no deficit was observed in acoustic startle response. In addition, we demonstrated the critical role of S1R in the ER stress response and mitochondrial activity. By analyzing the unfolded protein response (UPR) genes using qPCR, we observed that loss of S1R leads to decreased levels of ire1, xbp1s, perk and atf4b (in physiological condition) and increased upregulated levels of bip, atf6, perk ire1, xbp1s and chop after tunicamycin treatment (stressed condition). Finally, using a Seahorse analyzer, we observed that S1R deficiency leads to a decrease in basal respiration, ATP-linked respiration and non-mitochondrial respiration. In conclusion, this model will be highly valuable to decipher S1R cellular functions in vivo and a useful tool to deeply analyze the physiopathological roles of S1R.

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Poster

P233. Therapeutic Strategies: Preclinical Animal Models and Small Molecule Therapeutics III

Location: Virtual Only

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Program #/Poster #: P233.08

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R56AG05755501
Hunter RISE grant #2R25GM060665-21

Title: Inhibition of histone deacetylase mitigates spatial memory performance deficits and disease associated neuroinflammation in an Alzheimer’s disease transgenic rat model

Authors: *K. NDUKWE[1,2], L. XIE[4], P. SERRANO[3], P. ROCKWELL[2], M. E. FIGUEIREDO-PEREIRA[2];
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Abstract: Alzheimer's disease (AD) is one of the most common causes of dementia. In the United States, AD affects about 5.8 million Americans and is projected to reach 13.8 million individuals by 2050. Epigenetic alterations such as histone-modifications play a role in memory function, and disruption in the epigenetic processes is linked to the pathogenesis of neurodegenerative and neuropsychiatric diseases. Currently, there are no effective therapies for AD, and therapies targeting epigenetic mechanisms such as pharmacologic inhibitors of histone deacetylases (HDACs), are effective in improving cognitive performance in animal models of AD. The effects of histone deacetylase inhibitors on AD pathology and brain immune resident cells and neurons in an AD brain are not well understood. In this study, we treated Fisher transgenic 344-AD (TgF344-AD) rats, a model of AD, with the brain-penetrant inhibitor of histone deacetylase RG2833 for 5 months starting at the age of 6 months, and evaluated the rats for spatial memory performance and AD pathology. Using an active place avoidance test we identified a significant hippocampal-dependent spatial memory performance deficit in TgF344-AD rats compared to wild type littermates at 11 months of age. Notably, this deficit was mitigated in the RG2833 transgenic treated rats. Furthermore, RNAseq analysis of hippocampal tissue from TgF344-AD rats, revealed that treatment with RG2833 upregulated the expression of immediate early genes Arc and c-Fos, which are involved in synaptic plasticity and memory consolidation. We also found that RG2833 treatment failed to decrease amyloid beta or tau paired helical filament accumulation. However, RG2833 treated TgF344-AD rats exhibited a decrease in gliosis manifested by fewer amoeboid and ramified microglia in the hippocampal CA1 region compared to the untreated TgF344-AD littermate controls. These data indicate that histone modifying therapies can improve cognitive behavior by improving the expression of neuroprotective genes and by modulating the activation of immune cells and possibly dampening inflammatory response. Based on our data, we propose that RG2833 could be an effective therapeutic to treat disorders such as AD.

Disclosures: K. Ndukwe: None. **L. Xie:** None. **P. Serrano:** None. **P. Rockwell:** None. **M.E. Figueiredo-Pereira:** None.

Poster

P233. Therapeutic Strategies: Preclinical Animal Models and Small Molecule Therapeutics III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P233.09

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIA Grant RF1AG064859

Title: Inhibition of p38 alpha MAPK signaling provides benefit in a mouse model of mixed dementia

Authors: *D. J. BRAUN[1], M. J. COLEMAN[1], V. A. DAVIS[1], S. M. ROY[2], D. M. WATTERSON[3], L. J. VAN ELDIK[1];
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Abstract: Background: The p38α MAPK signaling pathway is a well-established regulator of neuroinflammation that is aberrantly activated in Alzheimer's disease (AD). However, there is a lack of insight into the p38α MAPK signaling pathway involvement in non-AD comorbidities found in roughly 80% of probable AD patients. The most common of these co-morbidities is vascular pathology. Therefore, we explored the potential pharmacodynamic effects of MW150, a p38α MAPK isoform selective inhibitor currently in clinical trials, on neuroinflammation in a co-morbid environment with both AD and vascular pathology. To this end, we used a mouse mixed dementia (MD) model that combines amyloid pathology with vascular pathology induced by hyperhomocysteinemia (HHcy). **Method:** The 5xFAD mouse model of amyloid pathology (MMRRC No. 34848-JAX) was used. 5-month-old animals were placed for 8 weeks on a B-vitamin deficient and methionine-supplemented diet (Envigo TD.97345) to induce HHcy and generate the MD model. In a preventative treatment paradigm (treatment before vascular pathology is present), the MD mice were given intraperitoneal injections of saline vehicle or varying doses of MW150. For baseline comparisons, WT littermates on a control diet and administered saline were included. At the end of the 8 weeks, mice underwent testing in the Morris Water Maze (MWM) at the Jackson Laboratory or were sacrificed in-house for pathological analyses, including measurement of pro-inflammatory cytokines and changes in various RNA transcript levels. **Results:** In the MD mice, MW150 was able to rescue deficits in the MWM task, prevent proinflammatory cytokine IL-1β overproduction, and reduce several markers of microglial activation in a manner consistent with its known anti-inflammatory activity in other models. **Conclusions:** These data indicate that early intervention with MW150 might be an efficacious strategy in the context of co-morbid amyloid and vascular pathologies. Work to test the efficacy of MW150 when administered in a therapeutic as opposed to preventative paradigm is currently ongoing.

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diversified mutual funds); Inventor on patent covering MW150, Scientific Founder of ImmunoChem Therapeutics, LLC, formed to commercialize MW150 and related compounds.

Poster

P233. Therapeutic Strategies: Preclinical Animal Models and Small Molecule Therapeutics III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P233.10

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Funding was provided by charitable contributions raised by Regions Hospital Foundation including a generous gift from the Sexton Family Foundation.

Title: Low-dose Intranasal Deferoxamine modulates memory, neuroinflammation, and the transcriptome in a rodent model of Alzheimer Disease

Authors: *J. KOSYAKOVSKY[1], J. M. FINE[2], W. H. FREY, II[2], L. R. HANSON[2]; [1]Sch. of Med., Univ. of Virginia, Charlottesville, VA; [2]Alzheimer's Res. Ctr., HealthPartners Neurosci. Ctr., Saint Paul, MN

Abstract: Intranasal (IN) Deferoxamine (DFO) has emerged as a remarkably promising candidate therapeutic across neurodegenerative and cerebrovascular disease. As an iron chelator, its mechanisms are multimodal and involve the binding of brain iron with the consequent engagement of several disease-nonspecific pathways. We and other research groups have shown that IN DFO rescues cognitive impairment in several rodent models of Alzheimer Disease (AD). We designed the present study in an effort to ease the translation of IN DFO to clinical trials, probing dosing regimes and mechanisms within the inducible intracerebroventricular (ICV) streptozotocin (STZ) rat model of AD. We found that a 1% solution of IN DFO rescued cognitive impairment caused by ICV STZ administration as measured by the Morris Water Maze (MWM) test. IN DFO rescued STZ-induced hippocampal neuron loss and decreased hippocampal apoptosis. Furthermore, IN DFO modulated several aspects of the neuroinflammatory milieu of the ICV STZ model which we assessed through a novel panel of brain cytokines and immunohistochemistry. Using RNA-sequencing and pathway analysis, we found that STZ induced several pathways of cell death and neuroinflammation, and that IN DFO engaged multiple transcriptomic pathways involved in hippocampal neuronal survival. In sum, to our knowledge this study is the first to assess the transcriptomic pathways and mechanisms associated with either the ICV STZ model or DFO treatment using RNA-sequencing and the first to demonstrate this therapeutic's efficacy at low doses. Additionally, we significantly elucidate the mechanisms and role of the ICV-STZ rodent model of AD.

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Poster

P245. Biomarkers in Neurodegenerative Diseases III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P245.01

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: JHU PMAP Initiative
NIH R21 AG070404

Title: Identification of alzheimer's disease subtypes using clinical structural MRI and EMR data

Authors: *H. P. COWLEY[1], M. S. ROBINETTE[1], J.-M. S. LEOUTSAKOS[2], A. MATHUR[3], P. P. ZANDI[5], M. A. NOWRANGI[2], K. OISHI[4];
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Abstract: A major challenge in effectively treating Alzheimer's Disease (AD) is the presence of disease subtypes with highly heterogeneous underlying biology. Identifying AD subtypes is critical for advancing patient care and subtypes related to cognitive and neuropsychiatric characteristics have been identified using data- and hypothesis-driven clustering approaches. Current approaches to subgroup identification, however, are based on research-quality neuroimaging limiting the feasibility, generalizability, and impact on decision-making in the clinical space. In this work, we process clinical structural brain MRI data via a high-throughput image parcellation algorithm and cluster the resulting parcellations along with readily available data from the electronic medical record. Our methods and results provide a connection between the identification of AD subtypes via neuroimaging and clinical data. Additional clinical features may be added to contextualize and operationalize meaningful AD subtypes towards informing and improving clinical care.

Disclosures: H.P. Cowley: None. **M.S. Robinette:** None. **J.S. Leoutsakos:** None. **A. Mathur:** None. **P.P. Zandi:** None. **M.A. Nowrangi:** None. **K. Oishi:** None.

Poster

P245. Biomarkers in Neurodegenerative Diseases III

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Program #/Poster #: P245.02

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Duke Department of Anesthesiology
International Anesthesia Research Society (IARS) Mentored Research Award (MB)
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NIH P30-AG028716 (MB)
K76-AG057022 (MB)

Title: Subgroup classification of delirium and CSF amyloid beta and tau status in relationship to preoperative and postoperative cognitive decline

Authors: *M. REESE[1], M. BERGER[1,2,5], J. N. BROWNDYKE[3,2,5,6], M. COOTER[1], W. M. BULLOCK[1], M. J. DEVINNEY[1], E. W. MORETTI[1], D. T. LASKOWITZ[1,1], L. SHAW[7], H. E. WHITSON[4,2], H. J. COHEN[4,2], J. P. MATHEW[1];
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Abstract: Introduction
Patients who experience postoperative delirium and those who meet ATN criteria established by the National Institute on Aging Alzheimer's Association are at increased risk of developing Alzheimer's disease and related dementias (ADRD). It is unclear whether delirium and ATN status are dissociable in their neurophysiological mechanisms, particularly in risk of postoperative cognitive decline associated with preclinical ADRD. Thus, existing data from preoperative and postoperative cerebral spinal fluid (CSF) AD biomarkers, the Confusion Assessment Method (CAM), and a neurocognitive test battery were used to characterize relationships between ATN status, delirium, and cognitive outcomes before to 6 weeks after surgery.
Method
Screening included 1,690 Duke University Medical Center English-speaking patients age ≥60 and scheduled for non-cardiac, non-neurologic surgery with a planned hospital stay of ≥1 night. 110 patients met inclusion criteria and completed baseline through 6-week CSF testing, a neurocognitive battery factor analyzed into 4 domains alongside the Mini-Mental State Exam

(MMSE), and CAM. ATN subgroups were identified at baseline using established immunoassay thresholds (Aβ <249 pg/ml or tau >93 pg/ml). The p-tau cutoff (>23 pg/ml) has low specificity for our AlzBio3 platform (N=66 p-tau+); thus, p-tau was excluded from subgrouping. Delirium+ included a positive CAM score or delirium chart review within 5 days post-surgery. Analyses included t-tests, ANOVAs, and non-parametric equivalents to study the association between subgroups and cognition from baseline to 6 weeks post-surgery.

Results

Delirium+ classification (N=13) was associated with worse cognition than delirium- (N=97) (baseline p=.030; 6-weeks post-surgery p=.020), particularly for visual memory (baseline p=.018; 6-weeks p=.015) and MMSE scores (baseline p=.088; 6-weeks p=.016). Baseline Aβ+ or tau+ patients (N=18) generally had a lower cognitive index (baseline p=.072; 6-weeks p=.021) and less visual memory (baseline p=.009; 6-weeks p=.002) than Aβ- and tau- patients (N=83). Delirium had no significant association with CSF Aβ, tau, or p-tau levels at any time point, and baseline CSF Aβ and tau status had no significant association with delirium incidence.

Conclusion

Exploratory analysis of subgroups differentiated CSF Aβ and tau from delirium, showing a 3 patient overlap and no significant association between baseline AD biomarkers or delirium status. Findings may suggest distinct underlying mechanisms for ATN and delirium despite similar patterns of cognitive impairment, which is an important consideration for future ADRD research.

Disclosures: M. Reese: None. **M. Berger:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Received EEG monitors for a post-operative recovery study in older adults from Masimo. F. Consulting Fees (e.g., advisory boards); Legal consulting fees related to post-operative cognition in an older adult. **J.N. Browndyke:** Other; funding from Claret Medical, Inc.. **M. Cooter:** None. **W.M. Bullock:** None. **M.J. Devinney:** None. **E.W. Moretti:** None. **D.T. Laskowitz:** Other; Officer of AegisCN, which is developing the ApoE mimetic peptide CN-105 for clinical use. **L. Shaw:** None. **H.E. Whitson:** None. **H.J. Cohen:** None. **J.P. Mathew:** None.

Poster

P245. Biomarkers in Neurodegenerative Diseases III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P245.03

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIA Grant R01 AG062306
BrightFocus Foundation
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Title: Identification and characterization of endogenous neuropeptide proteoforms, including VGF, in dementia pathophysiology

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Dept. of Neurol., Massachusetts Gen. Hosp., Boston, MA

Abstract: Neuropeptides are neurotransmitter-like molecules with multiple roles in neuronal activity that are processed into functional peptides (aka proteoforms) by prohormone convertases and as yet unidentified proteases. The neuropeptide neurosecretory protein VGF was the most nominated potential therapeutic target for Alzheimer's disease (AD) in a survey by the Accelerating Medicines Partnership-AD (AMP-AD), due to its levels being decreased in cerebrospinal fluid (CSF) and brain from patients with AD. Neuropeptides and their proteoforms remain understudied in their physiological processing pathways, their contribution to dementia pathogenesis and their potential use as a biomarker of dementia. Using mass-spectrometry and recombinant proteolysis assays we aimed to; 1) validate if specific neuropeptide proteoforms represent CSF biomarkers of synaptic health in dementia and 2) investigate their proteolytic processing. CSF was collected from 50 patients with AD, 64 with mild cognitive impairment (MCI), 18 who were cognitively resilient to AD pathology and 45 age-matched cognitively unimpaired controls. CSF samples were tandem mass tag (TMT)-labeled and pooled into thirteen 16-plex experiments. Pooled samples were size-filtered to eliminate peptides larger than 30kDa, and analyzed using a data-dependent acquisition workflow with a previously defined inclusion list of 241 endogenous non-tryptic neuropeptide proteoforms. Neuropeptide proteoforms from CHGA, CHGB, SCG2, SCG3, SCG5 and VGF were robustly detected (replicate coefficient of variation from 0.2-6.5%) from pooled samples run on every plex. Of those detected with less than 35% missing values across all samples, a majority were significantly associated with age, which was included as a co-variate in the linear modeling. 42 unique proteoforms detected had significant differential abundance between diagnostic groups, mainly between controls and AD. 7 VGF peptides and 5 SCG1 peptides were significantly different between AD and MCI while 9 VGF and 6 SCG1 peptides were significantly different between resilient subjects and AD. Using Proteasix, the MEROPS and human protein atlas database, 22 neuronally expressed proteases previously linked to dementia were identified that may cleave neuropeptides. Assays using recombinant proteins identified that ADAMTS4, calpain-1, cathepsin S, granzyme A and MMP-3 cleaved neuropeptides into N-terminal, mid-region, and C-terminal neuropeptide proteoforms. Overall, this work provides mechanistic insight into neuropeptide processing and how neuropeptide proteoforms could represent a novel biomarker for synaptic health in dementia.

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Poster

P245. Biomarkers in Neurodegenerative Diseases III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P245.04

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Associations between psychophysiological stress responses, inflammatory markers, and CSF biomarkers in the spectrum of Alzheimer's Disease

Authors: *H. OH[1], H.-C. LEUNG[3], T. CANLI[4], E. PARK[2];
[1]Psychiatry and Human Behavior, [2]Brown Univ., Providence, RI; [3]SUNY Stony Brook, Stony Brook, NY; [4]Dept Psychol, Stony Brook Univ. Dept. of Psychology, Stony Brook, NY

Abstract: Background: Although genetic factors associated with the aggregation of misfolded proteins clearly play a role in Alzheimer's disease (AD), several individual difference factors have been seen to confer an increased risk for the clinical manifestation of AD as well as the development of AD pathology. One of the key factors is an individual's susceptibility to psychological stress. The goal of this study was to examine the relationships between psychophysiological stress responses measured by plasma stress hormones and neuropsychiatric symptoms, inflammatory markers, and AD pathology along the spectrum of AD.**Method:** Alzheimer's Disease Neuroimaging Initiative (ADNI) consists of 819 adult subjects— 229 cognitively normal older adults (CNs), 398 participants with mild cognitive impairment (MCIs), and 192 patients with AD. Of these subjects, we included 58 CNs, 396 MCIs, and 112 AD patients whose plasma stress measures were available. Plasma cortisol and cytokines and CSF Aβ1-42, t-tau, and p-tau181 data were downloaded from ADNI. Neuropsychiatric Inventory (NPI-Q) total scores were used to indicate a psychological stress level.**Result:** AD stages were significantly related to the level of plasma cortisol and NPI-Q total scores showing that AD patients exhibit the highest level of cortisol and greater severity of psychological symptoms followed by MCIs and CNs. AD stages were also significantly related to the level of inflammatory markers measured by cytokines, although the level of inflammatory markers in relation to the diagnostic groups varied across different types of cytokines. Plasma cortisol levels did not relate to concurrent levels of CSF Aβ1-42 either across diagnostic groups or within diagnostic groups. Higher plasma cortisol, however, corresponded to higher CSF p-tau among CNs, but not in MCIs and ADs.**Conclusion:** Psychophysiological stress responses and inflammatory markers are significantly associated with the disease severity. A positive relationship between plasma cortisol level and CSF p-tau in CNs may indicate a role of individuals' susceptibility to stress in neuronal hyperexcitability in the early stage of AD and confer a risk of neurodegeneration and the development of AD. Alleviating stress and regulating stress responses may be important modifiable factors that help prevent AD.

Disclosures: H. Oh: A. Employment/Salary (full or part-time):; Brown University. **H. Leung:** None. **T. Canli:** None. **E. Park:** None.

Poster

P245. Biomarkers in Neurodegenerative Diseases III

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Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P245.05

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NMRC/CSA-SI/007/2016
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Title: Extracellular vesicle enriched miRNA-biomarkers show increased predictive potential for alzheimer's dementia and medial temporal atrophy

Authors: Y. CHAI[1], L. STROHM[1], Y. ZHU[1], R. CHIA[1], J. CHONG[1], L. ZHOU[1], H. TOO[1], M. LAI[1], E. KOO[1], C. CHEN[1], ***G. H. POPLAWSKI**[2];
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Abstract: "Liquid biopsies" have emerged as a new technique in the field of biomarker discovery. These non-invasive techniques, utilizing blood-derived extracellular vesicles (EVs), are especially promising in diagnosing dementia and neurodegeneration where brain biopsies are not practical. Investigation of blood EV-enriched biomarkers harbour the potential advantage of protecting miRNAs from degradation and could such increase the specificity of miRNA-biomarkers compared to serum-derived miRNAs. Here we show that indeed miRNAs isolated from blood-derived EVs are more predictive of Alzheimer's Disease as well as its neuropathological feature (namely medial temporal atrophy) compared to those isolated from serum samples. We further identified several novel miRNAs in AD patient derived EVs that have not been associated with AD beforehand. These data support the further utilization of EV-biomarker discovery for neurodegenerative diseases and provide potentially new biomarkers for the diagnosis of Alzheimer's Disease.

Disclosures: Y. Chai: None. **L. Strohm:** None. **Y. Zhu:** None. **R. Chia:** None. **J. Chong:** None. **L. Zhou:** None. **H. Too:** None. **M. Lai:** None. **E. Koo:** None. **C. Chen:** None. **G.H. Poplawski:** None.

Poster

P245. Biomarkers in Neurodegenerative Diseases III

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Program #/Poster #: P245.06

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: R01 AG055449
R01 AG068055
TL1 TR001997

Title: Enlarged perivascular spaces in the centrum semiovale predict MoCA scores among cognitively normal older adults

Authors: *T. J. LIBECAP[1], B. RODOLPHO[1], F. D. RASLAU[2], B. T. GOLD[1];
[1]Neurosci., [2]Radiology, Univ. of Kentucky, Lexington, KY

Abstract: Cerebral small vessel disease (cSVD) is an important risk factor leading to the development of vascular contributions to cognitive impairment and dementia (VCID). cSVD is characterized by several in-vivo neuroimaging biomarkers including enlarged perivascular spaces (ePVS). PVS are fluid-filled spaces believed to play a role in the glymphatic system's removal of waste from the brain. Reduced clearance may cause backup and enlargement of the PVS and subsequent accumulation of toxic solutes characteristic of neurodegeneration, including Aβ. Evidence supports ePVS role in aging and dementia, but the relationship between ePVS and cognitive function remains unclear**.** Due to their presence in brain regions that support cognition, we hypothesized that quantitative, cross-sectional ePVS counts in cognitively normal older adults would predict baseline cognitive performance. We explored the relationship between ePVS and the Montreal Cognitive Assessment (MoCA) in 71 cognitively normal older adults ranging in age from 60-86. Participants were scanned on a 3T Siemens Prisma scanner with a 64-channel head coil. All ePVS counts were performed on T1 MPRAGE and T2 FLAIR images by an experienced rater blinded to participant demographics and under the direction of a trained neuroradiologist. In line with previously established guidelines, ePVS were defined as regions of hypointensity less than 3mm in diameter on T1 imaging and were distinguished from lacunar infarcts by the absence of T2 FLAIR hyperintensity. ePVS were individually and manually counted in a single, representative slice in the axial plane of the white matter centrum semiovale (CSePVS), basal ganglia, hippocampus, and midbrain. Linear regression analyses controlling for age, sex, intracranial volume, and education demonstrated a significant, negative relationship between total ePVS counts combined across the four regions of interest and MoCA score ($\beta = -0.282$, $P = 0.022$). The strongest relationship was found among CSePVS and MoCA ($\beta = -0.337$, $P = 0.005$). These findings suggest that ePVS burden, driven primarily by CSePVS, is associated with cognitive performance at baseline. Ultimately, our results support the continued investigation of ePVS as an early neuroimaging biomarker of cSVD-related cognitive dysfunction. In particular, additional work that addresses the longitudinal relationship between ePVS and MoCA is needed to strengthen the findings that ePVS predict cognitive dysfunction.

Disclosures: T.J. Libecap: None. **B. Rodolpho:** None. **F.D. Raslau:** None. **B.T. Gold:** None.

Poster

P245. Biomarkers in Neurodegenerative Diseases III

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Program #/Poster #: P245.07

Topic: C.02. Alzheimer's Disease and Other Dementias

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the Collaborative Research Fund [C6027-19GF])
the Areas of Excellence Scheme of the University Grants Committee (AoE/M-604/16)
the Innovation and Technology Commission (ITCPD/17-9)

Title: Transcriptome analysis for AD-associated blood signatures in Hong Kong Chinese population

Authors: *H. ZHONG[1,2], X. ZHOU[1,2,3], Y. JIANG[1], Y. CHEN[1,3,4], N. LAI[1], E.-S. TONG[1], R. LO[1], T. KWOK[5], V. MOK[6], F. IP[1,2,3], K. MOK[1,3,7], J. HARDY[2,7,8], A. FU[1,2,3], N. IP[1,2,3];
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Abstract: Blood transcriptome has emerged as a powerful resource for studies of human diseases. However, the changes of blood transcriptome in patients with Alzheimer’s disease (AD) have not been well studied. We undertook transcriptome profiling of 422 participants 208 normal controls and 214 patients with AD) from the Hong Kong Chinese population. WGCNA (weighted correlation network analysis) was performed to construct co-expression network to identify modules that associated with AD. Co-expression modules are significantly associated with AD and plasma biomarkers of AD (i.e. Aβ and NFL). Pathway enrichment analysis revealed their possible involvement in specific biological pathways. Motif analysis identified potential mechanisms that drive the transcriptomic change in AD blood. Co-expression network analysis of AD blood transcriptome results in a better understanding on the disease mechanisms, as well as identification of potential targets for disease intervention and monitoring.

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Poster

P274. Huntington's Disease: Molecular Mechanisms II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P274.01

Topic: C.04. Movement Disorders other than Parkinson's Disease

Support: NIH R01NS110694
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Biomedical Research Awards for Interdisciplinary New Science (BRAINS)
NIH R21NS11626

Title: Striatal compartmentalization and clustering of different subtypes of astrocytes is altered in Huntington's disease

Authors: *T. BROWN[1], M. THAYER[2], N. ZARATE[1], R. GOMEZ-PASTOR[2];
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Abstract: Huntington's disease (HD) is a devastating neurodegenerative disease that manifests as motor, cognitive, and psychiatric impairments. HD preferentially affects medium spiny neurons (MSNs) of the striatum, but it also causes astrogliosis, which is defined by an increased density of astrocytes. Striatal astrocytes are not homogeneous, and they can be classified into different subpopulations characterized by the expression of different markers including glutamine synthetase (GS), glial fibrillary acidic protein (GFAP), and S100 calcium-binding protein beta (S100β). It has been suggested that these subpopulations have different functions and could differentially contribute to HD. We conducted a comparative analysis of the abundance of different subpopulations of astrocytes (GFAP, GS and S100β) in the heterozygous zQ175 HD mouse model. Our data using 2D fluorescent imaging showed increased marker expression and abundance of all three subpopulations in the striatum of zQ175 mice. To analyze whether there is a differential spatial distribution of any of these subpopulations, we developed an unbiased brain compartmentalization approach and found that while GS and S100β are more homogeneously distributed throughout the striatum, GFAP+ astrocyte accumulation is specifically enhanced in the dorsomedial striatum of HD mice, a region involved in integration of heterogeneous inputs from both thalamic and cortical subregions. In addition, GFAP+ astrocytes showed increased clustering in HD mice compared to wildtype, a parameter that may indicate areas of highly localized secretion of inflammatory molecules. Our data suggest a distinct striatal compartmentalization of different populations of astrocytes in HD that could

contribute to striatal dysfunction and behavioral deficits. These results have major implications for our understanding of functional compartmentalization of the striatum and its relevance in contributing to the different HD symptoms.

Disclosures: T. Brown: None. **M. Thayer:** None. **N. Zarate:** None. **R. Gomez-Pastor:** None.

Poster

P274. Huntington's Disease: Molecular Mechanisms II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P274.02

Topic: C.04. Movement Disorders other than Parkinson's Disease

Support: CHDI Foundation
Nebraska Tobacco Settlement Biomedical Research Development Funds LB692

Title: Genome-wide transcriptome profiling in striatum of HD mice exposed to a super-enriched environment

Authors: *H. KIM[1], S. H. WEE[1], J.-C. CHAI[2], T. YOO[1], R. ZUKIN[3], J.-Y. HWANG[1];
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Abstract: Huntington’s disease (HD) is an autosomal dominant inherited neurodegenerative disorder characterized by progressive motor, psychiatric and cognitive decline. Marked neuronal loss in the striatum and cerebral cortical structures is the main neuropathological hallmark of HD. Environmental factors can alter gene expression through epigenetic responses such as DNA methylation and histone modification, which are critical to genome reprogramming during brain development, tissue-specific gene expression, and global gene silencing under physiological and pathological conditions. An enriched environment has been shown to delay the onset and progression of motor symptoms and to improve neurological function and cognitive performance in animal models of HD. However, the impact of an enriched environment on transcriptional changes in the R6/2 transgenic mouse model of HD is, as yet, unclear. To address this issue, we performed genome-wide transcriptome analysis using next generation RNA-seq in striatal tissue from HD and wild-type mice reared in either a stark or super-enriched environment. Volcano plots demonstrate that an enriched environment has a very strong influence on gene expression changes in both HD and WT mice. The differentially expressed genes were analyzed by the Ingenuity Pathway Analysis software. Canonical pathway analysis reveals that autophagy related pathways such as elF2 signaling, mTOR signaling, and regulation of elF4 and p70S6K signaling are activated and mitochondrial dysfunction is inhibited in HD mice exposed to the super-enriched environment in comparison to HD mice reared in the normal environment. We

anticipate that the identification of genes and gene networks that are altered in HD mice in response to enriched environments will assist us in understanding how enriched environments can improve HD symptoms and neurological function and will contribute to the development of novel therapeutic strategies to ameliorate motor and cognitive deficits associated with this debilitating and devastating disease.

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Poster

P274. Huntington's Disease: Molecular Mechanisms II

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Program #/Poster #: P274.03

Topic: C.04. Movement Disorders other than Parkinson's Disease

Support: NIH Grant R01NS110694
American Cancer Society (ACS) 129819-IRG-16-189-58-IRG-107

Title: Tumor suppressor p53 regulates Heat Shock Factor 1 protein stabilization in Huntington's disease

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Abstract: In contrast to cancer, increased levels of tumor suppressor p53 have been reported in the brains of patients with Huntington’s disease (HD), a neurodegenerative disorder caused by a polyglutamine expansion in the Htt gene (mtHTT). It is known that elevated p53 plays a detrimental role in HD with evidence demonstrating that a systemic lack of p53 provides neuroprotection in a mouse model of HD. However, the molecular mechanisms that regulate p53 levels in HD and the cellular processes affected by p53 upregulation remain unknown. We have found that mtHTT enhanced p53 levels by altering the interaction between p53 and Mdm2, an E3 ligase responsible for p53 degradation, resulting in increased p53 stabilization. We also found that elevated p53 stabilization preferentially occurred in medium spiny neurons (MSNs) of the basal ganglia, a type of neuron highly susceptible to mtHTT accumulation and aggregation. Additional analyses revealed that increased p53 stabilization activated a unique transcriptional program that further contributed to enhance protein aggregation by promoting the degradation of the stress protective transcription factor HSF1. We developed a conditional deletion of p53 in MSNs of the heterozygous zQ175 HD mouse model and demonstrated that specific deletion of p53 in this cell type was sufficient to restore HSF1 levels, protein homeostasis, and synaptic

density. Overall, our data identified a novel molecular pathway connecting mtHTT with p53 and HSF1 protein stabilization in HD.

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Poster

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Program #/Poster #: P274.04

Topic: C.04. Movement Disorders other than Parkinson's Disease

Support: NIH grant R01NS116099
CIHR grant FDN 154278
HDSA grant

Title: Mutant huntingtin originating from the striatum contributes to its presence in the cerebrospinal fluid in Huntington disease

Authors: N. S. CARON[1], R. BANOS[2], A. E. ALY[1], Y. XIE[2], S. KO[1], N. POTLURI[2], C. ANDERSON[1], H. FINDLAY BLACK[1], B. GORDON[2], M. R. HAYDEN[3], ***A. SOUTHWELL**[2];
[1]Med. Genet., Ctr. for Mol. Med. and Therapeut., Vancouver, BC, Canada; [2]Univ. of Central Florida, Orlando, FL; [3]Univ. of British Columbia, Vancouver, BC, Canada

Abstract: Huntington disease (HD) is a neurodegenerative disease caused by a trinucleotide expansion in the *HTT* gene which codes for an elongated polyglutamine tract in the huntingtin protein (HTT). Expanded mutant HTT (mHTT) is toxic and leads to regional atrophy and neuronal cell loss in the brain, which is most pronounced in the striatum. Therapeutic lowering of mHTT in the central nervous system (CNS) has shown promise in preclinical studies, with multiple approaches currently in clinical development for HD. Concentration of mHTT in the cerebrospinal fluid (CSF) is being used in clinical trials as a pharmacodynamic biomarker of target engagement in the CNS. We have previously shown that the CNS is a major source of mHTT in the CSF, however, little is known about the specific brain regions and cell types that contribute to this pool of CSF mHTT. Therefore, a better understanding of the source of CSF mHTT is important for our interpretation of clinical CSF mHTT data since many of the mHTT lowering agents are expected to have limited distribution to the deeper regions of the brain, including the basal ganglia. Here, we use complementary pharmacological and genetic approaches to evaluate the contribution of the striatum to the pool of mHTT in the CSF. We show that viral expression of a HTT fragment with transduction restricted to the striatum leads to detectable mHTT in the CSF. We demonstrate that targeted delivery of an antisense

oligonucleotide against mHTT restricted to the striatum leads to a significant reduction of mHTT in the CSF of HD mice. Furthermore, using a transgenic mouse model of HD that expresses full length human mHTT and wild type HTT (Hu97/18), we show that genetic inactivation of mHTT selectively in the striatum results in a significant reduction of mHTT in the CSF. Taken together, our data support the findings that the striatum contributes sufficiently to the pool of mHTT in the CSF that therapeutic levels of HTT lowering in the striatum can be detected by this measure in mice, which suggests that CSF mHTT may represent a pharmacodynamic biomarker for therapies that lower mHTT in the striatum.

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Poster

P274. Huntington's Disease: Molecular Mechanisms II

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Program #/Poster #: P274.05

Topic: C.04. Movement Disorders other than Parkinson's Disease

Support: NIH NIDA IRP

Title: Sigma-1 receptor activation by pridopidine regulates TFEB-mediated autophagy via nucleoporin POM121 in NSC34 C9ALS/FTD model.

Authors: S.-M. WANG[1], ***H.-E. WU**[2], M. GEVA[3], M. HAYDEN[4], T. MAURICE[5], M. COZZOLINA[6], T.-P. SU[7];
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Abstract: Dysregulated autophagy relates to neurodegenerative diseases, in particular the *C9orf72*-ALS. In *C9orf*72-ALS, (G4C2)-hexanucleotide repeats expansion (HRE) does so by disrupting the nucleocytoplasmic transport of TFEB required for autophagy. However, the exact mechanism underlying the dysregulated autophagy caused by HRE remains unclear. The sigma-1 receptor (Sig-1R) is a multiple-functional chaperone protein that can affect the nuclear pore stability. Here we found that the Sig-1R regulates autophagy by chaperoning nucleoporin POM121 for the recruitment of importinβ1 that transports nucleus-bound TFEB. In NSC34 cells, the Sig-1R interacts and colocalizes with nucleoporin POM121 and Importinβ1. The Sig-1R as a chaperone stabilizes POM121 but not importinβ1, suggesting that Sig-1R interacts with POM121 that in turn binds importinβ1. HRE causes the dissociation of Sig-1R from POM121 and results

in a deficiency of nuclear importinβ1, TFEB, and an autophagy marker LC3-II. Overexpression of Sig-1R or POM121 rescues all of those deficits. Pridopidine is a potent and highly selective Sig-1R agonist currently under phase 2/3 clinical trials for ALS. But pridopidine's molecular mechanism of action against ALS remains unclear. We found that in the Sig-1R-BiP cellular assay, pridopidine acts as an agonist causing the dissociation of Sig-1R from BiP. In a chemical assay examining the chaperone activity of Sig-1R against the aggregation of citrate synthase, we found that pridopidine enhances the innate chaperone activity of Sig-1R and that the Sig-1R antagonist BD1063 blocks this action of pridopidine. Lastly, treatment of HRE-transfected NSC34 cells with pridopidine rescues all of the HRE-caused deficits mentioned above. Our results demonstrate that the Sig-1R is a ligand-regulated chaperone that controls autophagy by chaperoning a nucleoporin POM121. Results also suggest that pridopidine may be beneficial for the *C9orf72*-ALS patients through its action on the restoration of cellular autophagy. (Supported by the IRP, NIDA, NIH).

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Poster

P274. Huntington's Disease: Molecular Mechanisms II

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Topic: C.04. Movement Disorders other than Parkinson's Disease

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Title: RTP801/REDD1 is involved in neuroinflammation and modulates cognitive dysfunction in Huntington's disease

Authors: *L. PÉREZ-SISQUÉS[1], J. SOLANA-BALAGUER[1], G. CAMPOY-CAMPOS[1], N. MARTÍN-FLORES[1,2], A. SANCHO-BALSELLS[1,3,4], M. VIVES-ISERN[1], F. SOLER-PALAZÓN[1], M. GARCÍA-FORN[1,3,4,5], M. MASANA[1,3,4], J. ALBERCH[1,3,4,6], E. PÉREZ-NAVARRO[1,3,4], A. GIRALT[1,3,4,6], C. MALAGELADA[1];
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Abstract: RTP801/REDD1 is a stress-regulated protein whose levels are increased in several neurodegenerative disorders such as Parkinson's disease (PD), Alzheimer's disease (AD) and Huntington's disease (HD). Neuronal RTP801 downregulation *in vivo* ameliorates behavioral alterations in several mouse models of these disorders. Moreover, RTP801 silencing in hippocampal neurons in the 5xFAD mouse model of AD reduced several gliosis markers and inflammasome proteins, suggesting a novel role of RTP801 in neuroinflammation. In HD, RTP801 mediates mutant-huntingtin toxicity in cellular models and its levels are increased in human HD-iPSCs, postmortem putamen samples of HD patients and in striatal synaptosomes from mouse models. Moreover, RTP801 silencing in the striatum of the R6/1 mice prevented motor learning deficits and loss of synaptic proteins. Both HD patients and mouse models develop psychiatric and cognitive alterations before the onset of motor dysfunction. However, the potential role of RTP801 in the hippocampal pathophysiology in HD has not yet been elucidated. Hence, here, we investigated whether RTP801 is involved in hippocampal cognitive dysfunction and neuroinflammation in HD. We found that RTP801 levels are increased in the hippocampus of HD patients and that the levels of the protein correlate with gliosis markers. Although RTP801 expression is not altered in the hippocampus of the R6/1 model, RTP801 silencing in hippocampal neurons using shRNA-containing AAV particles ameliorates cognitive alterations. This recovery is associated with a partial rescue of synaptic markers and with a reduction of inflammatory events, particularly microgliosis. Altogether, our results indicate that RTP801 could be a biomarker for hippocampal neuroinflammation in HD patients and a therapeutic target to ameliorate the hippocampal-associated cognitive dysfunction in the disease.

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Poster

P274. Huntington's Disease: Molecular Mechanisms II

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Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P274.07

Topic: C.04. Movement Disorders other than Parkinson's Disease

Title: WWOX levels are increased in Huntington's disease post-mortem brains and ESC-derived cortical neurons

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Abstract: Huntington's disease (HD) is an autosomal dominant neurodegenerative disease caused by a cytosine-adenosine-guanine (CAG) trinucleotide expansion coding for a polyglutamine (polyQ) in the huntingtin (Htt) protein. Although the genetic mutation that causes HD is well-characterized, the pathogenic mechanisms leading to neuronal loss are not yet completely understood. It has been reported that alterations in mitochondrial function and oxidative stress play an early pathogenic role in HD. However, clinical trials involving antioxidant drugs in HD patients have been unsuccessful, highlighting the importance of unraveling the molecular pathways leading to increases in oxidative stress in HD. One such candidate is the WW domain-containing oxidoreductase (WWOX). WWOX is altered in Alzheimer's disease (AD) post-mortem brains, and mutations in the WWOX gene cause cerebral ataxia, seizures, and epilepsy. Additionally, WWOX plays a role in regulating the mitochondrial electron transport chain (mtETC) through its short-chain alcohol dehydrogenases (SDR) domain. Therefore, we hypothesized that alterations in WWOX may contribute to oxidative stress and mitochondrial dysfunction in HD. As described previously, we found a significant increase in complex I (NDUFB8) and III (UQCRC2) mtETC proteins in a large cohort of HD human post-mortem prefrontal cortex (PFC) samples, indicating increases in oxidative stress in HD brain. There was also a concomitant and significant increase in WWOX and its phosphorylated and activated form, pWWOX-Y33, in HD PFC compared to controls. Similarly, WWOX and pWWOX-Y33 levels were significantly increased in HD isogenic embryonic stem cell (ESC)-derived cortical neurons compared to control cells. Lastly, using a recombinant WWOX protein, we demonstrated a significant increase in cell death in vitro as measured by an MTT assay. Collectively, our findings suggest that increases in WWOX levels may contribute to mitochondrial dysfunction and neuronal loss in HD.

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Poster

P274. Huntington's Disease: Molecular Mechanisms II

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Program #/Poster #: P274.08

Topic: C.04. Movement Disorders other than Parkinson's Disease

Support: NHMRC Grant

Title: Spatial mapping of brain metabolism in a mouse model of Huntington's disease reveals markers of ER stress pre-symptomatically followed by neuronal deterioration

Authors: F. FARZANA[1], M. MCCONVILLE[2], T. RENOIR[1], *A. J. HANNAN[1], D. HATTERS[2], B. A. BOUGHTON[3];
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Abstract: A key pathological signature of Huntington's disease is the formation of intracellular inclusions formed by fragments of the huntingtin protein. In immortalized cell culture models, we previously showed that cells that had formed inclusions became functionally quiescent [Ramdzan et al., 2017, Cell Reports 19:919-927]. To determine whether inclusions played a role in establishing quiescence in vivo, we spatially monitored metabolism in the brains of a transgenic mouse model (R6/1) of HD at time points both before (6 weeks of age) and after inclusions manifested (12 and 16 weeks of age). MALDI-imaging mass spectrometry (MALDI-IMS) was used to monitor changes in metabolite abundances, and turnover, via deuterium isotope incorporation into lipids, introduced via drinking water and an intraperitoneal injection, in one brain hemisphere. The matched brain hemispheres were assessed in more detail by liquid-chromatography mass spectrometry (LC-MS). We found that in hippocampus regions dense with inclusions, there was a reduction in the relative abundance of neuronal-enriched lipids known to play a critical role in ER-stress protection, neurotransmission, synaptic plasticity, and neurogenesis. However, certain lipid synthesis rates were also increased in regions enriched with inclusions. These increased lipid synthesis rates were also apparent in mice at 6 weeks of age, which are asymptomatic and lack inclusions. Interestingly, elevated lipid synthesis has been previously recognized as a mechanism used by cells to cope with ER stress. Our data therefore suggests that increased lipid synthesis earmarks early-stage adaptive response to ER stress but, as HD progresses, a failure results in broader cellular collapse of neurotransmission, synaptic plasticity and neurogenesis. These findings provide new insight into the cascade of molecular events in brain regions spanning the development of inclusions.

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Poster

P297. Mechanisms of Neurotoxicity I

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Program #/Poster #: P297.01

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: DOD grant W81XWH-20-1-0278
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Title: Neurotrophic and epigenetic mechanisms underlying the antidepressant action of ketamine in a rat model for Gulf War Illness

Authors: A. C. R. RIBEIRO[1], E. H. HAWKINS[1], F. JAHR[2], J. L. MCCLAY[2], *L. S. DESHPANDE[1];
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Abstract: Over a third of the First Gulf War Veterans suffer from Gulf War Illness (GWI) with depression and memory impairment among its chief complaints. War-time exposure to organophosphate (OP) compounds has been implicated for GWI development. Following repeated, low-dose exposures to an OP and a nerve agent surrogate diisopropyl fluorophosphate (DFP), we previously identified signs of chronic depression in rats. We subsequently reported reduced levels of brain derived neurotrophic factor (BDNF) along with epigenetic histone dysregulations in this GWI model. Furthermore, ketamine (KET) boosted BDNF levels and produced an antidepressant effect in GWI rats. In this study, we further investigated the molecular consequences of reduced BDNF and investigated mechanisms by which KET was upregulating BDNF in our GWI model. Male Sprague-Dawley rats (3-m age) were injected with DFP (0.5 mg/kg, sc, 1x daily for 5-days). After about 6-m, rats received either a single dose of KET (10 mg/kg, ip) or saline. Twenty-four hours later, rats were either euthanized and rapidly dissected to obtain hippocampi for antibody-specific protein determinations or they were perfused with 4% paraformaldehyde, hippocampal slices subjected to Dil staining and confocal imaged to visualize dendritic spines. In agreement with our published data, GWI rats exhibited a significant decrease in BDNF levels that were significantly higher after KET treatment. Similarly, increased histone deacetylase (HDAC) activity along with a significant increase in HDAC1 and higher levels of HDAC3 and HDAC5 proteins were noted. In the KET-treated GWI samples, decreases in HDAC3 and HDAC5 levels were observed but only HDAC3 expression was deemed significantly lower. Confocal imaging revealed a significant decrease in dendritic spine density in GWI rats compared to age-matched control rats. Moreover, KET-treated GWI rats exhibited a significant increase in spine density over saline-treated GWI rats. BDNF has come to the forefront both as a molecule that not only affects spine maturation and synaptic mechanisms but also as a biomarker of depression and as a key transducer of antidepressant and memory effects. Our prior studies indicate that DFP exposures leading to GWI-related depression are associated with epigenetic HDAC modifications that affect BDNF protein expression. We now show that reductions in BDNF affect spine density that could underlie the pathological synaptic plasticity that precipitates as GWI neurological symptoms. KET may reverse this by decreasing HDAC activity, allowing for increased BDNF expression and spine density to produce long-lasting antidepressant effects in GWI rats.

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Poster

P297. Mechanisms of Neurotoxicity I

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Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: DoD Grant GW160151 (Qiang)
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DoD Grant GW170023 (Baas)
CURE Grant SAP Number: 4100072545 (Baas)
NIH Grant T32-MH079785 (Yates)

Title: A cellular approach to understanding and treating Gulf War Illness

Authors: P. L. YATES[1], A. PATIL[1], X. SUN[1], A. NICEFORO[1], R. GILL[1], P. CALLAHAN[2], W. BECK[2], E. PIERMARINI[1], A. V. TERRY, JR[2], K. SULLIVAN[3], P. W. BAAS[1], ***L. QIANG**[1];
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Abstract: Gulf War Illness (GWI), a disorder suffered by approximately 200,000 veterans of the first Gulf War, was caused by exposure to low-level organophosphate pesticides and nerve agents in combination with battlefield stress. To elucidate the mechanistic basis of the brain-related symptoms of GWI, human induced pluripotent stem cells (hiPSCs) derived from veterans with or without GWI were differentiated into forebrain glutamatergic neurons and then exposed to a Gulf War (GW) relevant toxicant regimen consisting of a sarin analog and cortisol, a human stress hormone. Elevated levels of total and phosphorylated tau, reduced microtubule acetylation, altered mitochondrial dynamics/transport, and decreased neuronal activity were observed in neurons exposed to the toxicant regimen. Some of the data suggest that neurons from veterans with GWI may be especially susceptible to developing GWI phenotypes compared to neurons from their counterparts without GWI. Wistar rats exposed to a similar toxicant regimen showed a mild learning and memory deficit, as well as cell loss and tau pathology selectively in the CA3 region of the hippocampus. These cellular responses offer a mechanistic explanation for the memory loss suffered by veterans with GWI and provide a cell-based model for screening drugs and developing personalized therapies for these veterans.

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Poster

P297. Mechanisms of Neurotoxicity I

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Program #/Poster #: P297.03

Topic: H.08. Learning and Memory

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Hungarian National Brain Research Program 2017-1.2.1-NKP-2017-00002
Hungarian Scientific Research Fund NKFIH-OTKA K 128016, NKFIH-OTKA PD 124148

Title: How to measure and modulate the rewiring of automatic behaviors by non-invasive brain stimulation

Authors: *T. VÉKONY[1], F. PEDRAZA[1], R. PHELIPON[1], K. JANACSEK[2], F. HAESEBAERT[1], B. TILLMANN[1], D. NEMETH[1];
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Abstract: Habits are recurring, automatic behaviors that strongly rely on statistical learning. To change the existing and sometimes harmful behaviors, we need to overwrite the old associations with new ones (rewiring). To date, little research has been done to explore the neurocognitive mechanisms underlying rewiring and possible approaches to facilitate successful rewiring. Previous research has shown an antagonistic relationship between executive, control function and statistical learning performance. The dorsolateral prefrontal cortex (DLPFC) has been proposed to play a "switching role" in this antagonistic relationship. This claim was supported by scientific evidence proposing that suppressing DLPFC-activity - thus, mitigating the control - might improve the acquisition of statistical knowledge. However, the question of whether we can influence the rewiring of already existing statistical associations by non-invasive brain stimulation of the prefrontal cortex remains elusive. Sixty-seven healthy young adults were enrolled in our study to test this question. On the first day of our experiment, participants practiced a probabilistic sequence learning task (Alternating Serial Reaction Time task) to measure their statistical learning performance. After a 24-hour delay period, they practiced the same task again with partly overlapping regularities, requiring them to overwrite their already existing statistical knowledge. During the acquisition of the new regularities (offline during breaks), we applied 1 Hz repetitive transcranial magnetic stimulation or sham stimulation over the left or right DLPFC. Our results demonstrate that participants were able to rewire their

knowledge of statistical regularities and acquire new associations. The old and the new knowledge can coexist, providing evidence for the compelling flexibility of the human mind in a changing environment. Moreover, we observed that the stimulation of the left DLPFC boosted the rewiring process by speeding up the acquisition of the new regularities. These findings are in line with previous results showing the mediating role of DLPFC in statistical learning. Our study provides further insights into the neurocognitive underpinnings of habit change and can contribute to a better understanding of the role of DLPFC in statistical learning.

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Poster

P297. Mechanisms of Neurotoxicity I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P297.04

Topic: H.08. Learning and Memory

Support: Gedeon Richter's Talententum Foundation
National Brain Research Program
Hungarian Scientific Research Fund
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Title: Does 1 Hz transcranial magnetic stimulation over the left DLPFC affect declarative and procedural knowledge?

Authors: *L. SZÜCS-BENCZE[1], T. VÉKONY[2], O. PESTHY[3,4], N. SZABÓ[1], D. NEMETH[2,4,5];
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Abstract: Different memory systems can support but also inhibit each other. Long-term memory representations of declarative memory can be detrimental in extracting completely new patterns from the environment, processes related to procedural learning. Previous non-invasive brain stimulation (NiBS) studies pinpointed the „neural switch" role of the dorsolateral prefrontal cortex (DLPFC) in this antagonistic relationship. They showed that inhibitory stimulation had improved implicit procedural learning. In contrast to previous studies that typically modulated the learning phase by NiBS, here, we examined the role of the DLPFC in the retrieval phase of already acquired declarative and procedural knowledge. In our study, participants performed

declarative learning (Paired Associate Learning Task) and implicit procedural learning (Alternating Serial Reaction Time Task) in the learning phase. After a 24-h offline period, we applied 1 Hz repetitive transcranial magnetic stimulation (rTMS) or sham stimulation for ten min over the left DLPFC just before retesting the declarative and procedural knowledge. A similar level of knowledge was measured following left DLPFC and sham stimulation for both tasks. Here, we focused only on the left hemisphere; the study of the right hemisphere and bilateral stimulation is ongoing. The lack of difference between groups may be due to interhemispheric compensatory mechanisms. More targeted NiBS studies are necessary to determine the functional role of the DLPFC in the retrieval of declarative and procedural knowledge.

Disclosures: L. Szücs-Bencze: None. **T. Vékony:** None. **O. Pesthy:** None. **N. Szabó:** None. **D. Nemeth:** None.

Poster

P297. Mechanisms of Neurotoxicity I

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Janos Bolyai Research Fellowship of the Hungarian Academy of Sciences (to K. J.)

Title: The role of the dorsolateral prefrontal cortex in the competition of memory processes - a transcranial brain stimulation study

Authors: *O. PESTHY[1,2], T. VÉKONY[3], Z. V. PESTHY[2], K. JANACSEK[2,4,5], D. FABÓ[6], D. NEMETH[2,3,4];
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Abstract: Behavioral memory performance can be a result of different interacting memory processes. This interaction can be cooperative, or competitive in nature. The balance of memory processes might depend on the dorsolateral prefrontal cortex (DLFPC). A recent study has shown that reduced functioning of the DLPFC results in better learning of probability-based regularities (i.e., statistical learning). However, the question remains if this improvement in statistical learning interferes with declarative learning and whether there is a hemispheric asymmetry in the role of DLPFC. Based on earlier neuropsychological studies, we hypothesized that disrupting the left DLPFC benefits statistical learning and inhibits declarative performance, while the right DLPFC has the opposite effect. Sixty-three healthy young adults participated in our sham-controlled, single-blind brain stimulation study. We used a probabilistic sequence learning task (Alternating Serial Reaction Time task) to measure statistical learning and the Paired Associate Learning task to measure declarative memory. We applied 1 Hz inhibitory repetitive transcranial magnetic stimulation over either the left or the right DLPFC throughout learning, during the breaks of the tasks. We tested the performance after a 24-hour delay period as well. We found that right DLPFC stimulation weakened statistical learning, but did not affect declarative memory. Furthermore, we did not find any effect of the left stimulation. Bayesian statistics also supported these results. Our findings are in line with previous literature on the benefit of the right DLPFC in statistical learning, however, we could not show evidence for competing memory processes on the behavioral level. We suggest that these results might be due to the compensation of the non-stimulated hemisphere.

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Poster

P418. Olfactory Receptor Neurons:?Function and Modulation

Location: Virtual Only

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Program #/Poster #: P418.01

Topic: D.05. Olfaction and Taste

Support: DFG grant STE531/20-1, SPP1392
DFG grant STE531/20-2, SPP1392

Title: The insect olfactory receptor-coreceptor Orco controls spontaneous activity and, thus, kinetics and sensitivity but not primary events of hawkmoth pheromone transduction

Authors: K. SCHROEDER[1], E. A. SOUFI[1], ***M. STENGL**[2];
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Abstract: The roles of Orco in insect odor transduction are still not completely comprehended. As established in *Drosophila* in the hawkmoth Orco is a chaperon that is obligatory for locating

and maintaining olfactory receptors (ORs) to ciliary membranes of olfactory receptor neurons (ORNs). Furthermore, if expressed in heterologous expression systems Orco from both *Drosophila* and hawkmoths forms a slow, spontaneously opening non-specific cation channel that fulfills criteria of pacemaker channels comparably to HCN channels. In contrast to *Drosophila* in the hawkmoth Orco is not involved in the primary events of pheromone transduction as ligand-gated olfactory receptor-ion channel complex. It activated about one second later during the late, long-lasting pheromone response, when membrane potential, Ca^{2+}, and cGMP levels are rising pheromone-dependently. Thus, we hypothesize that Orco acts as voltage- and second messenger-gated pacemaker channel that controls kinetics and sensitivity of pheromone responses via resonance. Indeed, with heterologous expression and extracellular tip-recordings of pheromone-sensitive antennal sensilla in *M. sexta in vivo* we found that Orco is a voltage-dependent non-specific cation channel. Since it opens at hyperpolarized potentials it drives spontaneously occurring membrane potential oscillations providing temporal selectivity for the detection of pheromone pulses that guide the males to the calling females. Furthermore, since hawkmoths' pheromone-guided mating behavior is under strict circadian control we performed long-term recordings over several days and established that spontaneous activity is controlled via a circadian clock in ORNs. Neither cAMP, nor phosphorylation via protein kinase C alone affect spontaneous activity, while after previous phosphorylation cAMP increases spontaneous activity only during the hawkmoth's activity phase. In addition, cGMP increases spontaneous activity phosphorylation dependently only during the resting phase. In summary, Orco is a target for second messenger- and circadian clock-dependent modifications to control resonance properties, sensitivity, and kinetics of hawkmoth ORNs.

Disclosures: K. Schroeder: None. **E.A. Soufi:** None. **M. Stengl:** None.

Poster

P418. Olfactory Receptor Neurons:?Function and Modulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P418.02

Topic: D.05. Olfaction and Taste

Title: Rapid transcuticular imaging of olfactory sensory neuron activity in the yellow fever mosquito Aedes aegypti

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Abstract: The yellow fever mosquito *Aedes aegypti* uses different physical and chemical cues emanating from the human body to locate a potential host. Carbon dioxide (CO_2) present in

human breath is an important behavioral activator, and this gas also synergizes with other volatile chemicals such as L-(+)-lactic acid found in human body odor to trigger mosquito host-seeking behavior. The dendrites of olfactory sensory neurons (OSNs) that detect these chemicals are housed in specialized sensilla on the mosquito antennae and maxillary palps. To date, electrophysiological approaches such as single sensillum recordings have only facilitated studies of the ligand tuning dynamics of a limited subset of OSNs from the *Aedes aegypti* olfactory system. This is primarily due to thick cuticle hampering electrode penetration of olfactory sensilla during these extracellular recordings, and the large number of sensilla present on the mosquito olfactory organs. Recent advances in mosquito genome editing allowed us to generate transgenic lines that express genetically encoded calcium indicators (GECI) targeted to specific tissues and permitted recording of activity from multiple neurons at the same time. Using the QF2-QUAS system we targeted the expression of GCaMP6s to a set of neurons that respond to CO_2 and another set that respond to carboxylic acids. We found that in peripheral sensory organs from wild-type mosquitoes, calcium-induced fluorescence is strongly attenuated given the highly melanized nature of the *Aedes aegypti* cuticle. To overcome this challenge, we introgressed our driver and effector lines to mosquitoes with a mutation that reduces melanization of the cuticle, which allowed for efficient detection of GECI fluorescence. This optimized method facilitates rapid transcuticular imaging of CO_2 receptor neuron responses and other OSNs that detect human related odorants such as L-lactic acid and butyric acid. It is further less invasive and enhances experimental throughput relative to other neurophysiological recording methods available in this non-model species. This technological advance stands to improve our fundamental understanding of the ligand tuning dynamics of the *Aedes aegypti* olfactory system.

Disclosures: D. Giraldo: None. **C.J. McMeniman:** None.

Poster

P418. Olfactory Receptor Neurons:?Function and Modulation

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Program #/Poster #: P418.03

Topic: D.05. Olfaction and Taste

Support: DBT/Wellcome Trust India Alliance IA/I/15/2/502091
SERB grant CRG/2020/004719

Title: Diversity of odor responses in maxillary palps of Aedes aegypti

Authors: *S. S. SINGH[1], A. AIRAN[1,2], S. GUPTA[1], N. GUPTA[1];
[1]Dept. of Biol. Sci. and Bioengineering, [2]Dept. of Physics, Indian Inst. of Technol., Kanpur, India

Abstract: Aedes aegypti mosquito is a vector for some of the deadliest diseases like dengue fever, zika fever, yellow fever, and chikungunya. Female mosquitoes use olfactory cues like body odors and carbon dioxide to find human hosts for blood-feeding. One of the main olfactory organs, the maxillary palp, detects these odors through several small sensilla present on its surface. These sensilla, known as the capitate pegs, house three sensory neurons within them. It is crucial to study the responses of these sensory neurons to understand how host odors are represented and processed in the mosquito brain. We recorded the responses of these neurons to carbon dioxide, 1-octen-3-ol, and 13 other odors, along with solvent controls, using single sensillum recordings (SSRs) in Aedes aegypti. We observed three types of spikes differing in their spike amplitudes and second-time derivatives within the SSR responses. Using second-time derivatives for thresholding, we characterized the spikes as A (biggest), B (middle-sized), and C (smallest), corresponding to the three neurons in the capitate peg sensilla. Consistent with previous reports, neuron A responded strongly to carbon dioxide, cyclopentanone, butanone, and 3-methyl cyclopentanone. There was some response to all the tested odors except L-lactic acid. In contrast, neuron B responded to only 4 out of 16 odors tested (4-methyl-cyclohexanol, triethyl amine, 1-hexen-3-ol, and (S)-limonene). Neuron C responded to all the odors but showed a relatively stronger response to 1-octen-3-ol. Interestingly, neuron A exhibited highly diverse responses to 1-octen-3-ol in different SSR recordings. These responses ranged from excitatory to inhibitory to no response and even temporally patterned responses in some cases. We found that this patterned activity was a combination of the intrinsic excitatory activity of neuron A and an inhibitory activity due to ephaptic coupling with neuron C. We observed reduced or no inhibition in the activity of neuron A of mosquitoes that lacked the receptors for 1-octen-3-ol. This result confirmed that the inhibition in neuron A was caused by ephaptic coupling in the sensillum through neuron C. The current study contributes to the understanding of the olfactory processing in the maxillary palps of Aedes aegypti. A deeper understanding of sensory processing may help in devising strategies to perturb mosquito-host interactions and control the spread of mosquito-borne diseases.

Disclosures: S.S. Singh: None. **A. Airan:** None. **S. Gupta:** None. **N. Gupta:** None.

Poster

P418. Olfactory Receptor Neurons:?Function and Modulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P418.04

Topic: D.05. Olfaction and Taste

Title: Dynamic switch between ON and OFF responses in an olfactory receptor neuron

Authors: *M. DEMIR[1,2], S. GORUR-SHANDILYA[1,3], T. EMONET[1,2,3,4];
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Abstract: Sensory neurons are either activated or inhibited by stimuli. Here we demonstrate the existence of a third response modality in the *Drosophila* olfactory receptor neuron ab2A, which in response to a single odorant can dynamically invert its polarity from excitatory to inhibitory. ab2A increases its spike rate in response to an initial increase in odor intensity, behaving like an "on" response. However, further increases in stimulus intensity elicit a decrease in the spike rate, whereas decreases in intensity elicit an increase in the spike rate, reminiscent of "off" responses. This on/off polarity inversion is dose-dependent and modulates olfactory behavior. It is specific to a subpopulation of acetate esters that are abundant in flies' natural settings, and is conserved across multiple *Drosophila* species that diverged at least 10 million years ago, suggesting broad occurrence and ethological relevance. Polarity inversion is independent of ephaptic coupling between sensory neurons, and does not rely on the most abundant odorant binding proteins and odor degrading enzymes. Inversion is observed at the level of signal transduction, upstream of action potential generation. Moreover, it is qualitatively recapitulated by a biophysical model with two binding sites, suggesting it might take place at the level of the receptor. Together, these results establish polarity inversion as an important property of olfactory receptor neurons, and provide a quantitative framework for future studies of this new dimension in odor coding.

Disclosures: M. Demir: None. **S. Gorur-Shandilya:** None. **T. Emonet:** None.

Poster

P418. Olfactory Receptor Neurons:?Function and Modulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P418.05

Topic: D.05. Olfaction and Taste

Title: Amplification of Olfactory Transduction Signals by Anoctamin 9/TMEM16J is Essential for Mammalian Olfaction

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Abstract: Sensing smells of food, prey, or predators determines animal survival. Olfactory sensory neurons(OSNs) in the olfactory epithelium(OE) detect odorants. cAMP and Ca^{2+} play a major role in transducing odorant inputs to electrical activity in OSNs. Here we show ANO9, a cation channel activated by cAMP/PKA pathway amplifies OSN responses. ANO9, expressed in OE, specifically the cilia of OSN, is activated by odorants through their respective receptors. *Ano9*-deficient mice elicited reductions in olfactory behavioral sensitivity, electro-olfactogram

signals, and neural activity in the olfactory bulb. ANO9 amplified the cAMP-evoked CNG currents in OSNs. Thus, the signal amplification by ANO9 contributes to mammalian olfactory transduction.

Disclosures: H. Kim: None. **H. Kim:** None. **N.T. Luan:** None. **T. Ha:** None. **U. Oh:** None.

Poster

P418. Olfactory Receptor Neurons:?Function and Modulation

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Topic: D.05. Olfaction and Taste

Support: E.E.Just Fellowship
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Amgen Scholars Program
Howard Hughes Medical Institute

Title: Inheritance of Learned Fear: cell proliferation in response to stress

Authors: *Y. R. AYMAN[1], E. JAEGER[2], H. LEE[3], C. LIFF[1], B. J. MARLIN[1], R. AXEL[1]; [1]Neurosci., Columbia Univ. Zuckerman Inst., New York, NY; [2]Columbia Univ., New York, NY; [3]Weill Cornell, New York, NY

Abstract: The precise mechanism behind epigenetic regulation in the olfactory epithelium is unknown although morphological changes in response to fear conditioning have been observed. Namely, after a chosen odor (acetophenone) is presented, an increase in the specific olfactory sensory neurons (OSNs) is observed in the parental generation (F0) and their offspring (F1) following a 3 day fear conditioning paradigm in which this odor is paired with a foot shock. It has been also shown that more receptors being expressed in response to an odor that is predictive of a shock may make the mice more sensitive to said odor. Here we provide evidence for the mechanism underlying these epigenetic changes by answering two key questions. Firstly, does fear conditioning extend the OSN lifespan or increase cell proliferation of the specific receptors to the conditioned odor. Secondly, is activation of the receptors to the conditioned odor necessary and/or sufficient for the epigenetic phenotype in F0 and F1. We use 5-ethynyl-20-deoxyuridine (EdU) to birth date OSNs at single cell resolution in the olfactory epithelium, and DREADDS (HM4Di and HM4Dq) to stimulate the OSNs in vivo in order to test the causal relations between the mature OSNs and the increase in OSN expression. Our data indicate a potential mechanism for how environmental signals are conveyed to somatic cells in sensory systems, before being transmitted to germ cells and subsequently to future generations.

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Poster

P418. Olfactory Receptor Neurons:?Function and Modulation

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Program #/Poster #: P418.07

Topic: D.05. Olfaction and Taste

Support: NIH RO11DC016222
NIH U19 NS112953
Simons Collaboration on the Global Brain
Brain Research Foundation
Tan Yang Center at Harvard Medical School

Title: A transcriptional rheostat couples past activity to future sensory responses

Authors: *T. TSUKAHARA[1], D. H. BRANN[1], S. L. PASHKOVSKI[1], T. BOZZA[2], S. R. DATTA[1];
[1]Dept. of Neurobio., Harvard Med. Sch., Boston, MA; [2]Neurobio. and Physiol., Northwestern Univ., Evanston, IL

Abstract: Animals traversing different environments encounter both stable background stimuli and novel cues, which are thought to be detected by primary sensory neurons and then distinguished by downstream brain circuits. Here we show that each of the ~1000 olfactory sensory neuron (OSN) subtypes defined by the expressed odorant receptors (ORs) in the mouse harbors a distinct transcriptome whose content is precisely determined by interactions between its odorant receptor and the environment. The expression of more than 70 genes relevant to sensory function (such as GPCR signaling factors, calcium binding proteins and ion channels) varies continuously and systematically across OSN subtypes. Consistent with a role in adaptation, levels of these genes accurately predict acute odor responses and dynamically adjust to novel odor environments over hours to days. The sensory periphery therefore separates salient signals from predictable background via a transcriptional rheostat whose moment-to-moment state reflects the past and constrains the future; these findings suggest a general model in which structured transcriptional variation within a cell type reflects individual experience.

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Poster

P418. Olfactory Receptor Neurons:?Function and Modulation

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Topic: D.05. Olfaction and Taste

Support: NIH RO11DC016222
NIH U19 NS112953
Simons Collaboration on the Global Brain
Brain Research Foundation
Tan Yang Center at Harvard Medical School

Title: Comprehensive characterization of chemical-receptor relationship in vivo in olfactory sensory neurons

Authors: *D. H. BRANN[1], T. TSUKAHARA[1], T. BOZZA[2], S. R. DATTA[1];
[1]Dept. of Neurobio., Harvard Med. Sch., Boston, MA; [2]Neurobio. and Physiol., Northwestern Univ., Evanston, IL

Abstract: Animals detect chemicals in the environment by odorant receptors (ORs) expressed in olfactory sensory neurons (OSNs). In contrast to other sensory systems, the olfactory system detects and distinguish between large numbers of volatile odorants, and these sensory stimuli span a high-dimensional chemical space. Mammalian genomes have a vast array of ORs to deal with this complexity (>1000 in mice and ~400 in humans). A comprehensive understanding of chemical-receptor relationships is essential for understanding how animals interact with the chemical world. However, due to large number of stimuli and ORs, most ORs have no known ligands, making it difficult to understand general questions about how the nose filters the olfactory world and how OR activation is linked to sensory perception. To overcome these issues, we applied single cell RNA sequencing (scSeq) to identify activated OSNs and ORs in vivo. Activation was OR-specific and different odors activated different sets of ORs, with overlapping sets or ORs for similar odorants. Additionally, exposure to odors consistently up or down regulated hundreds of genes in activated OSNs. scSeq reliably identified ~100 ORs activated by acetophenone (~15% of the detected ORs in our dataset), including almost all previously identified ones and many novel acetophenone-responsive ORs. Since scSeq captures the transcriptional changes for cells expressing each OR, we further developed a metric, activation score, to quantify the degree of odor response in an analog fashion on an OR-by-OR basis. Activation scores were reliable across animals, varied continuously across OSNs expressing different ORs, and were concentration-dependent. These results show that scSeq is a powerful tool to characterize how the entire repertoire of ORs is activated by odorants.

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Poster

P447. Auditory Processing: Deprivation

Location: Virtual Only

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Topic: D.06. Auditory & Vestibular Systems

Support: NIH NIDCD Grant R01DC014767

Title: Effects of early auditory deprivation on audiovisual processing: An EEG investigation of audiovisual stimulation in cochlear implant using children

Authors: *B. M. BORMANN, S. COFFEY-CORINA, L. M. MILLER, D. P. CORINA;
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Abstract: Early auditory sensory deprivation may have cascading effects on language and cognition in profoundly deaf children. Some studies have suggested these effects may be driven by cross-modal plasticity whereby auditory regions show additional preferential processing of visual information. However, little is known about how these children process concurrent auditory and visual stimulation. We make use of EEG to investigate the effects of concurrent sensory stimulation in CI using children (n = 39, m = 75 mos.) and typically developing controls (n = 30), m = 85 mos.). We recorded EEG while participants were presented with auditory only stimulation (continuous speech), visual stimulation (peripherally presented flashing checks) or these signals in combination. We characterized positive ERP responses in the time window of 80-210 msec, capturing responses known to be sensitive to auditory and visual stimulation. We examined audio only, visual only, and audiovisual stimuli in reference to two different time points. The first condition compared audio only stimuli to AV stimuli, time locked to the onset of the auditory events (AV-a). The second condition compared visual only stimuli to AV stimuli in reference to the onset of a visual events (AV-v). For both conditions, AV stimulation resulted in increased positivity for all groups. (Condition 1 p. < .009, Condition 2 p < .003). Our results indicate that for condition 1, CI using children show peak amplitude of the auditory-only response comparable to typically developing children (TD: 2.6 μV, CI: 2.4 μV, p = .306). We observed a greater positive amplitude for TD participants in this time window during concurrent audiovisual stimulation (AV-a) TD: 4.3 μV, CI 2.9, p < .008). In condition 2, a robust positive response for visual only stimuli was found for both groups of participants (TD: 7.5 μV; CI 4.8, μV p < .001). During concurrent audiovisual stimulation (AV-v) we observed a larger positive peak during this time window. This effect was more pronounced in TD subjects compared to CI subjects (TD:12.5 μV, CI 6.9 μV; p < .0001.). Taken together this data suggests that early auditory deprivation may differentially impact audiovisual processing.

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Poster

P447. Auditory Processing: Deprivation

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Topic: D.06. Auditory & Vestibular Systems

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Research Grant for the Next Generation Research Incubator from the Inst. for Global Prominent Research, Chiba Univ.

Title: Characteristics of speech perception by distantly-presented bone-conducted ultrasound assessed by phonetic-feature transmission analysis

Authors: *S. NAKAGAWA[1,4], K. DOI[2], S. OTSUKA[3];
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Abstract: Bone-conducted ultrasound (BCU) is perceived even by the profoundly sensorineural deaf. A novel hearing aid using the perception of amplitude-modulated BCU (BCU hearing aid: BCUHA) has been developed. In BCUHA, the vibrator is usually pressed against a part of the cranial bone behind the ear (mastoid process); however, BCU presented to parts of the body distant from the head can be perceived.In this study, to evaluate this “distantly-presented BCU” hearing, Japanese monosyllable articulation tests were conducted. Additionally, sequential information transfer analyses (SINFA) were conducted to determine what type of articulatory features were well transmitted.10 Japanese adults (males, 22-26 years), with normal-hearing participated in the monosyllable articulation test. 100-Japanese monosyllables recorded in a female voice were used as modulator signals. 30-kHz ultrasonic carrier was amplitude-modulated (AM) by the monosyllables and presented using a vibrator to following three locations: (a) mastoid process of the temporal bone, (b) sternocleidomastoid muscle (muscle of the neck), and (c) clavicle. The vibrator was fixed using an over-the-head-type steel band (mastoid), an elastic band (sternocleidomastoid muscle) or a shoulder supporter band (clavicle) with presentation pressure of 2 N (sternocleidomastoid muscle) or 5 N (mastoid and clavicle). Additionally, air-conducted (AC) monosyllables, i.e., the modulator signal itself, was presented to the same side as the BCU stimuli for comparison.In the monosyllable articulation test, the score for AC condition was higher than that of BCU condition. Confusion (miss-recognization of monosyllable) was more prevalent in the BC than in the AC condition. Differences in the percent correct between AC and BCU were significant in the clavicle for vowels and in all locations for consonants. Additionally, the phonetic features transfer ratios of vowels varied depending on the stimulus placements. Moreover, the “contracted sound” and “nasality” phonetic features of consonants were well transmitted when the stimuli were presented onto the sternocleidomastoid. The results obtained suggested that a practical speech transmission, comparable to ordinary

BCUs, presented onto the head, can be obtained by distantly-presented BCU. Additionally, it is also suggested that the feature information transmission may depend on the characteristics in the biological tissues such as attenuation rate and frequency characteristics.

Disclosures: S. Nakagawa: None. **K. Doi:** None. **S. Otsuka:** None.

Poster

P447. Auditory Processing: Deprivation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P447.03

Topic: D.06. Auditory & Vestibular Systems

Support: KAKEN Grant 21H03469

Title: Auditory perception elicited by the infrared neural stimulation of cochlear nerves in Mongolian gerbils

Authors: *M. UENAKA[1], Y. TAMAI[1], Y. ITO[1], K. TODA[2], S. HIRYU[1], K. I. KOBAYASI[1]; [1]Doshisha Univ., Kyoto, Japan; [2]Keio Univ., Tokyo, Japan

Abstract: Cochlear implants allow hearing-impaired individuals to regain auditory perception by electrically stimulating the cochlear nerves, bypassing damaged hair cells. One of the most significant drawbacks of the device is it requires surgical intervention for implanting. Infrared neural stimulation has been investigated for overcoming the disadvantage because the laser can elicit neural activity without contacting tissues. No study, however, has proven the feasibility of the laser auditory prosthesis at the perceptual level. The purpose of this study was to examine laser-evoked auditory perception using classical conditioning in head-fixed Mongolian gerbils (*Meriones unguiculatus*). A click-train of 4000 Hz (sound pressure level: 70 dB SPL) was presented as a conditioned stimulus for a reward (a drop of water), and licking behavior was recorded as a conditioned response. After the training was completed, an optic fiber was inserted into the gerbil's ear canal. The lateral side of the cochlea was irradiated with a 4000Hz pulsed laser from an outer ear (radiant energy: 0.2, 10.1, 19.1 mJ/cm^2) without the paired water. As results, laser stimulation increased the licking rate in the same way as auditory stimulation. Furthermore, an increase in the licking rate was observed with radiant energy of laser stimuli increased, which was similar to the increase in the licking rate with the sound pressure level of auditory stimuli increased. In a subsequent experiment, we trained head-fixed Mongolian gerbils with the 4000Hz repetitive infrared laser pulses (radiant energy:14.8mJ/cm^2) as a conditioned stimulus. After the training, white noise (sound pressure level: 35, 50, 65, 80, 95 dB SPL) was presented as a masking stimulus during the laser stimulating period. The laser-evoked licking significantly decreased as the masking stimulus intensified ($r=-0.70$, $p<0.001$). These results indicate that the infrared laser irradiation to the cochlea can evoke auditory perception, and the

perceptual level can be controlled by changing the radiant energy. This research will be an essential step for the clinical application of laser stimulation to cochlear implants.

Disclosures: M. Uenaka: None. **Y. Tamai:** None. **Y. Ito:** None. **K. Toda:** None. **S. Hiryu:** None. **K.I. Kobayasi:** None.

Poster

P447. Auditory Processing: Deprivation

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Program #/Poster #: P447.04

Topic: D.06. Auditory & Vestibular Systems

Title: Mechanisms Underlying the Enhancement of Medial Olivocochlear Reflex Associated with Rhythm-based Prediction: A Study Based on Temporal Correlation with Cortical Auditory Evoked Potential

Authors: *Y. ISHIZAKA, S. OTSUKA, S. NAKAGAWA;
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Abstract: Medial olivocochlear (MOC) fibers are efferent projections that emerge from the superior olivary complex and extend to the outer hair cells (OHCs). The MOC fibers are activated by acoustic stimulation and exert an inhibitory effect on OHC motility; this effect has been termed the medial olivocochlear reflex (MOCR). We have previously reported that MOCR is enhanced by anticipation based on the regularity of preceding sound sequence. Since there are efferent projections from the auditory cortex to the medial olivocochlear bundle, it is possible that processing at cortical levels involves the proactive control of MOCR. To elucidate the possibility, we measured the cortical auditory evoked potential (CAEP) and MOCR simultaneously and analyzed their temporal correlation. A broad-band noise for the MOCR and CAEP elicitation was presented in two conditions alternately. In the regular condition, interstimulus interval among the noises was fixed at 500 ms. In the irregular condition, the onset of the noise was randomly delayed or front loaded by 200 ms compared to the regular condition. The sound pressure level (SPL) and duration of the noise were set to 60 dB and 508 ms, respectively. Eight minutes-long measurements were performed, and the time fluctuations of the MOCR and CAEP were estimated.MOCR was assessed noninvasively using otoacoustic emissions (OAEs), which are sounds that originate in the cochlea and reflect OHC motility. We measured OAEs evoked by clicks presented right after the noise offset. Clicks were presented at the rate of 40 clicks per second and at a peak equivalent SPL of 60 dB. OAE level differences between the irregular and regular condition can be considered as MOCR changes associated with the timing predictability of the stimulus appearance (ΔMOCR_reg). CAEPs synchronized with the noise onset were measured. The recorded signals were band-pass filtered between 0.5-30 Hz to extract the cortical evoked response. The potential difference between the peaks of N1 and P2

was calculated from the average waveform.The time variation of ΔMOCR_reg was negatively correlated with that of N1-P2 amplitude: MOCR strength in the regular condition increases compared to the irregular condition, when N1-P2 amplitude decreases. This result suggests that the activity of auditory cortex is involved in the increase of MOCR strength associated with temporal expectation.

Disclosures: Y. Ishizaka: None. **S. Otsuka:** None. **S. Nakagawa:** None.

Poster

P447. Auditory Processing: Deprivation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

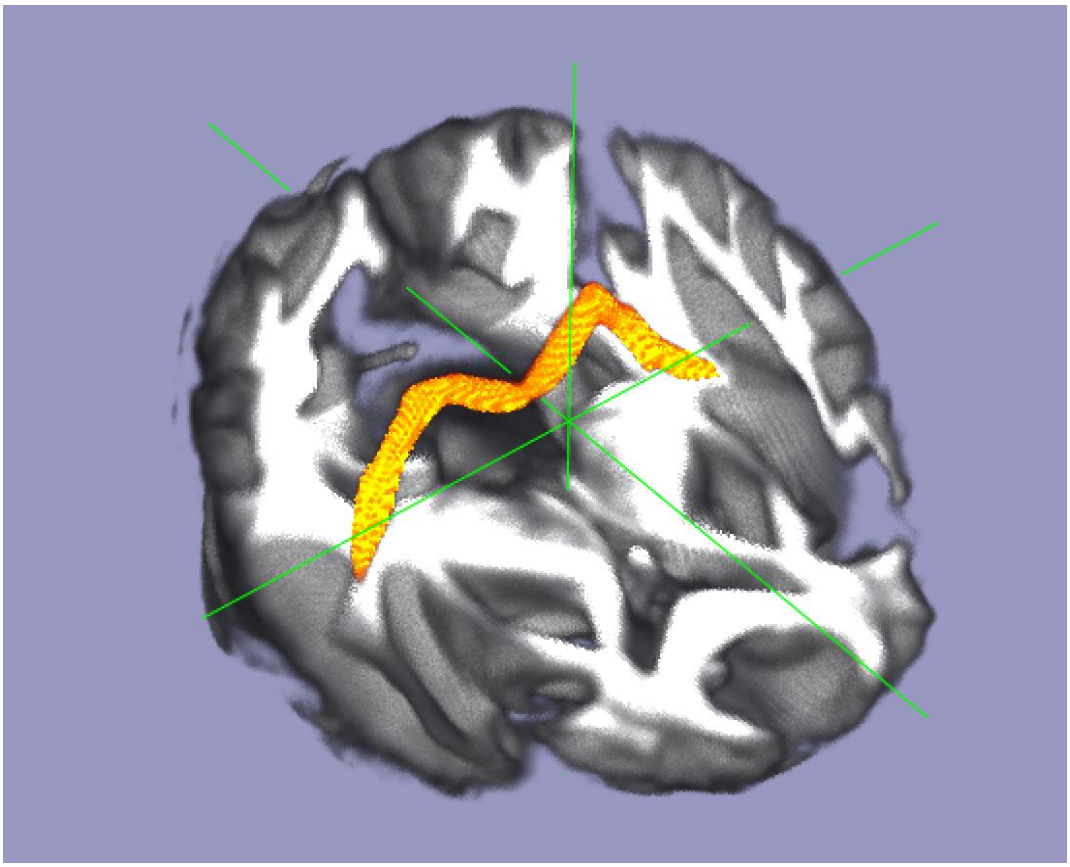
Program #/Poster #: P447.05

Topic: D.06. Auditory & Vestibular Systems

Title: Interhemispheric auditory white matter tract and impulsivity

Authors: T. E. STANSBERRY, ***T. IKUTA**;
Univ. of Mississippi, Oxford, MS

Abstract: The association between auditory processing and impulsivity has been consistently shown. However, the exact mechanism and pathological relations are not clear. The posterior part of the corpus callosum body has been repeatedly found to be affected in bipolar disorder, whose association to impulsivity is also well known. Although this part of the corpus callosum corresponds to the Interhemispheric Auditory white matter Tract (IAT), it has not been tested whether IAT is responsible for impulsivity. In this study, we first isolated the IAT and tested the association between IAT's integrity and impulsivity using diffusion tensor imaging (DTI) data from Nathan Kline Institute - Rockland Sample. Fractional Anisotropy (FA) was used as an index of white matter integrity. Out of 446 individuals, probabilistic tractography of IAT was successful in 390 individuals. FA of the IAT showed positive association with impulsivity scale, measured by UPPS (urgency, premeditation, perseverance, and sensation seeking) Impulsive Behavior Scale total score. Our results show that IAT is associated with impulsivity. Higher IAT FA is associated with higher impulsivity. This is inconsistent with the findings in bipolar disorder, where lower IAT FA is found and higher impulsivity is known. It is suggested that the IAT may not be a part of pathology in bipolar disorder.



Disclosures: T.E. Stansberry: None. **T. Ikuta:** None.

Poster

P447. Auditory Processing: Deprivation

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Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P447.06

Topic: D.06. Auditory & Vestibular Systems

Title: Optimized 40Hz ASSR in rodent cortical EEG - validation using a ketamine model of NMDA receptor hypofunction

Authors: *J. R. HUXTER, S. KANTOR, L. GIGGINS, M. DUXON;
Transpharmation Ltd., London, United Kingdom

Abstract: The auditory steady state response (ASSR) is impaired in patients with schizophrenia, reflecting a deficit in cortical-level sensory processing which may underlie common symptoms such as auditory hallucinations. ASSR is also easily measured in rodents, making it a powerful translational tool. Our aims were to 1) test ASSR detection in rats using near-midline frontal-parietal EEG, 2) to determine the sensitivity of ASSR to stimulus parameters, and 3) to test the

ability of atypical antipsychotics to reverse a ketamine-induced deficit in ASSR. Adult male Sprague-Dawley rats were implanted with epidural electrodes paired with wireless intraperitoneal transmitters (HD-S02, DSI USA). After recovery, animals were tested in sound-attenuating chambers in which blocks of click-trains were played for approximately 1 hour, using 1ms clicks with a 25ms inter-click interval. Our results indicate that by normalising responses to the 1-second baseline before each click-train, a robust ASSR signal (FFT-derived amplitude change at 40Hz, ~125% of baseline) emerged, even though the induced oscillations were not visually discriminable in the raw traces. The ASSR magnitude was replicated using tones as low as 25% of the volume typically used (75 dB) in human and rodent studies. Using a 75 dB stimulus, ketamine (0.3-10 mg/kg s.c.) induced a dose-dependent reduction in ASSR which was near 100% at 10mg/kg and returned to baseline after approximately 1 hour. The 10 mg/kg ketamine deficit was replicated in a separate cohort of animals, but not reversed by pre-dosing with aripiprazole (10 mg/kg s.c.) or clozapine (10 mg/kg s.c.), common antipsychotic medications. We conclude that with the appropriate stimulus and normalization, ASSR can be reliably detected using a frontal-parietal rodent cortical EEG montage, as opposed to recording from (or near) the auditory cortex itself. While ketamine-induced deficits were not reversed using atypical antipsychotics targeting 5-HT2A receptors, targeting alternative receptors and pathways may prove to be more effective. Alternatively, combining lower-amplitude stimuli with lower-doses of ketamine may yield a more reversible deficit.

Disclosures: J.R. Huxter: None. **S. Kantor:** None. **L. Giggins:** None. **M. Duxon:** None.

Poster

P447. Auditory Processing: Deprivation

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Program #/Poster #: P447.07

Topic: D.06. Auditory & Vestibular Systems

Support: ERC-STG-804029

Title: Modulating auditory perception using individualized tACS protocols

Authors: Y. CABRAL-CALDERIN, *M. J. HENRY;
Max Planck Inst. for Empirical Aesthetics, Frankfurt Am Main, Germany

Abstract: Auditory stimuli are often rhythmic in nature, and brain activity synchronizes to the rhythms in sounds. Stimulus-brain synchrony is advantageous for perceiving auditory information. Therefore, a clear hypothesis is that improving neural tracking, e.g., via transcranial alternating current stimulation (tACS), can aid auditory perception. In the present study we aimed to modulate auditory perception by using tACS to interfere with stimulus-brain synchrony. The effects of tACS on brain activity often show high inter-subject variability, due to

individual differences in brain anatomy and functional localization. Here, we used individualized electrode montages that were optimized to obtain the strongest electric field at individualized target regions determined based on functional imaging. In the first experiment, healthy human participants underwent an fMRI session where the BOLD signal was measured while they listened to frequency-modulated (FM) sounds. Regions showing higher activity during auditory stimulation when compared to rest, in bilateral temporal regions, were chosen as individual target regions for tACS. Individual FEM head models were created using structural MRI data and the electrode montage was optimized for each individual using Simnibs. In experiment 2, participants took part in two tACS sessions. In each session, participants listened to the same FM sounds modulated at 2Hz, while 2Hz tACS (using the individually optimized electrode montages) was applied at different phase lags relative to the auditory stimulus; we also included a sham condition. While listening to the sounds, participants detected silent gaps that were presented at different phases of the FM cycle. During the sham condition, gap detection was sinusoidally modulated by the FM-stimulus phase into which the gap fell. We hypothesized that, depending on the phase lag between tACS and the FM-stimulus, tACS could modulate gap detection in two different ways, 1) either by changing the FM-stimulus optimal phase for gap detection or 2) by changing the amplitude of the FM-stimulus-induced behavioral modulation. Cosine functions were fitted to hit rates to determine the optimal FM-stimulus phase and the amplitude of the stimulus-driven behavioral modulation. Best FM-stimulus phase for gap detection was not modulated by the tACS-sound phase lag. Compared to sham, we observed that amplitude of the FM-stimulus induced behavioral modulation was significantly smaller in the phase-lag opposite to the “best lag”. The results suggest that while tACS cannot overwrite neural tracking of the FM stimulus (no modulation of best phase), it can modulate its amplitude.

Disclosures: Y. Cabral-Calderin: None. **M.J. Henry:** None.

Poster

P470. Higher Visual Areas

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Program #/Poster #: P470.01

Topic: D.07. Vision

Support: NIH Grant EY029438
Whitehall Foundation (2016-08-18)

Title: Choice activity moderates sensorimotor associations within the V3A to CIP hierarchy

Authors: *R. DOUDLAH[1], T.-Y. CHANG[1], B. KIM[1], A. SUNKARA[2], A. ROSENBERG[1]; [1]Dept. of Neuroscience, Sch. of Med. and Publ. Hlth., Univ. of Wisconsin-Madison, Madison, WI; [2]WiSys Technol. Fndn., Madison, WI

Abstract: Visually guided behaviors require the brain to transform sensory signals into motor commands. To accurately and precisely represent an object, the visual system must solve the inverse problem of dissociating its three-dimensional (3D) orientation and position (i.e., 3D pose), which are confounded in retinal projections. To achieve desired goals, the 3D visual representations must then be associated with appropriate motor responses. We hypothesize that these transformations co-occur within a primary visual cortex (V1) to V3A to caudal intraparietal (CIP) area hierarchy. To test this, we used laminar probes to record from 692 V3A and 437 CIP neurons in three rhesus macaque monkeys while they simultaneously performed 3D orientation discrimination (3DOD) and visually guided (pop-up) saccade tasks. For the 3DOD task, planar surfaces were presented at different orientations and distances to investigate the sensory representation of 3D surface pose. Consistent with the hypothesized hierarchy, we found that 3D orientation and position were more dissociated in the responses of CIP neurons than V3A neurons. This finding implies that CIP encodes a higher-level, more robust 3D visual representation than V3A. Likewise, we found that a higher proportion of CIP neurons (44%) carried choice-related activity during the 3DOD task than V3A neurons (22%). Importantly, that difference was not explained by differences in inter-neuronal correlations within the areas. In both areas, neurons with more robust 3D selectivity tended to carry choice activity, suggesting that neurons which have resolved inherent sensory ambiguities may preferentially contribute to perceptual decisions. To explore potential sensorimotor associations, we first assessed if the neurons showed motor-related activity during the pop-up saccade task. Both areas contained similar proportions of neurons with saccade-related activity (V3A: 60%; CIP: 63%). Relative to the onset of the eye movement, saccade-related activity started in V3A (-108 ms) before CIP (-102 ms), suggestive of a feedforward saccade signal. We then compared the 3D orientation and saccade direction preferences to test for a sensorimotor association. In V3A, we found that sensory and motor preferences were aligned according to the task training, but only for neurons with choice activity. In CIP, both neurons with and without choice activity showed this association, but the relationship was stronger for neurons with choice activity. These findings imply a novel role for choice activity in moderating sensorimotor associations within the V3A to CIP hierarchy and call into question classical notions of sensory-motor dichotomies.

Disclosures: R. Doudlah: None. **T. Chang:** None. **B. kim:** None. **A. Sunkara:** None. **A. Rosenberg:** None.

Poster

P470. Higher Visual Areas

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Program #/Poster #: P470.02

Topic: D.07. Vision

Title: Protracted development of visual category representation in human prefrontal cortex underpinned by cortical tissue changes

Authors: *J. K. YAO, Z. YAZDANI, J. GOMEZ;
Princeton Neurosci. Inst., Princeton Univ., Princeton, NJ

Abstract: While occipitotemporal visual cortex has been the focus of studies examining how experience and development sculpt brain structure and function, little attention has been paid to the frontal lobe despite the known role it plays in visual processing. Here, we ask if prefrontal responses to ecologically relevant visual categories can be reliably observed relative to cortical folding patterns, and how functional responses may develop as children learn to read and improve face recognition skills. 29 children (ages 5-11) and 33 adults (ages 17-28) completed three runs of a visual category localizer while performing an oddball detection task. Contrast maps of faces, bodies, and words versus all other categories were produced for each subject, and volume of selectivity from thresholded t-value maps ($t>2.5$) on the cortical surface are quantified within regions of interest. We focus on ventrolateral prefrontal cortex (VLPFC), including cortex surrounding the precentral sulcus (PCS) and inferior frontal sulcus (IFS). We find that VLPFC is visually driven and shows prolonged functional development. Words and faces exhibit a hemispheric interaction: while face selectivity increases in the right hemisphere and is pruned from the left, word selectivity shows the opposite pattern ($F=8.7$, $p<0.005$). Body representation is larger in the right hemisphere but grows modestly in the left. A finer-grained anatomical analysis reveals that responses are not simply scaling with development, but that the distributed activation qualitatively changes across distinct anatomical segments of VLPFC. To quantify this, we divide VLPFC into anatomically-defined portions: inferior, superior, and dorsal PCS, and posterior (pIFS), middle (mIFS), and anterior IFS (aIFS). Face-related development is most pronounced in pIFS and dorsal PCS ($F's > 4.7$, $p's < 0.03$), while the mIFS shows the most body-related growth ($F=4.1$, $p<0.05$). For words, left mIFS and aIFS show the most striking developmental change ($F's > 6.2$, $p's < 0.01$). It is likely that such dramatic functional reorganization requires structural change. To verify this, all subjects also underwent quantitative MRI to acquire maps assessing brain tissue properties. We find strong evidence suggesting the underlying cortical tissue in these regions changes composition with development. For example, in right pIFS, where face responses undergo large functional change, the density of macromolecules grows into adulthood ($p's < 0.001$). Taken together, these findings reveal prefrontal structural-functional changes across development and stress the relevance of the frontal lobe in the maturation of visual processing.

Disclosures: J.K. Yao: None. **Z. Yazdani:** None. **J. Gomez:** None.

Poster

P470. Higher Visual Areas

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P470.03

Topic: D.07. Vision

Support: NSERC
FRQS

Title: Exploring multiple aspects of the mouse visual cortex functional organization using optogenetic patterned stimulation.

Authors: *V. CHOUINARD[1], M. P. VANNI[2];
[1]Dept. de neurosciences, [2]École d'Optométrie, Univ. de Montréal, Montreal, QC, Canada

Abstract: Multiple studies have shown that mouse higher visual areas are involved in discrimination task through intricate functional properties and complex networks. However, it remains enigmatic how this highly connected network defines specialized perceptual roles in higher visual areas and how these interact in mouse behavior. Here we demonstrate the potential of a novel method to study the connectivity and perceptual roles of these extrastriate areas using patterned optogenetic photostimulation combined with mesoscopic calcium imaging. Compared to galvo-based methods, this approach offers the potential to increase the sensitivity and specificity of the stimulation. (A) Our first goal was to implement an innovative system of photostimulation using a **D**igital **M**icromirror **D**evice (DMD) which uses mirrors to precisely shape a light beam at 475nm (10mW/mm^2) homogeneously into a large surface covering the entirety of the dorsal cortex of the mouse. Validation on Channelrhodopsin-expressing mice showed robust local and long-range activations using the red calcium indicator jrGECO1a. Our methods have also been very robust at evoking complex movement of the forelimb and vibrissae in all tested mice (n = 3). Such innovative technology opens new doors in optogenetics as brain areas often have complex shape and are interconnected with remote areas. Our second goal (B) was to apply this novel method of photostimulation to generate a photo-inactivation paradigm. We hypothesized that the inactivation of one or more extrastriate areas impacts the functional properties of adjacent areas and feedback connections to the primary visual cortex. To perform this experiment, we combined calcium imaging using jrGECO with the chloride channel iC+ expressed in excitatory neurons. We also studied how visual perception was affected during spatially integrated optogenetic inactivation by training mice to perform visual discrimination tasks. Overall, studying visual perception require rigorous knowledge on the function and the organization of visual areas and this research will have the potential to bring new light unto this interaction. Finally, we demonstrate how the simplicity and versatility of experimental methods can be used to study multiple aspects of vision and brain function. Supported by NSERC and réseau de recherche en santé de la vision (FRQS)

Disclosures: V. Chouinard: None. **M.P. Vanni:** None.

Poster

P470. Higher Visual Areas

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Program #/Poster #: P470.04

Topic: D.07. Vision

Support: NIH 2RO1 EY 022318-06
NSF DGE-1656518

Title: Cytoarchitecture scaffolds white matter connectivity of high-level ventral visual areas in children and adults

Authors: *E. KUBOTA[1], M. GROTHEER[2], J. GOMEZ[3], V. NATU[1], D. FINZI[1], A. REZAI[1], H. KULAR[1], M. NORDT[1], K. GRILL-SPECTOR[1];
[2]Dept. of Psychology, [1]Stanford Univ., Stanford, CA; [3]Princeton Univ., Princeton, NJ

Abstract: Classic theories of brain organization suggest that white matter connectivity, cytoarchitecture, and function all define a cortical region. Here, we use ventral temporal cortex (VTC) as a model system for testing the relationship between white matter connectivity, cytoarchitecture, and function. We tested if white matter tracts that connect to category-selective regions in VTC are organized by cytoarchitecture, function, or both using functional MRI (fMRI) and diffusion MRI (dMRI) in 27 children (ages 5-12) and 28 adults (ages 22-29). To test these hypotheses, in each participant we quantify the connectivity profile of white matter tracts that connect to each category-selective region of interest (ROI) in VTC in two ways: (1) the distribution of fibers across known bundles (dMRI models of anatomical fascicles) (2) the distribution of endpoints of white matter connections from each ROI to the rest of the cortical surface. We find that in both children and adults, connectivity profiles are similar between ROIs within the same cytoarchitectonic area, even if they are selective for different domains. Face (mFus-faces) and word (mOTS-words) selective regions in cytoarchitectonic area FG4 have similar connectivity to the posterior arcuate fasciculus (pAF), arcuate fasciculus (AF), and inferior longitudinal fasciculus (ILF). In contrast, we find that connectivity profiles are significantly different between ROIs in different cytoarchitectonic parcellations even if they are selective for the same domain. mOTS-words in FG4 has more connections to the pAF, while pOTS-words in FG2 has more connections to the vertical occipital fasciculus (VOF). Finally, we use an independent classifier to test if connectivity profiles can predict cytoarchitecture, category-selectivity, or ROI. Results show that in both children and adults connectivity profiles predict cytoarchitecture (mean classification accuracy = 0.87; SE = 0.02; chance level = .5) better than category-selectivity (mean classification accuracy = 0.61; SE = 0.03; chance level = .5) or ROI (mean classification accuracy = 0.55; SE = 0.03; chance level = .25). These findings show a tight coupling between white matter connections and cytoarchitecture and challenge the idea that regions selective for different categories have unique white matter connectivity profiles for category-specific processing.

Disclosures: E. Kubota: None. **M. Grotheer:** None. **J. Gomez:** None. **V. Natu:** None. **D. Finzi:** None. **A. Rezai:** None. **H. Kular:** None. **M. Nordt:** None. **K. Grill-Spector:** None.

Poster

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Topic: D.07. Vision

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NRF-2019M3E5D2A01058328
NRF-2021M3E5D2A01019544

Title: Spontaneous emergence of face-selective units in untrained deep neural networks

Authors: *M. SONG[1,2], S. BAEK[1], J. JANG[1], G. KIM[3], S.-B. PAIK[1,2];
[1]Dept. of Bio and Brain Engin., [2]Program of Brain and Cognitive Engin., [3]Dept. of Physics, KAIST, Daejeon, Korea, Republic of

Abstract: In the visual cortex of primates, face-selective neurons are observed and considered the basis of facial recognition (Tsao, 2006). The observation of this type of intriguing neuronal tuning in the brain has inspired neuroscientists, raising important questions about its developmental mechanism — whether face-selective neurons can arise innately in the brain or require visual experience, and whether neuronal tuning to faces is a "special" type of function distinctive from tunings to other visual objects. Recently, model studies have used biologically-inspired deep neural networks (DNN) to predict the response of the ventral visual stream and provide insight into the underlying mechanisms of brain functions. Using a DNN model of the ventral visual stream, AlexNet, we showed that face-selective units can arise spontaneously in the complete absence of training. We measured the response of the last convolutional layer to a stimulus set of face and 5 non-face classes. We found face-selective units in the untrained network, where the face-units are defined as a unit that shows a significantly higher response to face images than non-face images. We found that their face-selectivity indices are comparable to those observed with face-selective neurons in the brain. The emergence of face-selectivity is robust across various conditions of initial randomization of the network. Next, to characterize the feature-selective responses of these face-selective units qualitatively, we reconstructed preferred feature images of individual units using a reverse-correlation method and a generative adversarial network algorithm. The obtained preferred feature images show that face-selective units are selective for a face-like configuration, distinct from units with no selectivity. Furthermore, we also tested whether the selective responses of these face units could provide reliable information with which to distinguish between faces and non-face objects. We confirmed that face-selective units enable the network to perform face detection. Intriguingly, we

found that units selective to various non-face objects can also arise innately in untrained neural networks, implying that face-selectivity may not be a special type of visual tuning and that selectivity to various object classes can arise innately in untrained deep neural networks, spontaneously from random feedforward wirings. Overall, our results imply a possible scenario in which the random feedforward connections that develop in early, untrained networks may be sufficient for initializing primitive face-selectivity as well as selectivity to other visual objects in general.

Disclosures: M. Song: None. **S. Baek:** None. **J. Jang:** None. **G. Kim:** None. **S. Paik:** None.

Poster

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Program #/Poster #: P470.06

Topic: D.07. Vision

Support: NIH Grant EY029438
Whitehall Foundation (2016-08-18)

Title: Neuronal basis of dynamic, reliability-dependent cue integration for 3D visual perception

Authors: *T.-Y. CHANG[1], R. DOUDLAH[1], B. KIM[1], A. SUNKARA[2], A. ROSENBERG[1]; [1]Univ. of Wisconsin–Madison, Madison, WI; [2]WiSys Technol. Fndn., Madison, WI

Abstract: The visual system transforms two-dimensional retinal projections into robust (i.e., accurate and precise) three-dimensional (3D) percepts, allowing us to successfully interact with our surroundings. Constructing robust 3D representations requires the integration of distinct sensory signals, including stereoscopic and perspective cues. The reliability of these cues depends on the object's pose (orientation and position). Previous studies have shown that primates perceptually integrate these cues with weights that are dynamically set according to the pose conditions. However, it remains unclear where and how that neural computation is performed. We, therefore, trained macaque monkeys to perform a 3D orientation discrimination task using cue-isolated and combined-cue stimuli at two pose conditions with different relative cue reliabilities. Neuronal responses were then recorded from the caudal intraparietal (CIP) area using laminar probes while the task was performed. At each pose, we found that the number of neurons tuned for a given cue reflected the perceptual sensitivities, such that more neurons were tuned to the cue to which the animals were most sensitive. For neurons tuned to both cues, the cue-specific orientation preferences were highly similar. Consistent with cue integration and decoding models, combined-cue responses were greater in magnitude than cue-isolated responses, and neuronal response magnitude was highly correlated with behavioral sensitivity. To estimate the neuronal cue integration weights, we fit the combined-cue responses using a

linear weighted summation of the cue-isolated responses. The weights changed with the stimulus pose such that greater weight was assigned to the more reliable cue, mirroring the behavioral data. We then used an information-theoretic approach to compute the optimal cue weight ratio that would maximize the Fisher information of the combined-cue responses. The measured weight ratios from the linear fits and the optimal weight ratios were highly correlated, indicating that the information for performing the task was roughly maximized. These findings suggest that CIP neurons encode high-level 3D visual representations through dynamic, reliability-dependent reweighting of stereoscopic and perspective cues in an optimal manner.

Disclosures: T. Chang: None. **R. Doudlah:** None. **B. Kim:** None. **A. Sunkara:** None. **A. Rosenberg:** None.

Poster

P470. Higher Visual Areas

Location: Virtual Only

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Program #/Poster #: P470.07

Topic: D.07. Vision

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JSPS 26290011
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JSPS 372 C0219129

Title: Distributed context-dependent choice information in mouse dorsal-parietal cortex

Authors: *J. ORLANDI, M. ABDOLRAHMANI, R. AOKI, D. LYAMZIN, A. BENUCCI; RIKEN Ctr. for Brain Sci., Wakoshi, Japan

Abstract: Choice information appears in the brain as a distributed signal in multi-area networks, mixed with sensory, motor, and cognitive variables. In dorsal-parietal cortical regions—traditionally implicated in decision computations—the presence, strength, and area specificity of choice signals are highly variable, limiting a cohesive understanding of their computational significance.In this study, examining the mesoscale activity in mouse posterior cortex during a complex variant of a two-alternative forced choice visual discrimination task, we found that broadly distributed choice signals defined a decision variable in a low-dimensional embedding space of multi-area activations, particularly along the ventral visual stream. The decision variable was linked to neural trajectories for left and right choices that significantly separated at the onset of the animal's actions with a discrimination strength that ramped up before movement onset ($\Delta d' = 0.2$, $p < 0.001$). The embedding choice subspace obtained using a novel tensor decomposition technique, locaNMF, was near-orthogonal ($\Theta > 80°$, $p < 0.001$) to concurrently represented sensory and motor-related activations. The neural trajectories linked to the decision

variable were modulated by task difficulty and contextually by the animals' attention state, with easier trials during bouts of high sustained attention showing the greatest separation (d'=1.4 ± 0.1) and significantly higher than in low attention bouts (p = 0.003).To mechanistically relate choice representations to decision-making computations, we trained recurrent neural networks (RNN) with the animals' trial-by-trial decisions, including their mistakes, and found an equivalent decision variable whose context-dependent representational dynamics agreed with that of the neural data. The trained RNN reproduced the animals low-dimensional representation as well as their context-dependent modulations. In conclusion, our results demonstrated a decision variable broadly represented in the posterior cortex, controlled by task features and cognitive demands. Its dynamics reflected decision computations, possibly linked to context-dependent feedback signals used in variable animal-environment interactions.

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Poster

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Title: Processing of visual textures in primary and secondary visual cortex of the mouse

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Abstract: Textures are a useful class of visual stimuli for studying the circuit-level underpinnings of core object recognition along the visual stream. Textures are sufficiently complex to largely match the statistical properties of natural images, and, unlike natural images, they can be precisely parameterized. Examining the cortical processing of texture stimuli in mouse animal models is useful in light of the abundant tools available for dissecting neural circuits. However, whether the mouse visual system enables a differential encoding of texture

information (as it has been shown in primates) is largely unknown. We addressed this question by presenting mice with two types of stimuli, textures and spectrally matched noise (scrambles) whose low-level features matched those of the textures. While mice passively viewed the stimuli, we performed widefield and two-photon calcium imaging via virally expressed jGCaMP7f. At the widefield level we observed a differential modulation of textures relative to scrambles in the primary and secondary visual cortex (V1, LM); differences in modulation were larger in LM compared to V1 (LM = 21.3 ± 2.8 %, V1 = 13.6 ± 2.5 %, p = 0.013, t-test, n = 6 mice). In the two-photon experiments, we imaged large populations of neurons in areas V1 and LM, and found that the average single cell discriminability (d' measure) between textures and scrambles was significantly larger in LM than in V1 (d' = 0.26 ± 0.05 in V1, d' = 0.60 ± 0.06 in LM; p = 0.009, t-test, n = 5 mice). The difference between LM and V1 suggests a response modulation driven by the higher-order statistical features in texture stimuli. In conclusion, by presenting well-controlled and parameterized stimuli for which the main axis of variation depends on high-order statistical information, we found that the mouse ventral visual stream supports a hierarchical processing of textural information, suggesting that also in this species textures may be relevant for ethological behaviors.

Disclosures: F. Bolanos: None. **J.G. Orlandi:** None. **A.V. Jagadeesh:** None. **J.L. Gardner:** None. **A. Benucci:** None.

Poster

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Title: Probabilistic discrimination of relative stimulus features in mice

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Abstract: During perceptual decision-making, information about the stimulus and the upcoming decision is encoded in mixed representations within the same brain areas and neuronal populations. Studying decision computations in isolation thus requires tailored experimental design, such as one where decisions depend on a relative property of a stimulus. In this case, the

same amount of decision information can be given by different combinations of physical stimuli, neural representations of which can be factored out during data analysis. Mouse is a recently popular animal model in the research of visual decision-making thanks to the powerful genetic tools available for the dissection of brain circuits. However, whether mice can perform a relative visual discrimination task has not yet been fully established. Here we show that mice (n = 40, 1.3 million trials) can solve a complex version of orientation discrimination task, in which they indicate the more vertical of the two stimuli, and in which the same amount of sensory information can be given by many stimulus combinations as long as the difference in their verticalities is the same. Using this task, we show that mice have a typical orientation discrimination acuity of 9°, reaching 6° for one mouse tested with the smallest angular separation of 3°, which challenges the common belief that mice are poor visual discriminators. To reach these conclusions, we introduced a novel probabilistic choice model that explained behavioral strategies and demonstrated that the circularity of the stimulus space serves as an additional source of choice variability for trials with a fixed difficulty. The model explained additional 9.03% ± 1.48% of deviance fraction in comparison to a classic psychometric curve. Furthermore, the model accounted for history-dependent choice biases (explaining additional 5.07% ± 0.98% fraction of deviance) and demonstrated their dependence on the level of animal's task engagement: whenever an animal was less engaged in the task, as indicated by very short (within 10th percentile) reaction times possibly corresponding to compulsive choices, the decisions were driven by history biases more strongly than on trials with intermediate reaction times. Our results demonstrate that mice can be trained in a relative orientation discrimination tasks to a high discrimination acuity and that they adopt a diverse set of individual strategies that may co-vary with their cognitive state.

Disclosures: D. Lyamzin: None. **R. Aoki:** None. **M. Abdolrahmani:** None. **A. Benucci:** None.

Poster

P497. Human Perception II

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Program #/Poster #: P497.01

Topic: H.02. Perception and Imagery

Title: Co-speech gestures impact the whole-brain distribution of brain regions involved in the semantic representation of words.

Authors: *B. WANG[1], S. ALIKO[1,2], J. I. SKIPPER[1];
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Abstract: Some co-speech gestures contribute meaning to spoken words, resulting in better comprehension. Previous studies demonstrate that the processing of these involves 'typical'

language regions, reducing semantic selection and retrieval demands as indexed by processing in the inferior frontal gyrus (IFG). Work on the semantic representations of words demonstrates that they are distributed across the entire brain in a manner that mirrors their sensorimotor properties. Does the semantic content conveyed by gestures interact with that of words only in the IFG or across these more distributed regions? We address this question in a preliminary study, analyzing the fMRI data of six participants from the Naturalistic Neuroimaging Database (3 female; mean age = 22.17 yrs old) who watched the same movie. We analyzed the patterns of brain activity for spoken words and co-speech gestures in the movie using a combination of automated gesture annotation, voxelwise modeling, principal component analysis and clustering. Our cluster analysis revealed three groups of spoken words that were related to actions (e.g., 'think', 'watch', 'listen'), objects (e.g., 'mayor', 'clock') and numbers, and syntax (e.g., 'the', 'for', 'that'). These groups involved a distributed set of brain regions, including the right posterior fusiform gyrus, left central sulcus and inferior precentral and postcentral gyri, and mid-superior parieto-occipital sulcus, respectively. We also identified a number of cortical regions involved in gesture processing that overlap with 'canonical' language regions, including Heschl's gyrus, planum temporale, right STG, and left middle temporal gyrus. They also recruited the left inferior pars triangularis and right inferior temporal gyrus for 67% of participants. In addition, gestures and action words colocated in a further set of distributed regions related to actions in the left anterior angular/parietal cortex. Gestures and object/number words recruited Heschl's gyrus, STG, left planum temporale, and left inferior temporal sulcus. Overall, preliminary results suggest that 'typical' language regions play a crucial role in processing co-speech gestures including the IFG. However, we also show that both words and gestures share a distributed semantic representation in brain regions in and beyond these language hubs. Thus, these results suggest that meanings of co-speech gestures more than impact the selection and retrieval of words but also impact the semantic representation involved in word processing, involving the whole brain.

Disclosures: B. Wang: None. **S. Aliko:** None. **J.I. Skipper:** None.

Poster

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Program #/Poster #: P497.02

Topic: H.02. Perception and Imagery

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James S. McDonnell Foundation

Title: Exploring the hierarchical organization of the infant visual system with model-based representational similarity

Authors: *C. T. ELLIS[1], T. S. YATES[1], L. J. SKALABAN[1], A. J. BRACHER[2,3], N. B. TURK-BROWNE[1];
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Abstract: What is the nature of infant visual experience and how is this content represented in their visual system? In the mature human brain, there is a hierarchy of selectivity across visual areas, from low-level features (e.g., edges, luminance) in V1, to mid-level features (e.g., shapes, parts) in V4, to high-level features (e.g., object identity, category) in lateral occipital cortex. It is unclear how and when during development the visual system acquires this organized selectivity. In adults, prior studies have revealed the hierarchical organization of the visual system by comparing different layers of a convolutional neural network model with patterns of fMRI activity in visual areas. We applied this model-based approach to three cohorts of awake infants watching different movies during fMRI. The first cohort was used to optimize the analysis pipeline and included 15 sessions from infants aged 3-36 months. This pipeline was then applied to a second (38 sessions with infants aged 4-18 months) and third cohort (16 sessions with infants aged 4-32 months). For each cohort, we collected fMRI data from adults as a mature baseline. These data were analyzed by feeding frames of the movies into the VGG-19 computer vision model and calculating a time-by-time similarity matrix using the activation patterns for each layer. Using analogous time-by-time similarity matrices from the patterns of fMRI in different visual areas, we measured representational similarity with the model layers to determine the expression of low-, mid-, and high-level features in each area. In both infants and adults, VGG-19 explained significant amounts of variance in the fMRI data. Different from adults, infants did not show evidence of hierarchical organization: V1 and lateral occipital cortex represented features of all levels of complexity similarly. The lack of a hierarchy of selectivity in the infant visual system indicates that such specialization emerges later in development. This in turn may explain the dramatic changes in visual abilities that occur throughout childhood.

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Poster

P497. Human Perception II

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Program #/Poster #: P497.03

Topic: H.02. Perception and Imagery

Support: NSERC

Title: The temporal profile of dynamic facial expression perception as revealed through EEG-based decoding and movie reconstruction

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Abstract: Extensive research has investigated the perception of facial expressions, typically focusing on only a handful of basic and static emotional expressions. However, in our daily experiences we rarely encounter static expressions that perfectly fit these stereotypical expressions. Specifically, this study aims to provide a more complete understanding of the representational nature and temporal signatures underlying dynamic facial expression perception. We address these gaps through EEG-based decoding and movie reconstruction. EEG data were collected from healthy adults who viewed 1-second videos each displaying 1 of 24 dynamic facial expressions from two identities while performing an oddball task (i.e., angry expression recognition). Each stimulus began with a neutral expression and proceeded to a peak expression display over the course of ten 100ms frames. In addition, participants performed a behavioral pairwise similarity rating task with the same 24 expressions. First, we used EEG-based pattern analyses for expression decoding, then conducted a detailed investigation into the temporal dynamics of expression decoding to better profile dynamic expression perception. Lastly, we relied on pattern analysis techniques to conduct movie reconstruction from EEG data to uncover and visualize the structure of neural representations underlying expression recognition. Our investigation shows that the broad representational space of facial expressions derived from behavioral and neural data is generally accounted for by the classical dimensions of valence and arousal. Second, the temporal profile of EEG-based decoding shows a gradual increase in classification accuracy after 350ms reaching a plateau between roughly 600-1200ms following stimulus onset. Further, a frame-by-frame analysis indicates that peak decoding accuracy is reached at an intermediate step (i.e., the 6th frame of the video), prior to the apex expression (i.e., the 10th frame). Third, movie reconstruction is successfully achieved from EEG signals and, consistent with our decoding results, shows that reconstruction accuracy is highest after the midway point of a dynamic stimulus. Our results shed light on the neural and temporal representational structure of dynamic facial expressions. Our results characterize the time course of expression perception and reveal that peak discrimination occurs prior to the availability of a maximally informative peak expression. Of note, our reconstruction results elucidate the pictorial content of a dynamic expression with exceptional detail while also providing evidence for the possibility of EEG-based dynamic stimulus reconstruction.

Disclosures: T. Roberts: None. **G.C. Cupchik:** None. **J.S. Cant:** None. **A. Nestor:** None.

Poster

P497. Human Perception II

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Program #/Poster #: P497.04

Topic: H.02. Perception and Imagery

Title: Exploration path trajectory during self-guided viewing of complex photographs and paintings predicts brain activity patterns in the default network

Authors: *X. LI, E. MUSZ, B. BELLANA, Z. ZUO, J. CHEN;
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Abstract: Studies of scene perception often focus on what can be gleaned from a brief (<1 second) visual presentation, such as the general category or objects within a scene. However, in real life, people may take several seconds, or even minutes, to make sense of a complex scene. As each person takes their own "exploration path" through a visual scene, how does the history of their path influence their understanding of the next moment? How do neural responses reflect the process of making sense of a complex scene over time? In this fMRI study, we tested whether exploration paths during extended perception of complex images could predict neural responses within and across individuals. We recorded brain activity while participants viewed a set of five images and described aloud what they saw (N=17, 6-31 min). For analysis, each of these complex scenes was segmented into constituent people, objects, and environmental landmarks; each participant's utterances and corresponding brain data were labeled according to these segments. We first tested whether each image segment elicited a distinctive multi-voxel activity pattern that was replicable across individuals, regardless of temporal order. In both early visual cortex and in the default network, we observed segment-specific activity patterns—i.e., patterns were more correlated across people during descriptions of matching versus non-matching image segments, irrespective of the order in which the segments were mentioned. Next, we tested whether brain activity was sensitive to this exploration trajectory. For each subject we determined the order in which segments were described, and calculated similarity between all possible pairs of subjects; these correlations showed high similarity across subjects' exploration paths (Spearman's r= .47). Comparing the subject pairwise similarity matrices of these paths, we also found that relationships between subjects were consistent across images (Pearson's r= .70). The path similarity matrix was then used to predict subject-pairwise pattern similarity (inter-subject representational similarity analysis, IS-RSA; Chen et al., 2017; Finn et al., 2018; Nguyen et al. 2019) in selected brain regions (N=12). Results showed that path similarity predicted brain pattern similarity in posterior medial cortex (p=.03) and the entire default network (p=0.05), but not in early visual cortex (p=.16) or early auditory cortex (p=.26). Next, we will test whether subjects with similar exploration paths evince increasingly similar brain activity patterns over time as their comprehension of each scene matures, followed by an analysis of inter-subject similarity during cued recall of the images.

Disclosures: X. Li: None. **E. Musz:** None. **B. Bellana:** None. **Z. Zuo:** None. **J. Chen:** None.

Poster

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Title: Encoding and decoding models of music perception in the human brain

Authors: *L. BELLIER[1], A. LLORENS[1], D. MARCIANO[1], P. BRUNNER[2], G. SCHALK[3], R. T. KNIGHT[4], B. N. PASLEY[1];
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Abstract: Music is core to human experience and multiple brain regions have been reported to be engaged in music perception. However, the temporal dynamics of regional neural activity during music processing (e.g., rhythm) are largely unknown. To better understand the neural code for music, we used predictive modeling tools applied to intracranial EEG data recorded from 2,668 electrodes across 29 neurosurgical patients, who passively listened to a song (Another Brick in the Wall, Part 1; Pink Floyd). We used encoding models to identify electrodes encoding acoustic information about the song and to analyze their tuning patterns. We then performed an ablation analysis by selectively removing sets of music-active electrodes from linear decoding models, to assess how anatomical and functional regions represent information about the song's acoustics. Finally, we explored to what extent we could reconstruct an intelligible song from neural activity and quantified the impact of methodological factors on decoding accuracy.
We focused on high-frequency activity (HFA; 70-150 Hz) which is linked to neural population activity. Using Spectro-Temporal Receptive Fields (STRFs) as encoding models combined with HFA recording, we confirmed previous findings showing a central role of bilateral Superior Temporal Gyri (STG) in music perception. We also highlighted the involvement of bilateral Sensory-Motor Cortices (SMC) and Inferior Frontal Gyri (IFG) and observed a right hemispheric dominance for music perception. Using both a component analysis and temporal modulations, we found these cortical regions to be tuned to specific musical elements (presence of vocals, onsets of lead guitar motifs, sixteenth notes of the rhythm guitar). Within bilateral STG, we observed partially overlapping neural populations also representing these elements.
We performed an ablation analysis which is equivalent to virtually lesioning elements of the

music network. Compared to a baseline model using all 347 significant electrodes, we found that ablating electrodes from left or right hemispheres both deteriorated decoding accuracy, suggesting they each contain important, unique information and documented a right hemispheric dominance for music perception.
By exploring methodological factors impacting the performance of decoding models, we found a logarithmic relationship between decoding accuracy and either the number of electrodes or the dataset duration. Lastly, only non-linear decoding models were able to reconstruct an intelligible song from neural activity.

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Poster

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Program #/Poster #: P497.06

Topic: H.02. Perception and Imagery

Title: Measurement of mismatch negativity for changes of melody contour : Effects of contour shape and music training

Authors: *U. NAITO[1], R. SHIBA[2], S. OTSUKA[1,3,4,5], S. NAKAGAWA[1,3,4,5];
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Abstract: It is well-known that music evokes our emotions. Although such properties of music have been applied to music therapy, the neural basis of music perception and cognition is still unclear. Melody is one of the three main elements of music and includes several factors such as pitch, contour, tonality, and rhythm. The “contour” means a temporal pattern of ascendance and/or descendance of pitch. Humans are thought to perceive melodies as two pieces of temporal information; the contour and the interval (difference of pith between successive two sounds). However, there have been few studies on brain functions related to the perception of the melody contour and the neural mechanisms remain unclear. On the other hand, many studies have reported that musical experiences (trainings of music) affect the perception characteristics and neural responses to sounds, and even the anatomy of the cortex. Therefore, similar changes depending on the musical experiences may be observed also in the melody perception. We measured mismatch negativities (MMNs) for changes of melody contours and pitch in musicians and non-musicians. Five-note tone sequences were used as stimuli. First, four types of standard stimuli with different melody-contour shapes (rising-falling/falling-rising/rising/falling) were

generated by changing the pitch of each note. Second, seven types of deviants were generated by changing the pitch of the third note of each standard stimulus. MMN measurements were conducted 4 times in each subject with different combinations of a standard and seven deviant stimuli. As results, in the musicians, larger MMNs appeared for the change of the melody contour, i.e., a shape of pitch change of the entire sequence, than for the pitch change of the third tone. In term of effects of the melody-contour shape, MMN amplitude were significantly larger for the rising-falling and the falling-rising shape. In addition, the latency of the MMN was shorter in the musician compared to the non-musician. In contrast, in the non-musicians, larger MMNs tended to appear when the pitch change of the third note was maximum . No particular tendency common to all the contour shape was observed.These results indicated that musical experience enhances the ability to perceive sound sequences as a group and consequently improves the discrimination of melody contours, and increases the speed of melody processing. It is also shown that more complicated melody contour elicits larger MMN in the musicians, whereas no significant effects of the melody contour were observed in the non-musicians.

Disclosures: U. Naito: None. **R. Shiba:** None. **S. Otsuka:** None. **S. Nakagawa:** None.

Poster

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Topic: H.02. Perception and Imagery

Support: Kyushu University Fund Support Grant Program

Title: Investigation of effects of the aesthetic preference on the perception of human walking: a preliminary study using event-related potentials

Authors: *S. HAYASHI[1,2], Y. NISHIMURA[3,2], Y. IKEDA[4,2], Y. EGASHIRA[1], M. UKEZONO[1], S. UONO[1], T. OKADA[1], S. HIGUCHI[2];
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Abstract: We have an aesthetic preference for daily motion (i.e., walking) as well as artistic activities (i.e., dance performances). This study investigated the effect of the aesthetic preference on visual processing of human walking using event-related potentials (ERPs). Twenty male university students observed point-light movies representing human walking (like a runway model, with good posture, and with bad posture) and their scrambled motion. They were asked to discriminate whether the stimulus is human walking or not. The ERPs at left-/right-occipitofrontal areas (N200 and N300), reflecting the perceptual and attentional processing of visual stimuli, were measured. Participants' evaluation of stimuli showed that they aesthetically

prefer movies especially of model and good posture walking. The analysis of variance with the dot type (biological / scrambled), walking type (model / good posture / bad posture) and laterality (right / left) as within-participant factors for the N200 and N300 revealed significant main effects of dot type, which were qualified by a significant dot type × walking type interaction. The follow-up analysis showed the N200 elicited by the biological motion was larger than that by the scrambled motion for the good and bad posture, but not for the model. The analysis also showed N300 for biological motion were larger than that for scrambled motion regardless of walking types, though the simple effect of walking type was observed for the scrambled motion. The results suggests that the N200 reflect visual processing of biological motion, which is disturbed when the stimuli represent unusual walking (i.e., model walking), while the N300 reflect recognition of integrated human walking regardless the walking type. We also conducted exploratory regression analyses with the aesthetic-preference score of the stimuli as independent variables for the ERPs. The N300 for biological but not for scrambled motion were negatively deflected with increasing the aesthetic-preference scores. There is no significant effect of the aesthetic-preference scores for the N200. The results suggest that the N300 also reflect aesthetic preference when the stimuli represent human motion. In summary, this study suggests that the N200 was involved in processing of familiar biological motion, while the N300 was involved in integration of forms and optic-flow/motion information of biological-human motion as well as extracting the aesthetic information of biological motion even without detailed featural information of the walkers.

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Poster

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Topic: H.02. Perception and Imagery

Title: Predictive neural representations of sensory input revealed by a novel dynamic RSA approach

Authors: *I. E. J. DE VRIES, M. F. WURM;
Ctr. For Mind/Brain Sci., Rovereto, Italy

Abstract: To successfully navigate dynamic environments, we need to constantly update our inner representations of external dynamic information, such as actions of others. This poses a fundamental problem: how does the brain cope with a stream of constantly changing input? It takes time to transmit and process each retinal image. Consequently, do we represent the world in a lagged manner, or does the brain generate predictions of unfolding dynamics? Our capacity

to react to dynamic stimuli in a timely manner indeed demonstrates that our brain predicts future events. However, the representational nature of such predictions remains largely unexplored. One method that considers neural representations of sensory input is representational similarity analysis (RSA), which typically uses static models to investigate how stimulus features (e.g. color, category) are represented in the brain. We developed a dynamic extension to RSA that uses temporally variable models, i.e. a unique model for each time point, to capture the representational nature of dynamic stimuli (dynamic RSA hereafter). At each time point, the model representation is not only compared to the neural representation at the same time point, but also at all other time points during stimulus presentation, resulting in a model-time by neural-time dynamic RSA. Here we applied this approach to MEG data of 21 healthy human subjects who observed 14 unique 5-sec-long videos showing a sequence of smoothly connected ballet figures (~35 repetitions per video). Dynamic RSA revealed unique insights into the representation of low-level visual, body posture and kinematic features: Both low- and high-level information was represented ~ 110 - 300 msec after the actual visual input, but with low-level information being represented most prominently in occipital areas, and higher-level information in slightly more anterior (parieto-temporal) areas. Strikingly, the motion of the ballet dancer was not only represented in a lagged manner, but also in a second distinct temporal window that preceded the actual input by ~ 140 - 180 msec, indicating that these neural representations predicted future motion. Taken together, dynamic RSA reveals delayed bottom-up and predictive top-down processing in naturalistic dynamic stimuli. As such, it offers a novel approach to investigate how and when our brain represents the dynamics of the world. More generally, it can be used to study interesting concepts such as predictive coding not only in naturalistic visual stimuli, but also across a wide range of other domains such as auditory perception, language or action planning.

Disclosures: I.E.J. de Vries: None. **M.F. Wurm:** None.

Poster

P497. Human Perception II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P497.09

Topic: H.02. Perception and Imagery

Support: NIH F32MH117933
Simons Center for the Social Brain Postdoctoral Fellowship

Title: Evidence for domain-specific neural adaptation reductions in autism spectrum disorder

Authors: *A. D'MELLO[1], I. R. FROSCH[2], S. L. MEISLER[3], H. GROTZINGER[4], T. K. PERRACHIONE[5], J. D. GABRIELI[1];
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Abstract: Autism spectrum disorder (ASD) is a neurodevelopmental disorder defined by impairments in social communication and the presence of restricted interests and repetitive behaviors. There is empirical and theoretical evidence that across domains of impairment, individuals with ASD may have a reduced capacity to predict events and use predictions to rapidly adapt to changing situations. It has been hypothesized that many core diagnostic features of ASD, both sensory and cognitive, may be associated with these difficulties in adaptation, and that adaptation may be a mechanism that supports prediction by distinguishing familiar from novel events. Here, we tested the hypothesis that adults with ASD exhibit reduced neural adaptation across a broad range of materials, and that reduced neural adaptation is associated with ASD behaviors.

We assessed neural adaptation with functional magnetic resonance imaging (fMRI) in n=26 neurotypical and n=29 ASD adults. Participants passively attended to repeating and non-repeating stimuli across four domains: faces, spoken words, text, and objects. Neural adaptation was measured by contrasting activation from repeating and non-repeating blocks (non-repeating > repeating). Across all participants, neural adaptation was strongest in stimulus-specific processing cortical regions. Importantly, group differences in neural adaptation were domain specific: individuals with ASD showed reduced neural adaptation to faces in the fusiform face area (FFA) but not for other stimuli such as objects, text, or speech. Reduced neural adaptation in the FFA was associated with greater social and communication difficulties in individuals with ASD. In addition, compared to neurotypical individuals, individuals with ASD showed reduced functional connectivity between the dorsolateral prefrontal cortex and FFA when attending to repeating faces.

These findings suggest that reduced neural adaptation in ASD may be specific to domains that are most relevant to the ASD phenotype (e.g., faces for social processing). Further, differences in neural adaptation in the FFA in ASD may be driven in part by reduced top-down influences from the prefrontal cortex.

Disclosures: A. D'Mello: None. **I.R. Frosch:** None. **S.L. Meisler:** None. **H. Grotzinger:** None. **T.K. Perrachione:** None. **J.D. Gabrieli:** None.

Poster

P497. Human Perception II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P497.10

Topic: H.02. Perception and Imagery

Support: Wellcome/Royal Society Sir Henry Dale Fellowship (218535/Z/19/Z)

Wellcome Trust (203147/Z/16/Z)

Title: Determining the direction of communication between sensory cortex and hippocampus during expectation signalling

Authors: *O. WARRINGTON, N. GRAEDEL, M. F. CALLAGHAN, P. KOK;
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Abstract: The hippocampus (HC) holds a central point of interest for many different cognitive functions such as memory, spatial navigation, perception, and mental imagery. Therefore, a principal aim of cognitive neuroscience is to determine the functional role of HC interactions with the cortex. Until recently, these investigations required a trade-off between high spatial resolution with invasive animal techniques or lower resolution in human neuroimaging studies. Now, we can combine advances in ultra-high field MRI of the medial temporal lobe with layer-specific fMRI analyses to answer questions about functional connectivity between the HC subfields and the neocortex. The key to inferring the direction of communication is the entorhinal cortex, as it serves as the primary relay between the HC and cortex. Feedforward connections from the cortex travel to CA3, CA1 & DG via the entorhinal cortex superficial layers, while feedback connections travel from CA1 & subiculum to the deep layers. Therefore, by dissociating the layers of the entorhinal cortex with layer-specific fMRI, we can infer the directionality of functional communication between the HC and sensory cortex. This method could be utilised in studies crossing cognitive boundaries, and here we present a case study asking how the HC communicates with the cortex during the prediction of visual shape. At 7T, we optimised BOLD-sensitive 3D gradient-echo EPI to perform fMRI in the medial temporal lobe, with particular focus on minimising distortion and signal dropout. Participants perform an established shape discrimination paradigm in which auditory cues validly predict the upcoming visual shape on 75% of trials. Crucially, we omit the expected shape entirely on the remaining 25% of trials, allowing isolation of the top-down expectation signal from bottom-up sensory input. This paradigm, in combination with multivoxel shape decoding, allows us to determine which HC subfields and entorhinal cortex layers represent the expected-but-omitted shape. Additionally, functional connectivity analyses using the layers of the entorhinal cortex as seeds enable charting the wider cortical network involved in communication to and from the HC. This cognitive neuroscience paradigm represents a big step forward in studying the communication between hippocampus and neocortex in the human brain, a topic of great relevance to many cognitive domains ranging from memory and navigation to perception and imagery.

Disclosures: O. Warrington: None. **N. Graedel:** None. **M.F. Callaghan:** None. **P. Kok:** None.

Poster

P507. Cross-Modal Processing In Humans IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P507.01

Topic: D.09. Multisensory Integration

Support: NIH R00DC013828

Title: Successful decoding of lipreading information in auditory areas.

Authors: *G. KARTHIKEYAN[1], A. JAHN[2], C. Z. CAO[2], C. FINE[2], D. BRANG[3];
[1]Univ. of Michigan Psychology Grad. Program, Ann Arbor, MI; [3]Psychology, [2]Univ. of Michigan, Ann Arbor, MI

Abstract: Sounds play a major role in the comprehension of spoken speech. However, meaningful information provided by visual cues (such as speech content, timing, and speaker identity) facilitates speech perception, perceptually enhancing relevant information for the listener. This information includes the speaker's voice as well as articulatory movements from the speaker's face and lips. While several studies have shown broadband activation of auditory regions in response to visual speech, the information represented by these activations remains poorly understood. Indeed, our recent electrocorticography research demonstrates that visual speech modulates auditory speech processing at multiple temporal, spatial, and spectral scales, indicating multiple discrete influences of vision on speech. As a first step to understand what visual information is encoded within the auditory system, we used functional magnetic resonance imaging (fMRI) to investigate if visual activation of auditory areas reflects linguistic content from visual speech. The present study collected data from a large sample of individuals (n=64, based on a power analysis and pre-registered at osf.io) who were presented with three phoneme exemplars (consonant-vowel pairs such as /fafa/) and three visemes (videos showing articulations for each phoneme) in a randomized event-related design. To understand the extent to which linguistic information is encoded in auditory regions during visual speech, we trained and tested classifiers to discriminate between (1) the three specific phonemes, (2) the three specific visemes, (3) as well as tested phoneme-trained models' ability to classify visemes. Analyses were performed using a whole-brain searchlight classifier as well as ROI-based analyses focusing on auditory regions. Results from the searchlight analyses showed significant above-chance classification accuracy in both the phoneme and viseme conditions in auditory regions, including the posterior superior temporal sulcus (pSTS) and superior temporal gyrus (STG). Results from the ROI analysis showed a similar pattern. These results demonstrate that visual speech crossmodally activates and encodes linguistic information in auditory regions, confirming that auditory activations by visual speech are not merely due to attention or arousal effects.

Disclosures: G. Karthikeyan: None. **A. Jahn:** None. **C.Z. Cao:** None. **C. Fine:** None. **D. Brang:** None.

Poster

P507. Cross-Modal Processing In Humans IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P507.02

Topic: D.09. Multisensory Integration

Title: Deaf people show a weaker flash-lag illusion

Authors: *M. AMADEO, A. TONELLI, C. CAMPUS, M. GORI;
Italian institute of technology, genova, Italy

Abstract: Deaf people show a weaker flash-lag illusion
SUMMARYAccording to the flash-lag illusion (FLI), a brief flash quickly presented with a moving target appears to lag behind the moving stimulus. Over the years, numerous theories have been proposed to explain FLI, discussing whether it is more a spatial or a temporal phenomenon. However, none of them has ever investigated whether and how auditory deprivation interferes with the development of this illusion. Studying how the lack of auditory stimulation influences a visual illusion may give important insights into its underlying mechanisms and the potential role of multisensory processes. Since deafness is a natural model to investigate how neural reorganization following hearing loss influences perception, we tested a group of 13 early deaf and 18 hearing individuals with a visual FLI paradigm. The experiment consisted of two vertically aligned bars moving rightwards or leftwards and a static bar that briefly flashed between them. The static bar could appear in two different portions of the visual field on the horizontal plane: i) within the fovea or ii) in the periphery of the visual field. The instruction was to judge whether the flashed bar spatially appeared to the left or the right with respect to the moving bars. Results showed a reduction of the flash-lag effect following early deafness, independently of the spatial eccentricity of the static bar (i.e., central and peripheral visual field). Moreover, only for deaf individuals, there was a positive correlation between the flash-lag effect in the peripheral and central visual field, suggesting that the mechanisms underlying the effect in the center of the visual field expand to the periphery following deafness. These findings suggest that plastic changes due to early auditory deprivation profoundly impact visual mechanisms underlying the FLI, and multisensory integration processes are involved in developing the illusion.

Disclosures: M. amadeo: None. **A. Tonelli:** None. **C. campus:** None. **M. Gori:** None.

Poster

P507. Cross-Modal Processing In Humans IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P507.03

Topic: D.09. Multisensory Integration

Support: JSPS Grant JPMJTR20RR

Title: Effects of presenting first-person camera images in the background of a book on motion sickness when reading with a head-mounted display

Authors: *T. SUWA[1], Y. SATO[2], M. MAKIDA[1], T. WADA[2];
[1]Ritsumeikan Univ., Kusatsu-Shi, Japan; [2]Nara Inst. of Sci. and Technol., Ikoma-Shi, Japan

Abstract: <META NAME="author" CONTENT="諏訪 司(is0414if)">With the development of virtual reality (VR) technology, head-mounted displays (HMDs) are often used to view or read 2D content, such as videos and books, in a vehicle; however, when the 2D content are displayed in a fixed position on the display (head coordinate system), the visual appearance does not change even with head movement, and the body movements cannot be perceived from visual sensation. This cause conflicts with the body-movement information obtained from vestibular sensation. This visual–vestibular conflict may also cause severe motion sickness. However, previous studies have not clarified the means to reduce motion sickness when watching 2D content on HMDs while riding in a vehicle. Therefore, the purpose of this study is to investigate whether the motion sickness in reading with an HMD can be reduced by allowing readers to perceive body movements through visual sensation in situations where the body vibrates, such as riding a vehicle. Participants performed 20-minute reading tasks in which they read a book displayed on a HMD while seated in a device that vibrated randomly in the pitch direction. Because the book was fixed to the head coordinate system of the HMD, the appearance of the book was constant, and the participants could not perceive body movements through visual sensation. Two experimental conditions were explored: the first was the camera background condition in which the user could perceive his or her body movements through peripheral vision via real-time images of the first-person viewpoint obtained from the front camera mounted on the HMD in the background of the book; the second was a white background condition in which the background of the book was completely white (no movement) and the participants could not visually perceive their movements. The misery scale of motion sickness after the reading task was evaluated using a 11-point questionnaire, and the results indicated that motion sickness was significantly lower in the camera background condition than the white background condition. This suggests that when watching 2D content on a HMD in situations where the body is in motion, such as riding a vehicle, the sensory conflict between visual and vestibular sensations can be resolved and motion sickness can be reduced, provided the user perceive his or her motion information in the peripheral vision through the camera image, even if the user gazes at the 2D contents fixed in the head coordinate system in the central vision.

Disclosures: T. Suwa: None. **Y. Sato:** None. **M. Makida:** None. **T. Wada:** None.

Poster

P507. Cross-Modal Processing In Humans IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P507.04

Topic: D.09. Multisensory Integration

Support: NIH R00DC013828

Title: Quick as a flash: Electrocorticographic (ECoG) recordings show that sounds influence visual perception by speeding up visual responses in motion sensitive visual cortex (hMT+)

Authors: *C. FINE[1], W. C. STACEY[2], D. BRANG[1];
[1]Psychology, [2]Neurol., Univ. of Michigan, Ann Arbor, MI

Abstract: The natural environment presents imperfect information to each of our sensory systems. To make sense of ambiguous signals, the brain readily integrates information from multiple modalities through crossmodal processes, weighting each sense by its reliability. However, the neural mechanisms through which crossmodal influences are coordinated in humans remain poorly understood, particularly when the senses receive conflicting information. The double-flash illusion (DFI; also known as the sound-induced illusory flash effect) is a powerful example of how sounds can bias perception and change what is seen, even when it results in lower perceptual accuracy. In this illusion, a single visual flash surrounded by two brief sounds evokes the experience of two discrete flashes. Previous research has shown that the predisposition to see an illusory flash is modulated by prestimulus activity, and that the illusion is typically associated with reduced alpha, increased beta, and increased gamma power, localized to early visual areas and the STS. However, non-invasive imaging (EEG, MEG, and fMRI) lack the combined spatial and temporal resolution necessary to definitively identify where and how in the brain sounds modulate visual perception during this illusion. To address this gap, we recorded neural activity during the DFI from two patients who were undergoing clinical evaluation for epilepsy using electrocorticography (ECoG). Intracranial electrodes implanted within visual motion cortex (hMT+) were analyzed across unisensory trials (auditory-alone, visual-alone) and crossmodal trials that included the illusion (one flash + two beeps) and control conditions. Results showed a consistent pattern across the two patients, such that sounds elicited greater alpha suppression, increased high-gamma power (a correlate of population firing rates), and earlier visual response latencies. Moreover, these enhancements were greater on trials in which the illusion was experienced (one flash perceived as two). We hypothesize that these power and latency effects are due to phase-resetting mechanisms in visual cortex, in which incoming sounds increase the excitability of visual neurons before the visual stimulus is processed.

Disclosures: C. Fine: None. **W.C. Stacey:** None. **D. Brang:** None.

Poster

P507. Cross-Modal Processing In Humans IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P507.05

Topic: D.09. Multisensory Integration

Support: NIH Grant R00DC013828

Title: Auditory Cortex Uses Visual Timing Information to Enhance Auditory Speech Perception

Authors: *C. CAO[1], K. GANESAN[1], W. C. STACEY[2], D. BRANG[1];
[1]Psychology, [2]Neurol., Univ. of Michigan, Ann Arbor, MI

Abstract: Seeing a speaker's face facilitates accurate speech perception. However, how such visual speech facilitation is achieved in the human brain remains poorly understood. Previous studies have shown that visual signals can modulate the excitability of auditory cortex. Indeed, recently our group observed that visual speech has spectrally, temporally, and spatially distinct influences on auditory processing, but the information encoded in the auditory system remains unclear. We hypothesize that several types of auditory information are recovered from visual signals, including categorical linguistic representations, predictive and ongoing timing information, relative pitch information, as well as speaker identity. The present study examines the prediction of auditory timing information (e.g. the onset/offset of phonemes and the general rate of speech) extracted from visual speech signals. Such visual timing information has been shown to be encoded by theta band oscillations in the anterior and posterior superior temporal gyrus (STG), but its influence on speech perception accuracy has remained less clear. To address this more directly, the current study examined the impact of visual speech information on auditory timing precision using intracranial recordings (electrocorticography; ECoG) in a large group of patients with epilepsy (n = 21). Group-level analyses using linear mixed-effect models showed that congruent visual speech elicited theta power increases in the posterior superior temporal gyrus (pSTG) following auditory speech onset compared with auditory speech alone. Separately, we observed increases in inter-trial phase coherence (ITPC) at the anterior STG prior to sound onset in response to visual lip-movements (potentially predicting phoneme onset time). Finally, ITPC was increased for congruent (relative to incongruent) auditory-visual trials and maximal when auditory-visual trials were correctly identified. We interpret these results to mean that visual speech is improving the precision of auditory speech in part through predictive timing information.

Disclosures: C. Cao: None. **K. Ganesan:** None. **W.C. Stacey:** None. **D. Brang:** None.

Poster

P507. Cross-Modal Processing In Humans IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P507.06

Topic: D.09. Multisensory Integration

Support: JST Grant JPMJTR20RR

Title: Effects of presenting motion prediction information using augmented reality on motion sickness

Authors: *Y. SATO[1], T. ISHIKAWA[2], T. WADA[1];
[1]Nara Inst. of Sci. and Technol., Ikoma-Shi, Japan; [2]Ritsumeikan Univ., Kusatsu-shi, Japan

Abstract: <META NAME="author" CONTENT="佐藤勇起">According to the sensory conflict theory, motion sickness results from a mismatch between sensed and expected motions. Previous studies have demonstrated that motion sickness in a car could be reduced by presenting motion prediction information, for example, in the peripheral visual field using LED lights. However, the effects of presenting predictive information using augmented reality (AR) on motion sickness have not been investigated. AR may be more intuitive since it can superimpose motion prediction information over real space for viewing in the central visual field without large eye movements. Therefore, this study aims to verify how motion prediction information presented in AR using a head-mounted display (HMD) affects motion sickness. A participant wearing a HMD sat on a seat rotated in a range of ±30° in the yaw direction for 10 min at a preset irregular rotational direction and velocity, so that the end of the current or upcoming motion cannot be predicted. This motion prediction information was presented as an arrow in the AR space of the HMD. The rotational direction was indicated by the arrow, and the stop position was indicated by the arrow tip. The arrows were arched and placed around the participant's head to enable viewing the motion prediction information in the central visual field without large eye or head movements. New arrows were presented once the stopped seats began moving again to indicate the next rotational direction and stop position. Two experimental conditions were considered, namely with and without the arrows in AR space. The misery scale (MISC) of motion sickness was evaluated every minute during the rotations using a 11-step questionnaire. The results showed that the average MISC was significantly lower with the arrow information than without ($p = 0.0039$, Wilcoxon signed-rank test). The results of the study suggest that motion sickness could be reduced by presenting motion prediction information using AR.

Disclosures: Y. Sato: None. **T. Ishikawa:** None. **T. Wada:** None.

Poster

P507. Cross-Modal Processing In Humans IV

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Program #/Poster #: P507.07

Topic: D.09. Multisensory Integration

Support: JSPS, 16H05958
JSPS, 19H00631
JST, JPMJCE1311

Title: Rubber hand illusion related EEG oscillation in real and VR environment

Authors: *N. KANAYAMA[1,2], M. HARA[3], K. KIMURA[1];
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Abstract: Virtual reality (VR) enables highly controllable experimental body image and body schema setting. The rubber hand illusion (RHI), which is a typical illusory experience induced by visuo-tactile stimulations, can be realized in VR settings. Also, some phenomena related bodily illusion, such as full-body illusions, require head-mounted display (HMD), which can show the VR environment. However, when we measure scalp EEG during VR experience, HMD must add any noise on the EEG waveforms. To elucidate the difference of RHI related neural oscillation between real and VR environment, we have to control the noise contamination caused by usage of HMD. In this study report, I have introduced the way to control HMD-related noise contamination on the scalp EEG recording. Then under this noise-controlled condition, we have recorded scalp EEG components related to the the multisensory integration process during RHI in VR. Multisensory integration process can be altered in VR compared to real environment, because the visual scene is a not ordinary one when we wear HMD. Especially, the visual stimuli are only presented through the lens located near eyes, implemented in HMD, and the light from the room lighting is fully occluded. The objects seen in the field of view of HMD was created in 3D computer graphics, which mimicked the real things but not fully photorealistic. Although the 3DCG VR environment mimicking the real experimental settings can be subjectively felt very similar to the real one, the information process of visuo-tactile integration in VR is possibly different from real one. One useful investigative approach is measuring brain activities related multisensory integration process during RHI. We compared EEG data obtained in both real and VR environments, in which RHI task using vibration elements and LED as visuo-tactile stimuli. As a result, we observed the gamma and theta band oscillatory activities. Importantly, we found statistically significant differences between congruent (RHI) and incongruent (not RHI) conditions in the real environment, which agrees with previous studies. Only late theta activity, which is increased to incongruent visuotactile stimulation preventing RHI in real condition, has difference between real and VR environment, suggests that we have any alternation of multisensory process. The theta activities can be considered as the cognitive load in the visuotactile integration process based on the previous studies. Thus, we have to consider such a difference when we model the multisensory integration in VR to investigate our bodily self-perception.

Disclosures: N. Kanayama: None. **M. Hara:** None. **K. Kimura:** None.

Poster

P539. Cortical Planning and Execution: Fronto-Parietal Stream: Neurophysiology: Animal

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P539.01

Topic: E.04. Voluntary Movements

Support: Banting Research Foundation (D.F.C.)
Natural Sciences and Engineering Research Council of Canada (NSERC RGPIN-2017-06434, D.F.C.)
Canadian Foundation for Innovation (CFI-JELF 37382, D.F.C.)

Title: A map of muscle coactivation in rat frontoparietal cortex revealed with intracortical microstimulation (ICMS)

Authors: J.-J. PÉREZ-CORDERO, M. HAFEZI, L. A. GROCHOWSKI, B. K. NG, S. R. U, ***D. F. COOKE**;
Biomed. Physiol. and Kinesiology, Simon Fraser Univ., Burnaby, BC, Canada

Abstract: Muscles and joints rarely operate in isolation. Consistent with this, intracortical microstimulation (ICMS) at single sites in motor and parietal cortex often activates muscles across multiple joints and the activity of single motor cortex neurons is correlated with movement of multiple muscles. Yet many studies focus on the single muscle or joint most strongly linked to a distinct location in cortex, assuming that more widespread ICMS activation of muscles is artifactual. In contrast, we have shown previously that ICMS evokes coordinated combinations of muscle activation closely resembling natural complex behaviors like reaching, feeding, and defensive movements. Moreover, by targeting an ICMS-defined defensive zone with GABAergic drugs, we could selectively up- and down-regulate natural defensive behavioral reactions, demonstrating a functional role in specific multi-joint behaviors. Thus, patterns of muscle coactivation by ICMS reflect localized coding of complex motor behaviors, a finding since replicated by others. With the past focus on a primary muscle or joint, however, little is known, about how multiple muscles are coordinated by motor and parietal cortex. A few studies have examined patterns of arm muscle activity in monkeys during ICMS, and we have previously studied complex ICMS-evoked movements in rats, but without data from individual muscles. Here we describe a systematic study of muscle activity evoked by stimulation at sites across frontoparietal cortex. We applied ICMS in anesthetized rats to evoke muscle movements while recording video and electromyographic (EMG) activity from multiple muscles ranging in size and spanning the face, forelimb, trunk, and hindlimb. ICMS comprised 500-ms trains of biphasic pulses presented at 200 Hz with current amplitude ranging from 5-320 μA. While 500-ms ICMS is “long train”, we also analyzed the initial “short train” 50-ms period of EMG. We optimized stimulation circuitry to minimize ICMS artifact in EMG signals. Stimulation was controlled by a novel closed-loop system that conducts preliminary EMG analysis in real time and pauses ICMS during spontaneous muscle activity. Combined EMG and video data showed

that ICMS of sites over a large proportion of excitable cortex in M1 and S1 evoked coactivation of multiple muscles, which together frequently acted on more than one joint or body part. EMG data provided more precise latency information (and thus relative timing of co-activated muscles). For the first time these muscle-specific data are presented for motor maps spanning the full body representation.

Disclosures: J. Pérez-Cordero: None. **M. Hafezi:** None. **L.A. Grochowski:** None. **B.K. Ng:** None. **S.R. U:** None. **D.F. Cooke:** None.

Poster

P539. Cortical Planning and Execution: Fronto-Parietal Stream: Neurophysiology: Animal

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P539.02

Topic: E.04. Voluntary Movements

Support: NSF Grant 1656882

Title: Signals from different layers of posterior parietal cortex area 5b to M1 during simple and vision-guided locomotion

Authors: *I. N. BELOOZEROVA[1], W. U. NILAWEERA[2];
[1]Georgia Inst. of Technol., Atlanta, GA; [2]Div. of Neurobio., Ellsworth Community Col., Iowa Falls, IA

Abstract: Area 5 of posterior parietal cortex is a part of the cortical pathway that processes visual information for action conveying it from visual to motor cortex (M1), the "dorsal stream". While a number of studies investigated transmission of information in the beginning of the dorsal stream, the composition of signals that area 5 ultimately conveys to M1 remained unknown. We investigated the activity of area 5b neurons from most rostral aspect of the suprasylvian gyrus that send axons to M1, cortico-cortical neurons (CCs). In cats, we compared their activity between locomotion on the flat surface and horizontal ladder, thereby identifying components of the activity related to visual processing versus limb movement. Activity of 54 CCs with somata in cortical layers II-IV, 21 in layer V, and 10 in layer VI was analyzed. To understand how well CCs reflect activity of the respective layers, we investigated activity of neighboring neurons with non-identified projection of axon (noIDs): 58 in layers II-IV,162 in layer V, and 3 in layer VI. During locomotion on the flat surface, noIDs in layer V at ~13 spikes/s were twice more active than in layers II-IV. More neurons discharged one activity burst per stride than in layers II-IV, and less had two bursts. The activity of layer V groups was stride-related: one-bursters peaked during swing, two-bursters peaked during stance. The activity of layers II-IV groups was barely modulated. Discharge of individual CCs was stride-related showing same one-/two-burst patterns but the rate was only1/5th of noIDs. CC group activity was not stride-related in any layer. Thus,

these CC groups did not transfer any unified stride-related signal to M1, while individual CCs sent various low rate signals. Upon transition to the ladder, noIDs increased discharge, activity of more cells became stride-related. Proportion of two-bursters doubled, most gain occurring in layer V. In layer V, group of one-burst noIDs developed an additional activity peak during swing-to-stance transition, two-bursters formed an additional peak in early stance. Discharge of CCs doubled, proportion of two-bursters also increased and their activity became stride-related with two peaks that preceded peaks of two-burst noIDs by 15% of the stride. The group activity of one-burters remain steady. In layers II-IV, activity of two-burst noIDs became modulated with peaks during stance-to-swing and swing-to-stance transitions, while activity of one-bursters remained stride-unrelated. Activity of CC subgroups was barely modulated. Thus, layers II-IV CC groups did not transfer unified stride-related signal to M1, while layer V two-burst CCs transmitted an early vision-related signal.

Disclosures: I.N. Beloozerova: None. **W.U. Nilaweera:** None.

Poster

P539. Cortical Planning and Execution: Fronto-Parietal Stream: Neurophysiology: Animal

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P539.03

Topic: E.04. Voluntary Movements

Support: Ministero dell'Università e della Ricerca (PRIN-2017KZNZLN)
European Commission, H2020-EIC-FETPROACT-2019-951910-MAIA

Title: Anterior-posterior gradient in the joint processing of reach direction and depth in the macaque posterior parietal cortex

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Abstract: Accurate performance of arm movements depends on the integrity of a network of areas located in the parietal cortex that integrate visual information about the location of objects in space with visual and proprioceptive information about hand position, to guide arm movements. Although every arm movement happens in 3D space, it has become traditional among researchers to make a distinction between the direction of a movement and its depth. Psychophysical studies suggest that the direction and depth of reaching movements are likely to be specified independently, however there is no clear physiological evidence of this segregation. The present work addresses this gap of knowledge and provides with a comprehensive model of how movement-related areas contribute to different phases of complex 3D movements. Neural activity was recorded from 899 neurons in three distinct parietal cortex areas in *Macaca*

fascicularis monkeys while they fixated and reached towards visual targets located at different distances and directions from the body. We used information theory, singular value decomposition and dimensionality reduction methods and compared direction and depth strength and convergence within and across the three parietal areas. We found two major categories of cells: a) neurons that encoded both distance and direction information, and b) cells encoding only one type of signal. We compared direction and depth processing across task progress. While direction information was stronger than depth during early planning, depth signals became dominant towards movement execution. Going from anterior to posterior locations, cells that processed both signals consistently increased. These findings suggest a significant degree of sequential direction and depth processing that supports behavioral evidence. In addition, they reveal a gradient-like organization of joint versus independent control of reach parameters in parietal cortex that could be viewed in light of its role in visuo-motor transformations during actions.

Disclosures: K. Hadjidimitrakis: None. **M. De Vitis:** None. **M. Filippini:** None. **M. Ghodrati:** None. **P. Fattori:** None.

Poster

P539. Cortical Planning and Execution: Fronto-Parietal Stream: Neurophysiology: Animal

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P539.04

Topic: E.04. Voluntary Movements

Support: NIH Grant EY02686

Title: Thalamocortical and corticothalamic connections of functional domains in posterior parietal cortex of galagos

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Abstract: The posterior parietal cortex (PPC) of prosimian galagos (Otolemur garnetti) is subdivided into several functionally distinct domains from which specific types of complex movements (e.g., reach, climb, grimace) can be evoked by 500 ms long trains of electrical stimulation. Previous studies have shown that each PPC domain has unique corticocortical connection patterns. However, connections of these domains with the thalamus remain unclear. Here, we investigated corticothalamic and thalamocortical connections of movement-specific PPC domains in seven adult galagos. We made eight injections of retrograde and bidirectional tracers, including cholera toxin B subunit (CTB), biotinylated dextran amines (BDA) and fluoro-ruby (FR) in different movement domains (i.e. hindlimb, forelimb and face movements). Thalami were cut in horizontal, coronal and sagittal planes across cases to gain different view of

thalamic nuclei. Sections were divided into series, some of which were processed for Nissl substance, acetylcholinesterase or vesicular glutamate transporter 2 for architecture. Other series were processed either for CTB or BDA, or FR for labeled neurons and axon terminals. Plotting from brain sections revealed that all PPC domains investigated in this study received major inputs from the ventral lateral nucleus (VL), the posterior nucleus (Po) and the anterior pulvinar (APul). Additional labeled neurons were distributed in the ventral anterior (VA) and lateral posterior nuclei (LP). Occasionally, we observed labeled neurons in ventral posterior (VP), intralaminar (IL) and mediodorsal nuclei (MD). PPC domains mainly projected to VA, LP, VL, Po and APul, with the densest projections concentrated in VL and Po/APul. Most of densely labeled neurons and axon terminals were spatially overlapped, suggestive of reciprocal connections between PPC functional domains and these thalamic nuclei. These preliminary results from prosimian galagos appear to be largely consistent with related results from New world and Old world monkeys.

Disclosures: Q. Wang: None. **I. Stepniewska:** None. **C. Liao:** None. **J. Kaas:** None.

Poster

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Topic: E.04. Voluntary Movements

Support: NEI Grant EY012135
Washington University Cognitive Computational and Systems Neuroscience Fellowship

Title: Inter-areal communication in posterior parietal cortex during coordinated eye and arm movements

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Abstract: Behavior usually involves coordinated movements to different locations in space, and different parts of the nervous and musculoskeletal systems must communicate to plan and execute coordinated movements. However, the neural mechanisms of information exchange between different brain regions during eye-hand coordination are not fully understood. Posterior parietal cortex (PPC) in the macaque brain has been implicated in spatial processing during eye-hand coordination. A long-standing controversy concerns whether the lateral intraparietal area (LIP) in PPC is a locus for spatial attention or oculomotor control. Attention and eye movements

are confounded by the fact that we usually look at whatever we are attending to. We studied information flow between LIP and the parietal reach region (PRR), a nearby area involved in reaching. We recorded spikes and local field potential (LFP) in LIP and PRR while animals were instructed to make coordinated eye and arm movements. If LIP is an attentional filter for spatial information, then information should flow from LIP to PRR when a target for a reach is being identified. However, if LIP primarily subserves an oculomotor control, then information should flow from PRR to LIP so that animals can look at what they are reaching for: eye-hand coordination. We used two largely independent methods, spectral Granger causality between two LFP signals and spike-LFP coherence analyses in beta band, to assay information flow between the two areas during the movement preparatory period. Both methods suggest that information primarily flows from PRR to LIP rather than vice versa. Our results support the idea that LIP subserves primarily an oculomotor rather than an attentional role during eye-hand coordination.

Disclosures: J. Kang: None. **E. Mooshagian:** None. **L.H. Snyder:** None.

Poster

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Title: Predictive coding of tracking and intercepting inferred moving target in posterior parietal cortex

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Abstract: Benefit from “the predictive brain” which compensates for inevitable delays in sensory transmission and movement execution, we can survive in a dynamic world. Although the delay compensation has been widely studied, the underlying neural mechanism is still elusive. As an important sensorimotor interface, the posterior parietal cortex (PPC) has been found profoundly involved in sensory-guided movement. To further elucidate whether the PPC estimates current state of external stimulus or predicts consequence of an upcoming movement,

we recorded single-unit activity from the lateral intraparietal area (LIP, n=86) and parietal reach region (PRR, n=65) while a monkey (*Macaca mulatta*) performed the occluded manual interception task. During the task, the monkey initiated a trial by touching a central dot for 400 ms, then a target appeared at a random location of a circle and moved circularly with one of five angular velocities (0, static control; 120°/s or 200°/s clockwise, 120°/s or 200°/s counterclockwise) for 400-800 ms (cue period) and then disappeared. After the occlusion period of 500 ms, the center dot dimmed (GO cue) and the monkey was required to manually intercept the target inferred from the previous target motion during the cue period within 700 ms. The activity of LIP and PRR revealed a gradient from sensory to movement coding in the occlusion period. Most of neurons (~75%) in both areas were tuned to the instantaneous target location when it was visible. Once target disappeared, 45 (69%) PRR neurons and 39 (45%) LIP neurons were tuned to the upcoming reaching direction, whereas 28 (33%) of LIP represented inferred stimulus location, this type of neuron accounts for only 12 (18%) of PRR. In addition, a small proportion of neurons (~8% in each area) exhibited a transition from tracking of moving target to forming movement plan. However, the target tracking coding in above groups of neurons only lasted in the early occlusion period for ~200 ms. Therefore, during planning manual interception of inferred moving targets, most PRR neurons function as reach plan formation instead of instantaneous target estimation, while LIP neurons might subserve extrapolating stimulus state and forming motor plan. During the transformation process, we observed the interaction between sensory and movement modulation, indicating the role of evaluating or converting the sensory information into movement output in PPC. The encoding properties in PPC neurons revealed that it is not only involved in establishing the internal estimation of sensory state, but also forming impending motor plan in predictive sensorimotor control.

Disclosures: R. Zheng: None. **H. Cui:** None.

Poster

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Program #/Poster #: P539.07

Topic: E.04. Voluntary Movements

Support: NIH Grant EY030998
Schmitt Program on Integrative Neuroscience
Del Monte Institute for Neuroscience

Title: Do marmosets reach predictively for moving targets?

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Abstract: Visual information plays a critical role in the planning and on-line correction of coordinated movements. Primates especially utilize vision in reaching behavior. Studies of reaching in macaques indicate that the dorsal premotor and the medial intraparietal cortices are involved in on-line corrections that guide the hand towards dynamic targets. However, the specific behavioral contributions of circuit connections between these two areas have not been determined, due to technical challenges associated with circuit targeting and behavioral training in macaques. Marmosets provide an accessible experimental model for this research due to their lissencephalic brains that facilitate intraparietal targeting and innate insect hunting behavior that facilitate dynamic reach testing. We set out to generate high fidelity descriptions of marmoset hands as they reach for live crickets and ask whether unrestrained marmosets reach predictively for moving crickets. We developed a novel machine learning approach to achieve marker-less tracking of the hand and the cricket, and determined their spatiotemporal interplay during reaching. We utilized kinematic data and a multivariate linear modeling approach to demonstrate the influence of cricket motion on the future hand motion of reaching marmosets. Our findings suggest that marmosets incorporate cricket motion into their reaches in a predictive fashion. Furthermore, there is a specific time window when cricket motion strongly influences marmoset hand motion. These behavioral characterization and modeling results will provide a foundation for future research using optogenetic tools in marmosets to determine the circuit mechanisms underlying predictive reaching and online corrections.

Disclosures: L.H. Shaw: None. **K.H. Wang:** None. **J. Mitchell:** None.

Poster

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Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P539.08

Topic: E.04. Voluntary Movements

Title: Configuration of sensory, parietal, and frontal cortices during goal-directed virtual navigation in non-human primates

Authors: *J.-P. NOEL[1], E. BALZANI[2], E. AVILA[2], S. BRUNI[2], P. ALEFANTIS[2], C. SAVIN[2], D. ANGELAKI[3];
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Abstract: We do not understand how neural nodes operate within the recurrent action-perception loops that characterize our natural environment, nor how brain networks change with different computational demands. Here, we simultaneously record spiking activity from the dorso-medial superior temporal area (MSTd), area 7a in the posterior parietal cortex, and dorsolateral pre-frontal cortex (dlPFC) as monkeys attempt to “catch fireflies” in virtual reality. This task

required animals (1) to integrate optic flow signals into a position estimate over protracted periods of time, (2) to actively sample from a dynamic environment (e.g., via eye movements), and (3) to compute the evolving location of a latent variables - i.e., the angular and radial distance to target and from origin. We leveraged a novel Poisson Generalized Additive Model (P-GAM; Balzani et al., 2020) capable of inferring marginal confidence bounds for the contribution of each feature to neural responses. This technical innovation allowed us to identify a minimal set of task features that each neuron is responsive to while circumventing the need for computationally prohibitive model selection procedures (particularly given the number of variables in this naturalistic task). We observed mixed selectivity, with even MSTd tracking the latent variable. Strikingly, MSTd showed a global encoding profile akin to that of dlPFC and very distinct from 7a. We also observed significant unit-to-unit coupling (i.e., firing rate of one neuron impacting that of another) between MSTd and dlPFC. In contrast, single unit coupling between 7a and either dlPFC or MSTd was less common. Lastly, when sensory evidence was scarce and instead the task heavily relied on pre-established internal models, lateral connectivity within MSTd strengthened but its pattern remained fixed. On the other hand, the shape of the coupling filters within 7a and dlPFC adaptively re-configured when the task relied on internal models as opposed to sensory evidence. Together, these results highlight the distributed nature of naturalistic neural coding and suggest internal models may be housed in the pattern of fine-grain lateral connectivity within parietal and frontal cortices.

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Poster

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Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P539.09

Topic: E.04. Voluntary Movements

Title: Neural correlates of naturalistic decision-making in parietal and frontal cortices

Authors: *X. NIU, E. BALZANI, J.-P. NOEL, B. CAZIOT, E. AVILA, C. SAVIN, D. E. ANGELAKI;
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Abstract: Classical decision-making experiments usually involve an artificial stimulus (e.g., random dots), a predefined motor response (e.g., a saccade), extensive training, and thus poorly reflect behavior and neural circuitry in a naturalistic setting. To address these limitations, we modified a "firefly task" (see Noel, Balzani et al., SfN 2021) wherein monkeys actively path integrate to a briefly flashed target in a virtual reality environment. Specifically, after training with a single firefly, during testing we presented monkeys with two identical fireflies

simultaneously, so they now had to make a choice. We recorded single units from area 7a in the posterior parietal cortex, and dorsolateral pre-frontal cortex (dlPFC), which are involved in sensorimotor transformations and working memory. Not only did monkeys adapt to this new paradigm, but they also showed a preference for closer and more straight-ahead targets, a strategy that maximizes reward rate. To study the neural correlates of this decision-making process, we fit a Poisson Generalized Additive Model (P-GAM; Balzani et al., 2020) on smoothed spike counts. Most units exhibited mixed selectivity, with units in area 7a more tuned to sensorimotor variables (velocity, acceleration, target on/off and movement on/off) and LFP, and units in dlPFC more tuned to latent variables (distance traveled, distance to target). More units encoded the chosen target than those for the unchosen target, and this difference was larger when the target pair was far apart. To interrogate time-dependent effects, we trained a linear regression that decoded the distance to target from neural responses at each time point. Throughout the trial, the firing rates were better able to predict distance to the chosen target than the unchosen one, and the decoding performance peaked a short time after targets were extinguished. Interestingly, decoding showed another smaller peak when animals stopped moving, potentially revealing a "replay" or a different distance-coding mechanism when animals are stationary versus moving. Lastly, on a subset of trials monkeys appeared to change their mind, where they first leaned towards one target, then made an abrupt turn to approach the other target instead. Neurally, both targets could be decoded early in the trial, while decoding performance dropped quickly for the unchosen target and continued increasing for the chosen one. These results suggest an ongoing competition between the neural representations of two targets, which on occasions can lead to a behavioral change-of-mind consequence.

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Poster

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Title: Beta and gamma oscillations in marmoset frontal motor cortex during natural locomotor behavior

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Abstract: In arboreal environments, substrate orientation modifies body axis orientation and biomechanical constraints on fore- and hindlimbs, resulting in adjustments of postural and locomotor strategies. To characterize the role of cortical oscillations in integrating these demands, we recorded electrocorticographic activity from left dorsal premotor, primary motor, and supplementary motor cortices of three common marmosets moving on a branch-like small-diameter pole, fixed horizontally or vertically. Power spectrum was calculated in β (16-35 Hz) and γ (75-100 Hz) bands and differences between conditions were statistically assessed by aligned-rank-transform ANOVAs followed by Wilcoxon rank-sum tests. Results showed that animals displayed behavioral adjustments to the task in terms of preferred forelimb posture and gait. The horizontal condition mainly induced quadrupedal walk with pronated/neutral forelimb postures, whereas the vertical condition induced walk and bound gaits with supinated/neutral postures. Examination of cortical activity supports the view that β and γ oscillations fulfill partly different roles during the task. At rest, modulation of β power by substrate orientation (horizontal/vertical) could illustrate sensorimotor processes for postural maintenance, in line with close links between β oscillations and tonic muscle contraction. During locomotion, modulation of γ event-related synchronization by substrate orientation and epoch (preparation/execution) highlights precise tuning to the temporal dynamics of the task. By comparison, modulation of β event-related desynchronization by gait could depict a role in movement sequence and interlimb coordination; we do not exclude a possible similar role of γ frequencies, which could be addressed by future experiments. Overall, these results confirm that arboreal locomotion and postural maintenance in common marmosets rely on specific mechanisms involving adjustments of β and γ oscillatory activity to substrate orientation.

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Poster

P614. Stress and Cognition in Human Subjects

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P614.01

Topic: F.04. Stress and the Brain

Title: Detrended fluctuation and coherence under emotional burnout formation (Exhaustion stage)

Authors: *S. TUKAIEV[1], D. HARMATIUK[2], A. POPOV[2], M. MAKARCHUK[1];
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Abstract: The neurophysiological mechanisms of emotional burnout remain insufficiently studied. It was found that EEG changes during burnout differ from biomarkers of depression and chronic fatigue syndrome. To establish the EEG markers of burnout was our aim. 182 volunteers, first-third year students from the Taras Shevchenko National University of Kyiv aged 18 to 24 years participated in this study. The EEGs were recorded during the resting state (3 min, closed eyes condition) monopolarly using EEG 23-channel system Neurocom. To establish EEG correlates of emotional burnout during rest state we used a special software written in Python 3.6 to implement Power Spectral Density calculation, the interhemispheric and intrahemispheric average coherence and Detrended Fluctuation Analysis (DFA). We analyzed separate artifact-free EEG segments in all frequency bands from 0.2 to 45 Hz. Psychological testing was performed before the registration of EEG. To determine the formation of emotional burnout Boyko's "Syndrome of emotional burnout" Inventory was used. The Exhaustion phase of emotional burnout was formed in 13 participants, and it was under development in 47 participants. In background EEG activity during the development of the exhaustion variations in EEG spatial synchronization were observed in low- and high-frequency EEG components. Development of exhaustion includes breaking of parietal-occipital links (alpha2,3-subbands) and formation of interhemispheric prefrontal, anterior frontal, and frontal links (alpha1,2-subbands). There were interruptions of coherence in the beta1 (interhemispheric frontal-frontal and parietal-parietal links) and beta2 subbands (right interhemispheric frontal link). DFA describes the long-term temporal correlations in the cortex, which are involved in different aspects of brain functioning. We detected a high resting state DFA scaling exponent values (up to 0.70-0,95) under exhaustion development in the theta1 (left prefrontal area), theta2 (right frontal area), alpha3 (posterior regions), beta2 (left temporal region). Obtained values of DFA exponent and average coherence suggest the exhaustion formation is accompanied by the changes in visual and verbal processing, emotional processes (discretion and analysis).

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Poster

P614. Stress and Cognition in Human Subjects

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Program #/Poster #: P614.02

Topic: F.04. Stress and the Brain

Support: JSPS KAKENHI 17H01758

Title: Acute changes in default-mode functional connectivity by cognitive fatigue

Authors: *S. IWAKI[1,2], T. FUJIWARA[2,3];
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Abstract: Sustained engagement to a monotonous cognitive task causes a decrease of cognitive resources accompanied by subjective feeling of mental exhaustion, which is called “cognitive fatigue (CF)” (Pattyn, 2018). In this study, we aim to elucidate the CF-related changes in brain state by employing functional MRI to evaluate resting-state functional connectivity between brain regions to investigate changes in macroscopic brain networks under CF. Eighteen young adults (22.8±1.8 years of age, all right-handed) participated in the study. The experiment includes two days of lab visits, separated by more than a week. On each visit, the participants conducted two sessions of MRI data acquisition separated by either cognitive task sessions for fatigue loading (CF condition) or passive environmental video viewing sessions as control. The order of CF and control conditions was randomized between participants. The fatigue loading in the CF condition consisted of 4 sessions of 30-minutes continuous simple mental arithmetic task with 5 minutes of interval. MRI scans were conducted before and after the fatigue loading/control sessions, which include T1 structural scans and 12-minutes resting-state fMRI (rsfMRI) scans (TR=2,000 ms, TE=35 ms, FA=90 deg, 31 slices with 3.7 mm thickness) using 3 T scanner (Philips Ingenia 3.0T). CONN software (Whitfield-Gabrieli, 2012) was used to analyze the effective connectivity between brain regions. This study was approved by the internal review board of AIST. Results of the connectivity analysis of the rsfMRI data showed significant increase in effective connectivity between the posterior cingulate cortex (PCC), which is a part of the default mode network (DMN), and the bilateral inferior occipital gyrus (IOG) ($p < 0.05$; FDR corrected). The current results indicate the changes in resting-state brain network after the CF loading which might reflect the difficulty in allocating attentional resources to specific task performance under CF (Esterman, 2013).

References

Esterman, M. et al, *Cereb Cortex*, 23: 2712-2723, 2013.
Iwaki S. et al, *Adv Biomed Eng*, 2: 137-142, 2013.
Pattyn N. et al, *Front Psychol*, 9:1246, 2018.
Whitfield-Gabrieli S. et al, *Brain Connectivity*, 2: 125-141, 2012.

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Poster

P614. Stress and Cognition in Human Subjects

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Program #/Poster #: P614.03

Topic: F.04. Stress and the Brain

Support: Army Research Laboratory Cooperative Agreement Number W911NF2020067

Title: Activity variability as a neural marker of fatigue on sustained alertness tasks.

Authors: *I. LIMA DIAS PINTO[1], K. BANSAL[2], J. O. GARCIA[3];
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Abstract: The ability to maintain alertness and attention for a sustained amount of time is essential for a wide range of tasks, and has severe consequences on other aspects of cognition and decision making. Fatigue accumulation during a task has the potential to thwart this alertness and jeopardize performance. The psychomotor vigilance test (PVT) is one of the most used assessments of vigilance and alertness where subjects are asked to make a motor response as soon as they perceive a visual stimulus on the screen, this procedure is repeated for several minutes, and, importantly, inter-trial interval is random. From this task, alertness is often inferred from a score derived from the response time to the briefly presented stimulus. Previous research has shown that the subject's response tends to be slower as time progresses during the task (Basner, M. et al. 2011), suggesting behavioral sensitivity to fatigue accumulation. Recently, researchers have investigated the the ratio of the variability of BOLD activity between brain networks, containing somato-motor and -sensory regions of the brain, and investigated the relationship of this metric to the perception of the speed of time (Northoff, G. et al. 2018). Here we probe the effects of this balance of sensory and somatomotor networks on the performance of the subjects during the PVT in which the response time is essential. We examined the ratio of fMRI BOLD variability for visual and somatomotor networks whilst subjects were engaged in the PVT. Using the Schaeffer parcellation and network demarcation (Schaefer, A. et al. 2018) to determine networks of interest, we observed that the temporal evolution of the visual and somatomotor variability ratio decays over time and is highly correlated to the decay in performance after a rapid but brief transient period. Our results indicate that faster responses are observed when the balance of variation of the BOLD activity is associated with performance in the PVT, suggesting that the emergence of fatigue from time-on-task can alter this balance and affect the performance. This work suggests a new avenue of fMRI BOLD feature extraction that may be highly predictive of performance change in a variety of tasks and suggests the importance of the dynamic network tension in cognition.

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Poster

P614. Stress and Cognition in Human Subjects

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Program #/Poster #: P614.04

Topic: F.04. Stress and the Brain

Title: Investigating the effect of stress on prospection and intertemporal choice

Authors: *H. E. AHN, A. G. SHIVERS, G. J. NORMAN, A. BAKKOUR;
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Abstract: When we imagine events in our lives that haven't happened yet, we generate and evaluate mental representations of possible futures. What we are able to imagine seems to be constrained by what we have previously experienced, suggesting a close link between memory and prospection. Prior literature points to the deleterious effects of acute stress on memory, but the implied effect of stress on prospection is not well understood. Furthermore, humans tend to discount the value of future rewards over time during intertemporal choice. But having people imagine future events can bias them to make more farsighted, patient choices, thereby reducing delay discounting. However, having to consider tradeoffs between immediate and delayed outcomes might be particularly challenging under acute stress. Studies have shown that some individuals make more impulsive decisions when stressed out, but the question of whether stress affects decision making through its effect on prospection remains unclear.

We sought to investigate how acute stress might affect one's ability to prospect about their future and make future-oriented decisions accordingly. We hypothesized that stress influences decision making through its impact on memory and prospection. In order to test this, we conducted a two-visit online study: participants began one session with an online adaptation of the Trier Social Stress Test, and the other session with a no-stress control condition. In each session, participants were then asked to either imagine themselves spending money in a future event, or describe a picture of a scene. This 2x2 study design yielded four conditions for each participant: Stress-Imagine, Stress-Describe, No Stress-Imagine, No Stress-Describe. Following each Imagine/Describe trial, participants were prompted to make a choice between a larger future reward and a smaller immediate reward.

We predicted that the discounting parameter *k* (obtained from fitting a hyperbolic discounting model to the choice data) would be lower (less discounting of delayed reward) during prospection, but would increase (more discounting) in Stress sessions. Our results show a trend of less impulsivity for immediate rewards when participants prospected about the future under stress. These results weakly conform to our predictions. But the effects of stress on behavior were not clear in an online format, given that physiological measures of the stress response were not captured. As a whole, this study attempts to uncover the ways in which one can begin to view stress, memory, and decision making in tandem by examining how prospection influences future-oriented choice, and how stress might disrupt this relationship.

Disclosures: H.E. Ahn: None. **A.G. Shivers:** None. **G.J. Norman:** None. **A. Bakkour:** None.

Poster

P614. Stress and Cognition in Human Subjects

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P614.05

Topic: F.04. Stress and the Brain

Support: NIH/NIDA Grant R01DA045716
NIH/NICHD Grant 5T32HD091059

Title: Early-life stress, depression and acute stress are related to impaired selectivity for important information in a value-directed remembering paradigm.

Authors: *A. L. GORDON, T. K. PATTERSON, H. P. DE GUZMAN, B. J. KNOWLTON; Psychology, UCLA, Los Angeles, CA

Abstract: Early-life stress (ELS) is linked to lifelong increased risk for a broad spectrum of negative health outcomes including substance use disorders, sexually transmitted infections, and early mortality. However, the cognitive and neurological mechanisms producing these elevated risks are poorly understood. We have shown that ELS is associated with increased habits at the expense of goal-directed actions during instrumental avoidance learning. If this association is driven by impaired goal-directedness, we would expect ELS to be associated with other disrupted goal-directed behaviors such as selectivity for remembering important information under capacity constraints. To test whether this is the case participants (n = 100), half of whom underwent Trier social stress test induction, performed a value-directed remembering task in which they were asked to maximize their score by recalling words associated with arbitrary values. Half of the lists were performed under cognitive load. Patterns of recall were analyzed using hierarchical generalized linear modeling. Both ELS and acute stress were associated with diminished selectivity for high value words. Joint significance tests suggest that the effects of ELS are mediated by depressive symptomatology. These results have important implications for understanding how ELS has lifelong consequences for exposed individuals.

Disclosures: A.L. Gordon: None. **T.K. Patterson:** None. **H.P. De Guzman:** None. **B.J. Knowlton:** None.

Poster

P614. Stress and Cognition in Human Subjects

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P614.06

Topic: F.04. Stress and the Brain

Support: SR/SATYAM/33/2016

Title: Mind body techniques-based amelioration of stress and cognition through possible neurogenesis dependent pathway

Authors: *K. SHARMA[1], A. ANAND[2], S. GOEL[1], P. VERMA[3], K. SHARMA[4], K. MAITY[5], S. JINDAL[5];
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Abstract: Mind body techniques-based amelioration of stress and cognition through possible neurogenesis dependent pathway**Kanupriya Sharma*,** Akshay Anand, Sonu Goel, Prashant Verma, Kiran Sharma, Kalyan Maity, Sheetal Jindal**Conflict of Interest**: None**Funding Agency:** DST SATYAM**Abstract**The developing society has increasing severity of stress and anxiety-based disorders due to the current lifestyle and work life balance. Stress and anxiety can lead to cognitive decline and influence adult neurogenesis. Mind body techniques like Yoga have been known to reduce the stress and related complications. In order to understand the molecular mechanism through which Mind Body technique-based yoga protocol helps in amelioration of lifestyle stress-based cognitive decline, 45 women between the age of 20-55 were assessed, who were given 45 minutes of Yoga intervention for 3 months for 5 days a week. They were assessed for stress and anxiety scale through STAI (State Trait Anxiety Inventory) and their cognitive profile was assessed using neuropsychological scales, i.e., Digit Span test, Trail Making Test, Digit Symbol Substitution Test before and after the intervention. In order to understand the molecular aspects associated with cognitive benefits, we assessed the neurogenesis markers in blood through ELISA i.e., BDNF, VEGF and Angiogenin which are known to influence cognitive profile through the neurogenesis mechanism. After 3 month of Yoga practice, reduction in the stress and anxiety with improvement in the cognitive profile was observed. Neurogenesis and Angiogenesis markers also showed alterations after Yoga practice. These results call for comprehensive study to examine whether Yoga based mind body techniques can help in ameliorating lifestyle stress-based cognition through the neurogenesis.

Disclosures: K. Sharma: None. **A. Anand:** None. **S. Goel:** None. **P. Verma:** None. **K. Sharma:** None. **K. Maity:** None. **S. Jindal:** None.

Poster

P614. Stress and Cognition in Human Subjects

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P614.07

Topic: F.04. Stress and the Brain

Support: Z-28015/48/2020-HPC(EMR)/AYUSH

Title: Neuropsychological effects of mindfulness among quarantined HCWs, COVID-19 positive and COVID-19 convalescent patients: A Randomized clinical trial

Authors: *K. MAITY[1], M. RAIN[2], A. BHALLA[3], G. PURI[4], P. AVTI[5], V. KAUSHAL[6], P. MAHAJAN[6], P. VERMA[13], S. J. GUPTA[1], P. MEHRA[13], P. NADHOLTA[2], N. DIXIT[13], M. SINGH[7], R. AGARWAL[8], N. PANDEY[6], P. MALHOTRA[9], S. GOEL[10], K. SONI[11], N. SACHDEVA[12], B. SUBRAMANIUM[14], A. ANAND[2];
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Abstract: Individuals having low immunity and other comorbidities are more vulnerable to COVID-19. Healthcare system plays important role to rescue and provide the primary care to the patients suffering from deadly virus. Thus, COVID positive, recovered patients and healthcare workers have higher levels of distress, anxiety, and depression while dealing with such situations that has mental effects. Mental stress and depression are vulnerable conditions to increase the risk of acute respiratory infections by lowering the immunity. Here, we report that yoga practice may improve neuropsychological parameters. Study subjects were recruited from Postgraduate Institute of Medical Education and Research (PGIMER), Chandigarh, India from 13th October, 2020 to 7th January 2021. The three groups were 1) COVID-positive group 2) COVID-recovered group 3) Health care workers (HCW) group. Each group was randomly divided into intervention or yoga group and non-intervention or control group (n=12 each). The Yoga intervention was given for 15 days. Neuropsychological parameters included Perceived Stress Score (PSS), Profile of Mood States (POMS). JOY subscale (DPES-Joy), Warwick-Edinburgh Wellbeing Scale (WEMWBS), WHO quality of life (WHOQOL). COVID positive Yoga (CPY) group showed non-significant reduction in PSS and tension whereas, vigor was significantly increased in both CPY ($p=0.01$) and COVID positive control (CPC) group ($p=0.004$) as compared to baseline. Physical, psychological & environmental health were increased in both the groups as compared to the baseline. Whereas, social relationship was significantly reduced in CPY ($p=0.05$) as compared to baseline. After 15 days, physical health improved whereas, social relationship domain of WHOQOL scale reduced in COVID recovered Yoga (CRY) group. In HCW Yoga group tension, anger, fatigue, vigor, and overall wellbeing were non-significantly reduced and overall quality of life increased. Yogic practices can be considered as therapeutic intervention to prevent and manage COVID-19 related stress and improve overall wellbeing.
Ethical Statement: Ethical approval was taken from the Institutional Ethics Committee of PGIMER, Chandigarh. The ethical approval number is IEC-05/2020-1646 **Conflict of Interest:**

None ***Corresponding Author:*** Akshay Anand, Ph.D. Professor Neuroscience Research Lab Department of Neurology, PGIMER, Chandigarh, India. **Email:**akshay1anand@rediffmail.com

Disclosures: K. Maity: None. **M. Rain:** None. **A. Bhalla:** None. **G. Puri:** None. **P. Avti:** None. **V. Kaushal:** None. **P. Mahajan:** None. **P. Verma:** None. **S.J. Gupta:** None. **P. Mehra:** None. **P. Nadholta:** None. **N. Dixit:** None. **M. Singh:** None. **R. Agarwal:** None. **N. Pandey:** None. **P. Malhotra:** None. **S. Goel:** None. **K. Soni:** None. **N. Sachdeva:** None. **B. Subramanium:** None. **A. Anand:** None.

Poster

P614. Stress and Cognition in Human Subjects

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Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P614.08

Topic: F.04. Stress and the Brain

Support: Psi Chi Research Grant
Albright Creative Research Experience

Title: Examining stress markers during restricted environmental stimulation

Authors: *S. M. KING, K. A. FEIGENSON;
Albright Col., Reading, PA

Abstract: This study examined the impact of restricted environmental stimulation technique (REST) on stress biomarkers, and how effectively it could be used as a form of meditation. We hypothesized that the sensory restriction treatment would lower stress levels but increase anomalous sensory experiences. The methodology used was an alteration to traditional sensory deprivation techniques, where instead of a complete absence of sensory input there were incomprehensible sensory inputs (such as white noise). We assayed stress biomarkers before and after the treatment, focusing on salivary cortisol levels, which were examined using enzyme-linked immunosorbent assay. Randomized treatment participants relaxed on an air mattress for 20 minutes wearing noise cancelling headphones playing white noise and Ganzfeld-like goggles, whose purpose was to obscure light. The control group procedure mimicked the physical sensations of the treatment group, wearing goggles and headphones but without the sensory restriction elements. We quantified participants' sensory experiences as well as the average galvanic skin conductance for 20 minutes. Our experiment used established sensory perception techniques (e.g. Ganzfeld like goggles, headphones playing white noise, salivary cortisol levels) and combined them with measures to validate the reporting of the anomalous sensory events. This was created by having participants press a handheld button when they experienced a percept, and this was compared against a record the experimenter kept if they perceived any added stimuli during that time (e.g. external noise, gust of air, light shining into the room).

Additional variables collected included age, sex, race/ethnicity, in addition to self-reported scores on questionnaires on the Revised Hallucination Scale, the Peters Delusional Inventory, and the Multidimensional Schizotypy Scale. The anticipated results were that the cortisol levels and skin conductivity would be lower with the use of REST techniques compared to the control group. Results instead suggested that there were no significant differences by condition, but there was an across group decrease in cortisol levels from the start of the experiment to the end. Additionally, results show that the more sensory experiences an individual reported, the higher their cortisol levels at the end of the experiment. These findings suggest both REST and short term meditation reduce stress to similar degrees, but these effects are weakened when individuals experience anomalous sensory events.

Disclosures: S.M. King: None. **K.A. Feigenson:** None.

Poster

P614. Stress and Cognition in Human Subjects

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P614.09

Topic: F.04. Stress and the Brain

Title: Effects of yoga on memory, cognitive levels, overall quality of life and lipid profile in Police recruits

Authors: *P. VERMA[1], P. AVTI[2], A. ANAND[2], G. SINGH[1], K. SONI[3], K. MAITY[4], S. JINDAL[4], K. SHARMA[3], S. PAL[1], N. KAUR[1], P. BALI[5], S. JYOTI[6], S. KAUSHAL[3]; [1]Panjab Univ., CHANDIGARH, India; [2]P.G.I.M.E.R, Chandigarh, India; [3]P.G.I.M.E.R, CHANDIGARH, India; [4]S.V.Y.A.S.A., Bengaluru, India; [5]I.I.S.E.R., Mohali, India; [6]Haryana Yoga Ayog, Panchkula, India

Abstract: Effects of yoga on memory, cognitive levels, overall quality of life, and lipid profile in Police recruits

Authors: Prashant Verma[1],* Pramod Avati[2], Akshay Anand[3] Gurmeet Singh[4], Krishan Kumar Soni[5], Kalyan Maity[6], Sheetal Jindal[7], Kanupriya Sharma[8] Surinder Kaushal[9], Navneet Kaur[10], Parul Bali[11], Saras Jyoti[12], Sushant Sharma[13],

Abstract

Stress and health issues are common among police professionals and it also affects the mental status of the police and deteriorates the police-public relations due to their job profile which is full of challenges. Hence, there is an urgent need to develop validated mind-body techniques in their daily routine to improve their overall health and improve the compassion and mind-body balance among professionals. A few recent studies suggest that yoga aids in reducing stress and Meditation is functional in reducing stress and anxiety; in addition, it improves memory and attention. This study included additional parameters than past studies. 478 recruits from the

police department, UT Chandigarh, India were recruited for the study. The recruits were divided into two major groups as Group A and Group B. Group A practiced yoga with common yoga AYUSH protocol and their basic exercises for 3 months and Group B did not practice yoga but followed only traditional training exercises i.e. regular physical exercise regimen, Running, etc. during these 3 months. To examine the effects on memory, attention, and their overall health TMT A and TMT B, Digit memory test, WHO-QOL Bref, BMI, lipid profile tests were carried out at baseline and 3 months. In between the groups and within the group comparison were done in between Group A and Group B. Group A and Group B were analyzed according to age and gender after dividing them into 4 groups (comparison between 20-24 age males, 20-24 age females, 25-30 age males, 25-30 age females of Group A and Group B). Group A showed significant improvement in BMI, memory, executive functioning, cognitive processing speed, lipid profile, and overall quality of life, in all age groups and gender. This data supports that Yoga can be helpful in reducing stress during training and future studies may use this study to launch larger studies.

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Poster

P630. Thermoregulation, Respiration, and Exercise

Location: Virtual Only

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Program #/Poster #: P630.01

Topic: F.07. Autonomic Regulation

Support: JSPS-18K19262
JSPS-18KK0190

Title: Establishment of mPGES-1 deficient rats lacking LPS-inducible hyperthermia

Authors: *T. MATSUWAKI[1], A. SANDO[2], S. KAKUTA[2], K. YAMANOUCHI[1];
[1]Dept. of Vet. Physiol., [2]Dept. of Biomed. Sci., The Univ. of Tokyo, Tokyo, Japan

Abstract: Prostaglandin E_2 (PGE_2) is a strong pyrogen working in the CNS under infectious conditions. It has been demonstrated that mice lacking the inducible PGE_2 synthase, microsomal prostaglandin E synthase-1 (mPGES-1), show no endotoxin-dependent fever. In addition, we have recently reported that female mPGES-1 deficient (KO) mice showed strong hypothermia in response to injection of lipopolysaccharide (LPS) only when they are on proestrus stage. These results suggested that the endocrine system strongly affects hyperthermic and/or hypothermic response to the immune challenge. In the present study, thus, we produced mPGES-1 KO rats using Cas12a/CRISPR genome editing system for the future investigation of the possible

involvement of the endocrine system in thermoregulation under infectious conditions. The KO animals are lacking the splice acceptor site of intron1 of mPGES-1 locus. Reverse transcription PCR using their hypothalamus showed that they produce a short form of mRNA of mPGES-1 without exon2. First, we implanted a temperature transponder in the abdominal cavity of male rats and started measuring body temperature every minute immediately after implantation. Two weeks later, we injected LPS intraperitoneally to the animals. Subsequently, we treated another group of rats with LPS and obtained their CSF from the cisterna magna 5 hours after injection to measure the concentration of PGE_2. KO rats presented a regular circadian rhythm of body temperature as WT rats, suggesting mPGES-1 plays no role in regulation of body temperature under normal conditions. On the other hand, LPS-inducible hyperthermia was observed only in WT but not in KO rats. Moreover, PGE_2 concentration in the CSF was remarkably enhanced by LPS injection also only in WT rats. We have established a novel line of mPGES-1 KO rats and successfully reproduced the feature of mPGES-1 mice concerning the thermoregulation under infectious conditions. These animals will be of good use for studying the role of PGE_2 in the endocrine system that requires serial samples or samples of large amount.

Disclosures: T. Matsuwaki: None. **A. Sando:** None. **S. Kakuta:** None. **K. Yamanouchi:** None.

Poster

P630. Thermoregulation, Respiration, and Exercise

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Program #/Poster #: P630.02

Topic: F.07. Autonomic Regulation

Support: ERG Grant H2020-ERC-2017-COG
Heiderberg Klinik Grant D.10092820
Physician Scientist Program of the Medical Faculty of Heidelberg

Title: A synaptic mechanism of hypothalamic temperature detection

Authors: *G. KAMM[1], J. BOFFI[2], K. ZUZA[1], S. NENCINI[1], J. CAMPOS[3], K. SCHRENK-SIEMENS[1], I. SONNTAG[4], B. KABAGLOU[1], M. ABD EL HAY[1], A. TAPPE-THEODOR[1], C. ACUNA[3], T. KUNER[4], J. SIEMENS[1];
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Abstract: The preoptic area (POA) of the hypothalamus is an interoceptive brain structure able to detect changes in local temperature. It has been proposed that the ion channel TRPM2 is part of the machinery detecting warmth in the POA. However, the mechanism underlying TRPM2-mediated temperature detection in the POA is not fully understood.
Here, we show that TRPM2 facilitates whole-body cooling upon POA warming in mice. At the cellular level, TRPM2 enhances the thermosensitivity of preoptic warm-sensitive neurons (WSN). TRPM2 achieves this by modulating in a temperature-dependent manner the synaptic inputs that arrive at WSN. Altogether, our data suggests that TRPM2 mediates direct warmth detection in the POA. We further provide evidence that preoptic WSN are indeed a neuronal population detecting warmth in the POA and that WSN thermosensitivity can be conferred synaptically by temperature sensors, like TRPM2, acting extrinsically to WSN.

Disclosures: G. Kamm: None. **J. Boffi:** None. **K. Zuza:** None. **S. Nencini:** None. **J. Campos:** None. **K. Schrenk-Siemens:** None. **I. Sonntag:** None. **B. Kabaglou:** None. **M. Abd El Hay:** None. **A. Tappe-Theodor:** None. **C. Acuna:** None. **T. Kuner:** None. **J. Siemens:** None.

Poster

P630. Thermoregulation, Respiration, and Exercise

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Program #/Poster #: P630.03

Topic: F.07. Autonomic Regulation

Support: H2020-ERC-2017-COG

Title: Molecular characterization of preoptic area warm sensitive neurons

Authors: *K. ZUZA, G. B. KAMM, J. E. SIEMENS;
Inst. of Pharmacol., Heidelberg Univ., Heidelberg, Germany

Abstract: Body temperature is a dynamic homeostatic property, tightly regulated by the autonomic nervous system. How the brain coordinates maintenance of the body temperature within a narrow range of 37 °C, a condition which is needed for all enzymatic reactions and cellular processes to function properly, remains unclear. Among the brain regions that have been implicated in thermoregulation is the hypothalamus, more precisely its anterior portion, the preoptic area (POA). Early experimental studies have implemented *in vivo* heating or cooling of the POA which resulted in activation of thermoregulatory effector mechanisms. Additionally, electrophysiological studies have found neurons within the POA to respond to temperature stimuli suggesting that these neurons, known as warm sensitive neurons (WSNs), are the cells which detect deep brain temperature and might integrate temperature information from the periphery. The lack of specific molecular markers to identify preoptic WSNs limits our capacity to study how the POA acts as a body thermostat. Effort has been put in investigating those cells,

with several molecular candidates emerging, such as TRPM2 ion channel found to play a role in POA temperature detection and hypothermia. Therefore, our aim was to characterize WSNs of the POA at the molecular level. To achieve this goal we used primary POA cell culture from 9 days old C57BL/6 N mice (P9) and performed calcium imaging while applying a temperature stimulus of 45 °C in the presence and the absence of the TRPM2 antagonist, 2-Aminoethoxydiphenyl borate (2-APB). We identified and hand-picked temperature responding cells (n=37), cells which had temperature response blocked by 2-APB (n=34) and temperature non-responding cells (n=22) for the single-cell RNA-sequencing (scRNA-seq). Analysis of the scRNA-seq data pointed to the limitations of the P9 cell culture used. Majority of the temperature non-responding cells sequenced expressed glial marker genes together with neuronal markers, a combination not found *in vivo*. We concluded that genetic heterogeneity of the sequenced cells was too large and putative WSNs' molecular markers identified from cultured neurons would be ambiguous. In order to disentangle the neuronal circuits involved in body temperature homeostasis, further effort needs to be invested in search for WSNs' markers. Alternative ways include combining low throughput RNA-seq experiments coupled with physiological characterization of neurons, such as Patch-seq (Cadwell et al., 2016, Fuzik et al., 2016), with large scale scRNA-seq data sets, such as the POA genetic atlas (Moffit et al.,2018).

Disclosures: K. Zuza: None. **G.B. Kamm:** None. **J.E. Siemens:** None.

Poster

P630. Thermoregulation, Respiration, and Exercise

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Program #/Poster #: P630.04

Topic: F.07. Autonomic Regulation

Support: ZIA DK075062
ZIA DK075063
ZIA DK075088
ZIA DK075064
ZIA DK070002
ZIA DK075087

Title: Preoptic BRS3 neurons increase body temperature and heart rate via multiple pathways

Authors: *R. PIÑOL, A. MOGUL, A. SAHA, C. LI, V. SKOP, H. PROVINCE, S. KULKARNI, C. XIAO, O. GAVRILOVA, M. KRASHES, M. REITMAN;
NIH-NIDDK, Bethesda, MD

Abstract: The preoptic area (POA) is a key brain region controlling whole-body metabolism and body temperature (Tb), dictating thermogenic, cardiovascular, and behavioral responses to

regulate Tb. Ultimately survival depends on proper Tb regulation, making it critical to understand the physiology of this motivational need of the body. Known POA neuronal populations reduce Tb when activated; a population that increases Tb upon activation has not yet been reported. Using mice, we now identify bombesin-like receptor 3 (BRS3)-expressing POA (POA^{BRS3}) neurons as having this missing functionality. BRS3 is an orphan receptor that regulates energy and cardiovascular homeostasis, but the relevant neural circuits are incompletely understood. We demonstrate that POA^{BRS3} neuronal activation increases Tb, heart rate, and blood pressure sympathetically, and stimulation of projections to the paraventricular nucleus of the hypothalamus and dorsomedial hypothalamus does the same. Acute POA^{BRS3} inhibition reduces Tb. Long-term inactivation of POA^{BRS3} neurons increased Tb variability and produced exaggerated Tb changes, overshooting both increases and decreases in Tb set point, with RNA expression profiles suggesting multiple types of POA^{BRS3} neurons. Thus, POA^{BRS3} neuronal populations regulate Tb and heart rate, contribute to cold-defense, and fine-tune feedback control of Tb. These findings advance understanding of homeothermy, a defining feature of mammalian biology

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Poster

P630. Thermoregulation, Respiration, and Exercise

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Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P630.05

Topic: F.07. Autonomic Regulation

Support: DFG Sonderforschungsbereich

Title: Secretome analysis of hypothalamic neuroendocrine circuits regulating core body temperature in mice

Authors: *C. ARAUJO-SOUSA[1,2], D. COLOMBO[3], G. B. KAMM[1], S. CALDERAZZO[3], J. KRIJGSVELD[3,4], J. SIEMENS[1,2];
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Abstract: Surviving environmental challenges requires flexible neural systems. In this context, neuromodulators, including neuropeptides and some peptide hormones, are important players in tuning neural activity. These molecules were shown to regulate several physiological processes, such as sleep, mating, behavior, and energy balance. The core body temperature (T_{core}) in

mammals is maintained by the central thermoregulatory circuitry, which has its center at the preoptic area (POA) of the hypothalamus. Brain injection of peptides and differential expression (DE) analyses suggested a role of neuromodulators in energy balance, especially at the POA. Such studies implicated different peptides in the control of T_{core}, for instance: pituitary adenylate cyclase-activating peptide (PACAP) and leptin. However, these studies have limitations. The injections required previous knowledge about the identity of the peptides, and many did not account for other parameters that influence T_{core}, such as locomotor activity. And none of the DE analyses evaluated the peptides at the protein level. Therefore, the molecular mechanisms that modulate body temperature during thermal challenges remain unclear. Thus, we performed an unbiased screening of peptides released under thermal challenges. The peptides were collected from the POA of freely behaving adult male mice using microperfusion under warm and cold temperature stimulation. The control group was kept at room temperature. The samples were analyzed in an unbiased manner by liquid chromatography and mass spectrometry (LC/MS) and label-free quantification. With this, we aimed to investigate which peptides are released at the level of the POA during these stimuli, and how they help to integrate thermal information and to control T_{core}. We were able to detect, in unstimulated animals, peptides part of 29 proneuropeptides and propeptide hormones. In total, we identified twenty peptides more in comparison to a previous unbiased screening of brain secretome. Also, comparing samples of unstimulated and stimulated animals, we detected peptides from six proneuropeptides that were significantly regulated in warm-exposed animals. With the cold stimulation, we found ten peptides from neuromodulators that were differentially regulated. Further on, we plan to investigate if the identified peptides modulate neural activity and T_{core} by *in vivo* and *in vitro* studies. Hence, the identified peptides could contribute to a better understanding of thermoregulatory mechanisms. And considering the impact that T_{core} has on mammals, this project could have meaningful outcomes for healthy and diseased states.

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Poster

P630. Thermoregulation, Respiration, and Exercise

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P630.06

Topic: F.08. Biological Rhythms and Sleep

Support: R35GM133737

Title: Electrophysiological characterization of circadian clock neurons entrained to temperature cycles

Authors: *N. SONI, S. YADLAPALLI;
Cell & Developmental Biol., Univ. of Michigan, Ann Arbor, MI

Abstract: Abstract: Fruit fly *Drosophila melanogaster* offers an excellent model to study the distinct roles of different clock neuron classes in circadian timekeeping and sleep regulation. Flies central circadian clock consists of a network of approximately 150 clock neurons, each of which contains a molecular clock that generates gene expression oscillations with a period of approximately 24 hours. Circadian rhythms are self-sustained in constant conditions, but they must be entrained by external cues such as light and temperature cycles to maintain a precise 24-hour period. While we know a lot of how light entrains the clocks, the neural and molecular mechanisms underlying temperature entrainment are largely unknown. To address these critical gaps, here we employed electrophysiology, behavioral, and other genetic techniques to gain a deeper insight into how clocks are entrained to temperature cycles. Specifically, to understand the nature of change in membrane potential of DN1p in temperature cycle, we employed in vivo whole-cell voltage patch-clamp to measure the resting membrane potential (RMP) from all DN1p clock neurons; and found that RMPs measured in temperature cycle were not aligned to a common pattern usually observed in these neurons in LD cycle (Flourakis *et. al*., Cell 2015). Based on RMP measured in temperature cycle we observe three major clusters of DN1p. A recent finding suggests the presence of molecular heterogenicity within the DN1p population (Ma *et. al*., elife 2021), therefore, to maintain homogeneity in our recordings, we used a very specific subset of DN1 cells tagged with the GFP. We used DN1p cells positive for CnMa peptide. We observe that in LD cycle DN1pCnMA change in RMP follows a similar pattern observed previously (Flourakis *et. al*., Cell 2015). However, we observe that in temperature cycle there is a ~6 hour phase difference in the oscillations of neuronal depolarization of DN1pCnMA cells compared to that of LD cycles. Together, these results provide the first comprehensive measurement of DN1p change in RMP in temperature cycle. This study greatly expands our understanding of change in clock neurons electrophysiological behavior in two naturally occurring circadian changes in LD as well as in temperature and allow researchers to consider that clock neurons are not homogeneous in their electrical rhythm when we consider different entrainment.

Disclosures: N. Soni: None. **S. Yadlapalli:** None.

Poster

P630. Thermoregulation, Respiration, and Exercise

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P630.07

Topic: F.07. Autonomic Regulation

Support: FAPESP

CNPq
CAPES-Proex

Title: Post-inspiratory activity elicits by activation of phox2b-expressing retrotrapezoid nucleus neurons depends on the integrity of the Kolliker-Fuse region

Authors: *K. FLOR[1], L. M. OLIVEIRA[2], A. C. TAKAKURA[2], T. S. MOREIRA[1];
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Abstract: Considering that the retrotrapezoid nucleus (RTN) would be an important center in the central nervous system (CNS) involved in the maintenance and modulation of respiratory activity, we hypothesized that neurons in this nucleus would also be involved in the post-inspiratory phase of the respiratory cycle. Here we performed pharmacogenetic manipulation (AAV-hM3D(Gq)-mCherry or AAV-hM4D(Gi)-mCherry) in VGlut2-cre,Ai6 conscious mice to evaluate breathing parameters through whole body plethysmography. Under baseline conditions, RTN stimulation by intraperitoneal administration of CNO (90 µg/mL - 0.1 mL/animal) produced a long-lasting increase in respiratory frequency (f_R), tidal volume (V_T) and minute ventilation (V_E), as well as reduction in the duration of the post-inspiratory (T_{PI}) phase. The increase in V_T (3.3 ± 0.47 vs. RTN stimulation: 5 ± 0.39 ml/kg) and V_E (1108 ± 201 vs. RTN stimulation: 1595 ± 276 ml/min/kg) were reduced in a condition of pharmacogenetic inhibition of Kolliker-Fuse (KF) glutamatergic neurons. The reduction in TPI (152 ± 42 vs. RTN stimulation: 123 ± 16 ms) was prolonged after inhibition of KF neurons. After the inhibition of KF glutamatergic neurons, RTN stimulation produced a further increase in f_R (416 ± 36 vs. RTN stimulation: 284 ± 23 bpm). The phox2b-expressing RTN neurons seem to be involved in the modulation of respiratory activity and depend on the integrity of the glutamatergic neurons in the KF region.Financial support: FAPESP, CNPq, CAPES-Proex

Disclosures: K. Flor: None. **L. M. Oliveira:** None. **A. C. Takakura:** None. **T. S. Moreira:** None.

Poster

P630. Thermoregulation, Respiration, and Exercise

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Topic: F.07. Autonomic Regulation

Support: Landis-Berkman Family Fund (Private Donation)

Title: A Randomized Control Pilot Study of the Efficacy of Qigong vs Exercise-Nutrition Training on Improving Mood and Cardiorespiratory Measures

Authors: *C. S. ZIMMERMAN[1], D. DANIELS[1], C. PENNER[1], S. TEMEREANCA[2], C. E. KERR[3], S. JONES[4];
[1]Neurosci., [2]Dept. of Neurosci., [3]Brown Univ., Providence, RI; [4]Neurosci., Brown Univ., Brown University, RI

Abstract: Cancer-related fatigue is a debilitating, often long-term side effect of cancer treatment typically accompanied by significant mood disturbance. Since the autonomic nervous system governs overall body homeostasis and energy regulation and is intimately related to limbic and affective processing, cardiorespiratory measures of autonomic output such as heart rate variability (HRV) and respiratory sinus arrhythmia (RSA) may also be important for understanding overall mood and fatigue burdens. Exercise has emerged as an important contributor to alleviating fatigue but can be prohibitively intense for someone struggling with fatigue. Qigong is a form of meditative movement deriving from Chinese medical traditions and offers a feasible and gentle alternative to exercise. Additionally, the meditative awareness fostered in Qigong may have added benefit for mood-related symptoms of fatigue. We therefore assessed the effect of exercise vs qigong training on fatigue reduction in a 10-week pilot randomized control trial (n=22). We present secondary outcome measures related to mood changes assessed through multiple self report questionnaires (Profile of Mood States, Perceived Stress Scale, Multidimensional Assessment of Interoceptive Awareness, Difficulties in Emotion Regulation Scale) and alterations in cardio-respiratory measures of HRV and RSA (as assessed through Biopac MP150 3-lead ECG). Analysis of our primary fatigue reduction endpoint indicates that while both exercise and Qigong significantly reduced fatigue levels pre-post intervention, there did not appear to be a significant group x time interaction on ANOVA, suggesting that one may not be superior to the other in terms of fatigue reduction. Improvements in mood related function pre-post intervention were statistically significant in the Qigong but not the exercise group, suggesting that while both groups reduce fatigue, Qigong may additionally address mood-related symptoms. Results will be presented in relation to our ongoing analysis of HRV and RSA. This study suggests Qigong may be an effective therapy for improving mood and cardiorespiratory health, and helps identify changes in these factors in both exercise and qigong training that can influence fatigue reduction in cancer survivors with fatigue.

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Poster

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Program #/Poster #: P630.09

Topic: F.07. Autonomic Regulation

Support: NIH Grant UH3AT009145

Title: Effects of mindfulness-based hypertension intervention on resting state functional connectivity

Authors: J. POLCARI[1], R. CALI[3], B. NEPHEW[1], *J. CIROLI[1], F. SAADEH[4], E. LOUCKS[4], J. A. KING[2];
[2]Dean of Arts & Sciences, Professor of Biol. & Biotech., [1]Worcester Polytechnic Inst., Worcester, MA; [3]Martinos Ctr. for Biomed. Imaging, Boston, MA; [4]Brown Univ., Providence, RI

Abstract: Hypertension related illnesses are a leading cause of disability and death in the United States. Due to the high prevalence of adverse effects of pharmacological interventions for hypertension, there is increased interest in the development of nonpharmacological interventions. A mindfulness-based blood pressure reduction program (MB-BP) is effective in attenuating hypertension, but the neural correlates of this intervention are unknown. The objectives of the present study were to identify functional neural correlates of MB-BP using functional resting state magnetic resonance imaging (MRI) and assess potential correlations with key clinical outcomes. In subset of participants from a larger randomized controlled trial, MB-BP participants exhibited increased mindfulness and decreased depression compared to controls. Analyses of functional MRI data revealed significant group differences in several regions in the limbic system and/or associated with hypertension. Specific changes in functional connectivity were significantly associated with clinical measures of blood pressure, depression, anxiety, mindfulness, and emotional regulation. It is concluded that MB-BP has extensive and substantial effects on neural functional connectivity which could mediate beneficial changes in mindfulness, depression and related clinical measures in individuals with hypertension.

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Poster

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Program #/Poster #: P630.10

Topic: F.07. Autonomic Regulation

Support: ANR-17-ce37-0017-01

Title: Bodily Arousal Fluctuations During Task Performance: evidence from tonic and phasic electrodermal activity

Authors: T. GUIEYSSE[1], A. DOUBLIEZ[1], M. HOUOT[2], F.-X. LEJEUNE[2], S. RAZAFIMAHATRATRA[1], T. MEDANI[3], A. KLARSFELD[4], G. DREYFUS[4], ***K. ANDRADE**[1];
[1]Salpêtrière Hosp., Paris, France; [2]Paris Brain Inst., Sorbonne University, France; [3]Signal & Image Processing Inst., University of Southern California, CA; [4]ESPCI-Paris PSL, Paris, France

Abstract: Background: Our ability to learn from errors and flexibly adapt ongoing behavior is critical for an optimal performance. Fluctuations in physiological arousal, as revealed by both tonic and phasic components of electrodermal activity (EDA), typically accompany changes in attentional and emotional states with impact on task success. Nevertheless, the specific role of each of these EDA components during task performance is still imprecise. Objective and methods: Our main goal was to study whether tonic (activated arousal during the task) and phasic (electrodermal responses, EDR, to the stimuli) EDA components co-vary with each other and how they evolve during task performance. To this end, we recorded and analyzed the fluctuations of these components in nineteen healthy elderly subjects while they performed two consecutive sessions (A and B) of an experimental task composed by modified versions of the Go/No-go and Stroop tests. These tests were characterized by distinct workload conditions within and between sessions: the instructions of the Go/No-go test remained unchanged between sessions, while the instructions of the Stroop test were inversed in session B. Results: We observed a habituation effect related to stimulus repetition as revealed by a significant decrease in the amplitude of activated arousal between sessions (B < A) and within each test (end < beginning) in each session, as well as by a similar decrease in the amplitude of EDR on correct responses (session B < A). However, the amplitude of EDR on errors did not vary between sessions for any of the tests. Accordingly, the significant correlation between activated arousal and EDR amplitude observed on correct responses for each test, separately, disappeared with errors. Last, we observed higher EDR amplitudes in the Go/No-go test than in the Stroop test, as well as higher EDR amplitudes on erroneous (versus correct) responses only in the Go/No-go test, thus suggesting that subjects were less aware of their errors while performing the Stroop test (that is, the test more demanding in terms of mental effort and working memory). Conclusion: Our results suggest that fluctuations in tonic and phasic EDA are differently influenced by stimulus habituation and type of response (correct versus erroneous), which is in line with the role of EDR as a physiological signal of error awareness. Importantly, they further suggest that awareness of having committed an error (as revealed by EDR amplitude) might depend on the level of cognitive load associated with the task.

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Poster

P633. Sleep and Rhythms: Cellular and Molecular Contributors II

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Program #/Poster #: P633.01

Topic: F.08. Biological Rhythms and Sleep

Support: NIH Grant F31AG066385
NIH Grant F31NS115299
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Title: Examination of sex differences and diurnal variation in hippocampal neurophysiology and spatial memory

Authors: *L. GOODE[1], A. FUSILIER[1], N. REMISZEWSKI[1], K. L. GAMBLE[2];
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Abstract: Circadian rhythms are biological processes that cycle across 24-hours and are known to regulate many aspects of neurophysiology, including the processes of learning and storing memories. While there is well-documented evidence of diurnal variation in performance on spatial memory tasks, the effect of sex has not been as thoroughly examined across time of day. Moreover, there is little known regarding the impacts of time of day on hippocampal neuronal physiology. Here, we investigated the effects of both sex and time of day on hippocampal neurophysiology and hippocampal-dependent spatial memory in C57BL/6J mice. We found that performance on a hippocampal-dependent object location memory (OLM) task differs across circadian time of day and across sex. Using whole-cell patch-clamp, we measured spontaneous and mini excitatory and inhibitory postsynaptic currents in CA1 pyramidal cells in acute hippocampal slices from male and female mice during both the day and night. We found that the frequency and amplitude of both excitatory and inhibitory postsynaptic currents differed depending on time of day. We also measured active and passive properties of CA1 pyramidal cells and found differences in pyramidal cell excitability across time of day. Given that many neurological disorders impacting the hippocampus also display circadian disruption (e.g., Alzheimer's disease and epilepsy), understanding how hippocampal physiology and function are impacted by time of day can provide insight into the development of therapeutics targeting circadian disruption and inform the appropriate timing of existing treatments.

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Poster

P633. Sleep and Rhythms: Cellular and Molecular Contributors II

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Support: DFG grant STE 531/18-2
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DFG grant STE531/27-1

Title: The circadian coupling signal pigment-dispersing factor modulates visual sensitivity and locomotor output in the Madeira cockroach Rhyparobia maderae

Authors: *J. A. PLATH[1], **H. ZOLMON**[1], J. GESTRICH[1], P. ROJAS[2], W. ALAKA[1], M. BARTHOLMAI[1], T. VÖLKER[1], R.-M. VON BERLEPSCH[1], M. GARCIA[2], M. STENGL[1]; [1]Animal Physiology, Univ. of Kassel, Kassel, Germany; [2]Theoretical Physics, Univ. of Kassel, Kassel, Germany

Abstract: Circadian clocks are organized as dual morning and evening oscillator circuits of mammals and insects orchestrate sleep-wake rhythms entrained to external light dark cycles. We investigate how the circadian clock in the nocturnal Madeira cockroach gates and modulates sensory inputs and locomotor outputs for orchestration of sleep-wake cycles via timed neuropeptide release. The clock network was identified as multiple-peptide-expressing clock neurons which innervate the accessory medulla (AME) - a small neuropile in the optic lobes. The AME receives indirect visual input from the compound eyes and sends outputs to the midbrain to regulate rhythms of physiology and behavior. A key neuropeptide in insect circadian clock networks is pigment-dispersing factor (PDF), which controls sleep wake cycles via mostly unknown processes. Previous physiological recordings of the cockroach circadian clock suggested that PDF release at dawn activates morning-oscillator cells that promote sleep and inhibits evening oscillators promoting activity, thereby keeping sleep and activity promoting neuronal ensembles in stable antiphase. Here, in electroretinogram recordings from the compound eye we examined whether PDF as clock output controls circadian rhythms in visual sensitivity. Our preliminary results indicate that in the early day, when cockroaches sleep, PDF applications reduced the response to visual stimuli. Furthermore, in running-wheel studies of PDF-precursor/PDF-receptor RNAi double knockdown animals under constant conditions amount and rhythmicity of locomotor behaviour was strongly reduced. Surprisingly, rhythmicity was only moderately compromised in PDF-precursor or PDF-receptor single knockdown animals. We further characterized the activity of the clock network in long-term extracellular recordings of clock cells *in vivo*, which revealed dominant activity peaks at dawn, midday, dusk, and midnight consisting of electrical events at multiple scales. We are currently investigating whether these activity peaks are indicative of circadian clock-dependent release of PDF and other neuropeptides that together gate sleep wake rhythms.

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Poster

P633. Sleep and Rhythms: Cellular and Molecular Contributors II

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Program #/Poster #: P633.03

Topic: F.08. Biological Rhythms and Sleep

Support: NIH Grant 1R15GM134528

Title: Circadian gene expression of the endocannabinoid system in the mouse suprachiasmatic nucleus

Authors: *T. D. NIEPOKNY[1], I. D. PRATT[2], E. M. MINTZ[3];
[1]Biol. Sci. & Biomed. Sci., [2]Biol. Sci., [3]Col. of Arts & Sci., Kent State Univ., Kent, OH

Abstract: The endocannabinoid and circadian systems regulate a variety of physiological and behavioral processes in mammals; however, there is relatively little known about the interplay between the two. Some components of the endocannabinoid system, such as anandamide and 2-arachidonoylglycerol concentrations, are under rhythmic control in parts of the brain and body. Receptors and enzymes for endocannabinoid synthesis and degradation have recently been elucidated in the rat suprachiasmatic nucleus (SCN). A preliminary study of our own conducted at two timepoints, zeitgeber time (ZT) 4 and ZT16, confirms the presence of mRNA expression of many of the same components in the mouse SCN. We looked for 6 different receptors targeted by cannabinoids, cannabinoid receptor 1 (*Cnr1*), cannabinoid receptor 2 (*Cnr2*), vanilloid receptor 1 (*Trpv1*), and several orphan G-protein coupled receptors: *Gpr18, Gpr55,* and *Gpr119*. Of these, we only detected the presence of *Cnr1*. Gene expression was detected for each of the endocannabinoid synthesis and degradation enzymes we investigated: N-acyl phosphatidylethanolamine phospholipase D (*Nape-pld*), diacylglycerol lipase alpha (*Dagl-a*), diacylglycerol lipase beta (*Dagl-b*), glycerophosphodiester phosphodiesterase 1 (*Gde1*), abhydrolase domain-containing 4 (*Abhd4*), abhydrolase domain-containing 6 (*Abhd6*), abhydrolase domain-containing 12 (*Abhd12*), fatty acid amide hydrolase (*FAAH*), monoglyceride lipase (*Mgll*). The mRNA of some of these components are rhythmic in the rat SCN, and we hypothesize that they are also rhythmic in the mouse SCN. Experiments currently in progress are using laser capture microscopy and quantitative real-time PCR to assess gene expression patterns of the detected endocannabinoid system components in the SCN of male and female mice around the 24-hour cycle. These data will further establish whether necessary cannabinoid system components are rhythmically expressed in the SCN and help determine their role in clock function.

Disclosures: T.D. Niepokny: None. **I.D. Pratt:** None. **E.M. Mintz:** None.

Poster

P633. Sleep and Rhythms: Cellular and Molecular Contributors II

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Topic: F.08. Biological Rhythms and Sleep

Support: NIH Grant 1R15GM134528

Title: Components of the endocannabinoid system in the mouse dorsal and median raphe nuclei

Authors: I. D. PRATT[1], T. D. NIEPOKNY[2], *E. M. MINTZ[2];
[1]Biol. Sci., [2]Biol. Sci. & Biomed. Sci., Kent State Univ., Kent, OH

Abstract: Activity in the dorsal raphe (DR) and the median raphe (MR) influence circadian rhythms. The MR has a direct serotonergic projection to the suprachiasmatic nucleus (SCN), while the DR innervates the intergeniculate leaflet (IGL) as well as having connections to the MR. The IGL then innervates the SCN. Several molecular components of the endocannabinoid signaling, synthesis, and degradation systems have previously been identified in the SCN, DR, and MR. This experiment aimed to comprehensively identify gene expression of components of the endocannabinoid system in these regions, with a focus on future identification of the role of these components in the regulation of circadian rhythms. Male and female mice housed on a 12:12 light/dark cycle were sacrificed at two time points 12 hours apart (4 hours each after lights on and lights off). Brains were extracted and frozen for analysis. Sections were collected containing the MR and DR. The MR was removed from these sections with the use of a tissue punch and the DR was removed by dissection. Quantitative PCR was used to determine the presence or absence of numerous genes thought to be involved in the endocannabinoid system. B-actin was used as a control gene and tryptophan hydroxylase was used as a positive control for raphe tissue. Significant expression was detected in the DR for *Cnr1*, *Trpv1*, *Napepld*, *Dagl-a*, *Dagl-b*, *Faah*, and *Mgll*, while in the MR *Cnr1*, *Gpr18*, *Gpr119*, *Napepld*, *Dagl-a*, *Dagl-b*, *Faah*, and *Mgll* were detected. Ongoing experiments are evaluating whether any of these genes have a circadian rhythm of expression. The presence of the identified endocannabinoid system components suggests that endogenous cannabinoid signaling may modulate the influence of the raphe nuclei on circadian clock function.

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Poster

P633. Sleep and Rhythms: Cellular and Molecular Contributors II

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Topic: F.08. Biological Rhythms and Sleep

Support: MRC Grant MR/S01134X/1
BBSRC funded Oxford Interdisciplinary Bioscience DTP BB/M011224/1

Title: Investigating cell-specific neuronal ER stress after prolonged wakefulness

Authors: *A. CHAKRABARTY[1], S. E. NEWEY[1], L. DENLY[1], M. PROMI[1], J. PRIUS-MENGUAL[1], D. MUNRO[1], L. B. KRONE[1], L. E. MCKILLOP[1], A. HOERDER-SUABEDISSEN[1], Z. MOLNAR[1], S. PEIRSON[1], N. NAIDOO[2], V. V. VYAZOVSKIY[1], C. J. AKERMAN[1];
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Abstract: The restorative nature of sleep is supported by subjective experience as well as its homeostatic and local regulation in the mammalian cortex. Waking periods are believed to result in challenges to cellular physiology that leads to cellular stresses, and which sleep then serves to restore. For example, periods of extended wakefulness and sleep disruption have been linked with the accumulation of misfolded proteins in the endoplasmic reticulum lumen ("ER stress") in mouse brain tissue (e.g. Naidoo et al., 2006). Yet, it remains to be determined whether neuronal cell populations undergo different degrees of ER stress as a function of their activity and sleep-wake history. To address this question, we developed a quantitative immunohistochemical analysis pipeline to measure ER stress in neuronal subtypes in cortex after a period of prolonged wakefulness. We used cortical tissue from male C57BL/6 mice following a 6h period of sleep deprivation (SD) from light onset or an equivalent period of undisturbed sleep. In the first application of our pipeline, we used anti-NeuN and anti-PV antibodies to identify all neurons and parvalbumin-expressing fast-spiking interneurons, respectively. We stained for the major protein folding chaperone BiP/Grp78, an established marker of ER stress, with an anti-SEKDEL antibody. In frontal cortex, NeuN neurons showed a 15% increase in SEKDEL reactivity following 6h SD, whereas PV neurons did not. PV neurons showed a lower level of SEKDEL reactivity than neighbouring non-PV neurons. NeuN neurons from somatosensory cortex did not show a significant effect with SD. Together, these results indicate that there may be cell type-specific and region-specific differences in ER stress levels following prolonged wakefulness. In order to be able to monitor ER stress dynamically in further defined neuronal populations, we have also developed a novel fluorescent reporter based on the BiP/Grp78 promoter. In NIH3T3 mouse fibroblast cells challenged with low doses of the ER stress inducer, Tunicamycin, the BiP/Grp78 transcriptional reporter showed substantial changes in fluorescence compared to DMSO control. The novel reporter also reflected the temporal dynamics of ER stress following a 2h pulse treatment with Tunicamycin, with the reporter's fluorescence peaking at 12h and returning to baseline by 72h. These properties validate the reporter as a useful tool for monitoring patterns of physiological ER stress, with the potential to genetically target specific cell populations. Such an approach could be used to further investigate the relationship between cell-specific ER stress and sleep-wake history.

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Poster

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Program #/Poster #: P633.06

Topic: F.08. Biological Rhythms and Sleep

Support: NIH R01 NS118440

Title: Sleep deprivation-induced impairments in hippocampal memory and plasticity might be mediated by somatostatin interneurons

Authors: *F. RAVEN[1], J. DELORME[1], L. WANG[1], F. ROIG KUHN[3], V. K. KODOTH[2], J. MA[4], J. D. MARTINEZ[1], B. A. TOTH[5], V. BALENDRAN[1], A. VEGA MEDINA[1], K. SCHMIDT[6], S. JIANG[1], S. J. ATON[1];
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Abstract: Sleep loss is an increasing phenomenon, and can harm both physical and mental health. Even a single night of sleep deprivation (SD) can result in disturbed cognition, and as little as 6h of SD affects the dendritic spine density within the hippocampus, which is crucial for memory storage. Recent work from our lab has demonstrated that activity-dependent phosphorylation of ribosomal protein S6 (pS6) is decreased in dentate gyrus (DG) granule cells, and increased in somatostatin-expressing (SST+) interneurons, after just a few hours of SD. We find that chemogenetically activating SST+ interneurons in the mouse DG disrupts sleep-dependent fear memory consolidation. Conversely, inhibition of this specific cell population during post-learning sleep promoted fear memory consolidation, and increases pS6 labeling of granule cells. More recently, we have tested whether SD also affects SST+ interneurons at the structural level, using quantitative analysis of Cre-dependent Brainbow labeling in DG and other hippocampal structures after a period of *ad lib* sleep or SD. Our data indicate that the negative consequences of SD on hippocampus-dependent memory and hippocampal plasticity might be mediated by local SST+ interneurons. This finding may aid in the development of new therapeutic strategies to fight disorders that are associated with both sleep loss and memory deficits or structural disruption in the hippocampus, such as Alzheimer's disease.

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Poster

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Support: VA I01 BX001404 (RB),
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Title: Interactive role of basal forebrain neurons in sleep homeostasis

Authors: *R. BASHEER, C. YANG, H. BOUAOUDA, E. L. HODGES, D. S. UYGUN, J. T. MCKENNA;
VA Boston Healthcare System-Harvard Med. Sch., West Roxbury, MA

Abstract: Previous work by us and others suggested that basal forebrain (BF) cholinergic (ChAT+) neurons are important for the homeostatic sleep response (HSR) and the localized elevation of extracellular levels of the sleep-promoting neuromodulator, adenosine (ADex). Immunohistochemical and *in vitro* electrophysiological studies provide evidence for local interaction of cholinergic neurons with neighboring parvalbumin expressing GABAergic (PV-GABA) and vesicular glutamate transporter 2 expressing glutamatergic neurons (vGluT2+). Furthermore, blocking cholinergic receptors in BF during optogenetic cholinergic stimulations attenuated arousals suggesting that, within the BF, acetylcholine induces arousal indirectly via its action on neighboring non-cholinergic neurons. However, direct tests of the role of cholinergic vs. non-cholinergic systems for eliciting the HSR are lacking. To test the effect of individual neuronal subtype on HSR, we injected AAV-ChR2-EYFP virus into BF of ChAT-Cre, PV-Cre or vGluT2-Cre male and female mice also outfitted with EEG and EMG electrodes and optodialysis probe. Each set of mice were subjected to EEG/EMG recording and in vivo microdialysis from ZT0-ZT9 on (1) baseline day (no stimulation); (2) sleep deprivation (SD) day- SD from ZT3-7; and (3) stimulation day-opto-stimulation from ZT3-7 with or without pharmacological intervention. Levels of ADex were measured during SD or opto-stimulation. NREM sleep time and NREMδ (0.5-4Hz) were measured post-SD or post opto-stimulation from ZT 7-9.Preliminary results (N=3 for all) show both ChAT and vGluT2, but not GABA-PV, similar to SD effects, optogenetic stimulations led to increase in ADex levels (ChAT: 213%; vGluT2: 150%); and in post-stimulation, NREM sleep time and NREMδ (ChAT: 12.2±3% and 22±7% and vGluT2: 18.3±14% and 40±6%, respectively). Reverse microdialysis of a cocktail of cholinergic receptor antagonists during BF-ChAT+ neuronal stimulation did not attenuate ADex increase. On the contrary, administration of a cocktail of glutamate receptor antagonists attenuated ADex increase (62%) during BF-vGluT2+ neuronal stimulation. These preliminary observations suggest that cholinergic neuronal activity leads to increase in ADex, whereas the glutamatergic neurons elicit ADex increase via activation of cholinergic neurons.

Disclosures: R. Basheer: None. **C. Yang:** None. **H. Bouaouda:** None. **E.L. Hodges:** None. **D.S. Uygun:** None. **J.T. McKenna:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Merck MISP, Boston VA Research Institute.

Poster

P633. Sleep and Rhythms: Cellular and Molecular Contributors II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P633.08

Topic: F.08. Biological Rhythms and Sleep

Support: JSPS KAKENHI 18J21517

Title: Sik3 regulates sleep need via glutamatergic neurons in cerebral cortex in mice

Authors: *F. ASANO[1], T. FUJIYAMA[1], C. MIYOSHI[1], T. KITAZONO[1], N. HOTTA-HIRASHIMA[1], M. KAKIZAKI[1], A. IKKYU[1], S. KANNO[1], S. NAKATA[1], N. ASAMA[1], S. MIZUNO[2], F. SUGIYAMA[2], S. TAKAHASHI[1,2], H. FUNATO[1,3], M. YANAGISAWA[1,4,5]; [1]Intl. Inst. for Integrative Sleep Med. (WPI-IIIS), Univ. of Tsukuba, Tsukuba, Japan; [2]Fac. of Med., Univ. of Tsukuba, Tsukuba, Japan; [3]Dept. of Anat., Toho Univ., Tokyo, Japan; [4]Life Sci. Center, Tsukuba Advanced Res. Alliance, Univ. of Tsukuba, Tsukuba, Japan; [5]Dept. of Mol. Genet., Univ. of Texas Southwestern Med. Ctr., Dallas, TX

Abstract: Sleep is a fundamental behavior conserved from vertebrates to invertebrates. Although the “two-process model” has been proposed as a conceptual framework of sleep regulation, the molecular mechanism for homeostatic regulation of sleep and the neural substrate of sleep need remain unknown. *Salt-inducible kinase 3* (*Sik3*) is a member of AMP-activated protein kinase family, implicated in sleep need and circadian rhythm regulation. The gain-of-function *Sleepy* mutation in the *Sik3* gene increases sleep need and NREM sleep amounts. Most of systemic *Sik3* null mice die neonatally, but the few survived mice exhibit longer circadian period lengths. It remains unknown as to whether endogenous SIK3 is involved in sleep/wake regulation and, if so, which neuronal groups are responsible for the SIK3-dependent regulation. Here, we examined the sleep/wakefulness of neuronal type-specific SIK3-deficient mice. We established *Sik3-flox* mice, which abolished SIK3 expression in a Cre recombinase-dependent manner. *Sik3-flox* mice were crossed with various type of Cre driver mice to search for the SIK3-dependent sleep-regulating brain regions. We found that SIK3 deficiency in glutamatergic neurons (*Vglut2-cre; Sik3-flox* mice) resulted in reduced NREM sleep amounts. *Vglut2-cre; Sik3-flox* mice exhibited decreased delta power (1-4 Hz) during NREM sleep, which is a marker of sleep need, throughout the day. SIK3 is expressed broadly in brain and *Vglut2* positive cell lineage also covered many brain regions. Through further screening of various Cre lines, we found that *Vglut1-cre; Sik3-*

flox mice, whose SIK3 expression was diminished in glutamatergic neurons mainly in the cerebral cortex, showed reduced NREM sleep time and delta power in NREM sleep. We also confirmed that SIK3 deficiency in GABAergic neurons (*Vgat-cre; Sik3-flox* mice) did not change sleep time or delta power in NREM sleep. Instead, *Vgat-cre; Sik3-flox* mice showed a phase delay in sleep/wake pattern and longer free-running period length in constant dark conditions as previously described in *Sik3* null mice. Our results indicate that SIK3 regulates homeostatic sleep need through glutamatergic neurons in the cerebral cortex, and sleep need regulation was independent from the regulation of circadian rhythms by endogenous SIK3.

Disclosures: F. Asano: None. **T. Fujiyama:** None. **C. Miyoshi:** None. **T. Kitazono:** None. **N. Hotta-Hirashima:** None. **M. Kakizaki:** None. **A. Ikkyu:** None. **S. Kanno:** None. **S. Nakata:** None. **N. Asama:** None. **S. Mizuno:** None. **F. Sugiyama:** None. **S. Takahashi:** None. **H. Funato:** None. **M. Yanagisawa:** None.

Poster

P717. Pain, Fear, and Anxiety: Animal Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P717.01

Topic: G.04. Emotion

Title: Effect of 5HT7R agonist and antagonist on biological markers and pathophysiological mechanisms in neuropathic rats

Authors: *V. GOURA[1], P. JAYARAJAN[1], A. KISHORE[2], R. NIROGI[1];
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Abstract: Clinically it is acknowledged that treating neuropathic pain is most difficult due to comorbidities. To treat neuropathic pain, physician must evaluate all aspects of pain comorbidities like depression, anxiety, sleep and cognition of patients. As per literature, biological and neuroimaging data show that common brain areas are involved in the modulation of pain and its comorbidities. Improved understanding of the analgesics, its effect on biological markers of pain, and its comorbidities may offer promising avenues to treat neuropathic pain. We have tried to understand the effect of a newly identified G-protein coupled receptor (GPCR), the serotonin receptor subtype 7 (5-HT7). In this study, LP-211 a 5-HT_7R agonist and SB-269970 a 5-HT_7R antagonist were evaluated to find their role on modulation of monoamines, neuroendocrine markers, neurotrophic markers and immune-modulators in neuropathic rats. Histological studies were performed to evaluate its effects on the sciatic nerve myelin sheath. We also performed *in-vivo*electrophysiology study in spinal dorsal horn neurons to support further pathophysiological mechanisms. In the current study, we tested LP-211, 3mg/kg, *i.p*., LP-211, 3mg/kg, *i.p.* + Gabapentin (GBP) , 10mg/kg, *i.p.*, SB-269970, 3mg/kg, *i.p.* and LP-211, 3mg/kg,

i.p. + SB-269970, 3mg/kg, *i.p.*, on partial sciatic nerve ligation induced neuropathic pain (PSNL) and streptozocin induced diabetic neuropathic pain (DNP) rats. Von Frey filaments were used for evaluating pain, monoamines (serotonin (5-HT), norepinephrine (NE) and dopamine (DA)) were estimated using high performance liquid chromtography (HPLC), neurotrophic markers, immune- modulators using ELISA kits, and neuroendocrine markers using liquid chromtography mass spectrometry (LC-MS/MS). Monoamines were significantly decreased in pain animals when compared to control animals. LP-211 and LP-211 + GBP combination showed significant increase in 5-HT, NE levels were maintained equivalent to control animal values and there is no improvement was observed on levels of DA. LP-211 and LP-211 + GBP combination showed significant increase in neurotrophic markers. Only LP-211 + GBP combination showed decrease in immune modulators and substance P. The decreased myelin sheath density was restored by LP-211 + GBP combination. Dorsal horn neuronal firing induced by mechanical stimulation in PSNL rats was significantly decreased by LP-211 + GBP combination. SB-269970 did not show any significant affect on biological markers in the tested animal models. Thus, LP-211 a 5-HT_7R agonist in combination with GBP would be a promising therapy for treating neuropathic pain and its comorbidities.

Disclosures: V. Goura: A. Employment/Salary (full or part-time):; full time employee. **P. Jayarajan:** A. Employment/Salary (full or part-time):; full time employee. **A. Kishore:** None. **R. Nirogi:** A. Employment/Salary (full or part-time):; full time employee.

Poster

P717. Pain, Fear, and Anxiety: Animal Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P717.02

Topic: G.04. Emotion

Title: Inoculation mouse model of endometriosis creates a lowered abdominal pain threshold and signs of anxiety and depression, secondary to chronic pain, in the home-cage and group housed conditions.

Authors: *P. J. SWEENEY;
Naason Sci. Inc., Osong, Korea, Republic of

Abstract: Endometriosis is a condition that involves the ovaries, fallopian tubes and pelvic tissue. The condition rarely spreads outside of the pelvis and is characterized by pain of varying degrees and can result in debilitation from chronic pain and infertility if left untreated. There are currently treatments available, but these often don’t fully address the condition in women suffering from it. We attempted to model endometriosis in a group (N=20) of Balb/C mice by creating an age- matched donor group that was hormonally primed to induce the onset of menstruation and the subsequent generation of menstrual tissue that, after decidualization, was

extracted, post euthanasia, from the donor mice. This tissue was then introduced to the uterus of the donor mice (N=20) and resulted in endometriosis like tissue changes with a large percentage (80 %) of the animals developing a cystic like response to IP injection of the inoculate. After the inoculation animals were observed in their home cages using a unique home cage monitoring system that allowed for temporal observation of locomotor function and behavior in group housed conditions. RESULTS: Animals who received the inoculate of donor tissue tolerated the IP injections of this donor tissue very well with no mortality in the 72 hours after IP injections. Animals who received the inoculate were seen to have a slower overall motility in the home-cage and a higher degree of thigmotactic behavior than control animals subjected to a suture in the colonic mesentery which would produce pain and a degree of discomfort. Animals in both groups, control and tissue receivers, showed signs that could be considered to be symptomatic of abdominal pain with tactile allodynia measured with abdominal von Frey filament showing a significantly lowered pain thresholds in those mice that had the inoculation procedure performed. This threshold was in the region of 50% lower in gram value - control and sham operated groups did not show this decreased pain threshold. Upon closer inspection of the tissue pathology it was found that the lowered pain threshold persisted in those animals that received the inoculate but was also significantly lower in the groups that went on to develop cysts in the peritoneum that were visible to the naked eye. Conclusions: The presence of an endometriosis-like condition imposed in the Balb/c mouse creates an increased sensitivity to pain as measured by von Frey filament as well as locomotor and suspected psychological symptoms associated with chronic neuropathic pain. Further investigation into the group housed social behavior and chronic pain associated behaviors is warranted.

Disclosures: P.J. Sweeney: None.

Poster

P717. Pain, Fear, and Anxiety: Animal Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P717.03

Topic: G.04. Emotion

Support: Grant-in-Aid for Scientific Research from the Japanese Ministry of Education, Culture, Sports, Science, and Technology (16K01505)
Research funds from Japanese Physical Therapy Association (JPTA19-A36 and JPTA20-A23)

Title: Alteration of emotional behavior and associated neuroanatomical and immunohistochemical properties in the corticolimbic system of OLETF rats regardless of diabetic stage

Authors: *R. OCHI[1], N. FUJITA[1], N. GOTO[1], H. NISHIJO[2], S. URAKAWA[1];
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Abstract: Metabolic disorder, such as diabetes, can be accompanied with psychiatric disorders. We have reported that 20-week-old Otsuka Long-Evans Tokushima fatty (OLETF) rats, a model of progressive type 2 diabetes with age, exhibit increased anxiety-like behavior with reduced brain area and increased cholecystokinin (CCK)-positive neurons in the corticolimbic system. Parvalbumin (PV)-positive neurons in the corticolimbic system are also reportedly involved in anxiety. However, those alterations in different stages of type 2 diabetes remain unclear. Some of the diabetic animals reportedly show increased neuronal apoptosis and activating transcription factor 4 (ATF4), involving apoptosis accompanied by caspase-3 activation. However, these apoptosis related properties in OLETF rats remain unclear. We aimed to investigate emotional behaviors and its possible mechanisms at different diabetic stages in OLETF rats. Eight- and 30-week-old OLETF rats were used as diabetic animal models at the prediabetic and progressive stages of type 2 diabetes, and age-matched Long Evans Tokushima Otsuka (LETO) rats were served as non-diabetic controls. Glucose levels were higher in OLETF rats than in LETO rats at both ages, and those increased with age in OLETF rats. To examine the emotional states, we performed the open field test. Locomotion in the center zone was less and latency to leave the center zone was longer in OLETF rats than in LETO rats at 8 and 30 weeks old, respectively. To examine possible mechanisms of altered emotional behaviors in OLETF rats, we measured brain regional areas and performed immunohistochemistry. The area of the medial prefrontal cortex was smaller in OLETF rats than in LETO rats at both ages. The densities of CCK-positive neurons of OLETF rats were higher than those of LETO rats in the lateral and basolateral amygdala at only 8 weeks old, and in the anterior cingulate and infralimbic cortices and hippocampal cornu ammonis area 3 at both ages. The densities of PV-positive neurons of OLETF rats were lower than those of LETO rats in the prelimbic and infralimbic cortices at both ages, and in the hippocampal cornu ammonis area 2 at only 8 weeks old. To examine the mechanisms of those area and neuronal reductions, we performed immunohistochemistry for caspase-3 and ATF4. No caspase-3-positive cell was detected in both strains, but the percentage of ATF4 co-expression in CCK- and PV-positive neurons was higher in OLETF rats than in LETO rats at both ages in the anterior cingulate cortex and basolateral amygdala, respectively. These results suggest neurological changes in the corticolimbic system affect in emotional behavior already at prediabetic stage of OLETF rats.

Disclosures: R. Ochi: None. **N. Fujita:** None. **N. Goto:** None. **H. Nishijo:** None. **S. Urakawa:** None.

Poster

P717. Pain, Fear, and Anxiety: Animal Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P717.04

Topic: G.04. Emotion

Support: NIDA DA041781
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NIDA DA045463

Title: Pain increases inhibitory tone from RMTg onto VTA dopamine neurons to drive anhedonia-like behaviors

Authors: *T. MARKOVIC[1], N. MASSALY[1], Y. VACHEZ[1], C. MURPHY[1], K. ABIRAMAN[1], B. RUYLE[1], H. YOON[1], S. M. CONWAY[1], M. MIKATI[1], R. AL-HASANI[3], M. CREED[2], J. MORON-CONCEPCION[4];
[2]Dept. of Pharmacol., [1]Washington Univ. in St. Louis, Saint Louis, MO; [3]Dept. of Anesthesiol., St. Louis Col. of Pharm. & Washington Uni, Saint Louis, MO; [4]Anesthesiol., Washington Univ. In St Louis, St Louis, MO

Abstract: The persistence of negative affect in pain leads to co-morbid symptoms, such as anhedonia and depression, representing a major health issue in the US. However, the neuronal circuitry and contribution of specific cellular populations underlying these behavioral adaptations remains unknown. A common characteristic of negative affect referred as anhedonia is a decrease in motivation to initiate and complete goal-directed behavior. Here, we found that the resting membrane potential of VTA DA neurons, which are critical mediators of motivational and hedonic states, was decreased in pain animals while the rheobase was significantly higher. We next assessed the contribution of inhibitory inputs to this decrease in excitability, by washing on $GABA_A$ receptor antagonist picrotoxin onto the slice. While the application of picrotoxin had negligible effect on the control group, it depolarized the VTA DA cells from the pain group to levels comparable to control animals.Next, we measured the spontaneous inhibitory postsynaptic currents as a reflection of spontaneous neurotransmitter release. We found that pain increases the frequency of spontaneous events indicating enhanced pre-synaptic release and an increase inhibitory drive onto VTA DA neurons.One of the main sources of inhibitory drive onto the VTA DA neurons is rostromedial tegmental nucleus (RMTg) and inhibition of VTA DA neurons is known to mediate anhedonia-like behaviors. Thus, we hypothesized that pain enhances RMTg GABA input onto VTA DA neurons. Indeed, we show that pain increases release probability of RMTg GABA terminals onto VTA DA neurons. Furthermore, we show that activation of RMTg GABA neurons in naïve animals is sufficient to induce a decrease in consumption of sucrose solution in two-botte choice task similar to pain-induced anhedonia-like states. Lastly, we demonstrate that chemogenetic inhibition of RMTg GABAergic input onto VTA DA neurons is sufficient to prevent anhedonia-like behaviors in animals in pain.Previous findings indicate that pain induces dysfunction of μ opioid receptor (MOR) system within VTA. In addition, RMTg represents the major opioid sensitive inhibitory input onto VTA DA neurons. Thus, the ongoing experiments utilizing pharmacology and in vivo microdyalisis will assess the effects of pain on MOR system function and whether these changes contribute to the increased inhibition from RMTg onto VTA DA neurons in pain. In conclusion, our results provide further understanding of

pain's regulation of the inhibitory input onto VTA DA neurons and its consequences on anhedonia-like behaviors.

Disclosures: T. Markovic: None. **N. Massaly:** None. **Y. Vachez:** None. **C. Murphy:** None. **K. Abiraman:** None. **B. Ruyle:** None. **H. Yoon:** None. **S.M. Conway:** None. **M. Mikati:** None. **R. Al-Hasani:** None. **M. Creed:** None. **J. Moron-Concepcion:** None.

Poster

P717. Pain, Fear, and Anxiety: Animal Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P717.05

Topic: G.04. Emotion

Support: UMSNH/CIC

Title: Anxiety: ethanol and chlorazepate oral consumption after reward loss

Authors: *L. MANZO[1], E. MAGAÑA[2], A. C. TAFOLLA[2];
[2]Postgrado, [1]Univ. Michoacana De San Nicolas De Hidalgo, Morelia, Mexico

Abstract: Male rats from the Wistar strain received training in an instrumental extinction (reward loss) task in a straight alley after acquisition with continuous reinforcement (CR; 12 pellets per trial) vs. 50% partial reinforcement (PR; 12 or zero pellets per trial in random sequence). Extinction after CR training is known to induce a higher level of emotional activation than extinction after PR. Increased emotional activation is reflected in enhanced behavioral suppression (i.e., faster extinction) during episodes of reward loss after CR than after PR acquisition. We measured the impact of the reward loss experience in a 2-h preference test administered immediately after extinction sessions. increased preference for ethanol after being exposed to sessions of appetitive extinction (Manzo et al., Physiol Behav 2014 123:86-92). Such increased fluid preference did not occur after acquisition (reinforced) sessions or in control groups with postsession access to water, rather than ethanol. Because ethanol has anxiolytic properties in tasks involving reward loss, oral consumption after extinction sessions was interpreted as anti-anxiety self-medication. To test this hypothesis, Wistar rats exposed to a 12-to-zero pellets event in a PR situation were given a two-bottle, 2-h preference test immediately after extintion sessions. One of the two bottles contained 2% ethanol, 1.5 mg/kg chlorazepate, or water for different groups (the second bottle contained water for all groups). Chlorazepate is a potentially addictive benzodiazepine anxiolytic used in the psychiatric treatment of individuals suffering from anxiety disorders. Three additional groups received the same postsession preference tests, but were always exposed to 12 pellets during CR. Rats showed during extinction session effect, suppressing behavior after the downshift relative to unshifted controls. This effect was accompanied by a selective increase in both ethanol and chlorazepate oral intake

during the initial downshift sessions. Downshifted animals with access to water or unshifted controls with access to the anxiolytics failed to exhibit any changes in preference during the postsession test. This study shows for the first time that a prescription anxiolytic supports enhanced voluntary consumption during periods of emotional distress triggered by reward loss. These results support the hypothesis that anti-anxiety self-medication may provide insights into the early stages of addictive behavior.

Disclosures: L. Manzo: None. **E. Magaña:** None. **A.C. Tafolla:** None.

Poster

P717. Pain, Fear, and Anxiety: Animal Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P717.06

Topic: G.04. Emotion

Support: 1R15 AG060461-(01)

Title: Effects of forebrain NMDA receptor chimeric GluN2 subunit expression on anxiety behavior

Authors: *J. I. ESCOBAR[1], T. C. DUMAS[2];
[2]Psychology, [1]George Mason Univ., Fairfax, VA

Abstract: Title: Effects of forebrain NMDA receptor chimeric GluN2 subunit expression on anxiety behavior **Authors:** J. I. Escobar; Perdomo, Fares, Wild, Mastracchio, Keith, T. C. Dumas **Theme & Topic**:Theme G: Motivation and EmotionSubsection 4: EmotionsSub-subsection C: Emotion – Fear, anxiety, and painTheme B: Neural Excitability, Synapses and GliaSubsection 2: Ligand-gated ion channelsSub-subsection C: Ionotropic glutamate receptors **Grant Support:** 1R15 AG060461-(01)**Key Words (3):** NMDA Receptors, Transgenic Mice, Anxiety**Date of Submission:** July 15th, 2021**Abstract (Maximum 2300 Characters – Not including spaces):** Open channel blockers of N-methyl-D-aspartate receptors at excitatory glutamatergic synapses have been shown to ameliorate the symptoms of depression and anxiety in animal models and human beings. The current presumption is that drugs like ketamine antagonize extra synaptic NMDARs while causing a burst of release of glutamate, which returns depressed synapses to normal synaptic strength and completes circuits required for optimal mood regulation. An alternative explanation is that ketamine causes an increase in nonionotropic NMDAR signaling that permits subsequent activity-dependent synaptic strengthening. The latter notion has not been tested. However, nonionotropic NMDAR signaling influences on anxiety can be tested in mice with modified NMDAR subunits. GluN2 subunits are proposed to mediate nonionotropic signaling via their intracellular carboxy termini (CTD). By switching the CTDs of GluN2A and GluN2B it is possible to gain some insight into the distinct signals produced by

each CTD. Our “ABc” chimeric mice express the GluN2A amino terminal domain and transmembrane domain with the GluN2B CTD driven in the forebrain through the alpha calcium-calmodulin kinase II (CaMKII) minimal promoter. Conversely, our “BAc” chimeric mice express GluN2B amino terminal domain and transmembrane domain with the GluN2A CTD. Using the Open Field Maze (OFM) and Elevated Plus Maze (EPM), anxiety levels assessed in part through measures of latency to enter and dwell time in open regions of the mazes. Tests on young adult chimeric and wildtype mice on the EPM and OFM revealed no differences in anxiety levels across genotypes, which suggests that that GluN2 CTD signaling alone is likely not responsible for mediation of anxiety behaviors. Developmental experiments are underway to better understand how alterations in NMDAR signaling impact development of anxiety regulating systems. Results from these studies may reveal novel targets for the treatment of human anxiety disorders.**Current Character Count**: 1698

Disclosures: J.I. Escobar: None. **T.C. Dumas:** None.

Poster

P717. Pain, Fear, and Anxiety: Animal Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P717.07

Topic: F.04. Stress and the Brain

Support: NIH Grant MH114184

Title: Contribution of Neuropeptide Y (NPY) Y5 receptors in the basolateral amygdala to social interaction in male and female rats.

Authors: *J. H. URBAN[1], J. E. VANTREASE[2], M. R. DEJOSEPH[3], C. SAGARTZ[4], W. F. COLMERS[5];
[1]Ctr. for Neurobio. of Stress Resilience and Psychiatric Disorders; Physiol & Biophys, Rosalind Franklin Univ., North Chicago, IL; [2]Cell. and Mol. Pharmacol., Rosalind Franklin Univ. of Med. & Sci., North Chicago, IL; [3]Physiol & Biophy, Rosalind Franklin, North Chicago, IL; [4]DePaul Univ., Chicago, IL; [5]Univ. Alberta, Edmonton, AB, Canada

Abstract: Neuropeptide Y (NPY) is associated with resilience in humans and behavioral resistance in animal models of stress. Within the basolateral amygdala (BLA) both acute and repeated activation of NPY receptors increases social interaction (SI) and buffers against the deleterious effects of stress (Sajdyk et al., 2008). One of the striking effects of repeated NPY injections includes the induction of neuroplasticity of BLA neurons that is associated with persistent increases in SI and stress resistance as well as decreased freezing during fear extinction in male rats. Of the multiple NPY receptors expressed within the BLA, activation of either Y1R or Y5R increases SI acutely, and activation of Y5R, not Y1R, underlies the

generation of the persistent effects of NPY thereby supporting a novel role for the Y5R subtype (Michaelson et al., 2020). These findings provided an interesting dichotomy to the functions of Y1 and Y5 receptors in the BLA and put a new focus on the importance of the relatively understudied Y5R. This study was designed to examine the contribution of Y5R to the enhancement of SI and alterations in stress and BLA-related behaviors. Adult male and female rats received injections of shRNA-Y5R (lentiviral vector; Addgene) into the BLA and SI behavior was monitored weekly at 2-4 weeks post-injection. Behavior on the elevated plus maze (EPM) was also assessed at 4 wks. At the end of the experiment, brain tissue was collected to verify injection placements and degree of Y5R knockdown using Western analysis and immunohistochemistry. Using Western blot analysis, we observed decreases (40-60%) in Y5R protein after shRNA-Y5R treatment when compared with uninjected control BLA tissue. SI was decreased in both male and female rats 2 weeks post injection and decreased further at 3 wks ($p<0.05$, ANOVA) compared to their baseline SI (prior to virus injection). There were no significant changes in open arm time in the EPM, although a more complete analysis is being finalized. These data indicate that the Y5R plays an important role in regulating SI behavior in male and female rats and support the development of future studies to further characterize the circuitry involved in these NPY-mediated responses.

Disclosures: J.H. Urban: None. **J.E. Vantrease:** None. **M.R. DeJoseph:** None. **C. Sagartz:** None. **W.F. Colmers:** None.

Poster

P725. Acquisition I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P725.01

Topic: G.02. Reward and Appetitive Learning and Memory

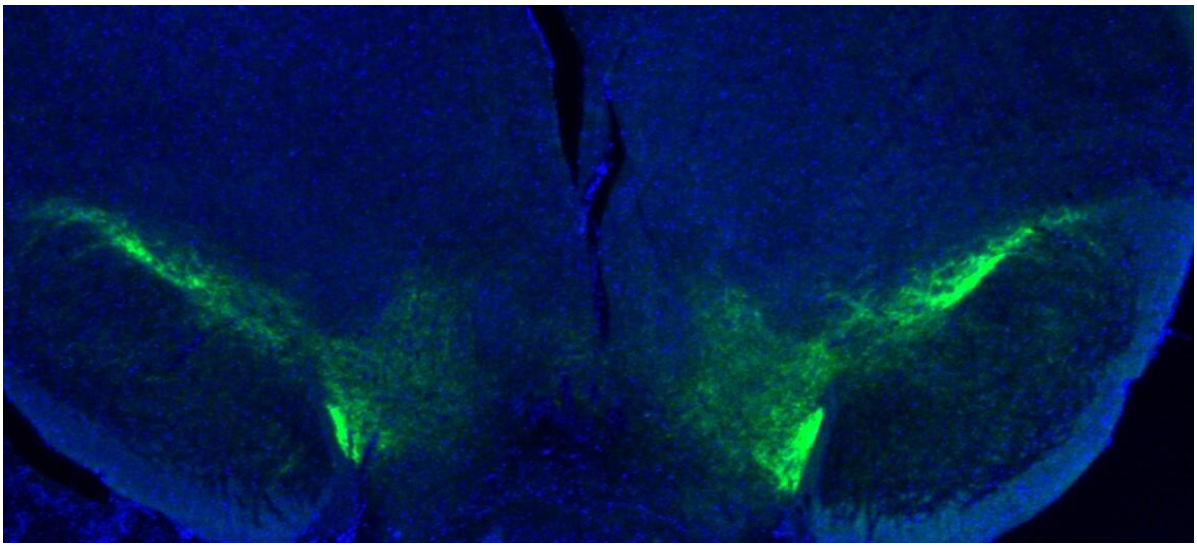
Support: NIH Grant R15MH122729-01

Title: Investigating the effect of differential striatal dopamine projections on reversal learning and immediate early gene (IEGs) expression

Authors: *R. K. VAN DER MERWE, M. GHANEM, J. A. NADEL, D. COPES-FINKE, S. PAWELKO, J. S. SCOTT, M. FOX, C. MOREHOUSE, R. MCLAUGHLIN, C. MADDOX, G. MALAKI, C. D. HOWARD;
Oberlin Col., Oberlin, OH

Abstract: Behavioral flexibility is key to survival in a dynamic environment. One form of flexibility is reversal learning, or the ability to switch between opposing actions following a change in outcome. Key to this process is the striatum, a subcortical structure tasked with action selection and the learning of novel motor behaviors. Early development of motor behaviors is

dependent on the dorsomedial striatum, which later becomes dependent on lateral striatum as behaviors become inflexible. Similarly, dopamine release in striatum mirrors this medial-to-lateral transition, suggesting that lateral dopamine release may be a key mechanism in establishing inflexible behaviors late in learning, though this has not been directly tested. To investigate this, we bred DATcre x Ai32 mice to allow selective optogenetic activation of dopamine terminals within medial, lateral, and ventral striatum. These mice performed an intracranial self-stimulation reversal task in which they received optogenetic stimulation to medial (n=7), lateral (n=7), or ventral (n=8) dopamine inputs when they pressed one of two levers, before the active lever was switched. Consistent with presumed ventromedial/lateral striatal function, we found that mice self-stimulating ventral or medial striatal dopamine were flexible following reversal, while activation of lateral striatal dopamine impaired reversal. This effect was associated with no difference in place preference or locomotion in an open field across groups. In addition, pilot studies suggest optogenetic activation of medial and lateral dopamine modifies reversal for food rewards, and this manipulation may be associated with striatal immediate early gene (IEG) expression, which support synaptic plasticity and learning. This work contributes to our general understanding of how dopamine modifies subcircuits within the striatum to switch between different behavioral strategies, and may provide insight into how drugs of abuse promote inflexible, habitual behaviors in drug addiction.



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Poster

P725. Acquisition I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P725.02

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIDA R01 DA044960

Title: Variation of the inter-trial interval during associative conditioning produces sex differences in sign-and goal-tracking behavior

Authors: *F. CZESAK[1], J. MORROW[2];
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Abstract: Variation of the inter-trial interval during associative conditioning produces sex differences in sign-and goal-tracking behaviorWhen associating a neutral stimulus (CS) with a reward (US), individual differences can be revealed in the propensity to attribute incentive salience to the CS. When the presentation of a lever (CS) is paired with a food reward (US), some rats will travel immediately to the food magazine to obtain the reward. This is called goal-tracking behavior, and these animals seem to interpret the CS simply as a predictor for the US. Other rats, upon associating the lever with the food reward, will interact with the lever itself, thus exhibiting sign-tracking behavior and suggesting an attribution of incentive salience to the CS. The behavior a rat will express has been found to differ depending on the length of the interval between associations of the CS and US. Here we asked how altering this inter-trial interval (ITI) between associations affects the attribution of incentive salience to reward cues in both males and females. Rats were trained using the PCA paradigm in experimental groups with different ITI ranges – 20-30s, 30-60s, or 30-260s. The results showed that increasing the ITI during PCA training causes increased sign-tracking behavior in all animals, and thus a greater likelihood of attributing incentive salience to the cue. Most importantly, females were significantly more likely to increase sign-tracking behavior with an increased ITI than males. These data suggest that males and females differ in their propensity to attribute incentive salience to cues, and highlight the importance of considering ITI length when studying Pavlovian conditioning in both sexes.

Disclosures: F. Czesak: None. **J. Morrow:** None.

Poster

P725. Acquisition I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P725.03

Topic: G.02. Reward and Appetitive Learning and Memory

Support: JSPS KAKENHI 18K03182
JSPS KAKENHI 24530917

Title: Delayed reward presentation among well-trained mice with lever preferences: Examining sign- and goal-tracking behaviors

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Abstract: When the presentation of an anticipated reward is delayed in an instrumental learning situation, despite the correct behavioral response for reward acquisition, various behaviors occur that do not directly contribute to the appetitive results. In experimental sessions with delayed reward presentation, some of these behavior patterns might facilitate or hinder reward acquisition. Thus, in this study, we examined a small sample of well-trained mice in a Skinner box. We focused on their preference between the right or left lever and measured both their lever-pressing and nose-poke responses to a pellet under five different time-lengths from lever-pressing to the delayed food pellet presentation (in seconds): 0 (no delay), 10, 20, 30, and 40. For this purpose, we measured the number of lever-pressing and nose-poke responses, of seven well-trained C57BL/6N mice, to acquire a pellet. Animals could press the levers in the predefined delay duration, but this action did not result in acquiring a food pellet. Response latency to the lever-presentation after predetermined delay was also measured for each trial. Moreover, the differences observed in the number of responses between the trials at 10 and 20 seconds, 20 and 30 seconds, and 30 and 40 seconds, were used to estimate the frequency of lever-pressing and nose-poke responses at the second, third, and fourth 10-second intervals, respectively, within the five reward delay trials. The animals were divided into two groups based on their lever preference: 1) the high preference group pressed either of the two levers at a rate of 70% or more and 2) the low preference group pressed both the levers at a rate of less than 70%. An exact probability test indicated that the high preference group tended to acquire more reward pellets in the overall sessions. In addition, the high preference group showed more correct responses, thereby acquiring more food pellets, than the low preference group within a test session. The high preference group also produced a relatively higher frequency of lever-pressing that did not contribute to appetitive results at the first 10-second reward delay interval, but not at any other intervals. Furthermore, the response latency from the lever presentation to pressing it was shorter in the high preference group. However, there were no significant group differences in the frequency of nose-poke responses during the delay intervals. The high preference group seemed to acquire more food pellets due to short latency to lever press, and to display a higher frequency of sign-tracking behavior immediately after the reward delay commencement.

Disclosures: T. Sato: None. **T. Yamakuni:** None.

Poster

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Program #/Poster #: P725.04

Topic: G.02. Reward and Appetitive Learning and Memory

Support: BG: Basic Science Program of the NRU Higher School of Economics
BSG: ANR-17-EURE-0017, and ANR-10-IDEX-0001-02
MR,TH: DA925634

Title: Homeostatically regulated reinforcement learning accounts for sodium balance-restoring behavior and state-dependent taste-specific dopamine signaling.

Authors: A. DURIEZ[1], T. HSU[2], M. F. ROITMAN[3], ***B. GUTKIN**[1,4];
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Abstract: Seeking nutritive outcomes in anticipation of homeostatic need is crucial for survival. Stimuli predictive of nutritive outcomes invigorate approach towards obtaining the nutritive outcomes with behaviour driven by learned hedonic values even in absence of need. The taste of nutritive stimuli acquires value that appears to depend on the internal state of the organism. Sodium appetite is an excellent demonstration of this, where only sodium-depleted animals seek out and consume hypertonic sodium solutions and not other salts (e.g. potassium chloride). Experiments showed that dopamine signals triggered by sodium stimuli depend both on the internal state and taste. How such modulations of dopamine signalling may generate sodium-motivated behaviour, in an interplay of innate and learned processes, has been an open question. Classical reinforcement learning (RL) models have not been able to account for appetite dependent changes in responding, nor for how internal needs modulate dopaminergic signals. A recent homeostatically regulated reinforcement learning theory (HRRL) has proposed a solution to this problem by defining the origins of rewards in a homeostatic manner: as outcomes that tend to re-establish homeostatic stability. Hedonic values are then learned for nutritive outcomes and assigned to stimuli that predict them. The theory also shows how responding is modulated by internal states and predicts that dopaminergic activity that signals the reward-prediction errors (among other things) is modulated as a function of the internal state. Last but not least, the theory suggests that sensory properties of the consumed goods can be a predictor of their impact on the internal state. We apply HRRL to sodium-seeking behavior, showing that it can fully account for the experimental data. We show that our simulated sodium-depleted HRRL agents replicate conditioned responding for sodium and stimuli predictive of sodium delivery, but not to potassium. We further show that this responding vanishes as the agent is satiated. The models explain how the experimentally observed modulation of dopamine signalling may reflect the state-dependence of the reward-prediction errors at consumption and predictive hedonic values at salient stimulus presentation. Finally we suggest that orosensory-based predictions of postingestive impact drive the state-dependent dopamine signals within the Nucleus accumbens. In sum, we suggest that sodium appetite is a paradigmatic example of homeostatically motivated

learned pre-emptive behaviour. Furthermore, we propose that taste is at least partially learned to act as an unbiased predictor of the state-dependent hedonic value.

Disclosures: A. Duriez: None. **T. Hsu:** None. **M.F. Roitman:** None. **B. Gutkin:** None.

Poster

P725. Acquisition I

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Program #/Poster #: P725.05

Topic: G.02. Reward and Appetitive Learning and Memory

Support: CIHR Grant 401526

Title: Chemogenetic activation of Prelimbic to Dorsomedial Striatum Pathway during acquisition attenuates goal-directed control

Authors: *M. L. SHIPMAN, J. A. EMTAGE, L. H. CORBIT;
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Abstract: The dorsomedial striatum (DMS) has been well established as a mediator of goal-directed behaviour, since lesion or inactivation before training or testing result in an insensitivity to reinforcer devaluation. Increased spontaneous activity in the same region following cocaine leads to a similar behavioural deficit suggesting that dysfunction, whether due to decreased or increased activity in the region, result in the same reduction in goal-directed control. We therefore aimed to evaluate whether tonically activating specific inputs to the DMS would produce deficits in goal-directed control. The prelimbic cortex (PL) sends robust glutamatergic afferents to the DMS and is also important in goal-directed responding. Because of this, it is a likely candidate for the increased glutamatergic input that impairs goal-directed control with drug and diet manipulations. We therefore utilized a dual-vector chemogenetic approach to activate the PL-DMS pathway while rats were trained on two levers with unique outcomes. We hypothesized that tonic activation of this pathway during training would interfere with the acquisition of specific response-outcome associations resulting in impairments in subsequent devaluation tests. Subjects were 8 male and 8 female Long Evans rats. Half of the rats received an active cre-dependent DREADD vector (AAV-hSyn-DIO-hM3D(Gq)-mCherry) while the other half received a control vector (AAV-hSyn-DIO-mCherry) in the PL in addition to all receiving a retrograde Cre vector (AAV-hSyn-HI-eGFP-Cre) in the DMS. Training began with two levers present, both associated with a common outcome. Once initial responding was acquired, training progressed where each lever now resulted in a specific outcome. Rats were injected with Clozapine dihydrochloride peripherally 30 min prior to each specific outcome acquisition session (6 sessions; 3 RR5 and 3 RR10). Choice extinction tests were then conducted following devaluation of one outcome by specific satiety. Histology and c-fos staining was

utilized to confirm activation of striatal MSNs following clozapine application and rats without expression or evidence of activation were removed from analysis (final N=5 for DREADDs and N=5 for controls). We found that responding was reduced for both the devalued and nondevalued reinforcers in the group trained under DREADDs activation suggesting reduced goal-directed control. These results suggest that DREADDs-mediated release of glutamate within the DMS during instrumental learning interfere with normal encoding instrumental associations. Ongoing studies are increasing sample size to examine potential sex differences.

Disclosures: M.L. Shipman: None. **J.A. Emtage:** None. **L.H. Corbit:** None.

Poster

P725. Acquisition I

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Program #/Poster #: P725.06

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIDA Grant R01 DA035943

Title: Mesostriatal dopamine is sensitive to Pavlovian information loss

Authors: *E. GARR[1], Y. CHENG[1], S. BROOKE[2], P. H. JANAK[1];
[1]Psychological and Brain Sci., [2]Dept. of Neurosci., Johns Hopkins Univ., Baltimore, MD

Abstract: Decades of research suggest that temporal contiguity between events—how close they occur in time—is not sufficient to explain Pavlovian learning. Even when the interval between cue and reward is held constant, the rate of learning is nevertheless stunted by free rewards in the absence of the cue and shortened inter-trial intervals (ITIs). This has been taken as evidence that Pavlovian learning is a function of the mutual information between cue and reward—the degree to which a cue signals a change in reward rate relative to the overall rate. Despite the importance of information in learning, investigations of mesostriatal dopamine function continue to emphasize a role in learning based on value, which is orthogonal to information. We therefore sought to find out whether mesostriatal dopamine dynamics are sensitive to Pavlovian information loss. We used fiber photometry to record calcium transients in dopamine neurons in the ventral tegmental area (VTA) during magazine approach conditioning followed by selective contingency degradation in TH-Cre+ rats (n = 8). Anticipatory magazine approaches increased during conditioning and decreased during contingency degradation. We also tested whether behavior was similarly attenuated during ITI shortening in a separate cohort (n = 8), but could not find supporting evidence. Calcium transients showed dynamics consistent with reward prediction error signaling during conditioning, and during contingency degradation the dopamine response to cue and reward during degraded trials was overall lower than during non-degraded trials. Dopamine response to reward on degraded trials did not significantly differ when the same

reward was delivered non-contingently during the ITI. In a separate experiment, we asked whether degrading the contingency between an audiovisual cue and optogenetic VTA dopamine neuron stimulation would attenuate conditioned locomotion. During conditioning, conditioned locomotion increased over time in TH-Cre+ rats (n = 15), but not in TH-Cre- rats (n = 8). During contingency degradation—when stimulation was delivered randomly during the ITI but conditioning trials remained unchanged—conditioned locomotion in TH-Cre+ rats was attenuated compared to a group of TH-Cre- rats and a group of TH-Cre+ rats that did not experience contingency degradation. These results suggest that VTA dopamine neuron activity within and outside conditioning trials can support sensitivity to contingency degradation.

Disclosures: E. Garr: None. **Y. Cheng:** None. **S. Brooke:** None. **P.H. Janak:** None.

Poster

P725. Acquisition I

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Program #/Poster #: P725.07

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIMH R01MH108729
NSF IOS-1656488

Title: Chemogenetic suppression of the perirhinal and postrhinal cortex impairs value inference in appetitive sensory preconditioning

Authors: *X. PENG, D. L. POETA, M. SLUSAREWICZ, R. D. BURWELL;
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Abstract: Complex goal-directed behavior is often supported by internalized models representing associative relationships in the environment. Building such models often requires integrating memories from separate events. Recent evidence suggests that the hippocampus plays an important role in model-building across multiple episodes. However, it is unclear whether other medial temporal lobe (MTL) structures also contribute to this process. This study employed the Designer Receptors Exclusively Activated by Designer Drugs (DREADDs) technique to investigate the involvement of two parahippocampal structures, the perirhinal and postrhinal cortices, in memory integration and value inference. Adult male rats received either sham surgery or bilateral viral injections into the perirhinal or postrhinal cortex. Then they were trained and tested in a three-phase appetitive sensory preconditioning procedure. In phase one, rats received presentation of two pairs of auditory stimuli, each containing one preconditioned stimulus and one conditioned stimulus (CS). In phase two, one auditory stimulus (CS+) in one pair was paired with pellet delivery, whereas one stimulus (CS-) in the other pair was presented alone. Finally in phase three, the preconditioned stimulus from each pair was presented to the

rats, and their responding was recorded as the dependent measure. All rats received intraperitoneal injections of clozapine N-oxide before behavioral sessions. This led to temporary suppression of neural activity in the perirhinal or postrhinal cortex for the experimental groups but not the control group. Preliminary results show that the experimental group had lower levels of responding to the preconditioned stimulus than the control group, indicating that suppressing either the perirhinal or postrhinal cortex impairs the ability to infer stimulus value based on its associative relationships. These findings update our existing knowledge of how various MTL structures contribute to goal-directed behavior and point to possible cooperation between them and other cortical areas.

Disclosures: X. Peng: None. **D.L. Poeta:** None. **M. Slusarewicz:** None. **R.D. Burwell:** None.

Poster

P725. Acquisition I

Location: Virtual Only

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Program #/Poster #: P725.08

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NICHD Grant 5R01HD088411-05
NINDS Grant 5U19NS107616-04

Title: Instrumental conditioning of video-playback of maternal behavior

Authors: *K. QUIÑONES-LARACUENTE[1], N. LÓPEZ CARABALLO[2], A. T. SEIDENBERG[2], O. M. MAVRIDIS[2], R. C. FROEMKE[3];
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Abstract: Certain aspects of mammalian parental behavior may be innate, while other behaviors can be learned by experience (Dulac et al. Science 2014). One form of maternal care in mice involves retrieval of isolated pups, in which experienced care-takers respond to ultrasonic distress calls and other sensory cues to move pups back to the nest. Non-biological adults can become alloparents after experience, and begin retrieving pups after co-housing (Marlin et al. Nature 2015). Recently, we found that alloparenting behaviors can be learned by virgin female mice visually observing experienced mothers retrieving pups. Visual input from the superior colliculus likely provides a visual signal to hypothalamic oxytocin neurons, promoting onset of alloparenting in virgin female mice (Carcea et al. Nature 2021). Here we have begun to determine the specific sensory cues and features that enable learned alloparenting, asking which aspects of maternal behaviors virgins might be observing. These studies build on ongoing work from our lab showing that pup retrieval onset is accelerated in head-fixed virgin female mice passively observing video-playback of dams and pups (López Caraballo et al. SfN Abstracts

2021). We presented head-fixed virgin females with the option to trigger the playback of a pup retrieval video with the press of a lever. This design allows us to ask about decision-making processes and the 'effort' animals are willing to expend to view or not view specific videos. Head-fixed mice were trained to press a lever for a 10 s video-playback of a dam retrieving a stray pup back into the nest. By the third day, mice were pressing the lever on their own (1.0 ± 0.06 presses/min, N=8 virgin female mice). The number of presses after a video started decreased over days (day three: 5.2, and day four: 2.9 average presses after video-playback started) The same animals were then presented with video playback that was controlled to match the color palette and luminescence levels of the pup retrieval video (abstract video), but with scrambled visual features. After four days, these mice were instead pressing at a much lower rate for these scrambled videos (0.2 ± 0.06 presses/min). A separate cohort of mice were trained with bar presses solely for the scrambled video. Lever presses were lower than in the group that could press for maternal retrieval videos (day three: 0.3 ± 0.06 presses/min, day four: 0.2 ± 0.06 presses/min, N=4). These data suggest that mice can exert effort to watch videos of conspecifics, and that video playback of certain behaviors might be rewarding.

Disclosures: K. Quiñones-Laracuente: None. **N. López Caraballo:** None. **A.T. Seidenberg:** None. **O.M. Mavridis:** None. **R.C. Froemke:** None.

Poster

P767. Mechanisms of Attention: Human Studies I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P767.01

Topic: H.01. Attention

Support: IBS-R015-D1

Title: The effect of temporal expectation on motion direction representation of an occluded object

Authors: *S. SON[1,3], J. MOON[1], J. LEE[1,3,2];
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Abstract: When a moving object briefly disappears, people track the occluded object with the inner eye and predict when and where the object will reappear. Previous studies showed that the timing of the occluded object's reappearance was represented in the suppression of the brain wave's alpha oscillation (Rohenkohl and Nobre, 2011; Samaha et al., 2015). Other studies also suggested that the temporal expectation enhances certain features of the occluded object, such as contrast sensitivity (Rohenkohl et al., 2012) or resistance over distractors (Bonnefond and

Jensen, 2012; van Ede et al., 2018). However, little is known about how the temporal expectation dynamically interacts with the motion information of the temporarily disappeared object. In this study, we estimated the motion information of the occluded moving object from the spatially distributed pattern of electroencephalography (EEG) recordings. The participants were asked to pay attention to a Gabor stimulus moving from the periphery towards the center of the screen while maintaining fixation at the central fixation point. A moving Gabor was turned on briefly then disappeared for a fixed amount of time or a randomized duration. When it reappeared at the center of the screen, the participants were instructed to smoothly track the moving stimulus with their eyes until it stopped moving. To estimate the motion direction information while the stimulus was occluded, we used an inverted encoding model (Brouwer and Heeger, 2009). The result showed that the neural representation of motion direction expectation was highest at the expected timing of the occluded stimulus's reappearance but barely existed for the rest of the time. Also, the strength of the neural estimate of the direction information was predictive of the behavioral biases induced by the direction expectation. Further analysis showed that the motion direction information of the occluded stimulus was most evident in EEG data recorded at the anterior channels. At the same time, the attenuation of alpha oscillation by temporal expectation mainly occurred at the posterior channels. Significant modulation of motion direction representation by the temporal expectation was only observed from EEG recordings in the vertex region. Taken together, the neural representation of temporal expectation was spatially distinct from the representation of motion direction expectation, and the integration of the two appeared to occur at the brain regions close to the vertex channels.

Disclosures: S. Son: None. **J. Moon:** None. **J. Lee:** None.

Poster

P767. Mechanisms of Attention: Human Studies I

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Program #/Poster #: P767.02

Topic: H.01. Attention

Support: 100014_159506 of the Swiss National Science Foundation
100014_178814 of the Swiss National Science Foundation

Title: Using SSVEP to evaluate attentional modulation in Action Video Game Players during facial emotion discrimination

Authors: A. CIOBANU[1], K. SHIBATA[1], L. ALI[2], K. RIOJA[1], S. K. ANDERSEN[3], D. BAVELIER[4], ***B. BEDIOU**[1];
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Abstract: Attentional mechanisms have been posited to mediate benefits in decision making. We investigated whether increased attentional modulation is associated with enhanced discrimination of facial emotions. We compared behavioural and neural correlates of attention to emotion in Action video game players (AVGP) known to have enhanced attentional processing, with non-video game players (NVGP) (ages 18-35). AVGP (n=36) and NVGP (n=32) underwent an emotion discrimination task which required detecting target emotions in one of two laterally presented streams of faces flickering at different frequencies (2Hz/2.5Hz). Steady-state visual evoked potentials (SSVEP) were used to quantify the neural activity corresponding to the processing of attended and unattended emotional streams. AVGP outperformed NVGP at detecting target emotions in the attended stream regardless of emotion valence (Hedge's g = 0.51, p = 0.04). As expected, SSVEP amplitudes extracted from parieto-occipital electrodes revealed group differences in attention (Group x Attention interaction: $F(1, 1054) = 7.949$, p = 0.005, $\eta^2 = 0.007$), driven by non-significant trends for both greater attentional enhancement and greater attentional suppression in AVGP. Importantly, the amplitude difference between attended and unattended SSVEP amplitudes correlated with behavioral performance (r = 0.35, p=0.0031), indicating an association between neural and behavioral measures of attention. In sum, we find enhanced attentional modulation in AVGP to be associated with enhanced discrimination of facial emotions. Additional unregistered exploratory analyses revealed the expected classical ERP components associated with target detection (larger P3 to targets vs. non-targets, p<.0001) and error processing (larger Error Related Negativity to false alarms vs hits, p<.0001). Neither of these effects interacted with group (all p's > 0.28), although AVGP displayed overall larger amplitudes than NVGP to both targets and non-targets in the P3 range. In conclusion, this study provides evidence for enhanced attentional processing in AVGP that extend to the processing of emotional stimuli.

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Poster

P767. Mechanisms of Attention: Human Studies I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P767.03

Topic: H.01. Attention

Support: NIH Grant R01-DA046410

Title: Primary rewards and aversive outcomes have comparable effects on attentional bias

Authors: *H. KIM, B. ANDERSON;
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Abstract: Reward learning imbues initially neutral stimuli with value, such that they come to predict reward and capture attention even when non-salient and task-irrelevant. The same is true for aversive conditioning; stimuli that signal threat also bias attention, suggesting that motivational salience guides attention. Most research that supports this conclusion has manipulated monetary gain, a secondary reinforcer, for reward learning and electric shocks, a primary punisher, for aversive conditioning, making it difficult to directly compare their influence on attention. Therefore, we matched for reinforcer dimensions by using primary taste as reinforcers/punishers and assessed their influence on attention.
Participants first completed a decision-making task to determine their point of subjective equality. On each trial, a certain amount of sweet juice and salt water each ranging from 0.1 ml to 1 ml were indicated. Participants had to decide whether to place a bet. One of the two solutions, in the amount indicated, was delivered following a decision to place a bet (each equally-likely); no solution was delivered if participants chose not to place a bet. We fit a regression and determined the amount of each solution to be used in a training phase on the next day.
In the training phase, participants had to make a saccade to a peripherally-presented square target, which could appear in one of three colours. One colour was associated with sweet juice (reward) and another with salt water (aversive) on 80% of trials, in the amounts determined in the decision-making task. A third color was never followed by liquid (neutral). In a test phase, a square (distractor) and a circle (target) were presented simultaneously; the same colours were used as during training, fully crossed and counterbalanced with respect to target and distractor status. Participants had to make a saccade to the circle.
We analysed response times (RTs) and error rates from the test phase. Error rates were defined as the proportion of trials containing an initial saccade to the distractor. RTs slowed down marginally when an aversive distractor was presented with a neutral target than when an aversive target was presented with a neutral distractor. Error rates were higher when there was a reward distractor than when there was a reward target. Importantly, a direct comparison of reward and aversive stimuli revealed no significant differences in RT or error rate. These results suggest that when matched for reinforcer dimensions, reward and aversive stimuli both bias attention in a similar manner and that their effects are comparable, providing further evidence in support of the motivational salience account.

Disclosures: H. Kim: None. **B. Anderson:** None.

Poster

P767. Mechanisms of Attention: Human Studies I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P767.04

Topic: H.01. Attention

Title: Direct recordings of sustained attention dynamics in the human brain

Authors: *M. T. DEBETTENCOURT[1], N. HAKIM[2], T. XIE[1], M. PADMANABAN[1], E. AWH[3], E. K. VOGEL[1], P. C. WARNKE[1];
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Abstract: In our daily life, attention inevitably fluctuates over time, encompassing highly attentive moments to potentially catastrophic lapses of attention. These attention dynamics have been characterized using sustained attention to response tasks, in which behavioral responses predict forthcoming attention lapses. Studies have implicated particular subcortical areas, including the basal ganglia and thalamus, as important for successfully performing these tasks. In this study, we examined neural signatures of attentional dynamics by directly recording from the human brain. We collected data from 9 patients who were awake and performing a sustained attention task while undergoing surgery for deep brain stimulator implantation. The task required patients to click a button to frequently presented circle stimuli (80% of trials) and withhold a response to infrequent square stimuli (20% of trials). We found that this task elicited canonical behavioral signatures of sustained attention performance. Patients demonstrated attention lapses (29% lapse rate), operationalized as failures to inhibit the prepotent response to the infrequent (i.e., square) trials. Furthermore, when responding more quickly, patients were more likely to lapse to an upcoming square trial that required deviating from the prepotent response (mean RT=50 ms faster prior to lapses). Next, we examined whether these behavioral signatures were reflected in electrode recordings from the subthalamic nucleus (STN) of the basal ganglia and ventral intermediate (VIM) nucleus of the thalamus. Indeed, using cluster-based permutation tests, we observed that total power in low-frequency power bands (e.g., alpha, 8-12 Hz) was significantly higher during frequent than infrequent trials. We also demonstrated that worse attentional states, as indexed by RT, correlated with lower beta power (12-20 Hz). Furthermore, we used multivariate pattern analyses to classify high vs. low attentional states. We reliably decoded attentional state from time-frequency decompositions of macroelectrode recordings ($p<0.05$). Finally, we used microelectrode recordings to identify single units with different responses during sustained attentional states. Overall, these results provide comprehensive insight into how moment-to-moment fluctuations of sustained attention are reflected in the brain.

Disclosures: M.T. deBettencourt: None. **N. Hakim:** None. **T. Xie:** None. **M. Padmanaban:** None. **E. Awh:** None. **E.K. Vogel:** None. **P.C. Warnke:** None.

Poster

P767. Mechanisms of Attention: Human Studies I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P767.05

Topic: H.01. Attention

Support: Office of Naval Research PR# 1400850872

Title: Impacts of cognitive fatigue on attentional control and evoked neural activity

Authors: *D. MCHAIL, K. PETTIJOHN, K. J. BLACKER, S. SHERWOOD;
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Abstract: The warfighter must often exercise attention and inhibitory control for prolonged periods to achieve mission success. However, performance in cognitive domains, including attention, is known to degrade with increased time on task. It is therefore critical to understand how brain regions underlying attentional control (i.e., the prefrontal cortex, or PFC) respond to cognitive fatigue. In this study, we leverage validated attentional control tasks to assess the impacts of cognitive fatigue on task performance and associated changes in PFC activity. Twenty-six participants age 18-40 were recruited for the study, which utilized a counterbalanced, within-subjects design. On two separate days, participants performed computerized versions of two tasks that assessed attention and cognitive control through inhibition. One variant required a response to rare stimuli based on the preceding stimulus, and the other required frequent responses to common stimuli while withholding responses to rare stimuli. Prior to task performance, participants in the experimental fatigue condition completed the Psychomotor Vigilance Task (PVT) for 10 min. Participants in the non-fatigue control condition watched a short video in lieu of the PVT. PFC blood-oxygenation was measured with functional near-infrared spectroscopy (fNIRS) during task performance. Analyses compared the effects of the fatigue and non-fatigue conditions on task performance (i.e., accuracy, response time) and PFC blood oxygenation (deoxygenated and oxygenated hemoglobin changes). The results suggest that overall task performance did not differ between fatigue and non-fatigue conditions. However, a non-statistically significant trend for greater PFC activation (relative oxygenated hemoglobin) was apparent in the fatigue compared to the non-fatigue condition during the inhibitory control task. In contrast, PFC activation was marginally lower in the fatigue compared to non-fatigue condition during the vigilance task. This suggests that the PFC may increase activity to maintain performance in fatigued individuals for tasks that require attentional control through inhibition. Results from this study help clarify the impact of cognitive fatigue on attentional control and how the brain responds to maintain performance. Follow-on efforts will focus on individual differences in susceptibility to cognitive fatigue and its effect on other cognitive abilities (e.g. task switching). Ultimately, these efforts will help develop real-time cognitive fatigue alerting systems and countermeasures to maximize warfighter performance during sustained attention tasks.

Disclosures: D. McHail: None. **K. Pettijohn:** None. **K.J. Blacker:** None. **S. Sherwood:** None.

Poster

P767. Mechanisms of Attention: Human Studies I

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Topic: H.01. Attention

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Basic Science Research Program through the National Research Foundation of Korea funded by the Ministry of Education 2020R1I1A1A01052810

Title: Rhythmic phase modulation on divided attention sampling

Authors: *J. CHUNG, D.-J. YI;
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Abstract: Recent studies have demonstrated that sustained attention has rhythmic temporal structures. When participants allocate attention to a spatial location or salient stimulus, target detection fluctuates at around 8 Hz. Interestingly, this sampling frequency decreases when multiple locations or stimuli are attended. Here, we explored the phase relationship between divided attention samplings by a sequential presentation of two stimuli with distinct visual features. We hypothesized that the prior temporal status of sampling determines the subsequent phase relationship between multiple samplings. In two experiments, participants detected color saturation of moving dots. In Experiment 1 (N = 31), the fluctuation of target detection was first explored when a single stimulus was attended (1-Cloud task). Target onsets were varied across 250 to 750 ms with a 34-ms gap. We found a near 8-Hz fluctuation of hit rates along with the target onsets. We fitted it with a sinusoidal function individually to extract three time points (two at the minimum and one at the maximum peaks). A 2-Cloud task was then performed, which sequentially presented two stimuli, with the second stimuli onsets corresponding to the three time points. Target onsets were the same in the 1-Cloud task. Fluctuation frequencies were consistent across the different onsets (around 3 Hz for the first and 5 Hz for the second stimulus). However, the phase difference between the first and second stimulus fluctuations was different across the onsets. The phase differences of the minimum-peak-onset conditions showed anti-phase relations (180°) but not in the maximum-peak-onset condition. In Experiment 2 (N = 25), a 2-Cloud task was performed with varied second stimulus onsets across 250 to 750 ms with a 34-ms gap. We found that hit rate differences between the first and second stimulus along the second stimulus onsets show rhythmic fluctuations (4 Hz). The findings indicate that the second stimulus onset affects the phase relationship between divided attention samplings. The phase modulation was rhythmic depending on the pre-division sampling. This study suggests that the temporal structure of divided attention would differ depending on the prior attention state.

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Poster

P767. Mechanisms of Attention: Human Studies I

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Topic: H.01. Attention

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Title: Human population motor fields and population motor attention fields as they relate to saccadic eye movement accuracy

Authors: *W. E. HUDDLESTON[1], A. E. CATTERSON[1], M. J. PENNING[1], O. L. SWANSON[1], A. S. GREENBERG[2], E. A. DEYOE[3];
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Abstract: Attentional selection of task-relevant information is fundamental to our ability to interact with our complex natural environment. This process operates through modulation of multiple topographically organized brain maps. We hypothesize that the attentional control computations demonstrated in the visual system may be generalizable to information selection in other modalities (e.g., motor domain), suggesting these modulations may apply widely throughout the brain. In Experiment 1, we assessed saccade crowding limits and found wide variation among participants (between 3-12°) in their ability to plan separate saccades to two possible targets with variable spacing. In Experiment 2, we tested a second group of participants performing two separate tasks while undergoing fMRI. In the first task, participants made saccades between two diametrically opposed targets slowly rotating clockwise around fixation. From this saccade localizer task, we calculated population motor fields in maps across cortex. In a second task, participants completed a delayed saccade task while we concurrently collected eye movement data to determine saccade accuracy. Participants selectively attended to a centrally presented RSVP stream surrounded by 24 static peripheral targets (radius 8°). Every 4 s, an 'N" appeared in the central RSVP stream to cue the participant to prepare a saccade to the next target in the clockwise direction thereby creating a focus of motor attention (i.e., saccade intention) drifting through associated cortical saccade-target maps. However, participants delayed saccade execution until a rarely presented 'X' appeared in the RSVP stream (approx. 5 times per run). Two times per cycle, a number (12 or 6) in the central RSVP provided a cue indicating the correct 'clock' location to be attended at that moment to ensure accurate shifting of motor attention. fMRI data from the Attentional Drift Design task just described were then used to constrain a computational model of the size and shape of the focus of motor attention, the voxel's population motor attention field. Consistent with the variation of saccade crowding limits seen in Experiment 1, population motor attention fields not only varied among participants, but were uniquely selective for spatial locations within individual participants, indicating promise as a neural substrate of eye movement behavior. Establishing a direct link between brain activity

and behavior is a crucial first step on the development of treatment interventions for patients with attentional deficits.

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Poster

P767. Mechanisms of Attention: Human Studies I

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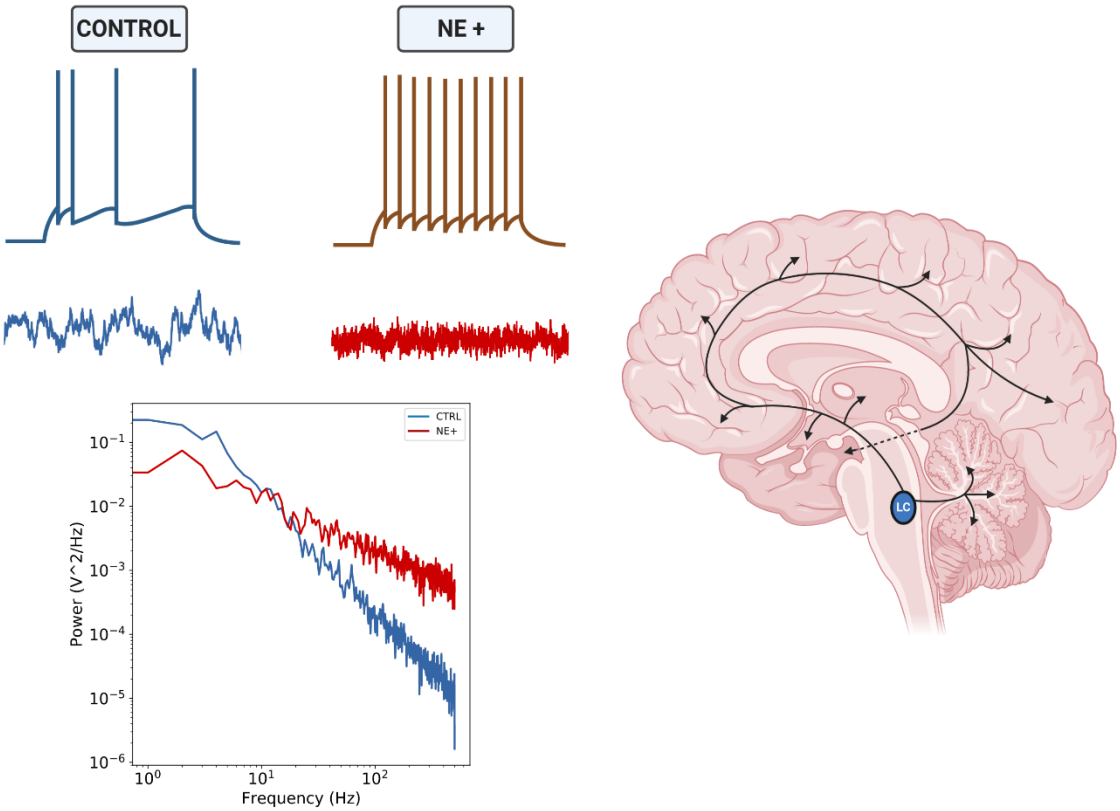
Support: BECA ANID N° 21180871
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FONDECYT N° 1160736
FONDECYT N° 3200248

Title: Interpreting the neuromodulation of 1/f aperiodic activity through the arousal-related switch of adaptation currents

Authors: *V. MEDEL[1], M. IRANI[1], B. MUNN[2], J. VALDÉS[1], F. GONZÁLEZ[1], B. FOLLET[1], G. WAINSTEIN[2], G. BONCOMPTE[1], J. M. SHINE[2], N. CROSSLEY[1], T. OSSANDÓN[1]; [1]Pontificia Univ. Católica de Chile, Santiago, Chile; [2]Univ. of Sydney, Sydney, Australia

Abstract: To actively engage in ever-changing environmental demands, animals dynamically switch between a variety of behavioral states that differentially affect neural responsiveness and the ability to optimally process information. Computational approaches have shown that the range of these activity patterns can be achieved by simulating a network of neurons with balanced excitatory and inhibitory synaptic activity modulated by activity-dependent adaptation on excitatory cells which dynamically close K+ channels, From this perspective, in order to shape the cortical activity and transition the cerebral cortex to an optimal state, the brain depends on global variables, such as relative levels of excitation and inhibition and the level of activity-dependent adaptation on excitatory populations. Interestingly, there is strong evidence showing that extracellular application of arousal-related neuromodulators, such as noradrenaline, eliminates the activity-dependent adaptation. This suggests that diminishing adaptation through neuromodulation could facilitate E/I balance and suggests a mechanistic role of ascending arousal system in regulating E/I balance and cortical state. Here we directly test this hypothesis by using a validated measure of E/I balance -the power spectral 1/f slope- and, with both a computational modelling and a human and non-human primate experimental approach, demonstrate a close relationship between activity-dependent adaptation and the 1/f slope of electrophysiological signals. Finally, we will show that fluctuations of arousal-related

neuromodulation (as measures by pupil diameter) track time-resolved changes of 1/f slope, suggesting a dynamic neuromodulatory mechanism shaping E/I balance and aperiodic activity in the cerebral cortex.



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Poster

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Support: Israel Science Foundation (ISF 526/17)
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Title: Closed-loop EEG-TMS modulation of frontal-occipital communication enhances visual perception

Authors: *S. KLORFELD-AUSLENDER[1], C. ZRENNER[3], S. YUVAL-GREENBERG[1,2], U. ZIEMANN[4], N. CENSOR[1,2];
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Abstract: Visual perception is constantly shaped by top-down mechanisms, such as attention. Indeed, it was shown that engagement of frontal and parietal regions that coactivate with the visual system, successfully prioritize processing of visual information. Previous studies suggested that these co-activations are expressed by lateralized alpha oscillations, measured at occipital electrode sites contralateral to an attended location, reflecting frontal-occipital communication. The goal of the current study was to causally reveal how communication patterns between frontal and occipital brain regions can facilitate visual performance. To address this question, we used a combination of electroencephalography (EEG) and non-invasive transcranial magnetic stimulation (TMS), in a closed-loop EEG-TMS procedure (Zrenner et al., 2018). TMS was delivered over the right frontal eye field (FEF) according to the phase of occipital alpha oscillations, measured in real-time with scalp EEG. This rationale was based on the notion that phase-locked oscillatory activity enables interregional synchronization and thus effective communication between anatomically distinct regions (Fries, 2005). Accordingly, we reasoned that enhanced frontal-occipital co-activation with closed-loop EEG-TMS would facilitate visual perception and its corresponding electrophysiological signatures. Subjects underwent a classical visual detection-attention task, known to require the engagement of frontal and occipital regions, before and after the closed-loop procedure. In the task, subjects were required to detect a peripheral low contrast target that appeared after the onset of either a valid or neutral cue. To assure there are no test-retest improvements that may be attributed to other factors, participants in a pilot no-stimulation condition completed the same task, without the stimulation procedure. Preliminary results show that frontal-occipital closed-loop EEG-TMS enhanced visual sensitivity in the neutral condition relative to performance improvements without stimulation. Consistently, the closed-loop stimulation to the right FEF induced greater right occipital alpha power suppression at the post-cue interval during the neutral, but not the valid, cue condition. If validated, these results suggest that real-time modulation of frontal-occipital communication can enhance visual perception.

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Poster

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Sloan Research Fellowship

Title: Disentangling the impact of top-down spatial attention and bottom-up stimulus drive on estimates of spatial selectivity in human cortex

Authors: *T. C. SPRAGUE, K. VU-CHEUNG, D. D. THAYER;
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Abstract: Visual attention enables the selection of relevant information in the environment for further processing. In single neurons, attention to a location of the visual field results in a warping of spatial receptive fields (RFs) measured from extrastriate visual cortex (e.g., Womelsdorf et al, 2006; Anton-Erxleben et al, 2009; Anton-Erxleben & Carrasco, 2013). Similarly, functional MRI experiments have identified changes in the spatial tuning of populations of neurons within voxels in retinotopic cortex whereby the location and/or size of voxel RFs (vRFs; Dumoulin & Wandell, 2008; Mackey et al, 2017) change as the locus of attention is manipulated between scanner runs (Klein et al, 2014; Sprague & Serences, 2013; Vo et al, 2017; van Es et al, 2018; Sheremata & Silver, 2015; Kay et al, 2015). In many of these studies, attention was directed either to the vRF mapping stimulus or the fixation point, which typically results in an increase in measured vRF size in extrastriate cortex when attending the mapping stimulus (Sprague & Serences, 2013; Kay et al, 2015; Sheremata & Silver, 2015; van Es et al, 2018). In other studies, attention was directed to a fixed location while vRFs are measured with an ignored mapping stimulus, which typically results in a shift in the vRF position with a change in the locus of spatial attention (Klein et al, 2014; Vo et al, 2017). However, in all these studies, the entire mapping stimulus was either attended or ignored. Thus, it remains unknown how vRFs depend on the locus of attention to a subpart of the mapping stimulus. Here, we acquired fMRI data while participants viewed a traversing bar vRF mapping stimulus broken into 3 segments. Each segment contained black and white dots moving in one of two directions. Across scanning runs, we manipulated which segment(s) of the bar participants attended using a central cue, and participants reported the direction of dot motion in the attended bar segment. Importantly, the stimulus sequence was fixed across scanning runs, so any changes in observed signals necessarily result from a change in the locus of attention. Direct comparison of activation timeseries revealed similar spatial selectivity, but a dynamic reweighting of activation levels at each voxel's preferred position as a function of which bar segment was attended. This reweighting was more pronounced in extrastriate visual cortex as compared to V1, consistent with previous reports that attentional modulation of vRF properties is smallest in V1. In ongoing work, we are fitting a variety of nonlinear vRF models (Kay et al, 2013; Mackey et al, 2017) incorporating the stimulus drive, attentional locus, and a combination of these factors.

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Poster

P770. Mechanisms of Attention: Human Studies II

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Title: Divide and Conquer: Own-Name detection during ecological concurrent speech processing

Authors: *D. PINTO, M. KAUFMAN, E. ZION-GOLUMBIC;
The Gonda Ctr. for Multidisciplinary Brain Research, Bar Ilan Univ., Ramat Gan, Israel

Abstract: A well-known phenomenon is the experience of noticing one's own name spoken by unattended speaker. This "Cocktail Party Effect" has fueled debates regarding the extent to which so-called "unattended" speech is processed. However, it is difficult to reliably gauge the prevalence of this effect since most studies use selective attention tasks, where detection of unattended words is assessed indirectly. For example, using subject-report measures, only ~33% of participants report noticing their name. This limits the ability to draw theoretical inferences regarding the depth of processing of concurrent speech. We address this from a different perspective by employing a Divided attention task. We ask two questions: What is the capacity for detecting individual words from background speech, while primarily engaging in processing a natural speech narrative? And does detecting one's own name enjoy a unique status? Participants were presented with two concurrent speech streams: A **Narrative** stream, which was a natural recording of continuous speech, and a **Starbucks** stream, that simulated a barista calling out orders, including names and food items. Participants were told to listen to the Narrative and answer comprehension questions. In addition, they were also required to monitor the Starbucks stream and detect a specific target-name, which could be their own name or a different name. Neural activity was measured using EEG during the task, as well as changes in skin-conductance (GSR). Accuracy in both tasks was high, with target detection rates near ceiling (>90%). Also, there were no apparent behavioral tradeoffs between performance on the two tasks. This suggests that individuals have a relatively good capacity for monitoring background speech and detecting specific words, without compromising comprehension of a main narrative. When comparing detection rates of the two targets, performance was better for detecting one's own name vs. a control name. This was accompanied by increased neural responses (in a time-window corresponding to the P600 component) as well as heightened GSR responses to one's name. These results provide new perspectives about the capacity for processing concurrent speech and indicate that it is possible to monitor background speech for specific information while also processing continuous speech. This is in line with 'late-selection' models and invites fine-tuning of theoretical accounts of 'bottlenecks' for processing concurrent speech. In addition, the advantage for detecting one's own name is in line with previous findings, indicating an advantage for processing personally meaningful information in noisy, multi-speaker contexts.

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Poster

P770. Mechanisms of Attention: Human Studies II

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Topic: H.01. Attention

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POS_FCE_2020_1_1009198

Title: The enhancement effects of background knowledge on selective auditory attention: an analysis of cortical responses in the cocktail party problem

Authors: *T. SÁNCHEZ-COSTA, A. CARBONI, F. CERVANTES CONSTANTINO; Facultad de Psicología, Univ. de la República, Uruguay, Montevideo, Uruguay

Abstract: When chats overlap, listeners will ignore so they get to hear the more interesting story. To solve this 'cocktail-party' problem, gain mechanisms by selective attention operate on the neural representations of individual speech objects. Foreground enhancement by attention has been observed to peak around 125 ms during online processing of speech in auditory cortex. In sensory cortex, it has been suggested that attentional and predictive processes interact to boost even further a foreground when it is predictable. Priming studies, by contrast, indicate that familiarity with a stimulus may produce lower neural responses than when it is novel. In a cocktail party, does prior experience of a foreground boost or revert foreground enhancement? Would background foreknowledge help reduce its representation during a subsequent selection? To address these questions, 27 participants attended individual speakers from brief (7-9 s) acoustic mixtures of two while undergoing electroencephalography (EEG). Each mixture was presented once and immediately preceded by a solo speech segment which could be identical to the foreground (attended known 'AK') or the background (unattended known, 'UK'), or to an unrelated stimulus (no prior, 'NP'). Every unique mixture was built anew from a diverse database of over 250 natural speech segments, per participant. Subjects identified aspects such as topic, keywords, and sex and age groups of the attended speaker.
Temporal response function (TRF) estimates of the auditory response were used to assess single-trial cortical encoding of speech across conditions. Attentional enhancement of the neural representation of foreground speech, observable around 140 ms (N1 component), improved in UK over AK conditions. Background knowledge boosted differential encoding more effectively than foreground knowledge did, even as UK trials appeared to be more difficult than AK behaviorally. The UK lead at this stage was associated to a significantly delayed, but also increased, P1 peak corresponding to the known unattended stream. Such changes also resulted in observable neural segregation effects by attention as early as 60 ms for UK conditions only.

The results indicate that while neural stream segregation normally occurs at the N1 stage without any prior knowledge, knowledge of the background is incorporated into segregation mechanisms much earlier. By contrast, foreground knowledge appears to reduce the problem of active segregation, at either the P1 or N1 stage, as listeners only verify a narration already heard. It is suggested that prediction is adaptively involved in the cocktail party problem via gain magnitude and delay mechanisms.

Disclosures: T. Sánchez-Costa: None. **A. Carboni:** None. **F. Cervantes Constantino:** None.

Poster

P770. Mechanisms of Attention: Human Studies II

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The KMUTT's Startup Fund for junior researchers

Title: Conflict dynamically shapes the focus of visual attention

Authors: *P. SOOKPRAO[1], P. WIWATPHONTHANA[1], K. LERTLADALUCK[2,1], T. WILAIPRASITPORN[5], N. DILOKTHANAKUL[5], I. CHATNUNTAWECH[6], C. CHUNHARAS[7], J. SERENCES[8,9,10], S. ITTHIPURIPAT[1,3,4];
[1]Neurosci. Ctr. for Res. and Innovation, Learning Inst., [2]Gifted Educ. Office, Learning Inst., [3]Big Data Experience Ctr., [4]Futuristic Res. in Enigmatic Aesthetics Knowledge Lab., King Mongkut's Univ. of Technol. Thonburi, Bangkok, Thailand; [5]Sch. of Information Sci. and Technol., Vidyasirimedhi Inst. of Sci. and Technol., Rayong, Thailand; [6]Natl. Nanotechnology Ctr., Bangkok, Thailand; [7]Neurology, department of Intrnl. Med., King Chulalongkorn memorial hospital, Chulalongkor, Bangkok, Thailand; [8]Neurosciences Grad. Program, [9]Dept. of Psychology, [10]Kavli Fndn. for the Brain and Mind, UCSD, La Jolla, CA

Abstract: The ability to deal with conflict from processing incompatible sensory information is fundamental to adaptive behaviors in humans and other primate species. It is generally known that conflict reduces the speed and efficiency of sensory information and decision-making processes. One prominent theory posits that such behavioral interference is due to an additional step required for individuals to focus their attention on task-relevant sensory inputs while filtering out task-irrelevant distractors. Recent studies have directly tested this hypothesis using a variety of conflict tasks but many of them failed to provide positive evidence. Here, we examined changes in the spatial scopes of attention in human subjects performing a variant of the Eriksen flanker tasks where they discriminated the shape of the cued target surrounded by congruent or incongruent distractors. The spatial scopes of attention in the congruent and incongruent conditions were indexed by the spatial reconstructions based on the modeling of spatially specific alpha band activity (EEG oscillations at 8-12Hz), which have been shown to track the precise location of covert attention. We found that the scopes of the alpha-based spatial reconstructions were smaller in the incongruent conditions compared to the congruent conditions and these conflict-induced modulations occurred ~100-250 ms after stimulus onset. Moreover, we found that the degree of the reduction in the spatial scope of attention predicted the degree of behavioral interference. These results provide neural evidence implicating the important role of early attentional selection in conflict resolution at least in the context of our experimental design.

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Poster

P770. Mechanisms of Attention: Human Studies II

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NIH/NIBIB P41-EB018783 (PB)
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Title: Cross-frequency phase-amplitude coupling as a mechanism for temporal expectation: a human intracranial recording study

Authors: *N. TAL-PERRY[1], D. MARCIANO[3], L. BELLIER[3], J. J. LIN[5], P. BRUNNER[6,8,7], R. T. KNIGHT[3,4], S. YUVAL-GREENBERG[1,2];
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Abstract: Expectations allow our perceptual system to guide itself through the everyday information deluge. One form of expectation is temporal expectation, based on predictions about the timing of relevant events. Despite abundant evidence on the behavioral benefits of temporal expectation, little is known about its underlying neural mechanisms. Scalp-EEG studies provided evidence for anticipatory oscillatory activity accompanying temporal expectation and correlated with target predictability and behavioral performance. Based on these findings we hypothesized that temporal expectation is driven by cross-frequency coupling (CFC) between the phase of a low-frequency oscillation and the amplitude of high-frequency activity: cycles of low frequencies oscillations are required for timing interval durations, whereas their coupled high-frequency bursts are involved in performing the local computations required for motor preparation for the upcoming event.
Here, we examined this hypothesis using invasive electrocorticography (ECoG) and stereoencephalography (sEEG) in epileptic neurosurgical patients (n=5). We analyzed 66 electrodes implanted in pre-motor area (in 5/5 patients), supplementary motor area (SMA; in 2/5 patients), and primary motor area (M1; in 2/5 patients). In each trial, participants heard an auditory cue (4 kHz pure tone for 50 ms) followed by a vocalization target ("goo" or "ga" presented for 100 ms). The interval between cue and target (the foreperiod) was manipulated between blocks, as either 2 s throughout a block (fixed condition), enabling anticipation of target onset, or changing randomly between 1, 2, or 3 s between trials (random condition), making prediction more difficult. Participants were asked to indicate which of the two syllables was presented via a speeded button press. CFC between low-frequency (1-6 Hz) phase and high-frequency (40-150 Hz) amplitude was computed during the foreperiod (+300 ms relative to the cue and -50 ms relative to the target).
Results showed evidence for enhanced cross-frequency phase-amplitude coupling in the fixed relative to the random condition in the premotor area, SMA, and M1 (all p < .05). Specifically, we found coupling between (i) delta phase (1-3.5 Hz) and low-gamma (40-70 Hz) amplitude in 12 channels (3 patients) of the premotor area; (ii) delta phase and high-gamma (120-140 Hz) amplitude in 7 channels (2 patients) of the SMA; and (iii) delta phase and broadband gamma (40-120 Hz) amplitude in 8 channels (2 patients) of M1. These findings provide the first human intracranial evidence that the construction of temporal expectation is driven by delta-gamma CFC in the motor cortices.

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Poster

P770. Mechanisms of Attention: Human Studies II

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Title: Differential neural mechanisms underlie top-down attentional modulation mediated by cortical alpha-band oscillations

Authors: *X. YANG[1,2], I. C. FIEBELKORN[1,3], J. PARVIZI[4], O. JENSEN[5], R. T. KNIGHT[6], S. KASTNER[1,2];
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Abstract: Selective attention relies on neural mechanisms that facilitate processing of behaviorally relevant sensory information and suppress processing of irrelevant information. M/EEG studies in humans have repeatedly linked increases in alpha-band oscillations in the visual attention network to the suppression of potentially distracting information, thus facilitating the processing of target stimuli. However, M/EEG data provide a limited window into the spatial selectivity and network-level dynamics of such alpha-related attentional modulation. We therefore analyzed cortical alpha-band activity from intracranial electrodes implanted in 8 epilepsy patients who performed a spatial attention task. Following a spatial cue and a variable delay, subjects differentiated between two shapes at the cued location in an array of distracters. We observed a cortical dissociation between attention-modulated alpha desynchronization and synchronization in preparation for an upcoming, spatially predictable target, with dorsal cortical areas primarily demonstrating attention-related alpha desynchronization (at attended locations) and ventral cortical areas primarily demonstrating attention-related alpha synchronization (at unattended locations). We next probed the relationship of cortical alpha-band activity and high-frequency broadband (HFB) activity (a proxy for population neuronal spiking) by measuring within-region cross-frequency coupling (CFC) and directionality (CFD) between alpha phase and HFB amplitude. We found that areas along the intraparietal sulcus (IPS) exhibited significant group-level increases of within-region alpha-HFB coupling relative to baseline at attended and unattended locations. Our findings of negative mean CFD further suggest that population spiking modulates the phase of alpha-band activity during attentional deployment. Finally, we investigated whether IPS alpha activity mediates top-down attentional modulation across the visual system by measuring between-region CFC/CFD and found that IPS alpha phase modulates

cortical excitability of the visual cortex in directing attention-related enhancement to facilitate the processing of upcoming target. Results of Granger causality analysis further suggest that IPS areas influence visual cortex through the alpha-band channel in directing attention-related suppression to attenuate the processing of distracting information. Our findings uncover the differential neural mechanisms underlying target enhancement and distractor suppression in directing top-down attentional modulation to prepare the sensory system for upcoming target detection.

Disclosures: X. Yang: None. **I.C. Fiebelkorn:** None. **J. Parvizi:** None. **O. Jensen:** None. **R.T. Knight:** None. **S. Kastner:** None.

Poster

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Support: DGAPA-PAPIIT UNAM IN 102420
Conacyt CB A1-S-10610

Title: Balanced scaling of horizontal excitation and inhibition in primary visual cortex as a plausible mechanism for fine visual resolution modulation by exogenous attention

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Abstract: Recent experiments have revealed how exogenous attention modulates in time and space fine spatial vision across the central fovea. At short delays, visual resolution is locally enhanced at the cued location and decreased at un-cued locations. At long delays, the pattern is reversed. The first contribution of our work is to explain mechanistically how vision of detail can be controlled in this highly localized fashion. We show that a balanced scaling of horizontal excitation and inhibition in V1 can focus (enhance the resolution of) or defocus (decrease the resolution of) the feedforward visual kernel originating in the upstream visual pathway. We illustrate our theory on a neural field model of V1. The second contribution is to show that the existence of an attention traveling wave, originating at the cued location and coordinating the E/I-balanced scaling of V1 horizontal connections, explains and reproduces the spatiotemporal dynamics of visual-resolution control. Lateral inhibition around the bump of the attention traveling wave explains the early resolution decrease at un-cued locations. The after-hyperpolarization period of the attention traveling wave explains the late resolution decrease at the cued location. Our modeling results constitute a novel mechanistic account of covert-

attention-related resolution changes in the visual pathway and reveal an overlooked connection between visual resolution control and the scaling of balanced horizontal excitation and inhibition in V1. Besides explaining a number of recent experimental observations, they allow us to formulate novel predictions about the mechanisms of fine visual resolution modulation by exogenous attention.

Disclosures: A. Franci: None. **Y. Zhang:** None. **M. Poletti:** None.

Poster

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ARL W911NF-19

Title: Brain signatures of active visual search in a free-viewing task

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Abstract: Visual search involves a series of mental processes that guide the eye movements through the visual scene. Early access to categorical visual information plays an important role during free-viewing in real-world behaviour. In fixed-gaze, there are well-established electrophysiological signatures of categorical processing. In particular, a hallmark of face processing emerges around 170 ms after stimulus onset with an increased EEG amplitude to faces. Previous evidence suggests the higher involvement of low-frequency oscillations in integrative brain processes, such as decision-making or top-down attention, in comparison to local processing of the stimuli.Here, we aimed to study how the sensitivity to category information extends to a free viewing paradigm by studying fixation-related potentials and brain oscillations. We co-registered brain activity and eye-tracking to investigate fixation-related potentials to pictures of different categories during visual search and exploration. Participants were asked to “observe” / “search for one target from” an array of faces and objects embedded in random noise. We hypothesized a larger evoked response to face fixations in comparison to objects. Moreover, we proposed that there are specific oscillatory brain signatures underlying active search for a target that is not present during exploration.Firstly, we show a stronger

activation for fixations to faces than to objects in both types of tasks. Secondly, exploiting a deconvolution approach of the electrophysiological signal, we observed significant early differences (100-200 ms after fixation) between the components elicited by fixations before the target was found in visual search and fixations during exploration. Moreover, differences between visual search and exploration also impacted low-frequency oscillations in a broader interval after fixation onset. These results generalize the characterization of early visual face processing to a wider range of experiments and show specific components potentially associated with eye movement guidance in active search.

Disclosures: J.E. Kamienkowski: None. **M. Da Fonseca:** None. **D. Care:** None. **B. Bianchi:** None. **M.J. Ison:** None.

Poster

P770. Mechanisms of Attention: Human Studies II

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Title: Self-control and its association with global/local attentional processing

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Abstract: Attention guides hierarchical (global/local) information processing and contributes to decision making and self-control processes. The later two processes are key to achieve long-term goals and resist short-term temptations. Individual differences in global/local processing may be associated with the primacy of self-control or its counterpart, impulsivity. The main goal of this study was to assess personality traits on the self-control/impulsivity continuum and their association with global/local information processing efficiency. Seventy-four young participants (38 women) responded to the five-factor model of personality, the UPPS-P: negative or positive Urgency (tendency to act hastily under negative or positive emotions), lack of Premeditation (tendency to act without considering the consequences), lack of Perseverance (difficulty for staying focused on a task), and Sensation seeking (tendency to seek new and exciting

experiences); and the Navon task (NT) which assesses global/local processing. In reaction times (RT) and percentage of correct responses (%CR), the effects previously reported in the NT were observed, with no differences identified as a function of the high or low self-control group. However, the inverse efficiency (IE) index (a relative measure which relates RT and %CR), revealed that low self-control was associated with higher efficiency solving the attention task, regardless of global/local processing, than those with medium or high self-control. These results suggest that being a low level self-control subject might be adaptively functional in some settings, since it guides attentional behaviors to generate quick responses to cope with sudden environment demands.

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Poster

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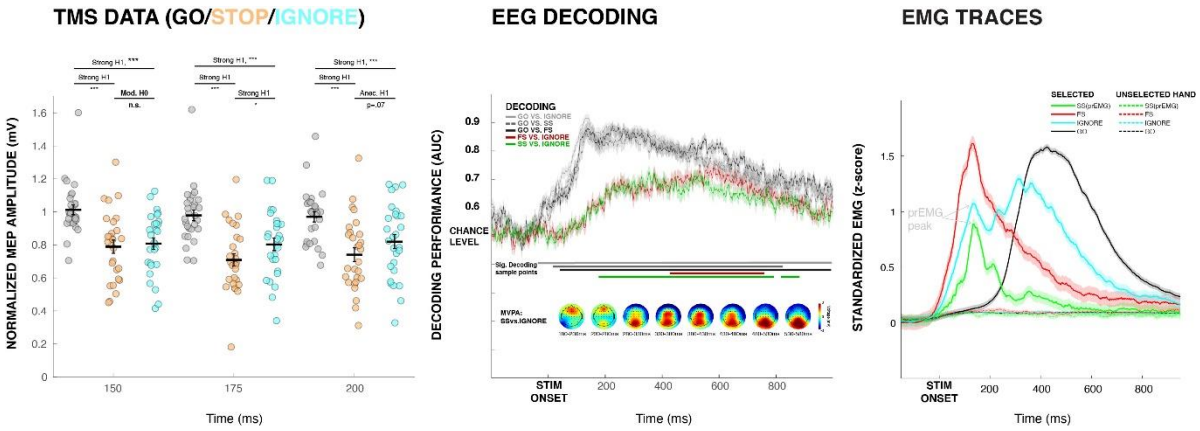
Support: NSF CAREER 1752355
NIH R01 NS102201

Title: Common and unique inhibitory control signatures of action-stopping and attentional capture suggest that actions are stopped in two stages

Authors: *C. SOH, J. R. TATZ, J. R. WESSEL;
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Abstract: The ability to stop an already initiated action is paramount to adaptive behavior. Much scientific debate in the field of human action-stopping currently focuses on two interrelated questions. First: Which cognitive and neural processes uniquely underpin the implementation of inhibitory control when actions are stopped after explicit stop-signals, and which processes are instead commonly evoked by all salient signals, even those that do not require stopping? Second: Why do purported (neuro)physiological signatures of inhibition occur at two different latencies after stop-signals? Here, we address both questions via two pre-registered experiments that combined measurements of cortico-spinal excitability (CSE), electromyography (EMG), and whole-scalp electroencephalography (EEG). Adult human subjects performed a stop-signal task that also contained 'ignore' signals - equally salient signals that did not require stopping but rather completion of the Go response. We found that both stop- and ignore-signals produced equal amounts of early-latency inhibition of CSE and EMG, which took place ~150ms following

both signals. Multi-variate pattern analysis of the whole-scalp EEG data further corroborated that this early processing stage was shared between stop- and ignore-signals, as neural activity following the two signals could not be decoded from each other until a later time period. In this later period, unique activity related to stop-signals emerged at fronto-central scalp sites, reflecting an increased stop-signal P3. These findings suggest a two-step model of action-stopping, according to which an initial, universal inhibitory response to the saliency of the stop-signal is followed by a slower process that is unique to outright stopping.
Figure1. Left Panel: Cortico-spinal excitability (CSE) recorded from the task-irrelevant hand; Middle Panel: Decoding results using the signals recorded from the whole scalp EEG electrodes; Right Panel: Trial-averaged continuous electromyography (EMG) traces for Go, Ignore, Successful (SS) and Failed Stop (FS) trials.



Disclosures: C. Soh: None. **J.R. Tatz:** None. **J.R. Wessel:** None.

Poster

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Title: A common factor for inter-subject correlation of brains, hearts, and eyes

Authors: *J. MADSEN, L. C. PARRA;
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Abstract: A wide range of neural, physiological and behavioral signals correlate between human subjects in a variety of settings. A great number of hypotheses have been proposed to explain this inter-subject correlation, however there is no clarity under which conditions or

circumstances it arises, for which signals, or whether there is a common underlying mechanism. In this work we hypothesized that similar cognitive processing of a shared stimulus is the source of inter-subject correlation. To test this we presented informative videos to participants in an attentive and distracted condition and subsequently measured recall memory. Inter-subject correlation was observed for electro-encephalography, gaze position, pupil size and heart rate, but not respiration and head movements. The strength of correlation was co-modulated in the different signals, changed with attentional state, and predicted subsequent recall of information presented in the videos. There was robust within-subject coupling between brain, heart and eyes, but not respiration or head movements. The theory that emerges is that inter-subject correlation is the result of cognitive processing of a shared stimulus, but can be observed only for those signals that exhibit a robust brain-body connection. While physiological and behavioral fluctuations may be driven by multiple features of the stimulus, correlation with other individuals is co-modulated by the level of attentional engagement with the stimulus.

Disclosures: J. Madsen: None. **L.C. Parra:** None.

Poster

P775. Imagery and Imagination

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Topic: H.02. Perception and Imagery

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NSF Grant 1632738
NIH Grant R21EY032713
NIH Grant R01EY014681

Title: The Neural Bases of Simulation in the Primate Brain

Authors: *A. AHUJA, T. M. DESROCHERS, D. L. SHEINBERG;
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Abstract: To engage with the world, we must regularly make predictions about the outcomes of scenarios. How do we make these predictions? Recent evidence points to simulation - the idea that we can internally manipulate rich, mental models of the world - as one possible explanation for how such predictions are accomplished. While theories based on simulation are supported by computational models, neuroscientific evidence for simulation is lacking and many important questions remain. For instance, do simulations simply entail a series of abstract computations? Or are they supported by sensory representations of the objects that comprise the scene being simulated? We posit the latter and suggest that the process of simulating a sequence of events is

likely to evoke imagery of those events. Using functional magnetic resonance imaging (fMRI), we demonstrate that when human participants predict how a ball will fall through an obstacle-filled display, motion-sensitive brain regions (such as area MT) are activated. This activity, which occurs even though no motion is being sensed, resembles activity patterns that arise while participants actually perceive the ball's motion. We also demonstrate that when a nonhuman primate (NHP) performs the same task in an fMRI scanner, motion-sensitive brain regions are similarly activated as he attempts to predict the ball's path. Together, these findings suggest that the process of simulating the ball's movement is accompanied by a sensory representation of this movement in human and nonhuman primates. Thus, these data may show that mental simulations can recreate sensory depictions of how a scene is likely to unfold.

Disclosures: A. Ahuja: None. **T.M. Desrochers:** None. **D.L. Sheinberg:** None.

Poster

P775. Imagery and Imagination

Location: Virtual Only

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Program #/Poster #: P775.02

Topic: H.02. Perception and Imagery

Title: Unrestricted eye movements strengthen causal connectivity from the medial temporal lobes to oculomotor regions during scene construction

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Abstract: There is an intimate relationship between the memory system of medial temporal lobes (MTL) and the oculomotor system. This relationship is not only reflected in the functional and anatomical connections between these systems, but also in eye movement behaviours. Visual exploration during perceptual and mnemonic viewing tasks has previously been shown to be associated with constructive processes in the MTL, including the hippocampus and parahippocampal place area. However, the directionality of connections between memory and oculomotor control regions, and how it relates to visual exploration, has not been studied directly in human neuroimaging. In the current study, we used dynamic causal modeling (DCM) for functional magnetic resonance imaging (fMRI) to investigate this relationship at a mechanistic level using a scene construction task in which participants' eye movements were either restricted or unrestricted. By omitting external visual input in our task design, we were able to determine the directionality of neural connectivity while participants mentally constructed scene representations. As opposed to when eye movements are restricted, allowing free eye movements during construction of novel scenes strengthens top-down connections from the MTL, to oculomotor regions, and to lower-level cortical visual processing regions. Meanwhile, our results

also demonstrated that bottom-up connections along the visual stream are suppressed. Taken together, these findings provide novel, non-invasive evidence for the causal architecture between the MTL memory system and oculomotor system associated with constructing vivid mental representations of scenes.

Disclosures: N. Ladyka-Wojcik: None. **Z. Liu:** None. **J.D. Ryan:** None.

Poster

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Topic: H.02. Perception and Imagery

Support: NSERC

Title: The neural basis of visual-orthographic processing in perception and mental imagery

Authors: *S. LING[1], A. C. LEE[1,2], B. C. ARMSTRONG[1,3], A. NESTOR[1];
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Abstract: Clarifying the neural and representational basis of mental imagery has elicited significant interest in the cognitive neuroscience of visual recognition. Recent work has suggested that visual perception and mental imagery are supported by overlapping regions of high-level and early visual cortex (EVC) and by similar patterns of activation at the level of these regions as captured by functional magnetic resonance imaging (fMRI) data. Here, we investigated the neural correlates of perception and imagery underlying visual-orthographic processing with the aid of fMRI.
To this aim, we collected data from 13 participants while they viewed or mentally imagined the appearance of four-letter nouns exhibiting the same CVCC (consonant-vowel-consonant-consonant) structure. Multivariate searchlight mapping revealed, consistent with previous findings, extensive and overlapping areas of the ventral stream capable of supporting pairwise word discrimination for perception and imagery. Yet, perception and imagery diverged in exhibiting more widespread reliance on occipitotemporal areas, for the former, and frontal areas, for the latter. Region-of-interest (ROI) analyses revealed significant levels of decoding for the visual word form area (vWFA) with higher accuracy for imagery than perception and the opposite pattern of results for early visual cortex (EVC). Notably though, decoding accuracy, as characterized by multivariate mapping, peaked in the vicinity of the vWFA at adjacent locations for perception and imagery. Further, we identified separate cortical networks accounting for visual-orthographic, phonological and sematic similarity across words in the two conditions.
In sum, the current study supports the idea of distinct but overlapping mechanisms for perception

and imagery of orthographic information, it points to the similar role of the vWFA in mediating these processes, and, more broadly, it sheds light on the neural architecture underlying these two types of visual processing.

Disclosures: S. Ling: None. **A.C. Lee:** None. **B.C. Armstrong:** None. **A. Nestor:** None.

Poster

P775. Imagery and Imagination

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Program #/Poster #: P775.04

Topic: H.02. Perception and Imagery

Title: Auditory-frontal and -motor networks during distinct neural subprocesses of auditory imagery

Authors: *S. SAKAMOTO, K. MATSUSHITA, A. KOBAYASHI, R. SHIMIZU, A. AOYAMA;
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Abstract: Neuroimaging studies in auditory imagery have reported regions associated with multiple neural subprocesses, including auditory information processing, memory retrieval, and mental manipulation. However, the distinct neural oscillations related to these subprocesses are less known; our objective was to investigate the oscillations associated with the memory retrieval and mental manipulation subprocesses during auditory imagery.
In the experiment, we first presented a reference sound at 880 or 220 Hz for 2 s and then a visual stimulus of either upward or downward arrow for the context of imagery. After an upward arrow, the participant imaged a sound octave higher than the reference sound (1760 or 440 Hz), and after a downward arrow, they imaged a sound octave lower than the reference sound (440 or 110 Hz). We analyzed 64-channel electroencephalogram data during imagery with two types of contrasts: the context contrast, which shared the imaged frequency of 440 Hz but had different contexts, and the content contrast, which shared the context of imagery but had different imaged frequencies. The context and content contrasts enable us to extract the mental manipulation and memory retrieval processes, respectively. All participants had music training experience (mean=10.8 years).
During imagery of sound, we found significant differences in the right frontal and temporal gamma power for the context contrast and right fronto-temporal delta phase coupling, left fronto-temporal delta-gamma phase-amplitude coupling (PAC), and right frontal-central delta-beta PAC for the content contrast.
Our finding of delta-gamma fronto-temporal PAC is convincing as frontal gamma and temporal delta power reflect memory load, suggesting that these oscillations also reflect the memory content during auditory imagery. Frontal and temporla gamma power in the context contrast

implies the importance of top-down manipulative process during imagery, as the same oscillation is involved during visual object rotation as well as the top-down process from frontal to temporal regions during auditory recall. Moreover, the distinction of two contrasts sheds light on the dual-pathway model of auditory cortical processing; anterior stream process spectral content while the dorsal stream process spectral motion and calculation. While more analysis is needed using methods with higher spatial resolution, our result in the auditory-frontal network possibly reflects the distinct oscillations of the two streams. Additionally, the delta-beta coupling in the content contrast plays a role in auditory-motor integration, implying possible relation of memory functions and motor functions.

Disclosures: S. Sakamoto: None. **K. Matsushita:** None. **A. Kobayashi:** None. **R. Shimizu:** None. **A. Aoyama:** None.

Poster

P775. Imagery and Imagination

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Program #/Poster #: P775.05

Topic: H.02. Perception and Imagery

Title: Behavioral and neural differences in imagery between a pair of aphantasic and non-aphantasic identical twins: a case study

Authors: *D. PRASAD, W. A. BAINBRIDGE;
Univ. of Chicago, Chicago, IL

Abstract: Aphantasia is a recently defined term characterizing people who lack voluntary visual imagery, despite otherwise normal performance in other cognitive abilities like perception and visual recognition (Zeman et al., 2015). Instead of experiencing perception-like imagery during visual recall, they often report semantic representations of their memories (Bainbridge et al., 2021). This atypical experience offers us a unique insight into the neural underpinnings of visual recall and what kinds of information these representations contain. Although the prevailing thought behind visual recall is that of sensory reinstatement, recent research has challenged this idea that brain regions activated during perception are similarly activated during recall, showing an anterior shift and representational transformation during recall versus perception (Bainbridge et al., 2020; Favila et al., 2020; Steel et al., 2021). Studying aphantasics could inform us on the nature of these newly defined recall regions - whether this anterior shift reflects representation of more abstract, conceptual information. Here, we present a case study with a pair of monozygotic, handedness discordant twins (Female, Age=31 years) where one twin reports having typical visual imagery (VVIQ=47, OSIQ Spatial=38, OSIQ Object=56) but the other twin reports lacking visual imagery (VVIQ=24, OSIQ Spatial=49, OSIQ Object=22). Using a paradigm adapted from Bainbridge et al. (2020), we examined whether there were differences in the

content of their visual recall. Participants studied naturalistic scene images, then drew them from memory and, later, from perception. Despite their drawings from perception having a similar amount of visual information, drawings from recall were starkly different between the aphantasic and non-aphantasic, with the aphantasic twin drawing fewer objects and using less color. To further examine this difference in recall ability, we conducted an fMRI study using two visual imagery tasks: imagery for novel object and scene images (Bainbridge et al., 2020), and imagery for familiar people and places (Steel et al., 2021). We also used a resting state scan to examine functional connectivity and a language localizer task to account for potential differences in lateralization (Fedorenko et al., 2010). Importantly, both twins experienced identical experiments, with identical familiar stimuli, allowing us to examine the ability to cross-decode stimulus representations between the two twins. We observe interesting differences between the aphantasic and non-aphantasic imagery representations, despite perceiving and recalling the same inputs across tasks.

Disclosures: D. Prasad: None. **W.A. Bainbridge:** None.

Poster

P775. Imagery and Imagination

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Program #/Poster #: P775.06

Topic: H.02. Perception and Imagery

Support: Future Learning Initiative ETHZ

Title: Intuitive physical inference: misconceptions, biases and neural bases.

Authors: *G. ZBÄREN[1], S. MEISSNER[1], V. GASHAJ[2], M. KAPUR[2], N. WENDEROTH[1,3]; [1]Neural Control of Movement Lab., Dept. of Hlth. Sci. and Technology, ETH Zurich, Zurich, Switzerland; [2]Inst. for Learning Sci. and Higher Educ., Dept. of Humanities, Social and Political Sciences, ETH Zurich, Zurich, Switzerland; [3]Future Hlth. Technologies, Singapore-ETH Centre, Campus for Res. Excellence And Technological Enterprise (CREATE), Singapore, Singapore

Abstract: *Introduction* Despite our ability to successfully interact every day with the physics of the world, our physical inferences are prone to errors. This can be seen in common misconceptions that arise when solving seemingly simple physics problems. A well-known example is the “straight-down misconception”, the erroneous belief that an object falling from a moving body falls straight-down.
We investigated (i) how common the straight-down misconception is when participants are exposed to a dynamical scene (ii) whether people who do *not* have a straight-down misconception still exhibit biases not in line with the laws of physics and (iii) what neural substrates support physical inference.

Methods 79 healthy volunteers (42 females; age = 24.13 +/- 4.17) took part in our study for which we created a computer-based, dynamical 3D physics world. In this 3D world, a horizontally moving object with a ball attached is displayed. The moving object comes either from the left or the right side of the screen, and its height and velocity vary across trials. At a certain point, the ball drops and participants are asked to indicate via button presses when and where they think the ball lands. Crucially, once the ball drops, the screen gets occluded such that the trajectory of neither the ball nor the moving object can be seen. In a next step, first pilot participants underwent a training session during which they performed the same physics task while receiving corrective feedback on their performance. This training was followed by a 3T fMRI experiment using the same task to measure BOLD responses during physical inference. *Results and Conclusion* Similar to previous studies using paper-pencil tests to assess the straight-down misconception, we found that 53.16% of the participants exhibited this misconception. Participants who correctly estimated the ball trajectory, tended to overestimate the falling time and mistakenly predict that the ball falls faster at higher velocities of the object. Thus, even when people do not exhibit a misconception, their inferences are prone to biases. Preliminary training and fMRI data suggest that errors greatly diminish when participants receive feedback. Neural activity underpinning physical inference of falling objects activates motion-responsive visual (MT/V5), motor and parietal regions when compared to a control task matched for visual input but not requiring physical inference. By applying multivariate pattern analysis to our fMRI data, we could successfully decode essential task parameters from visual and parietal regions. This suggests that participants use mental simulation to solve the task, which is line with current views on intuitive physics.

Disclosures: G. Zbären: None. **S. Meissner:** None. **V. Gashaj:** None. **M. Kapur:** None. **N. Wenderoth:** None.

Poster

P775. Imagery and Imagination

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P775.07

Topic: H.02. Perception and Imagery

Support: SNF Grant Spark CRSK-1_190836

Title: Can we modulate locus coeruleus activity via pupil-based neurofeedback?

Authors: *S. N. MEISSNER[1], M. BAECHINGER[1], A. TAUBNER[2], N. WENDEROTH[1]; [1]Hlth. Sci. and Technol., [2]Computer Sci., Swiss Federal Inst. of Technol. Zurich, Zurich, Switzerland

Abstract: ***Introduction*** The locus coeruleus (LC) in the brainstem contains noradrenaline (NA)-releasing neurons and sends widespread projections to most of the brain. The LC-NA system is suggested to be involved in the control of various behaviors. Recently, there is growing evidence for a link between LC activity and non-luminance dependent changes in pupil size. Building on this knowledge, we aimed to investigate whether changes in pupil size (i) can be *volitionally* induced by humans via pupil-based neurofeedback (pupil-NF) training, and (ii) are linked to changes in LC activity as measured with fMRI.
Method To investigate whether healthy volunteers can learn to volitionally modulate (i.e., *increase* or *decrease*) their own pupil size, 43 participants (pupil-NF group; 28 females; 24 +/- 3.60 years) received three pupil-NF sessions on three consecutive days. 27 control participants (11 females; 24 +/- 5.45 years) received the same amount of training but no veridical pupil-NF. Prior to training, all participants were familiarized with potential mental strategies to modulate pupil size. Pupil size was measured by means of an eye tracker and fed back to the participants during training. To further explore the link between pupil size changes and LC activity, first participants (n = 11) underwent follow-up sessions combining pupil-NF with 3T fMRI to measure BOLD responses in the LC. First-level analyses involved the construction of a general linear model (conventional block design: *pupil increase vs. decrease)*. To further extract responses from our region of interest, we delineated the LC for each participant by means of turbo spin echo (TSE) structural MRI scans (in-plane resolution: 0.7 x 0.88 mm).
Results Healthy volunteers were able to modulate (i.e., *increase* and *decrease*) their pupil size relative to baseline measurements (mixed-effects model main effect *session*; $p < .001$). Importantly, this self-modulation was clearly reduced in control participants who did not receive veridical pupil-NF (main effect *group*; $p < .001$). Our preliminary fMRI analysis further showed that volitional pupil modulation may be linked to activity changes in the LC and connected areas including the amygdala and thalamus: we found higher BOLD responses in these areas during volitional *increase* than *decrease* of pupil size.
Conclusions Our findings suggest that healthy volunteers can learn to modulate their own pupil size via pupil-NF. Furthermore, we provide new insights into the link between pupil dynamics and LC activity by offering first preliminary evidence that volitional pupil modulation may indeed be linked to activity changes in the LC and interconnected areas.

Disclosures: S.N. Meissner: E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Patent application filed for procedure related to work that will be presented. **M. Baechinger:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Patent application filed for procedure related to work that will be presented. **A. Taubner:** None. **N. Wenderoth:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Patent application filed for procedure related to work that will be presented.

Poster

P775. Imagery and Imagination

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Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P775.08

Topic: H.02. Perception and Imagery

Title: Alpha-band ERD in unimanual and bimanual motor imagery

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Abstract: <META NAME="author" CONTENT="梅野 和也">Bimanual movements have been reported to be effective for rehabilitation of stroke patients. However, few studies have examined the effects of bimanual motor imagery on brain activity and motor learning. This study investigated how the motor imagery of bimanual movements differs from that of unimanual movements in the effects on brain activity, by calculating ERD (Event-related desynchronization) recorded from motor-related areas. When one mentally simulates a physical action, called motor imagery, the amplitude of the α wave band in the motor-related areas decreases, which is called ERD and considered to reflect the quality of the motor imagery.In the experiment, twelve right-handed healthy students participated. The participants mentally simulated the action of rotating two balls (45 mm in diameter) on their palm without physical movements (ball rotation imagery task). They alternately performed the imagery task unimanually (left or right hand) or bimanually. One trial consisted of a rest period (physical activity was not allowed; 6 s), a preparation period (2 s), an imagery period (7 s), and a break period (5 s). Participants watched visual cues displayed on the computer screen during the task. The experiment consisted of 4 blocks, and 1 block consisted of 5 trials each of right-hand imagery, left hand imagery, and bimanual imagery (accordingly, 15 trials × 4 blocks in total). The participants took a two-minute break between blocks.OpenBCI Ultracortex Mark IV was used for the measurement of EEG. Eight channels of EEG data were recorded with the OpenBCI system. The EEGs recorded from the electrodes of C3 and C4 were analyzed. The reference and ground were placed at A1 and A2, respectively. EEG signals were filtered with a bandpass filter of 8 - 13 Hz to measure the amplitude of the α wave band. ERD was calculated by dividing the difference in the amplitude of the α wave band between the imagery and rest periods by the amplitude in the rest periods.The results showed that the amplitude of bimanual ERDs were higher than those of the unimanual ERDs. There was not a large difference in the amplitude of ERD between the left and right hand imageries. The amplitude of ERDs did not increase as practice proceeded. In addition, some participants did not show reliable ERDs through the experiments. The stronger ERDs observed in bimanual motor imagery compared to unimanual motor imagery may indicate that bimanual motor imagery helps neurofeedback beginners to efficiently evoke ERDs in the motor-related areas, which might also lead to the development of effective training for stroke patients.

Disclosures: K. Umeno: None. **Y. Itaguchi:** None.

Poster

P775. Imagery and Imagination

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Topic: H.02. Perception and Imagery

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ICNAS Produção

Title: The neural basis for visual effects induced by inhaled psychedelics:DMT leads to activation of extrastriate cortex concomitantly with behavioural effects

Authors: M. TEIXEIRA*[1], C. SOARES*[1], M. PAIS*[1], G. LIMA[1], S. MACHADO[2], ***M. CASTELO-BRANCO**[1];
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Abstract: Introduction: DMT is an indole alkaloid widely found in nature, best known by its presence in ayahuasca. It is a psychedelic tryptamine associated with intense visual phenomena, perception changes and profound spiritual experiences. Induced visual percepts are driven by environment but characterized by an increased mismatch between the physical objects and their internal representations. In this study we aimed to explore the neural correlates of these altered brain states and the role of the visual system, during the experience produced by inhaled DMT using fMRI and periodic visual stimuli. **Methods:** A within-subject design study with control and active treatment conditions is being conducted at CIBIT, in 2 sessions (different days): Control (no treatment) or Active treatment (DMT). 6 healthy volunteers (mean ages= 35YO, SD= 13; 4 Males; 2 Females) have been tested. The study was conducted in accordance with the tenets of the Declaration of Helsinki and all participants signed a written informed consent. History of neuropsychiatric or organic disorders was excluded. For the active condition, smoked DMT was self-administered by participants (~50mg), obtained by extraction from the root bark of Mimosa Hostilis in freebase form. In both sessions, pre and post measures of subjective experience were collected with the Spiritual Well Being Questionnaire(SWBQ), the Vividness of Visual Imagery Questionnaire(VVIQ-RV), the Mystical Experience Questionnaire(MEQ30) and the Hallucinogen Rating Scale(HRS). fMRI task consisted of a block design with a visual hallucination inducer paradigm, with visual stimuli containing parts with empty-field flicker. Data were pre-processed and analysed in BrainVoyager 21.4. **Results and Conclusion:** Multivariate ANOVA results with HRS, MEQ and Condition (active and control group as within fixed factors) suggest overall higher scores for all the items of both scales in the active condition with significant differences between conditions for HRS Total [marginally sig: F= 6,347 (1,4), p=0,65] and Intensity [F =25,6 (1,4), p< 0.05) factors, and Positive Mood factor of MEQ[F=

12,774 (1,4), p<0.05]. No differences were found for repeated measures ANOVA Pre and Post VVIQ and SWBQ. Exploratory analysis of brain activity measured with fMRI contrasting DMT vs Control conditions under visual stimulation, show increased BOLD activity in non-primary visual areas BA 18, 19 and Precuneus for active condition. These results suggest a neural basis for the visual effects evoked by Ayahuasca which occur concomitantly with significant behavioral effects. *equal contribution **Funding:** FCT/UID4950, ICNAS Produção and Bial Foundation 252/18

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Poster

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Topic: H.01. Attention

Support: Albertfonds
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Title: Mindfulness-induced white matter changes in breast cancer survivors with cognitive complaints: A preliminary longitudinal fixel-based analysis

Authors: *M. MELIS, J. BLOMMAERT, A. SMEETS, G. SCHROYEN, C. SLEURS, K. VAN DER GUCHT, S. SUNAERT, S. DEPREZ;
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Abstract: Cancer-related cognitive impairment affects 17-78% of cancer survivors and has been associated with white matter abnormalities. It might impact the quality of life, but unfortunately, there is currently no treatment available. Previous research showed that standardised mindfulness-based interventions (MBI) might have an impact on cognition and white matter structure. Therefore, we investigated the potential of MBI to reduce cognitive complaints and reverse chemotherapy-induced white matter changes. Thirty-three adult breast cancer survivors with cognitive complaints after chemotherapy (mean age (SD) = 45.2 (6.19)) were randomly allocated to a mindfulness (MBI; N=18) or waitlist control condition (WL; N=15). Twenty-three participants (N=12 MBI; N=11 WL) completed multi-shell diffusion-weighted imaging (DWI) before the start of the intervention, immediately after, and 3 months post-intervention. White matter micro- and macrostructure was assessed using a whole-brain longitudinal fixel-based analysis (FBA). In this analysis, significant changes in fibre density (FD), fibre cross-section (FC) and a combined measure of fibre density and cross-section (FDC) were evaluated using connectivity-based fixel enhancement (p_{FWE}<0.05). Spearman correlations (r_s) were calculated to

investigate the relationship between significant changes in FBA-outcomes and cognitive/emotional complaints. We found a significant group-by-time interaction effect; a bigger reduction in log FC in fixels (N=5) located in the right superior longitudinal fasciculus in the mindfulness compared to the control group immediately post-intervention. Within-group analyses showed that immediately after the intervention, MBI participants had a lower FC in a group of fixels (N=122) in the right superior longitudinal fasciculus and a higher FC in fixels (N=63) located in the optic radiation compared to baseline. The decrease in FC correlated with a reduction in cognitive complaints (r_s=.42). The participants in the waitlist group had a lower FDC in a group of fixels (N=19) in the genu of the corpus callosum at three-month follow-up versus baseline. This reduction correlated with an increase in emotional complaints (r_s=-.52). In conclusion, these results provide preliminary evidence for white matter micro- and macrostructural changes following MBI, which might aid in recovery from cancer-related cognitive impairment. Research with bigger sample sizes is necessary to evaluate the potential of MBI in targeting cognitive impairment after cancer treatment.

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Poster

P842. Dynamics of Memory Representations

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Topic: H.08. Learning and Memory

Support: NIH K99AG068425
Simons Collaboration on the Global Brain 500639
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Title: Hippocampal replay reflects specific past experiences rather than a plan for subsequent choice

Authors: *A. K. GILLESPIE[1], D. A. ASTUDILLO MAYA[1], E. L. DENOVELLIS[1,2], D. F. LIU[1], D. B. KASTNER[1], M. E. COULTER[1], D. K. ROUMIS[1], U. T. EDEN[3], L. M. FRANK[1,2]; [1]UCSF, San Francisco, CA; [2]HHMI, San Francisco, CA; [3]Boston Univ., Boston, MA

Abstract: Executing memory-guided behavior requires both the storage of information about experience and the later recall of that information to inform choices. Awake hippocampal replay, when hippocampal neural ensembles briefly reactivate a representation related to prior experience, has been proposed to critically contribute to these memory-related processes. However, it remains unclear whether awake replay contributes to memory function by promoting

the storage of past experiences, by facilitating planning based on an evaluation of those experiences, or both. We designed a dynamic spatial task which promotes replay before a memory-based choice and assessed how the content of replay related to past and future behavior. We found that replay content was decoupled from subsequent choice and instead was enriched for representations of previously rewarded locations and places that had not been recently visited, indicating a role in memory storage rather than in directly guiding subsequent behavior.

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Poster

P842. Dynamics of Memory Representations

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Topic: H.08. Learning and Memory

Support: Simons Foundation for the Global Brain Grants 542971
Simons Foundation for the Global Brain Grants 542981
HHMI

Title: Hippocampal replay of experience at real-world speeds

Authors: *E. L. DENOVELLIS[1], A. K. GILLESPIE[1], M. E. COULTER[1], M. SOSA[2], J. CHUNG[1], U. T. EDEN[3], L. M. FRANK[1];
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Abstract: Representations related to past experiences play a critical role in memory and decision-making processes. The rat hippocampus expresses these types of representations during sharp-wave ripple (SWR) events, and previous work identified a minority of SWRs that contain “replay” of spatial trajectories at ~20x the movement speed of the animal. Efforts to understand replay typically make multiple assumptions about which events to examine and what sorts of representations constitute replay. We therefore lack a clear understanding of both the prevalence and the range of representational dynamics associated with replay. Here we develop a state space model that uses a combination of movement dynamics of different speeds to capture the spatial content and time evolution of replay during SWRs. Using this model, we find that the large majority of replay events contain spatially coherent, interpretable content. Furthermore, many events progress at real-world, rather than accelerated, movement speeds, consistent with actual experiences.

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Poster

P842. Dynamics of Memory Representations

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Topic: H.08. Learning and Memory

Support: NIH NRSA 1F32MH107086-01
NIH R21EY028381

Title: Latent learning drives sleep-dependent plasticity in distinct CA1 subpopulations

Authors: *W. GUO, J. J. ZHANG, J. P. NEWMAN, M. A. WILSON;
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Abstract: Latent learning allows the brain the transform experiences into cognitive maps, a form of implicit memory, without reinforced training. Its mechanism is unclear. We tracked the internal states of the hippocampal neural ensembles and discovered that during latent learning of a spatial map, the state space evolved into a low-dimensional manifold that topologically resembled the physical environment. This process requires repeated experiences and sleep in-between. Further investigations revealed that a subset of hippocampal neurons, instead of rapidly forming place fields in a novel environment, remained weakly tuned but gradually developed correlated activity with other neurons. These ‘weakly spatial’ neurons bond activity of neurons with stronger spatial tuning, linking discrete place fields into a map that supports flexible navigation.

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Poster

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Topic: H.08. Learning and Memory

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Title: Selective disruption of hippocampal sharp-wave ripples leads to impaired object-place recognition memory

Authors: *S. DUTTA, J. ZHANG, C. KEMERE;
Rice Univ., Houston, TX

Abstract: Animals exhibit an innate curiosity towards novelty. Whether in the form of contexts or objects, the resultant behavior is quantified by them spending more time with novel or displaced objects or even novel locations as opposed to familiar ones. Extensive lesion studies have demonstrated this novelty preference to be hippocampally-dependent via a variety of object recognition test (ORT) paradigms. More recent work has correlated hippocampal CA1 LFP signatures, such as fast-gamma and beta-band oscillations, as potential neural substrates for object-place recognition memories and has demonstrated modulations in other transient hippocampal events, sharp-wave ripples (SWRs). Specifically, SWRs have been shown to increase after encoding of novel or displaced objects and general novelty in the testing paradigm; however, the concomitant spiking activity has not been correlated to preferentially code object-place pairings prompting the question of the role of these events in this curiosity driven task. To illuminate the role of SWRs in potentially driving the curiosity towards novelty, we selectively modulate SWR activity using our previously engineered open-source, closed-loop SWR detection system during a displaced ORT paradigm. Although behaviorally similar with regards to various exploration environment metrics, our results show statistically significant deficits in exploration of familiar objects in novel locations of SWR disrupted rats compared to control animals (post-SWR stimulation or non-implanted wild type rats). We look to further investigate the role of SWRs along with its co-occurring neural activity and potential causal changes that SWRs transiently cause within the hippocampal circuit that may lead to such a deficit in this novelty recognition memory through exploration of LFP metrics such as cross-frequency coupling, phase-amplitude comodulation, SWR rates, etc. Altogether, our preliminary findings suggest SWRs play a crucial role in curiosity driven memory tasks as opposed to just spatial memory recall tasks.

Disclosures: S. Dutta: None. **J. Zhang:** None. **C. Kemere:** None.

Poster

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Topic: H.08. Learning and Memory

Support: NIH
DARPA

Title: Using auditory and multisensory virtual reality to understand how spatial acoustic cues contribute to hippocampal place map formation

Authors: *S. GAO[1,2], Z. CHEN[2], M. J. MCGINLEY[3], C. KEMERE[2];
[1]Electrical And Computer Engin., Rice Neuroengineering Initiative, Houston, TX; [2]Rice Univ., Houston, TX; [3]Neurosci., Baylor Col. of Med., Houston, TX

Abstract: For decades, many neuroscientists have actively been uncovering how our brain recognizes the environment around us during spatial navigation. Traditionally, spatial navigation in animal models in virtual reality (VR) settings has been studied primarily using visual cues. There has recently been interest in exploring other sensory cues such as odor and sound. However, few studies have investigated VR navigation in environments promoting interactions between the auditory system and hippocampus. Here we present a novel virtual reality system defined exclusively by acoustic landmarks that are amplitude modulated based on the animal's position. We used modified UCLA Miniscope to chronically image the CA1 area of head-fixed mice performing an operant navigation task collecting water reward for up to two months. We report behavioral evidence that mice can learn to navigate in our auditory VR task. Namely, we observed anticipatory licking and slowing preceding the reward region. Furthermore, we introduced a signal-to-noise ratio (SNR) manipulation on well trained animals in order to study how precision of sensory stimulation is integrated with idiothetic cues. We observed population wise hit rate performance increase as the SNR increases. In addition, the distance between the reward location and the lick location increases as SNR decreases. Our results suggest that animals were able to form a cognitive map with only auditory landmarks but rely heavily on the ability to detect onset of target sound within a noisy sound background. Intriguingly, the number of highly spatially selected stable neurons observed were low, and we asked whether visual cues are necessary to engage a large population of stable place cells during navigation. To investigate further, we have developed a visual VR system that is synchronized with our previous auditory VR, and we are aiming to understand whether the inclusion of visual motion cues facilitate the formation of stable place cells defined by acoustic landmarks. We envision that combining our behavior paradigm with our novel sound and visual VR system has the potential to provide new insight on how multiple neural systems interact with each other and respond to stimuli using spatial aspects of sound in an environment.

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Poster

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Title: Associative learning dynamics across the dorsal/ventral axis of the hippocampus

Authors: *J. BIANE[1], M. LADOW[1], F. STEFANINI[2], S. BODDU[1], A. FAN[1], S. HASSAN[1], N. DUNDAR[1], D. APODACA[1], K. LITKE[1], N. WOODS[1], M. KHEIRBEK[1];
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Abstract: The dorsal hippocampus generates internal representations of cue-outcome relationships, while the ventral hippocampus has been implicated in orchestrating responses to emotionally salient cues. Thus, the hippocampus may be responsible not just for binding cue-outcome associations, but also for attaching and modifying the emotional significance of cues, and may do so differentially across its dorsal/ventral (d/v) axis. Using 2-photon imaging in mice we examined the evolution of stimulus encoding across the d/v axis of the hippocampus as mice learned to associate odor stimuli with different outcomes (reward or nothing). We found robust coding of odor identity in populations of dCA1 neurons across all phases of odor-reward learning (baseline, learning, extinction and reaquisition), indicating that odors are robustly encoded by dCA1, regardless of their value. In contrast, neuronal signals in vCA1 tracked the value of odorants assigned during learning. Analysis of the 2 sec trace period between odor offset and reward delivery found that responsivity to this period was virtually absent at the onset of training, but increased considerably with learning in both dCA1 and vCA1. This signal faded following extinction of the odor-reward contingency, but was rapidly restored when the contingency was reinstated. Moreover, in well-trained animals this reward anticipation signal was conserved across days, suggesting a stable representation for reward expectation exists across d/v regions of the hippocampus. We additionally examined the within-trial stability of population activity and found that, following learning, distinct ensembles of dCA1 neurons represented each component of the trial (odor, trace, reward). In contrast, in vCA1 we found overlapping representations to these components, which persisted for seconds after the trial concluded. This suggests that dCA1 neuronal ensembles represent the distinct elements of a trial while vCA1 can generalize across elements to generate a broad representation of trial identity. Collectively, these data further clarify how the hippocampus is recruited for reward seeking, showing both CA1 regions strongly encode reward expectation, but display a shift from externally based (stimulus ID) to internally based (value) encoding of environmental variables along the d/v axis. Further, the specific (dCA1) vs general (vCA1) encoding of task elements suggests the hippocampus may employ different levels of granularity to facilitate information coding and recall.

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Poster

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Title: Place fields can be formed by individual visual landmarks in CA1, but not in CA3, in the hippocampus in a VR environment

Authors: *H.-W. LEE[1], I. LEE, PhD[2];
[2]Brain and Cognitive Sci., [1]Seoul Natl. Univ., Seoul, Korea, Republic of

Abstract: We investigated the effects of visual landmarks on the formation the place field in the CA1 and CA3 subfields of the hippocampus. For this purpose, we recorded single units simultaneously from the CA1 and CA3 while rats (n = 4) experienced visual landmarks in a virtual reality (VR) environment. The rat was head-fixed and trained to run on a treadmill to obtain water rewards in random locations in the virtual track. Once the rat reached the end of the linear virtual track (3m in length), the rat was unknowingly teleported to the start of the track so that the animal could travel indefinitely along the track. Five different environments were used in the study, depending on the number of visual landmarks presented in the virtual environment: (a) no landmark (NL) - a black screen was presented along the sides of the track; (b) single landmark (SL) - one visual landmark was shown on one side of the track; (c) dual landmarks (DL) - one more visual landmark was added to the SL condition; (d) triple landmarks (TL) - the third visual landmark was added to the DL condition; (e) ensemble landmarks (EL) - the empty spaces among the three visual landmarks of the TL condition were filled with many visual landmarks to produce a scene-like stimulus. On the first day of recording, rats ran in the NL and EL conditions. On the next day, rats ran in the SL, DL, TL, and EL conditions in that sequence (30 trials/condition). Finally, rats experienced the same conditions in the reverse order (EL to SL). Place cells were recorded from the CA1 (n = 312) and CA3 (n = 175) while rats ran in those virtual environments. In the NL, place cells were hardly found in both the CA1 and CA3. However, adding a visual landmark produced a dramatic increase in the number of place cells. Specifically, in the CA1, the proportion of place cells was positively correlated with the number of landmarks (SL: 15.4%, DL: 28.5%, TL: 42.6%, EL: 47.1%). Surprisingly, place cells in the CA3 rarely responded when individual landmarks were added, but they dramatically increased their activities as the scene-like ensemble landmarks were presented in the EL condition (SL:

7.4%, DL: 7.4%, TL: 7.3%, EL: 20.6%). The difference in the proportion of place cells between the CA1 and CA3 was statistically significant ($p < 0.001$, logistic regression). Very similar trends were also found in other measures (e.g., spatial information, sparsity, etc.) to describe the behavior of the place cells, and no sequence effect was found between the SL-to-EL and EL-to-SL sessions. Our findings suggest that the CA1 represents individual landmarks, whereas the CA3 represents a complex visual scene more readily than the individual landmarks.

Disclosures: H. Lee: None. **I. Lee:** None.

Poster

P842. Dynamics of Memory Representations

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P842.08

Topic: H.08. Learning and Memory

Support: NRF 2017M3C7A1029661
NRF 2018R1A4A1025616
NRF 2019R1A2C2088799
the Ministry of Education and the NRF grant BK21 FOUR

Title: Theta phase-based multiple place fields of single-units in the subiculum represent multiplex information in a visual scene memory task

Authors: *S.-M. LEE[1], J.-M. SEOL[1], I. LEE, PhD[2];
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Abstract: The subiculum is located at the critical juncture to interface the hippocampus with the various cortical and subcortical areas. However, the exact roles of the subiculum in the hippocampal-dependent memory tasks remain largely unknown. One of the major obstacles to making analytical comparisons of neural firing patterns between the subiculum and the hippocampus is the broad and continuous firing fields of subicular neurons. To address this issue, we used spiking-phase relationships to parse the broad firing fields of a subicular neuron into multiple subfields to find the unique functional contribution of the subiculum in a hippocampal-dependent visual scene memory task. Specifically, rats were trained to choose either the left or right arm in a T-maze according to the visual scene stimulus presented on LCD monitors. Once the rat reached the performance criterion, the single-unit spiking activities and local field potential were recorded simultaneously in the dorsal subiculum and CA1 by an implanted multi-electrode drive equipped with 24 tetrodes. We found that some of the broad firing fields of the subicular neurons were successfully divided into multiple subfields by using the theta-phase precession cycle. The resulting phase-based fields in the subiculum were similar to those in CA1 in terms of the field size and phase-precession strength. Furthermore, this new field identification

method using phase code significantly improved the detection of task-relevant information (e.g., visual scene or choice response) in subicular cells without affecting the information content represented by CA1 cells. Notably, multiple fields of a single subicular neuron could carry heterogeneous task-related information such as visual scene and choice, which was rarely observed in the CA1. Our findings suggest that the subicular cells represent multiplex information and integrate them by using theta rhythm to associate an environmental context with action.

Disclosures: S. Lee: None. **J. Seol:** None. **I. Lee:** None.

Poster

P842. Dynamics of Memory Representations

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P842.09

Topic: H.08. Learning and Memory

Title: A modeling framework for studying dynamic memory representations in the hippocampus

Authors: *M. NATRAJAN, J. E. FITZGERALD;
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Abstract: A central question in neuroscience is how learning and memory are manifested by changes in the brain. In the hippocampus, a key center for learning and memory, connections from CA3 to CA1 region have been found to be highly dynamic, with spines continually appearing and disappearing (Attardo et al., 2015). Dynamics have also been seen at the representational level, as different assemblies of CA1 place cells are activated on different days, even when the animal is not learning or forgetting a task (Ziv et al., 2012). Some neurons lose selectivity for a place while others become selective, leaving many statistics of representation intact. This leads to our general question. How can spatial memory in the hippocampus be retained, or even improved, by circuit remodeling and representational drift? Since there is redundancy when a low-dimensional task is represented in high-dimensional neural activity, representational changes don't lead to memory changes if representational dynamics are limited to the null space (Rule et al., 2020). A Learning scheme from Kappel et al. (2015, 2018) provides a neural network framework for exploring multiple high-performance regions of the synaptic parameter space.
Here we built a hippocampus model with 3 layers: (i) CA3 with stable place representation; (ii) CA1; and (iii) abstract spatial decoding neurons. CA3-CA1 connections were dynamic, while connections from CA1 to decoding neurons were fixed. By adapting the Kappel et al. plasticity rules to CA3-CA1 connections, CA1 learned to inherit spatial information from CA3, and the system could maintain its representation despite ongoing changes in connectivity and CA1 representation. We analytically characterized the representational solution space in a general

case, as well as for a simpler toy model where the solutions could be visualized and intuitively understood. Using these analytical results, we developed a numerical method of diffusion with constraints to explore CA1 representations that generate exact position decoding. This simple model recapitulated the phenomena of fading in and out of the representation, with changes in cellular membership as observed experimentally. Though all CA3-CA1 connections were unstable, when a CA1 neuron dropped out and then dropped back in, it responded to the same angle that it used to respond to previously. Thus, simulations reproduce patterns seen experimentally and help us understand how long-term memory can exist without a stable representation. We are currently applying this framework to study potential benefits of representational drift including encoding uncertainty, timestamping, robustness, and learning multiple environments.

Disclosures: M. Natrajan: None. **J.E. Fitzgerald:** None.

Poster

P842. Dynamics of Memory Representations

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P842.10

Topic: H.08. Learning and Memory

Support: NRF 2017M3C7A1029661
NRF 2018R1A4A1025616
NRF 2019R1A2C2088799
the BK21 FOUR by the Ministry of Education and the NRF.

Title: Differential neural correlates of place cells between the CA1 and CA3 subfields of the hippocampus in response to visual novelty in a VR environment

Authors: *J. SHIN[1], S.-W. JIN[2], I. LEE, PhD[2];
[2]Brain and Cognitive Sci., [1]Seoul Natl. Univ., Seoul, Korea, Republic of

Abstract: Although it has been demonstrated that the dentate gyrus and CA3 subfields of the hippocampus process environmental novelty through pattern separation and completion, the roles of the CA1 remain largely unknown. To investigate the differential roles of the CA1 and CA3 in processing a modified environment in a more controlled setting, we recorded single units simultaneously from both subfields in a virtual reality (VR) system. Single units were also recorded simultaneously from the visual cortex (V2L) as the head-fixed rat navigated in different VR track environments by running on a cylindrical treadmill. In each environment, rats (n = 8) were trained to move forward along a linear track to obtain water rewards on the two random sites per trial (20 μL/site). Once the rat reached the criterion, data acquisition started. Robust place fields were found in our VR system (CA1: 27.72%; n = 168/606, CA3: 20.51%; n =

129/629). After experiencing the original environments for ~7 days, different conditions of fog trials (controlled by modifying the density of fog; e.g., 15% and 30% of fog) were introduced. When different densities of visual fog were pseudo-randomly introduced to the familiar environment after the rat finished the standard trials as a baseline, 37.0% (n = 80/216) of CA1 place cells showed global remapping instantly. In contrast, 88.3% (n = 98/111) of CA3 place cells retained their original firing fields. Place cells in CA1 exhibited significant global remapping than those in CA3 and V2L ($p < 0.0001$, Kruskal-Wallis test). In addition, when in-field firing rates were compared across the fog conditions, the subpopulation of CA1 cells that showed global remapping exhibited a significant increase in the firing rate across the fog conditions ($p < 0.0001$). Our findings suggest that most place cells in the CA3 preserve their memory representation of the original environment where there is a subtle visual mismatch between past and present VR environments. However, some place cells in the CA1 rapidly form new spatial representations and increase their firing rates over time, presumably to stabilize the neural firing patterns. Our prelimianary data suggest that the CA1 network is more biased to form a new spatial representation in response to visual novelty than the CA3 network. In contrast, the CA3 is more biased to maintain the neural representation for the original environment despite the novel visual changes in the VR environment.

Disclosures: J. Shin: None. **S. Jin:** None. **I. Lee:** None.

Poster

P882. Timing and Temporal Processing I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P882.01

Topic: H.10. Human Learning and Cognition

Title: Temporal recalibration occurs without awareness across motor and perceptual judgment tasks

Authors: *M. BUBNA[1], M. LAM[2], E. CRESSMAN[1];
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Abstract: Sensory stimuli occurring immediately after a voluntary action are typically interpreted as consequences of the action (e.g., one presumes a letter appearing on the screen arises due to the fact that they pressed a key on the keyboard). After exposure to a short, constant delay between voluntary movement and sensory stimuli, temporal recalibration (TR) occurs to realign asynchronous events such that they once again appear consistent with previous expectations. The current experiment looked to establish if awareness of the temporal lag between a motor response (i.e., a keypress) and a sensory event (i.e., a visual flash) is necessary for TR to occur. We further investigated whether the influence of awareness on TR is modulated

based on the motor and perceptual judgment task requirements. Two groups of 24 participants (Group 1: Aware and Group 2: Unaware) completed 8 blocks of trials, in which the motor tasks (e.g., a single or repetitive tap), perceptual judgment tasks (e.g., judging the temporal order of the keypress in relation to the visual flash or judging whether the two events were simultaneous or not), and fixed temporal lag between keypress and visual flash (e.g., a 0 ms or 100 ms lag) varied. The Aware group was told of the temporal lag between their keypress and visual flash at the beginning of each block, whereas the Unaware group was not. TR was determined by comparing judgments between corresponding blocks of trials with the same motor and perceptual judgment task requirements when the temporal lag was 0 ms and 100 ms. Results revealed that both the Aware and Unaware groups demonstrated a similar magnitude of TR across all motor and perceptual judgment tasks. However, accuracy of responses differed between the two judgment tasks, such that simultaneity of the flash and keypress was perceived to occur *before* true simultaneity in the temporal order judgment task, but *after* true simultaneity when judging whether the events were simultaneous or not. We propose that each judgment task engaged different cognitive processes that resulted in their respective response patterns. Together, these results suggest that awareness of a temporal lag does not influence the magnitude of TR achieved and that motor and perceptual judgment task demands do not modulate the influence of awareness on TR.

Disclosures: M. Bubna: None. **M. Lam:** None. **E. Cressman:** None.

Poster

P882. Timing and Temporal Processing I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P882.02

Topic: H.10. Human Learning and Cognition

Support: JSPS KAKENHI (19H01087)
JSPS KAKENHI (17KK0004)

Title: Testing the crossed-hands effect on motor-effector specificity for learning of multiple prior distributions in human coincidence timing

Authors: *Y. MATSUMURA[1], N. SHIMADA[2], R. SATO[1], J. HERON[3], N. ROACH[4], M. MIYAZAKI[2,1];
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Abstract: The brain can acquire a prior distribution of sensory inputs and use them to optimize performance on sensorimotor tasks. Recently, we found that in a coincidence timing task, participants can simultaneously acquire two different prior distributions when they are assigned to two different motor effectors such as the right and left hands (motor-effector specificity). In the present study we use the crossed-hands method (Yamamoto & Kitazawa. 2001, Shore et al. 2002) to investigate whether effector specific priors are organized according to intrinsic (anatomical) or extrinsic (extrapersonal) spatial coordinates. In our experiments, three sequential visual stimuli (S1→S2→S3) were presented on the right or left side of the fixation point. Participants pressed a key such that its onset coincided with the onset of S3 based on the time intervals between S1 and S2 (target intervals). Each participant performed 640 trials (40 trials/set × 16 sets) of the task. Target intervals were sampled from short-time (424-988 ms) or long-time (1129-1694 ms) prior distributions. Each of the two priors (short/long) was assigned to one of the two stimulus locations (right/left). In Experiment 1, participants performed the task while crossing their hands in all sets. For example, when stimuli appeared on the left side, participants pressed the left key using their right hand. Under these conditions, participants' temporal estimates were biased in a manner consistent with acquisition of both the long-time (sets 1-4 onwards) and short-time priors (sets 5-8 onwards). In Experiment 2, participants crossed their hands during the first half of testing (sets 1-8), then uncrossed them for remaining trials (sets 9-16). While similar results to Experiment 1 were found during the initial, crossed-hands trials, the influence of the short-time prior disappeared when the hands were uncrossed. These results suggest that multiple priors are acquired based on the mapping between stimuli and intrinsic, body-centered co-ordinates.

Disclosures: Y. Matsumura: None. **N. Shimada:** None. **R. Sato:** None. **J. Heron:** None. **N. Roach:** None. **M. Miyazaki:** None.

Poster

P882. Timing and Temporal Processing I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P882.03

Topic: H.10. Human Learning and Cognition

Title: Human's heterogeneous perception of time

Authors: M. POURRAHIMI[1], ***N. MIRHAKIMI**[1], N. ESHRAGHI[2], S. AGAHI[2], M.-R. A.DEHAQANI[1];
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Abstract: A great deal of research has been conducted to reveal the underlying mechanisms of time perception in humans, yet these mechanisms remain still in debate. One of the main observable characteristics of human timing behavior is the way time perception varies with variations in the time interval to be perceived (i.e. target time). Gaining an understanding of the pattern of dependency of time perception on the target time is proved to be a great facilitator in the development and assessment of time perception models as is greatly exploited in the investigation of the conformity of the Scalar Expectancy Theorem (SET) to human timing behavior. Although some research has been conducted to address this dependency through examining the validity of mean accuracy property (the mean of perceived times equals target time) and scalar property of time variance (the standard deviations of perceived times grow linearly with the increase in target time) presumed by the SET, the pattern of this dependency remains poorly understood. Moreover, there is mixed evidence about the validity of SET as a major theorem for human time perception. Therefore, we designed a sub-second fine resolution time production task to investigate the dependency of time perception, measured by participants' produced time (i.e. perceived time), on the length of target time and further study the ability of SET in explaining human timing behavior. Time perception was evaluated regarding three different aspects, namely, temporal accuracy, temporal variability, and temporal learning. The observations reveal heterogeneous dependence of human timing behavior on the target time, regarding all three aspects. These patterns are proved to be consistent for all participants through sessions of the experiment. Results pose a new challenge on the SET, opposing its assumption of scalar property, shedding light on our understanding of underlying time representations in the brain.

Disclosures: M. Pourrahimi: None. **N. Mirhakimi:** None. **N. Eshraghi:** None. **S. Agahi:** None. **M. A.Dehaqani:** None.

Poster

P882. Timing and Temporal Processing I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P882.04

Topic: H.10. Human Learning and Cognition

Support: K23NS110920
R01-DC012379
R00-NS065120
DP2-OD00862

Title: Human hippocampal surface micro-grid recordings reveal spatiotemporal timecourses of normal and abnormal neurophysiology

Authors: *J. KLEEN[1], K. K. SELLERS[2], J. ZHOU[3], M. TRIPLETT[3], K. LEE[3], A. TOOKER[3], R. HAQUE[4], E. F. CHANG[5];
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Abstract: Intracranial recordings in patients with drug-resistant epilepsy provide a novel approach to studying hippocampal neurophysiology in vivo. This includes how oscillations and other transient phenomena propagate within this structure. We recently described the existence of bidirectional traveling waves moving up and down the hippocampal structure using novel thin-film micro-grid arrays conformed over this structure's ventricular surface. Here, we extend our examination to the spatiotemporal properties of transient bursts of activity including both normal and abnormal (epileptiform) phenomena. Six participants undergoing epilepsy surgery (five with medically-refractory seizures and one with a circumscribed lateral temporal lobe glioma) underwent limited excision of the antero-lateral temporal lobe for clinical purposes (four under anesthesia and R-sided, two awake and L-sided). A 32-channel micro-grid array (4 x 8 layout, 2mm spacing, 1.22mm diameter platinum/iridium contacts) was carefully placed and conformed onto the lateral hippocampal surface over the CA1 region. Activity was recorded at 24 kHz from all 32 contacts for approximately 8-22 minutes for each participant. Using two-dimensional regression modeling, we found that traveling low-frequency (1-15 Hz) oscillations occurred in transient bouts, with medians ranging from approximately 1.2 to 3.0 seconds, and maximums from 3.4 to 9.3 seconds. There was no correlation between maximum bout duration and oscillation frequency ($p=0.141$, Spearman's rho). Normal physiological hippocampal ripples were also observed, though less frequently in participants with more severe hippocampal disease. Interictal epileptiform discharges (IEDs) were observed in five participants (2.13 to 15.71 per minute, median 5.97). Preliminary results using a template matching algorithm suggested IEDs were generated from one or occasionally two consistent spatial sources that were distinct for each participant. In summary, low-frequency traveling oscillations occur in short (seconds-long) bouts, translating to several or dozens of continuous cycles flowing toward either the septal or temporal hippocampal pole. These transient timescales may support information processing (online and offline) by facilitating frequent diverse exchanges between the hippocampus and cortical structures interconnected along its axis. The two-dimensional spatial perspective also suggested that IEDs are generated from intrahippocampal sources that show little spatial variation (within millimeters) suggesting relatively concrete pathophysiological circuitry.

Disclosures: J. Kleen: None. **K.K. Sellers:** None. **J. Zhou:** None. **M. Triplett:** None. **K. Lee:** None. **A. Tooker:** None. **R. Haque:** None. **E.F. Chang:** None.

Poster

P882. Timing and Temporal Processing I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P882.05

Topic: H.10. Human Learning and Cognition

Support: Institute for Biohealth Innovation (George Mason University)

Title: A fMRI-EEG investigation of feedback-driven improvements in visual time reproduction

Authors: *F. BADER[1], M. ALQURAISHI[2], M. WIENER[3];
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Abstract: Time perception is critical for everyday behaviors and cognitive functions ranging from speech to motor control. Errors may occur when learning to time durations; therefore, recognizing and correcting the temporal errors is essential for timing self-awareness. Electrophysiology and behavioral studies have revealed that humans possess a certain self-awareness of their individual timing ability; however, there are conflicting reports over whether humans can discern the direction of the timing error (earliness or lateness). Testing the impact of feedback on time perception is a way to understand the level of self-awareness of temporal errors. Recently, we performed a behavioral study using a visual temporal reproduction task and showed that temporal estimates were more accurate and precise when post-trial non-directional feedback on the estimates was provided. Even in the absence of feedback, subjects improved the accuracy of temporal estimates but were not more precise. Here, in the present study, we examined the neural underpinnings of our these behavioral findings by conducting a simultaneous fMRI-EEG study of eleven healthy adults while they performed a visual temporal reproduction task. Participants viewed a blue square for a set amount of time and then re-created the time the square was on-screen using a keypress and received adaptive non-directional feedback for their performance. Crucially, participants could repeat each trial following feedback, essentially allowing a “re-do” to learn from the successes or errors in the first trial. We observed two event-related potentials while participants underwent fMRI-EEG: the contingent negative variation (CNV), a timing-related signature that indexes time durations and is associated with temporal expectation and encoding; and the reward positivity (RewP), a feedback component reflective of outcomes in probabilistic trial-and-error learning tasks. During the reproduction phase of the task, significant EEG-informed fMRI activity was observed in the supplementary motor area (SMA), pre-motor cortex, and thalamus. With positive feedback, the EEG-informed fMRI activity due to the CNV from the SMA elicited a large RewP and was observed in the basal ganglia and thalamus. Conversely, negative feedback due to inaccurate responses led to greater activity within the SMA. The results demonstrate the importance of feedback valence in modulating region-specific responses to temporal judgements and in influencing the future processing of temporal information.

Disclosures: F. Bader: None. **M. Alquraishi:** None. **M. Wiener:** None.

Poster

P882. Timing and Temporal Processing I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P882.06

Topic: H.10. Human Learning and Cognition

Support: JSPS KAKENHI (19H01087)
JSPS KAKENHI (17KK0004)

Title: Inner-speech specificity for the acquisition of multiple prior distributions in human coincidence timing

Authors: *S. NATSUME[1], C. YAMADA[3], Y. MATSUMURA[1], R. SATO[1], M. MIYAZAKI[1,2]; [1]Grad. Sch. of Integrated Sci. and Technol., [2]Fac. of Informatics, Shizuoka Univ., Hamamatsu, Japan; [3]Advanced Res. Ctr. for Human Sci., Waseda Univ., Tokorozawa, Japan

Abstract: The brain can maximize precision in a sensorimotor task by optimally utilizing a prior distribution of the task. To deal with various events in daily life, we should acquire multiple priors in the real environment. Our group recently reported that if two priors are assigned to two different motor effectors (left hand/right hand), participants acquire the priors separately ("motor-effector specificity"). However, in daily tasks (e.g., baseball), we cannot always use different motor effectors according to differences in targets (e.g., fastball/slowball). Here, we focused on "inner speech." During timing in sports, we often chant "1, 2, 3" internally. We expect that if we assign different inner speeches to different priors, we may acquire the priors separately ("inner-speech specificity"). In our experiments, three sequential visual stimuli (S1, S2, S3) were presented on the left or right side of a fixation point. Based on the S1-S2 interval, participants anticipated the onset of S3 and pressed a key to temporally coincide with the onset of S3. The S1-S2 (= S2-S3) interval was randomly sampled from the short-time (424-988 ms) or long-time (1129-1694 ms) prior distribution. These priors were assigned to the left or right visual stimuli. The participants performed the task using only the dominant index finger. Each participant completed 640 trials (40 trials/set × 16 sets) of the task. In Experiment 1, participants (n = 12) had no instructions regarding inner speech. As a result, the participants only acquired the long-time prior but not the short-time prior. In Experiment 2, participants (n = 12) were instructed to perform the task with inner timing as "A, A, A" or "B, B, B" according to the stimulus positions. In the first-half (1-320) trials, the participants acquired the long-time prior but not the short-time prior. However, in the second-half (321-640) trials, they also acquired the short-time prior. These results suggest that we can acquire multiple prior distributions by assigning different inner speeches to the priors. Meanwhile, individual variation was also observed, although the results were statistically significant. Four of twelve participants did not acquire the short-time prior in the second-half trials, in which two participants acquired the short-time prior in the first-half trials, but this disappeared in the second-half trials. This may reflect that some participants could not continue the inner speeches with sufficient intensity over

a large number of trials. We should also investigate whether participants acquire multiple priors more robustly if they are assigned different explicit speeches to the priors.

Disclosures: S. Natsume: None. **C. Yamada:** None. **Y. Matsumura:** None. **R. Sato:** None. **M. Miyazaki:** None.

Poster

P887. Neural Circuits and Mechanisms II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P887.01

Topic: H.11. Language

Support: 108-2410-H-010-006-MY2
107-2410-H-010-006-

Title: Neural mechanism of language tone processing revealed by distributed source modeling of intracranial stereoelectro-encephalographic (sEEG) measurements

Authors: *H.-J. LEE[1,2], H.-Y. YU[3,5], C.-C. LEE[4,5], C.-C. CHOU[3], F.-H. LIN[1,2], W.-J. KUO[6,5]; [1]Physical Sci., Sunnybrook Res. Inst., Toronto, ON, Canada; [2]Dept. of Med. Biophysics, Univ. of Toronto, Toronto, ON, Canada; [3]Dept. of Neurol., [4]Dept. of Neurosurg., Taipei Veterans Gen. Hosp., Taipei, Taiwan; [5]Brain Res. Ctr., [6]Inst. of Neurosci., Natl. Yang Ming Chiao Tung Univ., Taipei, Taiwan

Abstract: Language tone is a suprasegmental speech unit and functionally serves word meaning distinction. Understanding how tone processing proceeds in the brain is essential because it can help resolve a paradox between linguistic pitch processing (i.e., tone) and non-linguistic pitch in the brain and link to research about how the brain develops to acquire a language. In this study, by taking advantage of the high spatiotemporal resolution of intracranial EEG data, i.e., the stereo-EEG (SEEG), we were allowed to profile the neural mechanism of language tone processing with precision higher than other neuroimaging tools, for example, fMRI. Specifically, we expect that linguistic (tone) knowledge would modulate brain activation of the frontal and temporal language areas, i.e., earlier and larger activation for language tone judgment (linguistic pitch) than for voice gender discrimination (non-linguistic pitch).
Twelve refractory epilepsy patients, who received depth-electrode implantation to monitor their epileptiform activities for surgery purposes, gave written informed consent for participation before joining the experiment. Each electrode (5-mm spacing between contacts) had 6 or 8 contacts. SEEG was sampled at 2,048 Hz. T1 MRI before and after electrode implantation was collected for localizing intracranial contacts. Two tasks were designed by using the same set of speech sounds. Participants performed tone judgment in one task and gender judgment in another one. Noise-normalized distributed source modeling of SEEG data was calculated at each time

point using the minimum-norm estimates.
The results for language tones indicated the estimated neural current starts at the left primary auditory cortex (A1) at 24 ms after the tone sound display and starts later with smaller amplitudes at the right A1 (34 ms). Then, the neural current spreads to the associated auditory cortex (A2) and medial temporal cortex at around 44 ms after the sound onset and subsequently to frontal and inferior temporal regions. For voice gender processing, the start of neural current occurs at about 63 ms of the sound onset. It spreads through bilateral A2 and medial temporal areas to the ventromedial prefrontal cortex and frontal pole. The activity for voice gender judgment is smaller and relatively dispersed than that for language tone discrimination.
In sum, our SEEG results indicated the brain networks recruited for process language tone (linguistic pitch) and voice gender (non-linguistic pitch). Neural activation for the former took place in temporal and frontal language networks with an earlier onset and a larger amplitude than that for the latter.

Disclosures: H. Lee: None. **H. Yu:** None. **C. Lee:** None. **C. Chou:** None. **F. Lin:** None. **W. Kuo:** None.

Poster

P887. Neural Circuits and Mechanisms II

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Program #/Poster #: P887.02

Topic: H.11. Language

Support: This research is supported by the Azrieli Centre for Autism Research (ACAR), the Fonds de recherche du Québec – Santé (FRQS
This research is supported by the Quebec Autism Research Training Program

Title: Lateralization during development as a predictor of language outcomes in ASD

Authors: *G. BLANCO-GOMEZ[1], C. O'REILLY[2], M. ELSABBAGH[2];
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Abstract: Autism Spectrum Disorder (ASD) is a neurodevelopmental disorder characterized by impaired social and communication skills also involving language impairments in 50% of the cases. It is also associated with reduced brain lateralization, but more research is needed to understand how a lack of lateralized dominance during language tasks in ASD contributes to language impairments. We hypothesize that abnormal lateralization in language areas will predict language outcomes. We used the EEG-IP cohort; a multi-site ASD sibling cohort (London: 7, 14 months; Seattle: 6, 12, 18 months) including 195 infants. We excluded 12 subjects after artifact rejection/correction (92 at familial risk; 91 controls; 88 males; 95 females). We estimated EEG cortical sources and computed brain lateralization based on spectral power

(SP) using (Left SP− Right SP)/(Left SP+ Right SP). Mullen Scales of Early Learning (MSEL) scores were also collected at 24 months. We used cluster-permutation to identify regions showing significant between-group differences in lateralization, in both sites independently. For this set of regions, we confirmed these associations by using a linear mixed-effect model with age and site as within-subject covariates. Controls showed leftward lateralization for all 3 selected regions (superior parietal gyrus: p=0.03, z=-1.91; middle temporal gyrus: p=0.04, z=-2.05; pericalcarine area: p=0.03, z=-2.21; n=164). Infants at risk showed reduced lateralization. To investigate interactions between lateralization and MSEL Expressive and Receptive Language scores, we supplemented this set of regions with 6 language-associated regions based on the literature. Using linear regression, lateralization was correlated with 24-month expressive language across EEG time points for the at-risk group for the inferior parietal region (Pearson's ρ= 0.62, 95% CI [0.57, 0.74], p=10−12). Other regions showed inconsistent patterns across diagnostic groups and time points. No significant correlations were found for receptive language. The inferior parietal region has been previously associated with syntax processing, sensorimotor integration and the perception of emotion, suggesting that disruption of these networks during infancy can contribute to expressive language impairments.

Disclosures: G. Blanco-Gomez: None. **C. O'Reilly:** None. **M. Elsabbagh:** None.

Poster

P887. Neural Circuits and Mechanisms II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P887.03

Topic: H.11. Language

Support: NIH Grant DC014989

Title: Neural mechanisms of sensorimotor control in the auditory dorsal stream of the macaque

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Abstract: Ventral and dorsal streams analogous to those in visual cortex have been established in the auditory cortex (AC) of humans and nonhuman primates using neuroimaging and electrophysiology (Rauschecker and Tian, 2000; Rauschecker and Scott, 2009). The auditory dorsal stream is known for its role in the processing of auditory space and motion, action understanding, and speech processing. What these seemingly heterogeneous functions have in common is the implementation of internal models, relating motor commands to their sensory outcomes. Using functional MRI, our lab has shown that motor regions along the dorsal stream and in the putamen are activated, when macaques are listening to auditory-motor sequences they had previously learned to produce by pressing levers with their hands ('monkey piano')

(Archakov et al, 2020). Specifically, the hand/arm regions of premotor cortex (PMC) displayed increased activity when the monkeys listened to self-produced sound sequences as opposed to sound sequences they could not produce. To characterize the role of dorsal-stream areas in auditory-motor control at the single-unit level, we are now performing electrophysiological recordings from auditory and motor regions in awake behaving monkeys. Animals are trained to produce an 8-note tone sequence on the monkey piano. During unit recordings, the animals are either listening to learned auditory sequences, or are engaged in the production of auditory-motor sequences. The results suggest that a predictive forward model alters sensory responses in AC and PMC based on task conditions. Auditory responses of single neurons in AC were suppressed during sequence-production compared to passive listening of the same learned sounds. Furthermore, when the piano produced incorrect notes, i.e. the sounds did not align with the animal's expectation, AC neurons responded to the sound more robustly, indicative of an error signal being generated. Similar error signals were also observed in PMC. Our findings support the notion that an internal model of expected auditory consequences of motor behavior is generated in PMC. This prediction signal is sent to AC, where it is compared to the actual auditory input. Prediction errors generate a signal in the AC, which is sent to motor regions. To confirm the directionality of these signals, we are planning simultaneous recordings in AC and PMC. Together, the results provide evidence for the existence of internal models in the auditory dorsal stream of old-world monkeys. Internal models are a prerequisite for speech. The fact that monkeys don't speak suggests that internal models have evolved to support their hand movements but not their vocalizations.

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Poster

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Title: Bidirectional Connectivity Between Broca and Wernicke's Areas During Interactive Listening

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Abstract: We investigated a directional connectivity network in canonical language areas of two brains during interactive and non-interactive verbal communication. The underlying hypothesis is that functional connectivity between canonical speech areas in the human brain will be modulated by social interaction. Thirty-three pairs of healthy adult participants alternately talked and listened to each other while performing an object naming and description task that was either interactive or not during hyperscanning with functional near-infrared spectroscopy (fNIRS)[1]. In the non-interactive condition, the speaker named and described a picture-object without reference to the partner's description. In the interactive condition, the speaker performed the same task but included an interactive response about the preceding comments of the partner. Directional connectivity networks of hemodynamic responses from Broca's and Wernicke's Areas were determined by comparing Granger causality measures between real, surrogate, and shuffled trials within dyads. We found valid directional connectivity within the speaker's and the listener's brains, while no statistically significant across-brains directional connectivity was found[2]. The interactive communication was characterized by bidirectional connectivity between Wernicke's and Broca's Areas of the listener's brain. Whereas, this connectivity was unidirectional in the speaker's brain. In the case of the non-interactive condition, both speaker's and listener's brains showed unidirectional top-down (Broca's Area to Wernicke's Area) connectivity. The directional connectivity analysis revealed bidirectional flow of neuronal information during dynamic two-person verbal interaction for processes that are active during listening (reception) and not during talking (production). Findings are consistent with prior cortical activity measures showing neural modulation of the receptive language system associated with Wernicke's Area during two-person live interaction.
1.Hirsch, J., Noah, J.A., Zhang, X., Dravida, S., Ono, Y. A Cross-Brain Neural Mechanism for Human-to-Human Verbal Communication. Social Cognitive and Affective Neuroscience. Volume 13, Issue 9, 11 September 2018, Pages 907–920. doi: 10.1093/scan/nsy070.2.Ono, Y., Zhang, X., Noah, J.A., Dravida, S., & Hirsch, J. Bidirectional connectivity between Broca's area and Wernicke's area during interactive verbal communication. Brain Connectivity. 2021 Jun 15. doi: 10.1089/brain.2020.0790. Online ahead of print.

Disclosures: Y. Ono: None. **X. Zhang:** None. **J.A. Noah:** None. **S. Dravida:** None. **J. Hirsch:** None.

Poster

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Topic: H.11. Language

Support: PICT 2018-02699
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ARL W911NF-19

Title: Differential brain activity on mnemonic predictions during natural reading: an EEG and eye tracking co-registration study

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Abstract: During reading our brain is continuously generating predictions on upcoming words. These predictions, if correct, could speed up the processing of the word when it is finally read. Also, they are a reflection of the ongoing integration of the text. It has been amply shown that Predictability (i.e. the estimated probability of guessing the next word) have an impact on how we move our eyes across the text: eye tracking (ET) experiments showed that more predictable words are fixated for shorter periods of time than less predictable words. Additionally, EEG experiments showed that the amplitude of the N400 potential, a hallmark of semantic processing, is modulated by the Predictability: the more predictable words are, the less amplitude the N400 has. Typically, EEG experiments were performed avoiding eye-movements, because the muscular contraction interferes with the brain electrical signal. But, during the last years, co-registration of EEG and ET have become a powerful technique for understanding our brain in more natural contexts, allowing eye movements during the task.
Previous ET studies have shown that mnemonic predictions increase the skip probability and decrease the gaze duration on words. Meanwhile, previous EEG studies during Serial Visual Presentations of words have shown that the N400 potential is only present for predictions performed based only on linguistic context. In the present work, we analyzed brain potentials and eye movements during natural reading of Common and Memory-Encoded sentences (like proverbs), with the goal of understanding how different sources of predictions are used during natural reading.
Firstly, we replicated the results from both experiments under the co-registration paradigm. Secondly, our results showed that during memory-encoded sentence reading there is also a decrease in regression and refixation probabilities. And thirdly, fixation-related potentials analysis using a novel Linear Mixed Models (LMM-CBP) approach showed that the classical N400 component is only present for common-sentence reading and for words previous from the point in which the Memory-Encoded sentences are recognized. These results indicate that the

N400 potential could be associated only with predictions performed based only on linguistic context.

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Poster

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Program #/Poster #: P887.06

Topic: H.11. Language

Support: NIH NIDCD R01 DC014589

Title: The critical role of the mid fusiform cortex in semantic word retrieval

Authors: *K. SNYDER[1], C. DONOS[2], K. FORSETH[3], P. ROLLO[4], J. BREIER[5], S. FISCHER-BAUM[6], N. TANDON[7];
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Abstract: Temporal lobe resections often result in significant confrontation naming decline, which can prevent patients from returning to independent functioning. While exact substrates responsible are unknown, it was previously assumed that injury to the superior temporal gyrus or resection size predicted these declines. A clear understanding of critical constituents might influence surgical strategies to minimize language declines. We used multivariate voxel based lesion symptom mapping (VLSM) to relate changes in language function using the Boston Naming Test (BNT) to dominant temporal lobe resections.
Data were obtained from 95 patients who underwent surgery in the dominant left temporal lobe for epilepsy. Patients underwent neuropsychological testing and MRI prior to and 6 months after surgery. Lesion masks were traced on postoperative MRIs and aligned to a normative space. The effects of preoperative scores were removed from postoperative scores via linear regression. VLSM using multivariate support vector regression was used to assess postoperative test scores. Beta maps were converted to p values and corrected using a permutation-based cluster level correction. In a subset of subjects with ECoG recordings, a surface-based mixed-effects multilevel analysis was used to estimate broadband gamma activity during picture naming, and the resulting maps were integrated with VLSM results. Additionally, DCM results were mapped to the cortical surface as percent of stimulations causing functional disruption, and regions associated with language disruption were compared with those found in VLSM. Lastly, fMRI

(picture naming) was used to assess SNR in significant regions.
VLSM results revealed that loss of basal temporal regions, primarily mid fusiform gyrus (mFus) and inferior temporal gyrus, was associated with a prominent decline on BNT ($p<0.005$). ECoG analysis showed a significant increase in activity in mFus during picture naming immediately preceding articulation. CSM of mFus caused disruption of visual naming without disrupting repetition. Many of these voxels were spatially correlated with the susceptibility artifact on fMRI perhaps explaining why this region has been underappreciated as the locus responsible for postoperative deficits.
The importance of the ventral temporal cortex in lexical access is shown using multivariate VLSM and further supported by both ECoG and CSM. While it has previously been assumed that mFus lesions do not cause pervasive impairment, our data and recent work (FATES study) support the role of mFus as a critical semantic access hub. As such, surgical approaches in temporal lobe epilepsy should strive to preserve mFus.

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Poster

P887. Neural Circuits and Mechanisms II

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Topic: H.11. Language

Support: CIHR Grant 148762

Title: Reading is fundamental: connectivity at rest between visual word form area and left inferior frontal gyrus predicts global language skills throughout childhood and adolescence

Authors: *N. DEOM, O. KHALIL, L. J. AYOUB, M. MCANDREWS;
Univ. of Toronto, Toronto, ON, Canada

Abstract: The language network includes a vast system of interconnected brain regions, involving most of the temporal lobe as well as frontal and parietal regions. Characterization of the individual role of key regions in the network is well advanced, but assessment of the importance of connectivity between regions in the network is still incomplete, especially in children and young adults. We used resting-state functional MRI (rsfMRI) to investigate the connectivity between regions of interest in Broca's area, Wernicke's area, and the visual word form area (VWFA) and the rest of the language network and assessed their relationship to performance on language tests. Analyses were conducted on publicly available data from the Child Mind Institute's Healthy Brain Network initiative. From the available dataset, 175 subjects (81 males), ranging from age 6 to 21 (mean 12.2, SD 3.6), with complete 10 minutes resting-

state scans were included. From the rsfMRI scans, resting-state connectivity was calculated ROI-to-ROI between BOLD timeseries extracted from all regions of the language network. Language ability was evaluated using the listening comprehension subtest of the Wechsler Individual Achievement Test (WIAT-III) and the Clinical Evaluation of Language Fundamentals (CELF-5) Screening Test which assesses a broad range of language functions including grammar, semantics, verbal comprehension, and auditory memory. We found a significant positive correlation between connectivity between the VWFA and the left inferior frontal gyrus (l-IFG) and performance on the CELF ($r(173) = .30$, $p < .05$ FDR-corrected for multiple comparisons). We also found a positive correlation only as a trend between the connectivity between the VWFA and the left supramarginal gyrus and the WIAT ($r(155) = .29$, $p < .10$ FDR-corrected). Thus, a stronger connectivity at rest between the VWFA and the l-IFG predicted a better global language performance. In addition, none of these connectivity measures significantly correlated with participants' age. Our findings suggest that greater network connectivity of a brain region primarily associated with visual word decoding has a broader influence on language ability than had been previously recognized.

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Poster

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Program #/Poster #: P887.08

Topic: H.11. Language

Support: Région Bourgogne-Franche-Comté ANER RobotSelf

Title: Narrative Event Segmentation in the Cortical Reservoir

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Abstract: During perception of movies or stories, humans cortical activity reveals hierarchical segmentation of event structure. Sensory areas display high frequency segmentation related to the stimulus, while semantic areas display a lower frequency segmentation related to transitions between events (Baldassano et al. 2017). These hierarchical levels of segmentation are associated with different time constants for processing. Chien and Honey (2020) observed that when two groups of participants heard the same sentence in a narrative, preceded by different contexts, neural responses for the groups were initially different and then gradually aligned. The time constant for alignment followed the segmentation hierarchy: sensory cortices aligned most quickly, followed by mid-level regions, while some higher-order cortical regions took more than 10 seconds to align. These hierarchical segmentation phenomena can be considered in the

context of processing related to comprehension. Uchida et al. (2021) recently described a model of discourse comprehension where word meanings are modeled by a language model pre-trained on a billion word corpus (Yamada et al 2020). During discourse comprehension, word meanings are continuously integrated in a recurrent cortical network. The model demonstrates two fundamental characteristics: real-world event semantics are represented in the word embeddings, and these are integrated in a reservoir network which has an inherent gradient of functional time constants due to the recurrent connections.
Here we demonstrate how this model displays hierarchical narrative event segmentation properties beyond the embeddings alone, or their linear integration. The reservoir produces activation patterns that are segmented by the hidden Markov model (HMM) of Baldassano et al (2017) in a manner that is comparable to that of humans. Context construction displays a continuum of time constants across reservoir neuron subsets, while context forgetting has a fixed time constant across these subsets. Virtual areas formed by subgroups of reservoir neurons with faster time constants segmented with shorter events, while those with longer time constants preferred longer events. This neurocomputational recurrent neural network simulates narrative event processing as revealed by the fMRI event segmentation algorithm of Baldassano et al (2017), and provides a novel explanation of the asymmetry in narrative forgetting and construction observed by Chien and Honey (2020).

Disclosures: P.F. Dominey: None.

Poster

P887. Neural Circuits and Mechanisms II

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Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P887.09

Topic: H.11. Language

Title: Distinct nodal responses for verbs in the production network

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Abstract: Aphasia often presents with selective impairments in the ability to produce specific word classes, namely nouns and verbs, suggesting that the retrieval and production of distinct word classes engage unique network components. However, the spatiotemporal dynamics that separate these two processes are not yet fully understood. We used electrocorticographic recordings in a large cohort during visually-cued object and action naming tasks (n = 10730, 93 pts) to characterize the cortical network dynamics during noun and verb production. A surface-based mixed-effects multilevel analysis (SB-MEMA) of broadband gamma activity (BGA)

yielded population maps of focal activity changes. We further explored effects of semantic complexity by performing an SB-MEMA of complex versus simple verb stimuli. A broad cortical network was recruited for both verb and noun naming; immediately following picture presentation, cortical activity propagated along dorsal and ventral streams to intraparietal sulcus (IPS) and mid-fusiform gyrus (mFus), respectively. Next, cortical activity was observed concurrently in posterior middle temporal gyrus (pMTG), angular gyrus (AG), and a constellation of frontal regions: pars Triangularis (pTr) and opercularis, superior frontal sulcus, and supplementary motor areas. Pre-articulatory activity was uniquely observed in pMTG and AG during verb naming ($p<10^{-3}$); post-articulatory pMTG activity was matched in both tasks. pTr exhibited similar temporal dynamics in both noun and verb production, but this activity was significantly enhanced during verb production ($p<10^{-3}$). Peaks in cortical activity were observed in mouth sensorimotor and auditory cortex after onset of articulation. With respect to semantic complexity, a gradient of increasing BGA from nouns, to simple verbs, to complex verbs was observed in pTr and mFus ($p<10^{-3}$), suggesting that engagement of these broader language areas is influenced by complexity demands. Finally, we found that posterior AG is sensitive exclusively to verb complexity, whereas more anterior portions were exclusively sensitive to word class. This points to a functional dissociation in AG with respect to both grammatical (anterior) and semantic (posterior) representations, providing a more fine-grained computational description of this region than previous neuroimaging research. Our results demonstrate that object and action naming networks are overlapping but differentially activated. This comprehensive view of category specific production networks may contribute to rehabilitative interventions for aphasia.

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Poster

P902. Schizophrenia: Molecular Mechanisms I

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Program #/Poster #: P902.01

Topic: H.13. Schizophrenia

Support: T32DC011499

Title: Schizophrenia-associated MAP2 phosphorylation affects interactions with microtubules and actin

Authors: *R. DEGIOSIO[1], P. G. NEEDHAM[2], O. A. ANDREWS[3], M. J. GRUBISHA[1], J. L. BRODSKY[2], R. A. SWEET[1];
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Abstract: Microtubule-associated protein 2 (MAP2) is the predominant dendritic cytoskeletal regulator, influencing dendritic arbor and spine morphology, which are altered in schizophrenia (SZ). MAP2 is regulated by multiple mechanisms, most prominently phosphorylation (MAP2-P). We have previously demonstrated that MAP2-P state is altered in SZ, with multiple phosphopeptides being upregulated in disorder. Moreover, a subset of identified phosphopeptides were negatively correlated with dendritic spine density in primary auditory cortex, suggesting that SZ-associated MAP2-P has consequences for protein function, in turn compromising neuronal morphology/function. Here, we set out to characterize the consequences of SZ-associated MAP2-P events on protein function by assessing the 1) tubulin polymerization, 2) microtubule-binding, and 3) actin-binding abilities of purified phosphomimetic MAP2c constructs. Phosphomimicry at multiple residues in the proline-rich and C-terminal domains- including T293, T300, S426, S439 and S443- were sufficient to delay microtubule nucleation. S426 additionally slowed polymerization rate. T293, T300, S426 and S439 inhibited actin-binding, while only S426 and S439 inhibited microtubule-binding. Other tested sites, including T249, S252, Y253, S297 and S446, showed no effects. These data indicate that SZ is characterized by both regulatory MAP2-P events and events which are irrelevant to its interactions with cytoskeletal filaments. We put forward MAP2-P-mediated changes to microtubule nucleation kinetics and microtubule/actin crosslinking as potential mechanisms for dendritic dysmorphogenesis in SZ.

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Poster

P902. Schizophrenia: Molecular Mechanisms I

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Program #/Poster #: P902.02

Topic: H.13. Schizophrenia

Support: NIH Grant R01 MH110701
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Title: Executive dysfunction and its relationship with psychosis risk in the schizophrenia-associated 3q29 deletion syndrome

Authors: *E. SEFIK[1], K. ABERIZK[2], R. ESPANA[2], R. GUEST[2], K. GOINES[2], D. NOVACEK[2], M. MURPHY[1], E. WALKER[2], J. MULLE[1];
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Abstract: The 3q29 deletion syndrome (DS) is caused by the recurrent hemizygous deletion of a 1.6Mb genomic interval spanning 21 protein-coding genes. 3q29DS is a major risk factor for

psychosis, as it confers a ~40-fold increased risk for developing schizophrenia. Recent findings by the Emory 3q29 Project indicate that executive function (EF) deficits, which affect higher-order cognitive processes required to control and coordinate behavior, also constitute a significant burden in this population. However, the extent of these EF deficits and their potential clinical correlates remain unknown. In the present study, we conducted the first in-depth investigation of EF in 3q29DS by direct and systematic evaluation of $N = 32$ patients with the deletion (mean age = 14.50 years, SD = 8.26; 63% male). We used the BRIEF as a standardized and ecologically valid measure of EF. Given previous evidence of EF deficits in idiopathic psychoses, we further tested the hypothesis that EF deficits in 3q29DS are associated with severity of florid and attenuated psychotic symptoms, rated by the gold-standard SIPS interview. Our results indicate that the mean *global executive composite* (GEC) score and the means of all BRIEF subdomain scores were elevated in 3q29DS subjects compared to the general population (T scores > 60). 81% of the subjects had clinically significant scores in at least one subdomain of EF (T Scores ≥ 70), with the *shift* subdomain having the highest mean score (T score = 68) compared to other subdomains. Multiple linear regression analyses indicated a significant and positive relationship between GEC scores and symptom severity in the positive and disorganization symptom dimensions of the SIPS ($p's < 0.05$), while covarying for age, sex and full-scale IQ. Overall, our findings confirm that executive dysfunction is a significant feature of 3q29DS that warrants attention. Our findings also indicate that while there is a global EF deficit in 3q29DS, this profile is especially marked by a relative weakness in the ability to shift flexibly between mental sets, tasks, or strategies to generate adaptive behavioral responses. Finally, our results reveal that higher levels of executive dysfunction indexed by the BRIEF is a significant predictor of florid and attenuated psychotic symptoms, beyond the variation explained by psychometric intelligence. EF deficits may represent a previously unknown intermediate cognitive endophenotype in 3q29DS that can help deconstruct the link between the 3q29 deletion and psychosis. Future work will employ performance-based measures of EF and investigate the neurobiological underpinnings of executive dysfunction via neuroimaging in this population.

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Poster

P902. Schizophrenia: Molecular Mechanisms I

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Program #/Poster #: P902.03

Topic: H.13. Schizophrenia

Title: An imbalance of IgG receptors may contribute to the increased neuroinflammation seen in the midbrain of a sub-group of schizophrenia patients

Authors: *A. PETTY[1], C. S. WEICKERT[2], L. J. GLASS[3], T. PURVES-TYSON[2], D. ROTHMOND[2];
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Abstract: There is growing evidence that neuroinflammation may contribute to schizophrenia pathology. Elevated proinflammatory cytokines are evident in the midbrain of schizophrenia subjects, and these findings are driven by a sub-group of patients, characterised as a "high inflammation" biotype. Cytokines trigger the release of antibodies, of which immunoglobulin G (IgG) is the most common. Testing whether IgG contributes to the neuroinflammatory abnormalities in this sub-group of schizophrenia patients may help direct novel treatment targets. Therefore, we measured IgG protein, IgG receptor transcripts, and IgG transporter levels in midbrain tissue from schizophrenia patients (including both high- and low-inflammation biotypes) and healthy control subjects. Fresh frozen midbrain samples were either homogenised for protein and mRNA analysis, or 14um sections were taken for immunofluorescent staining. IgG protein abundance (relative to β-actin) assessed by western blot was unchanged between groups, however preliminary immunofluorescence analysis indicated a significant difference in levels of parenchymal IgG in the substantia nigra between diagnostic/inflammatory groups, suggesting that differences may exist within brain tissue (p=0.03). In addition, mRNA levels of the pro-inflammatory IgG receptor, FcGR3A (also known as CD16) were increased in the high inflammation biotype schizophrenia subjects (p<0.0001) in comparison to low inflammation schizophrenia subjects, and healthy controls. In contrast, mRNA levels of the anti-inflammatory IgG receptor, FcGR2B (also known as CD32b) were unchanged between all diagnostic and inflammatory groups. Finally, while protein levels of the IgG transporter FcGRT were comparable between groups, mRNA levels of the transporter were elevated in the high inflammation schizophrenia subjects (p=0.02), suggesting that there may be changes in transporter turnover. Future work will quantify whether the anatomical distribution (as well as abundance) of IgG in the midbrain is altered between diagnostic and/or inflammatory groups. However, even if levels of IgG are comparable between groups, the imbalance between the pro-inflammatory FcGR3A and anti-inflammatory FcGR2B receptors suggests that "high inflammation biotype" schizophrenia patients may have an exaggerated response to IgGs. Future work will determine whether this elevation in FcGR3A is specific to a certain cell-type, thereby providing critical information regarding the nature of this neuroinflammatory abnormality in a sub-group of schizophrenia patients.

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Poster

P902. Schizophrenia: Molecular Mechanisms I

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Support: NSW Ministry of Health, Office of Health, and Medical Research
National Health and Medical Research Council (Australia) Principal Research Fellowship (PRF) #1117079

Title: Markers of inflammation define subgroups of individuals with schizophrenia and bipolar disorder in midbrain

Authors: *Y. ZHU[1], S. J. OWENS[2], K. AJULU[1], D. A. ROTHMOND[2], T. D. PURVES-TYSON[2,3], M. J. WEBSTER[4], C. S. WEICKERT[1,3,2];
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Abstract: Neuroinflammation is evident in the blood of ~40% of people living with schizophrenia and in postmortem samples from prefrontal cortex, striatum, and midbrain of ~40-50% of schizophrenia cases. Our recent study indicated that inflammation in the midbrain may play a role in dopamine dysregulation since we found 46% of schizophrenia cases in the Tissue Resource Center (TRC) cohort had elevated levels of pro-inflammatory cytokines in the midbrain. In this study, we aimed to replicate these findings in the larger Stanley Medical Research Institute (SMRI) cohort and to define a high inflammation subgroup in bipolar disorder. We investigated mRNA levels of 7 cytokine and immune transcripts in postmortem midbrain tissue (N=105, n=35 per diagnostic group) and performed two-step recursive clustering analysis to define inflammation subgroups. The final cluster predictors were TNFα, IL1β, IL6, and SERPINA3 (predictor importance >0.5). We identified 6% (2/35) as high inflammation controls (HI-CON), 46% (16/35) as high inflammation schizophrenia (HI-SCZ), and 29% (10/35) as high inflammation bipolar disorder (HI-BPD)($X^2 = 14.42$, $p<0.001$). SERPINA3 mRNA was elevated in schizophrenia compared to controls (>200%, $p<0.0001$) and bipolar disorder (>150%, $p<0.05$). However, IL6, IL1β, and TNFα mRNAs were unchanged across diagnostic groups ($p>0.2$). When stratified by inflammation subgroups, all 4 mRNAs used to identify the clusters were increased in HI-SCZ and HI-BPD compared to low inflammation controls (LI-CON) (SERPINA3 $p<0.01$; TNF $p<0.0001$; IL6 $p<0.05$; IL1B $p<0.001$). Protein levels of IL6 and IL1β were elevated in HI-SCZ compared to all low inflammation subgroups, and elevated in HI-BPD compared to LI-SCZ and LI-BPD (all $p<0.05$). In contrast to our expectations, TNFα protein was significantly decreased in HI-BPD compared to LI-CON and HI-SCZ ($p<0.05$). In conclusion, we replicated our previous findings and found the percentage of HI-SCZ in the SMRI cohort was consistent with the TRC cohort. In addition, we found the BPD cases have an intermediate level of high inflammation cases. Most inflammation mRNAs and proteins that we examined were significantly changed when stratified by inflammation subgroups. However, further studies are needed to determine why TNFα mRNA and TNFα protein are differentially expressed. These changes in cytokine levels proximal to dopamine cell bodies may contribute to the dopamine abnormalities that are found in schizophrenia.

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Poster

P902. Schizophrenia: Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P902.05

Topic: H.13. Schizophrenia

Support: NSW Health Funding for the Schizophrenia Research Lab

Title: Bdnf and trkb splice variants are altered in post-mortem midbrain in schizophrenia and exacerbated by inflammation

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Abstract: Background: Psychotic symptoms of schizophrenia likely stem from subcortical hyperdopaminergia in the nigrostriatal pathway. Elevated pro-inflammatory markers (SERPINA3, IL6, IL1β, TNFα) found in the midbrain of ~45% of schizophrenia cases may damage already hyperactive dopamine neurons. Dopamine neurons are not considered to be lost in schizophrenia, suggesting trophic factors such as Brain-Derived Neurotrophic Factor (BDNF), signaling via the full-length Tropomyosin kinase B receptor ($TrkB^{TK+}$), may promote dopamine neuron survival. However, BDNF binding to the truncated receptor ($TrkB^{TK-}$) on glial cells diverts trophic support away from neurons. BDNF transcription is regulated by nuclear receptor related-1 protein (NURR1) and its trophic effects are counteracted by its precursor form, proBDNF, binding to the apoptotic p75 receptor. We hypothesised that molecular measures of BDNF trophic support would be altered in the schizophrenia midbrain and be exacerbated in inflammatory states, reflecting the degraded health of dopamine neurons.
Methods: BDNF (exon IV), $TrkB^{TK+}$, $TrkB^{TK-}$, NURR1 and p75 mRNA levels were quantified by qPCR in post-mortem midbrain tissue (28 control/28 schizophrenia from the NSW Brain Tissue Resource Centre). Gene expression was analysed by diagnosis and by previously assigned high/low inflammation biotypes. $TrkB^{TK+}$ mRNA was visualized using *in situ* hybridization (20 control/10 schizophrenia from the Clinical Brain Disorders Branch).
Results: Gene expression of BDNF IV (31%), $TrkB^{TK+}$ (19%) and NURR1 (21.4%) was significantly reduced, while $TrkB^{TK-}$ (29%) and p75 (8.8%) mRNA levels were increased in the midbrain of schizophrenia cases compared to controls ($p<0.05$). Schizophrenia cases with a high inflammation biotype displayed the greatest degree of alterations in BDNF IV, $TrkB^{TK+}$, $TrkB^{TK-}$, NURR1 and p75 mRNA levels compared to low inflammatory controls ($p<0.05$). $TrkB^{TK+}$

mRNA was localised in midbrain neuromelanin-positive neurons in both schizophrenia and control cases. Males and females were included for both cohorts and no sex differences were observed.
Conclusion: We provide the first evidence for altered gene expression of NURR1, BDNF and its respective TrkB and p75 receptors in the schizophrenia midbrain, the magnitude of which is intensified in the presence of elevated inflammatory markers. These alterations do not reflect the hypothesized elevated trophic support to overcome damage by neuroinflammation. Instead, the lack of trophic support resulting from depleted $TrkB^{TK+}$ mRNA in midbrain dopamine neurons, may further diminish the health of these neurons, known to be dysregulated in schizophrenia.

Disclosures: J. Chandra: None. **T.D. Purves-Tyson:** None. **S.J. Owens:** None. **D.A. Rothmond:** None. **C.S. Weickert:** None.

Poster

P902. Schizophrenia: Molecular Mechanisms I

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Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P902.06

Topic: H.13. Schizophrenia

Support: NIDA U01DA043098
Office of Naval Research (ONR) 00014-19-1-2149
The Hope for Depression Research Foundation (HDRF)
The Pritzker Neuropsychiatric Research Consortium
University of Michigan Undergraduate Research Opportunities Program (UROP)
Michigan Research and Discovery Scholars (MRADS) program

Title: Neurotransmission-related gene expression in the frontal pole (Brodmann area 10) is altered in subjects with schizophrenia and bipolar disorder

Authors: *L. C. THEW FORRESTER[1], A. MEDINA[2], M. H. HAGENAUER[1], D. M. KROLEWSKI[3], M. WASELUS[4], E. HUGHES[6], M. P. VAWTER[7], D. M. WALSH[8], E. RICHARDSON[1], C. A. TURNER[5], W. E. BUNNEY[9], R. M. MYERS[10], J. D. BARCHAS[11], F. LEE[12], A. F. SCHATZBERG[13], H. AKIL[1], S. J. WATSON, Jr.[1];
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Abstract: BACKGROUND: Schizophrenia (SCZ) and Bipolar Disorder (BP) have long been associated with dysregulation in the frontal cortex. The gene expression underlying this dysregulation has been explored in detail using transcriptional profiling studies (microarray, RNA-Seq) in the dorsolateral prefrontal cortex, but the larger frontal pole region (Brodmann Area 10, BA10) has received less attention. Our study explored the association between SCZ and BP and the frontal pole using methods that provide a sensitive snapshot of neurotransmission-related gene expression. METHODS: To ensure a high concentration of neuron-derived transcripts, post-mortem dissections focused on grey matter. Only samples with agonal factor=0, pH>6.5, and RNA integrity>6.0 were included to exclude brains with prolonged hypoxia before death. Gene expression was measured using two sets of TaqMan microfluidic qPCR cards targeting transcripts related to signaling via 1) GABA and glutamate ("GABAGLU" experiment), 2) dopamine and serotonin ("DA5HT" experiment). In each experiment, the subject samples were run on at least two separate cards. After quality control (QC), the sample size was 69 subjects (control (CTRL): n=26, SCZ: n=22, BP: n=21) and included measurements from 111 targeted gene transcripts (GABAGLU: 82, DA5HT: 29). We normalized measurements using a panel of reference gene transcripts (GABAGLU: 11 genes, DA5HT: 17 genes). Differential expression (DE) was assessed via a multi-level model that included biological (gender, age, pH, post-mortem interval [PMI]) and technical (RNA concentration, integrity, card) co-variates. The results from the top DE genes (false discovery rate <0.10) were explored in relation to symptom profiles, medication, and drug and alcohol exposure. These results were compared to the results from two publicly available microarray datasets examining the influence of psychiatric diagnosis on gene expression in BA10 (GSE12654; GSE17612). We re-analyzed both datasets using a custom chip definition file with updated annotation. Our DE models controlled for large sources of noise as determined by relationships with top principal components of variation (pH, age, RNA degradation, PMI, rate of death). After QC, the sample size for GSE12654 was 50 subjects (CTRL: n=15, SCZ: n=13, BP: n=11, Depression: n=11); and for GSE17612 the sample size was 51 subjects (CTRL: n=23, SCZ: n=28). RESULTS AND CONCLUSION: Collectively, our results showed a similar pattern of DE gene expression in the frontal pole in both disorders, including genes directly involved in major neurotransmitter functions affecting the serotonergic, dopaminergic and GABAergic systems.

Disclosures: L.C. Thew Forrester: None. **A. Medina:** None. **M.H. Hagenauer:** None. **D.M. Krolewski:** None. **M. Waselus:** None. **E. Hughes:** None. **M.P. Vawter:** None. **D.M. Walsh:** None. **E. Richardson:** None. **C.A. Turner:** None. **W.E. Bunney:** None. **R.M. Myers:** None. **J.D. Barchas:** None. **F. Lee:** None. **A.F. Schatzberg:** None. **H. Akil:** None. **S.J. Watson:** None.

Poster

P902. Schizophrenia: Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P902.07

Topic: H.13. Schizophrenia

Support: Lieber Institute for Brain Development

Title: Electrophysiological measures from human IPSC-derived neurons are associated with schizophrenia clinical status and predict individual cognitive performance.

Authors: S. C. PAGE[1], S. SRIPATHY[2], F. FARINELLI[1], Z. YE[1], Y. WANG[1], D. J. HILER[1], E. A. PATTIE[1], C. V. NGUYEN[1], M. TIPPANI[1], R. L. MOSES[1], H.-Y. CHEN[1], M. N. TRAN[1,3], N. J. EAGLES[1], J. M. STOLZ[1], J. L. CATALLINI[1,9], O. R. SOUDRY[1], D. DICKINSON[10], K. F. BERMAN[11], J. A. APUD[12], D. R. WEINBERGER[1,4,5,6], K. MARTINOWICH[1,4,6], A. E. JAFFE[2,4,6,7,8], R. STRAUB[1], ***B. MAHER**[1,4,6];
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Abstract: Schizophrenia (SCZ) is a complex, polygenic disorder with marked clinical heterogeneity and no clear pathological mechanism or cellular pathology. The advent of human induced pluripotent stem cell (iPSC) technology has enabled the capture of the polygenic nature of SCZ *in vitro*. Several previous studies using iPSC to model SCZ have identified interesting phenotypes, but their relevance to the clinical illness in adults remains unclear. Here, we describe results of our investigation into the relationships between the electrophysiological properties of iPSC-derived cortical neurons and the individual's clinical status, severity of symptoms, and cognitive performance. Importantly, we look beyond case-control differences, and identify within-case patterns of association that could serve to illuminate dimensions of illness and subgroups of patients that may be suitable for targeted treatments. We studied cortical neurons from 13 SCZ patients with high polygenic risk scores (PRS) and 15 neurotypical individuals with low PRS with a battery of assays to identify cellular phenotypes, done in a double-blind manner. We correlated electrophysiology measures to the individual's rating on the Positive and Negative Syndrome Scale (PANSS), six cognitive domains (Verbal Memory, Nback, Visual Memory, Processing Speed, Card Sorting, and Digit Span), estimated IQ, and general cognitive ability composite ("g"). We observed five electrophysiological measures related to Na^+ channel function associated with diagnosis. Lines derived from SCZ donors showed an increase in membrane resistance, an increased number of Na^+ current peaks in response to a voltage ramp protocol, a decrease in the activation threshold of the second Na^+ peak, a significantly hyperpolarized shift in Na^+ channel activation and V1/2 activation voltage, and a significant hyperpolarized shift in the voltage dependence of inactivation and V1/2 inactivation voltage. In SCZ, the number of Na^+ peaks showed a directionally-consistent, positive association with some positive symptom ratings ($R=0.77$, $p=0.0033$), whereas there were no correlations with negative symptom ratings or cognitive measures. In striking contrast, sEPSC amplitude showed no correlation with positive symptoms, but showed a negative correlation ($R=-0.93$, $p=2.94E-05$)

with Wisconsin Card Sort Test performance. Our results demonstrate for the first time that common genetic variants associated with SCZ converge on electrophysiological mechanisms that relate to clinically relevant features of SCZ, and therefore underscore the potential of this approach for biomarker identification and perhaps downstream drug development.

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Poster

P902. Schizophrenia: Molecular Mechanisms I

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Program #/Poster #: P902.08

Topic: H.13. Schizophrenia

Support: R01MH106575
R01MH100915
R01AG063175

Title: Modeling the loss-of-function mutation of OTUD7A within the schizophrenia-associated 15q13.3 microdeletion in human neurons

Authors: *A. KOZLOVA[1], S. ZHANG[1], A. KOTLAR[2], J. MCDAID[1], M. FORREST[3], H. ZHANG[1], B. JAMISON[1], D. CUTLER[2], M. ZWICK[2], Z. PANG[4], A. SANDERS[1], S. WARREN[2], P. GEJMAN[1], J. MULLE[2], J. DUAN[1];
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Abstract: A number of large and rare copy number variants (CNVs) are found to confer strong risk to schizophrenia (SZ) and other neurodevelopmental disorders. However, identifying the causative gene(s) within a CNV is challenging since most CNVs encompass many genes. *Otud7a* heterozygous and homozygous knockout mice have been shown to recapitulate some phenotypes associated with the 15q13.3 microdeletion syndrome, leading to the hypothesis that *OTUD7A* is the driver of neuropsychiatric phenotypes. Here, by sequencing 1779 SZ cases and 1418 controls for SZ-associated protein-coding variants in genes encompassed by SZ-associated CNVs, we identified three different loss-of-function (LoF) mutations in the exon 5 of *OTUD7A in* 3 cases, but none in controls. We thus aimed to model the LoF of *OTUD7A* in human induced pluripotent stem cell (hiPSC)-derived induced excitatory neurons (iNs). We performed CRISPR/Cas9

editing to introduce the risk allele of a LoF mutation rs757148409 onto two control donor iPSC lines, each with three different clones, resulting in a heterozygous G to A change leading to a stop-gain mutation. By examining the isogenic iNs, we found that the introduced premature stop-gain mutation did not cause nonsense-mediated mRNA decay of *OTUD7A*, but instead resulted in ~50% decrease in protein expression. Consistent with the reduced *OTUD7A* expression, iNs carrying the heterozygous LoF mutation exhibited ~50% reduction in dendritic complexity and ~30% decrease in total dendrite length. Synaptic puncta analysis further showed a marked decrease in the puncta density and area of AMPA receptor subunit GluA1 and postsynaptic scaffold PSD-95 in iNs carrying the LoF mutation. Congruent with the observed phenotypic alterations associated with the *OTUD7A* LoF mutation, our transcriptomic analysis of the CRISPR-edited isogenic iNs showed that genes downregulated, but not upregulated, in the *OTUD7A* LoF iNs were significantly enriched for gene ontology (GO) terms relating to the synapse, synaptic vesicle cycle, and neurotransmitter release. In addition, microelectrode array analysis showed reduced neuron firing rate and network activity in iNs carrying LoF of *OTUD7A*. Taken together, these results suggest that the rs757148409 LoF mutation in *OTUD7A* impaired neurodevelopment, causing decreased dendritic complexity, synapse number, and expression of synaptic genes. Our study provides mechanistic insight into the possible role of *OTUD7A* in driving phenotypes associated with SZ and other neurodevelopmental disorders associated with the 15q13.3 microdeletion.

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Poster

P902. Schizophrenia: Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P902.09

Topic: H.13. Schizophrenia

Title: Progressive Brain Abnormalities in Schizophrenia Across Different Illness Duration: A Structural and Functional MRI Study

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Abstract: Schizophrenia has been regarded as a chronic and deteriorating brain disorder since it was named originally as dementia precox by Kraepelin in 1900s. However, when and how brain abnormalities occur and evolve in schizophrenia remain undetermined. The present study aimed

to explore progression of brain abnormalities on structural and functional MRI in patients with schizophrenia throughout the illness course by using the four analysis methods of voxel-based morphometry (VBM), fractional anisotropy (FA), global functional connectivity density (gFCD) and sample entropy (SampEn). We recruited 336 Han Chinese participants and stratified them into three age and sex-matched groups. The 5-year group (illness duration below 5 years) had 40 individuals with schizophrenia and 80 healthy controls, the 15-year group (illness duration between 12 and 18 years) had 33 individuals with schizophrenia and 66 healthy controls, and the 25-year group (illness duration over 25 years) had 39 individuals with schizophrenia and 78 healthy controls. All participants underwent resting-state scanning using a 3T MRI. T1-weighted images were preprocessed with the DARTEL algorithm for VBM analysis. FA values were calculated by fitting a tensor model to the data at each voxel obtained from diffusion tensor imaging. gFCD was defined as the number of Pearson's correlation r between BOLD signals at that voxel and those in the remaining gray matter voxels, which surpassed an arbitrary threshold ($r>0.6$). SampEn of the BOLD signal was computed for all gray matter voxels to create a whole-brain parametric map. We used T-test, Chi-square test, ANOVA and General Linear Model (GLM) to do statistical analysis. We found functional abnormalities occurs firstly in single brain areas in early course of schizophrenia, functional connectivity abnormalities and gray matter abnormalities ensue in middle course, and structural connectivity abnormalities manifesting with white matter tracts disintegration emerge at last. Our findings represent a novel approach to study progression course of brain abnormalities in schizophrenia. In conclusion, using structural and functional brain MRI analysis, we showed brain abnormalities occur in limited areas in early course of schizophrenia and progress into much widespread areas in later course.

Disclosures: C. Shen: None. **A.C. Yang:** None.

Poster

P936. Probe Design and Engineering: Sensors

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P936.01

Topic: I.04. Physiological Methods

Support: NIH BRAIN Initiative grant U01NS113358
Z18110200130000 Beijing Municipal Science & Technology Commission

Title: Improved GRAB sensors for monitoring dopamine dynamics in vitro and in vivo

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Abstract: Dopamine (DA) is a crucial monoamine neurotransmitter involved in many physiological and pathological processes, and the ability to directly monitor DA dynamics is essential for understanding its physiological function. Here by tapping into different species and dopamine receptor subtypes, we further developed a series of green fluorescent $GRAB_{DA}$ sensors (DA3m and DA3h), for improved signal-to-noise (SNR), sensitivity, selectivity and as well as diverse pharmacology properties in cultured cells. The improved $GRAB_{DA}$ sensors also exhibited superior properties in mouse acute brain slice, *in vivo* in flies and mice by using transgenic or virus-mediated expression. Specifically, the improved DA3m could stably report DA dynamics during sleep-wake cycles and selectively monitor DA changes under anesthesia condition induced by 1.5% isoflurane in the mice medial prefrontal cortex (mPFC). Apart from green DA sensors, we further optimized spectrum-compatible red dopamine sensors ($rGRAB_{DA}$) with higher brightness and larger fluorescent increase in DA detection after iterative engineering in fluorescent module and molecular scaffold based on the prototype, and also generated red DA sensors with different affinity range and pharmacology profile by introducing diverse GPCR backbone. In sum, by enabling multicolor imaging of DA and other signaling components, these $GRAB_{DA}$ sensors serve as powerful tools to study comprehensive neurochemical modulation in dopamine system.

Disclosures: Y. Zhuo: None. **H. Dong:** None. **T. Qian:** None. **Y. Wang:** None. **X. Yi:** None. **X. Xia:** None. **Y. Li:** None.

Poster

P936. Probe Design and Engineering: Sensors

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Program #/Poster #: P936.02

Topic: I.04. Physiological Methods

Support: NSFC Grant 31871087
NSFC Grant 31925017
NIH Grant U01NS103558

Title: Improved norepinephrine sensors shine lights on in vivo noradrenergic activities

Authors: *J. FENG[1], **R. CAI**[1], J. LISCHINSKY[2], J. ZHOU[3], H. DONG[1], H. WANG[1], F. DENG[1], H. XIE[4], Q. DAI[4], G. CUI[3], D. LIN[2], Y. LI[1,5,6];
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Abstract: Norepinephrine (NE) is a key biogenic monoamine neurotransmitter involved in a wide range of physiological processes. However, its precise dynamics and regulation remain poorly characterized. With the structural similarity and promiscuous patterns inside our brain for norepinephrine and dopamine, it is indispensable to tease apart these two monoamines during behaviors in vivo. Previously, we have developed prototype GPCR activation-based NE ($GRAB_{NE}$) sensors with either high sensitivity or high selectivity to NE. Here, we further optimized and characterized improved $GRAB_{NE}$ sensors (NE2m and NE2h) with higher responses. They exhibited both higher sensitivity and selectivity to NE compared with prototypes. Viral-expression of improved $GRAB_{NE}$ sensors was able to detect optogenetically and behaviorally triggered NE release in the locus coeruleus (LC) and hypothalamus of freely moving mice. Specifically, dual color fiber photometry recording of a transgenic mouse expressed both $GRAB_{NE2m}$ and jRGECO1a reveals NE and calcium dynamics during sleep and wake. Moreover, dual color 2-photon and large-scale imaging of $GRAB_{NE2m}$ and jRGECO1a reveals spatiotemporally dynamics of noradrenergic and calcium activity in awake mice. Additionally, we further optimized NE sensors by introducing backbone GPCR from Arabian Camel and screened a candidate with up to 10-fold ΔF/F0 and over 1000-fold selectivity to NE vs. DA in cultured cells. We also expanded the spectrum of the sensor by generating a prototype red NE sensor that shows 2-3 folds ΔF/F0 in cultured cells with good sensitivity and specificity, which offers the potential for further simultaneously multicolor imaging of NE with other biosensors. In sum, the improved $GRAB_{NE}$ family provides robust tools for spatiotemporally monitoring in vivo NE dynamics with high sensitivity and specificity in both physiological and pathological processes.

Disclosures: J. Feng: None. **R. Cai:** None. **J. Lischinsky:** None. **J. Zhou:** None. **H. Dong:** None. **H. Wang:** None. **F. Deng:** None. **H. Xie:** None. **Q. Dai:** None. **G. Cui:** None. **D. Lin:** None. **Y. Li:** None.

Poster

P936. Probe Design and Engineering: Sensors

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P936.03

Topic: I.04. Physiological Methods

Title: Local 5-HT signaling bi-directionally modulates the coincidence time window of associative learning

Authors: *X. LI[1,2], J. ZENG[1,2,3], Z. ZHANGREN[1,2], Y. LI[1,2,3];
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Abstract: In Pavlovian associative learning, the temporal coincidence between a conditioned stimulus (CS) and an unconditioned stimulus (US) is an essential component for memory formation. The coincident time window of associative learning has been observed in a variety of behavioral paradigms in a wide range of species, including the eye-blinking task in humans, the gill-withdraw reflex in Aplysia, and the olfactory learning in Drosophila. Despite the importance of this component, it remains unclear how does the coincidence time window is regulated in a molecular or circuit-based way. To address this question, we explored the role of the serotonergic dorsal paired (DPM) neuron by a sensitive 5-HT sensor, $GRAB_{5\text{-}HT1.0}$. We found the DPM neuron releases 5-HT at the mushroom body (MB) in a compartmentalized pattern in response to both an odorant (the CS) and electric shock (the US). This 5-HT signal is induced by local acetylcholine (ACh) release from MB intrinsic Kenyon cells (KCs) via nicotinic ACh receptors. Relatively, the KCs receive feedback inhibition from DPM via the inhibitory 5-HT1a receptor. By manipulating the 5-HT signals or the DPM activity with genetic or pharmacological methods, we found suppressing or promoting 5-HT release from DPM neuron shortens or prolongs the coincidence time window of the odor-shock pairing–induced change in synaptic plasticity and the resulting olfactory learning behavior. These results revealed the serotonergic feedback circuit in MB regulates the length of coincident time window, which provides a model for studying how an organism times the temporal continuity of environmental events and learns the underlying causal relationship.

Disclosures: X. Li: None. **J. Zeng:** None. **Z. Zhangren:** None. **Y. Li:** None.

Poster

P936. Probe Design and Engineering: Sensors

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Program #/Poster #: P936.04

Topic: I.04. Physiological Methods

Support: the State Key Laboratory of Membrane Biology at Peking University School of Life Sciences
the Peking-Tsinghua Center for Life Sciences
NIH BRAIN Initiative NS103558

Title: A series of genetically encoded GRAB sensors for detecting serotonin dynamics in vitro and in vivo

Authors: *F. DENG[1,2], J. WAN[1,2], G. LI[1,2], H. DONG[1,2], X. XIA[1,2,3], T. QIAN[1,2], Y. LI[1,2,3]; [1]Sch. of Life Sci., [2]PKU-IDG/McGovern Inst. for Brain Res., [3]Peking-Tsinghua Ctr. for Life Sciences, Acad. for Advanced Interdisciplinary Studies, Peking Univ., Beijing, China

Abstract: Serotonin (5-HT), a phylogenetically conserved monoamine neurotransmitter, is associated with a plethora of important physiological functions, such as sleep-wake cycles, appetite control, mood regulation, as well as learning and memory. Malfunction of the serotonergic system also involves in addiction, anxiety, depression and other neurological diseases. In order to understand the physiological and pathological roles of the serotonergic system under different conditions, it is essential to decipher the precise dynamics of 5-HT with high sensitivity, high spatiotemporal resolution and good cell specificity. Based on the GPCR Activation Based (GRAB) strategy, we systematically developed a series of genetically-encoded 5-HT sensors named GRAB$_{5\text{-HT}}$, including novel red 5-HT sensors and improved green 5-HT sensors, by inserting cpmApple or cpEGFP into the third intracellular loop (ICL3) of different 5-HT receptors. The HTR4 based sensors showed the largest response to 5-HT and were further improved by semi-rational engineering of GPCRs and cpmApple or cpEGFP. After characterization of the performance of these newly developed sensors in cultured cells and acute brain slices, we further demonstrated that these sensors could reliably detect the extracellular 5-HT *in vivo* in different organisms, including *Drosophila* and mice. Thus, these newly developed 5-HT sensors will facilitate our understanding of the serotonergic system both in health and in diseases.

Disclosures: F. Deng: None. **J. Wan:** None. **G. Li:** None. **H. Dong:** None. **X. Xia:** None. **T. Qian:** None. **Y. Li:** None.

Poster

P936. Probe Design and Engineering: Sensors

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P936.05

Topic: I.04. Physiological Methods

Support: NIH Grant 1UF1NS107713-01

Title: A DNA-based nano MRI contrast agent for detecting acetylcholine

Authors: *H. SEO[1], **K. MA**[1], E. FRIEDENTHAL[1], C. A. FLASK[2], H. A. CLARK[1]; [1]Northeastern Univ., Boston, MA; [2]Case Western Reserve Univ., Cleveland, OH

Abstract: Acetylcholine is a neurotransmitter that plays a key role in motivation, arousal, attention, learning, and memory in the brain. Since its discovery, numerous efforts have been poured into understanding the regulatory mechanism of acetylcholine towards brain function and how dysregulation can lead to brain disorders such as Alzheimer's disease, Parkinson's disease, and schizophrenia. However, elucidating the exact mechanism and function of acetylcholine-mediated signaling processes remains challenging. The solution to understanding acetylcholine could be solved with non-invasive tools with high spatial and temporal resolution for use *in vivo*.

Magnetic Resonance Imaging (MRI) is a radiation-free, non-invasive imaging modality that offers good spatial and temporal resolution, making this modality popular in neuroscience research and clinical work. For example, fMRI has been widely used to study brain activity in humans in real-time. Direct visualization of the molecular target in the brain, however, is limited on MRI without having a target-specific contrast agent. Gadolinium-based contrast agents (GBCA) have been used around 30% of clinical scans to improve the accuracy of diagnosis by enhancing the contrast at the accumulated area. To give the GBCA a target-specificity, developing bioresponsive contrast agents have gained much attention to enable modulation of MR contrast in the presence of molecular targets on MRI. In our previous study, we reported nanoparticle-based MRI contrast agents for *in vivo* detection of acetylcholine in rat brain. Immobilized acetylcholinesterase selectively converts acetylcholine to choline and acetic acid, decreasing the local pH and thus modulating the signal of co-immobilized pH-sensitive GBCAs. To further improve the homogeneity in size and functionalization of the nano-sized contrast agent, here we report a DNA nanostructure-based MRI contrast agent for monitoring acetylcholine. DNA nanostructure was programmed to have a dendrimer shape with ten peripheral functional sites. We conjugated pH-responsive contrast agents and acetylcholinesterase on the DNA scaffold and characterized the functionalization and structure by MALDI-TOF and gel electrophoresis. The synthesized DNA-based contrast agent induced brighter contrast on the T_1-weighted image in the presence of acetylcholine. The application will be further demonstrated in *in vivo* animal experiments. This research would benefit developing new tools to visualize acetylcholine to better understand acetylcholine-related signaling processes in the brain.

Disclosures: H. Seo: None. **K. Ma:** None. **E. Friedenthal:** None. **C.A. Flask:** None. **H.A. Clark:** None.

Poster

P936. Probe Design and Engineering: Sensors

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Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P936.06

Topic: I.04. Physiological Methods

Support: NIH BRAIN Initiative grant U01NS113358

Title: A family of sensitive and specific genetically-encoded GRAB sensors for measuring neuropeptides dynamics

Authors: *H. WANG[1], T. QIAN[1], C. WU[1], P. CHEN[2], L. GENG[1], J.-N. ZHOU[2], D. LIN[3], Y. DAN[4], Y. LI[1,5,6];
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Berkeley Dept. of Mol. and Cell Biol., Berkeley, CA; [5]PKU-IDG/McGovern Inst. for Brain Res., Beijing, China; [6]PKU-THU Ctr. for Life Sciences, Acad. for Advanced Interdisciplinary Studies, Peking Univ., Beijing, China

Abstract: Neuropeptides are key signaling molecules in endocrine and nervous system, regulating virtually all physiological processes, ranging from food ingestion, sleep & circadian rhythm, to pain sensation and social behaviors. To understand their important functions in vivo, it is critical to be able to monitor neuropeptide dynamics with selectivity, sensitivity and good spatiotemporal resolution. One major hindrance to the above goal is the lack of direct, sensitive and non-invasive probes to report these bioactive peptides *in vivo.* Here, by tapping into GPCRs for neuropeptides, we developed a toolbox of GRAB (GPCR activation based) sensors for neuropeptide Y (NPY), somatostatin (SST), corticotrophin releasing factor (CRF), neurotensin (NT), cholecystokinin (CCK), vasoactive intestinal peptide (VIP), substance P, vasopressin (AVP) and oxytocin (OXT). These fluorescent sensors utilize native GPCR ligand binding pockets as neuropeptide sensing modules and circular-permutated GFPs as optical reporters. We have developed sensors with ultra-good selectivity for neuropeptides, with nano molecular affinity, up-to 1500% dF/F fluorescence increase upon cognate ligand application. In addition, these GRAB sensors have minimal downstream G protein coupling, or arrestin coupling, compared to their original GPCR scaffolds. The neuropeptide GRAB sensors are able to reveal endogenous CRF and oxytocin release in acute brain slices and capture the CCK, OXT and AVP dynamics in behaving mice. These sensors are capable of detecting the relevant peptides with microendoscopic imaging across sleep wake cycles with good temporal resolution. Further development of these sensors and their deployment will likely significantly enhance our understanding of neuropeptide release mechanisms, neuropeptide functions and regulation in physiological and pathophysiological conditions.

Disclosures: H. Wang: None. **T. Qian:** None. **C. Wu:** None. **P. Chen:** None. **L. Geng:** None. **J. Zhou:** None. **D. Lin:** None. **Y. Dan:** None. **Y. Li:** None.

Poster

P936. Probe Design and Engineering: Sensors

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P936.07

Topic: I.04. Physiological Methods

Title: A Fluorescent Sensor for Real-Time Measurement of Extracellular Oxytocin Dynamics in the Brain

Authors: *D. INO, H. HIBINO;
Osaka Univ., Suita, Japan

Abstract: Oxytocin (OT), a neuropeptide initially discovered as an uterus-contracting hormone in the early 20th century, has been attracted a lot of attentions as a key regulator of many animal behaviors. In addition to the well-characterized roles as a peripheral hormone to promote childbirth and lactation, OT released as a neuromodulator within the brain participates in the regulation of diverse physiological functions from sensory processing, feeding control, social cognition, and emotion. To exert such central effects, OT primarily supplied from neurons in paraventricular nucleus and supraoptic nucleus of hypothalamus acts on the target brain regions such as limbic regions, sensory cortex, and brainstem, where a G-protein coupled receptor (GPCR) named as OT receptor (OTR) is highly expressed. OT release in the brain is evoked by external inputs such as sensory cues, food intake, and social rewards, and can also be controlled in an autoregulatory manner potentially by the signaling associated with biological clocks. Reportedly, the impairment of OT signaling in the brain may underlie the cognitive and emotional dysfunctions associated with psychiatric disorders (e.g. autism, schizophrenia) and brain aging. Despite its importance in health and disease, the relation between the animal behaviors and the spatio-temporal distribution of OT in the brain still remains largely elusive. Here, we describe MTRIA$_{OT}$, a GPCR-based green-fluorescent sensor that enables us to conduct real-time measurement of brain OT dynamics in living animals. MTRIA$_{OT}$ exhibit a large dynamic range, an optimal affinity, ligand specificity to OT orthologues, minimal effects on the downstream signaling, and long-term fluorescence stability in vitro. By combining viral gene delivery and fiber photometry-mediated fluorescence measurement, we demonstrate the utilities of MTRIA$_{OT}$ to track the OT level in the living brain. Moreover, we also demonstrate the potential of the fluorescent module of MTRIA$_{OT}$ for development of a variety of GPCR-based fluorescent sensors. MTRIA$_{OT}$ will be a powerful new tool to understand the OT functions in the brain.

Disclosures: D. Ino: None. **H. Hibino:** None.

Poster

P936. Probe Design and Engineering: Sensors

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Program #/Poster #: P936.08

Topic: I.04. Physiological Methods

Support: NIH 1R01GM139850-01
NIH 1R21DA051193-01A1
NIH P30DA048736-Pilot

Title: A highly sensitive mu-opioid peptide sensor engineered with a massively parallel GPCR optogenetic tool building platform.

Authors: *M. RAPPLEYE, S. J. WAIT, J. C. SIEBART, J. D. LEE, A. A. NGUYEN, N. SMITH, D. C. CASTRO, A. SUKO, J. D. DEEM, M. R. BRUCHAS, A. BERNDT;
Univ. of Washington, Seattle, WA

Abstract: The mu opioid receptor (MOR) plays a critical role for regulating neuronal networks involved in analgesia, addiction, reward behavior, and more. However, we currently lack a tool capable to investigate MOR activation with high spatio-temporal precision in vivo. Here, we present a protein-based mu opioid sensor engineered from a high throughput, optogenetic engineering platform. We constructed a genetic library with >1 million potential variants by randomizing DNA codons in key structural elements of the sensor. We express the library in a genetically engineered HEK 293T cell line that ensures each cell expresses a single sensor variant. In the next critical step, we then isolated the cells on our customized polydimethylsiloxane microwell array for subsequent sensor stimulation by the synthetic opioid peptide DAMGO and signal detection by fluorescent microscopy. Our transfection, expression, and screening techniques are streamlined and highly efficient, enabling us to screen hundreds of variants in parallel in a single field of view and tens of thousands of cells/sensor variants per day. Immediately upon signal detection on PDMS arrays, we quantify and rank each construct's performance with a custom, real-time analysis. Specifically, for the development of the mu-opioid sensor, we have screened >40,000 cells/variants, by far exceeding current throughput in sensor development. After recovering the gene, we validated the sensor's signal, biophysical characteristics (retaining MOR specificity, agonist, and antagonist ligand profiles) within mammalian cell culture populations. Importantly, we confirmed that the sensor does not internalize upon stimulation and does not induce intracellular signaling cascades. Furthermore, we confirmed that the sensor maintains the MOR's native hierarchy of affinities towards endogenous peptides (measured using epifluorescence microscopy in triplicate experiments for each replicate). After several rounds of unbiased high-throughput optimization, our platform produced an MOR-based sensor, able to detect endogenous neuropeptides in the nM range. Additionally, we confirmed these sensor features are maintained in cultured primary rat neurons. In summary, we demonstrated the ability to rapidly engineer and optimize neuropeptide sensors, showcasing our platforms abilities to develop highly specific and sensitive sensors through the directed evolution of a mu opioid sensor. Our ability to screen and identify variants specialized to detect low levels of agonist opens the field to the rapid creation of effective sensors for low concentration neuropeptide ligands.

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Poster

P936. Probe Design and Engineering: Sensors

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Program #/Poster #: P936.09

Topic: I.04. Physiological Methods

Support: Grant-in-aid for Scientific Research from MEXT Japan
Doshisha Harris Grant

Title: Monomeric green fluorescent probe gCarvi is optimal in cyclic adenosine monophosphate imaging of live neuron

Authors: S. KAWATA, *N. SAITOH;
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Abstract: The second messenger cyclic adenosine monophosphate (cAMP) plays regulatory roles in various neuronal functions. Although molecular mechanisms underlying cAMP-mediated signaling pathways are well clarified, spatio-temporal dynamics of intracellular cAMP remain largely unknown. To address this issue, we have developed a new monomeric green fluorescent cAMP indicator named “gCarvi”. gCarvi is composed of cAMP binding domain of E coli cAMP receptor protein, conjugated with circular permutated GFP. This probe detects cAMP within 0.2-20 μM with a Kd of 2 μM and has a 2.5-fold dynamic range. This probe rapidly detects cAMP with a Kon of $1.13 \pm 0.19 \times 10^6$ mol^{-1} s^{-1} and Koff of 2.19 ± 0.16 s^{-1}. Unlike previous cAMP probes, this cAMP probe has more than 100-fold specificity in cAMP detection over cGMP or other nucleotides. Since presynaptic cAMP is thought to be involved in synaptic plasticity at cerebellar granule cell - Purkinje synapses, we asked whether presynaptic elevation of cAMP might affect the efficacy of vesicle cycling. rAAV-gCarvi expressed granule cells (ICR mice, 15 DIV) were stimulated by depolarization with a high K^+ solution in the presence (load) of FM 4-64 dye, and were simultaneously applied with 10 μM forskolin and 50 μM ibudilast to stimulate cAMP production and inhibit cAMP degradation, respectively. The presynaptic cAMP elevation of the granule cell boutons showed a positive correlation to the magnitude of FM 4-46 changes ($r = 0.84$, $P < 0.001$). These results suggest that intra-bouton cAMP elevation can increase the recycling synaptic vesicle size in individual boutons. Next, gCarvi and jRCaMP1b (a red fluorescent Ca^{2+} probe) were co-expressed in hippocampal neuron (ICR mice, 17 DIV). After confirming fluorescent cAMP and Ca^{2+} signals in hippocampal neurons, we bath-applied 10 μM forskolin and 100 μM IBMX (a phosphodiesterase inhibitor), which caused a gradual increase in the somatic gCarvi fluorescence and reached a plateau within 2 min. Dual imaging of cAMP and Ca^{2+} revealed that forskolin-induced cAMP increase caused increases in Ca^{2+} signals in hippocampal neurons. We utilized this probe to measure basal [cAMP]i in hippocampal culture neurons. The mCherry fused ratiometric gCarvi expressed neurons were permeabilized using 30 μM escin and exposed to cAMP at various concentrations (1-100 μM). The basal [cAMP]i estimated from the titration curve was 1.25 ± 0.20 μM. This concentration is slightly below the level required for PKA activation. The Kd of gCarvi for cAMP is just above the basal concentration, indicating its suitably for imaging dynamic changes of neuronal [cAMP]i.

Disclosures: S. Kawata: None. **N. Saitoh:** None.

Poster

P936. Probe Design and Engineering: Sensors

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Program #/Poster #: P936.10

Topic: I.04. Physiological Methods

Support: NIH 1R01GM139850-01
NIH 1R21DA051193-01A1
NIH P30DA048736-Pilot
NIH UW ISCRM Pilot Award
NIH UW Royalty Research Fund

Title: Massively parallel, high-throughput engineering of optogenetic biosensors for neuronal signaling.

Authors: M. RAPPLEYE, J. LEE, S. WAIT, J. SIEBART, A. NGUYEN, D. CASTRO, N. A. RICHARDSON, N. SMITH, P. P. GUNDAPANENI, A. SUKO, C. CHAVKIN, J. D. DEEM, M. BRUCHAS, ***A. BERNDT**;
Univ. of Washington, Seattle, WA

Abstract: The use of genetically encoded sensors, combined with in vivo microscopy and fiber photometry, has revolutionized the access to and dissection of neuronal circuits that control every aspect of information processing in the brain. The complexity and the sheer number of biochemical signaling pathways in the brain calls for an equally precise array of sensor technology. However, the development of novel sensor proteins is significantly slowed down by the low throughput of current validation methods that test engineered optogenetic sensors sequentially and one-by-one. Consequently, only a few hundred new variants can be tested within reasonable timeframes risking to miss superior sensors that match the sensitivity and specificity necessary to detect neuronal signals in vivo optically. Therefore, we developed a novel high-throughput engineering platform that significantly accelerates the screening process. We can generate sensor libraries with up to several million variants and functionally test a large number of them (>40,000) in a novel parallel approach. Here, individual sensor variants are expressed in individual mammalian host cells (HEK293), and hundreds of them are simultaneously stimulated on Polydimethylsiloxane (PDMS) arrays. The resulting change in signal (dF/F_0) is tracked using fluorescent microscopy and analyzed in real-time. Furthermore, our unbiased approach provides a high degree of freedom as we can screen the protein libraries at low ligand concentrations and use varying ligands, which is especially useful for neuropeptide sensors that are activated by several different agonists. As a result, more extensive sensor protein libraries can be screened in shorter timescales under relevant physiological conditions. Importantly, we confirmed the high accuracy and reproducibility of our platform. For example, the coefficient of variation of the fluorescent output during the screen is below 10% of the total signal (dF/F_0). Furthermore, the selected sensor variants produce equivalent signals in

mammalian cell culture populations. We will showcase the development of improved sensors for monoamine neurotransmitters, neuropeptides, and intracellular signaling pathways. The sensor ligand sensitivities and specificities have been validated in mammalian cell populations in replicates of at least three independent double-blinded experiments, each totaling >100 individual cells. In summary, our novel engineering platform can significantly accelerate sensor development at a lower cost which enables us to adopt them from the whiteboard to neuroscience applications quickly.

Disclosures: M. Rappleye: None. **J. Lee:** None. **S. Wait:** None. **J. Siebart:** None. **A. Nguyen:** None. **D. Castro:** None. **N.A. Richardson:** None. **N. Smith:** None. **P.P. Gundapaneni:** None. **A. Suko:** None. **C. Chavkin:** None. **J.D. Deem:** None. **M. Bruchas:** None. **A. Berndt:** None.

Poster

P944. Drug Delivery and Formulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P944.01

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: Vascular Dementia Research Fondation
Synergy Excelenece Cluster Munich (SyNergy)
Solorz-Zak Research Foundation
European Research Council (ERC), grant agreement No.865323

Title: Single-cell Precision Detection of DNA Nanotechnology in Whole Body

Authors: *M. MOLBAY[1,2], B. KICK[3], S. ZHAO[1,2], M. I. TODOROV[1,2], T.-L. OHN[1], S. ROTH[2], I. KHALIN[2], H. S. BHATIA[1,2], A. LIESZ[2], N. PLESNILA[2], F. HELLAL[1,2], H. DIETZ[3], A. ERTURK[1,2];
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Abstract: Nanomedicine have emerged as a promising and innovative approach for targeted drug delivery and treatment of numerous diseases including CNS related diseases. Among them DNA nanotechnology assembles versatile, biocompatible, and programmable structures enabling selective accumulation of drug molecules in targeted sites. However, despite the importance of the investment in the development of these technologies, most are on hold at the *in vivo* evaluation stage. Lack of tools enabling the investigation of cellular level biodistribution hinders the fine tracking of these structures in the whole body. Recent developments in tissue clearing and light sheet microscopy have made it possible to investigate large volumes at cellular resolution. Here, we took advantage of our newly designed whole-mouse body clearing and

imaging technologies (vDISCO) to investigate the cellular level distribution of DNA origami. Towards this goal, we generated DNA origami rods (8nmx80nm) carrying fluorescent dyes. The production efficiency and stability were assessed *in vitro* prior *in vivo* evaluation of the immune compatibility, half-life, targeting efficiency and bio-distribution. DNA origami administered intravenously (iv) or per gavage inCD1 mice (n=5). The DNA origami treatment did not alter immune cell counts up to 7 days post-injection demonstrating the immune safety of the treatment. Brain surface imaging using intravital 2-photon microscopy enabled to show clearance of the DNA origami from the meningeal blood circulation less than 20 min after injection. We did not see leakage of DNA origami into the brain parenchyma. In the periphery, the DNA Origami was gradually cleared from the liver until 24h post-injection. We then compared the biodistribution and targeting potential of iv injected DNA origami coupled with antibodies. After 20 minutes circulation, the intact whole mouse bodies were subjected to vDISCO clearing and imaged using light sheet microscopy. DNA origami coupled with CX3CR1 antibody, specific to immune cells was detected in lymph nodes, spleen as well as in kupffer cells in liver. We also coupled the DNA origami with human carbonic anhydrase (CA) XII-specific antibody (6A10), a cancer therapy antibody. We analyzed the targeting efficiency in mice that injected with MDA-MB-231metastatic cancer cells. While the DNA origami coupled with CX3CR1 preferentially targeted immune organs the 6A10 coupling directed the DNA origami to the lung were it co-localized with metastases. So far, our approach enables the assessment of biodistribution in the intact body with sensitivity down to the single-cell level, revealing DNA origami's feasibility for drug targeting.

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Poster

P944. Drug Delivery and Formulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P944.02

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: Basic Science Research Program through the National Research Foundation of Korea(NRF) funded by the Ministry of Education(2020R1A6A1A03047902). Global R&D Activities through the Korea Center for Women In Science, Engineering and Technology(WISET) funded by Ministry of Science and ICT

Title: Feasibility study of magnetic induced intranasal dug delivery

Authors: *E. YE[1], E. PARK[2], S.-M. PARK[1];
[1]Pohang Univ. of Sci. and Technol., pohang, Korea, Republic of; [2]Med. Device Resear Ctr., Samsung Med. Ctr., Seoul, Korea, Republic of

Abstract: Drug delivery to the brain tissue is key for the treatment of multiple central nervous diseases. However, drug therapies for central nervous diseases has the lower success rates compared to other therapeutic fields due to the poor transport across the blood-brain barrier (BBB). Therefore, one of primary needs for brain drug delivery is overcoming the BBB, which prevents entry into the brain of most drugs. Intranasal route has emerged for brain drug delivery with a benefit of circumventing the BBB, via neural pathways in the olfactory region. However, when drugs administrated intranasally, extremely low amount of drugs can reach the olfactory region, because it located the upper and posterior region of the nose. Hence, strategies to improve intranasal drug delivery are critically needed. In this study, we proposed a new delivery approach based on the hypothesis that magnetic nanoparticles (MNPs) can be guide in a desired direction such as olfactory region by applying a proper magnetic field. We demonstrated the feasibility of our concept using the *in vivo* experiments via rat models. To deliver intranasally administrated MNPs to brain, we designed the magnetic field device that can generate strong static magnetic fields (<175 mT) in the superior direction. During injecting fluorescence MNPs, the rat's head was fixed inside our designed device and exposed to superior magnetic field. After the magnetic exposure, the brain was extracted and fluorescently imaged to identify brain delivery (n= 3~5). For finding appropriate delivery conditions, we tested various magnetic field exposure times (90~150 min). Our results showed that our proposed strategy enhance the delivery efficiency of MNPs from nose to brain by the magnetic force. Although there were slight differences in the results depending on the magnetic exposure time, brain delivery was markedly increased compared to the sham control. From these results, we confirmed the feasibility of our proposed magnetic induced drug delivery concept. In the next phase of the study, we intend to conduct a parameter study to establish the conditions for drug delivery by the intranasal pathway using drugs containing MNPs.

Disclosures: E. Ye: None. **E. Park:** None. **S. Park:** None.

Poster

P944. Drug Delivery and Formulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P944.03

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: Feedback control on the pituitary-thyroid hormone axis by thyroid hormone agonists correlates with peripheral exposure as opposed to central exposure and on-target activity

Authors: D. A. MACKENNA[1], J. M. BACCEI[1], *M. B. WOERNER[1], A. ALAVI[1], M. W. AKHTAR[1], T. S. SCANLAN[2], B. A. STEARNS[1], J. R. HARRIS[1];
[1]Autobahn Therapeut., SAN DIEGO, CA; [2]Oregon Hlth. & Sci. Univ., Portland, OR

Abstract: Thyromimetic small molecule agonists are known to perturb the thyroid hormone axis (THA) at pharmacologically relevant doses. Established biology suggests that this may occur centrally in the hypothalamus, through thyrotropin releasing hormone in the periventricular nucleus, or peripherally in the pituitary, through thyroid stimulating hormone, or both centrally and peripherally to varying degrees.
We are developing brain-directed thyromimetic prodrugs that are activated by fatty acid amide hydrolase (FAAH). FAAH is ubiquitously expressed but is enhanced in the CNS and gastrointestinal tissues. The objective of these studies was to utilize our novel delivery technology to elucidate mechanisms by which thyromimetics disrupt the THA. Is it possible to separate on-target activity in the brain from that on the THA? Studies were performed using recombinant FAAH, tissue-derived S9 fractions, in vivo tissue distribution (TD), gene expression in brain and liver and effects on T4 in mice as a marker of THA disruption.
ABX prodrugs enhance delivery of active metabolite to the brain by >30x with brain-to-plasma ratios >1. T3-regulated gene expression in the brain increased at doses 30-fold lower than that of the metabolite with little difference in potency in liver and heart. Both prodrug and active metabolite reduce T4 levels after 7 days of treatment. Effects on THA and liver gene expression parallel plasma distribution more closely than exposures in the CNS suggesting this may be primarily a pituitary-driven effect.
We characterized both centrally-penetrant and peripherally-restricted FAAH inhibitors (CFI & PFI, respectively). PFIs have a >30-fold dose window in mice for inhibiting FAAH peripherally vs. centrally, while CFIs inhibit FAAH globally. Using the CFI, potency shifted by ~35x for all endpoints including TD, gene expression in brain and liver and THA. In contrast, PFI administration reduced potency of prodrugs on activation of T3-regulated genes in the liver and heart by >10x, without affecting activity or exposure in the brain. PFIs also decreased potency on the THA, consistent with negative feedback based on circulating peripheral metabolite rather than brain exposure. Thus, use of a PFI allowed separation of on-target brain effects from those on THA.
Negative regulation of T4 by thyromimetics does not appear to be predominantly centrally-mediated because the effects on THA and liver gene expression parallel plasma distribution more closely than exposure or activity in the CNS. As such, the combination of a thyromimetic prodrug and a PFI may further enhance delivery of our thyromimetics to the brain and maximize centrally-targeted distribution.

Disclosures: D.A. MacKenna: A. Employment/Salary (full or part-time):; Autobahn Therapeutics. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Autobahn Therapeutics. **J.M. Baccei:** A. Employment/Salary (full or part-time):; Autobahn Therapeutics. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Autobahn Therapeutics. **M.B. Woerner:** A. Employment/Salary (full or part-time):; Autobahn Therapeutics. E. Ownership Interest (stock, stock options, royalty, receipt of

intellectual property rights/patent holder, excluding diversified mutual funds); Autobahn Therapeutics. **A. Alavi:** A. Employment/Salary (full or part-time):; Autobahn Therapeutics. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Autobahn Therapeutics. **M.W. Akhtar:** A. Employment/Salary (full or part-time):; Autobahn Therapeutics. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Autobahn Therapeutics. **T.S. Scanlan:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Autobahn Therapeutics. F. Consulting Fees (e.g., advisory boards); Autobahn Therapeutics. **B.A. Stearns:** A. Employment/Salary (full or part-time):; Autobahn Therapeutics. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Autobahn Therapeutics. **J.R. Harris:** A. Employment/Salary (full or part-time):; Autobahn Therapeutics. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Autobahn Therapeutics.

Poster

P944. Drug Delivery and Formulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P944.04

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: Microglia Depletion using Unformulated PLX5622 in Rodent Diet

Authors: *B. L. WEST[1,2], A. MARIMUTHU[2], S. YEUNG[3], M. PELLIZZON[3], K. N. GREEN[4]; [1]CIMA, Newark, CA; [2]Cytoscient, Newark, CA; [3]Res. Diets, Inc, New Brunswick, NJ; [4]Neurobio. and Behavior, Univ. of California, Irvine, Irvine, CA

Abstract: Microglia survival in the adult mouse depends on signaling through the CSF-1 receptor (CSF-1R). Pharmacological inhibition of the CSF-1R causes microglial death, and this method can be used to investigate microglial functions in both health and disease. PLX5622 is a potent and selective inhibitor of the CSF-1R that can be used to lower microglial number. The extent of microglial depletion depends on the concentration of PLX5622 reached in the test animal. Like most drugs with very low solubility, specialized formulation strategies are required for PLX5622 to reach sufficient concentrations in circulation. Mixing of high concentrations of PLX5622 into the AIN-76A diet has successfully yielded microglial depletion at >90% levels. To date, most or all of the successful examples use the PLX5622 drug product which was professionally pre-formulated before incorporation into the AIN-76A diet, raising the question of the necessity of the pre-formulation, and whether a more streamlined production

method is possible.
In the current studies, neat PLX5622 purchased from a chemical vendor and without any pre-formulation was incorporated into the AIN-76A purified rodent diet at a 1200 ppm drug load. This diet was fed ad libitum for 14 days to a cohort of 3 male C57Bl/6N 8-week old mice, as was a drug-free AIN-76A diet fed to 3 control mice. On the morning of day 15, Na2-EDTA plasma samples were taken, diets removed, mice sacrificed, and brains removed for fixation in 4% paraformaldehyde. After 48 hrs fixation brains were transferred to 30% sucrose for histology. Brain sections were imaged for microglia using anti-Iba1 immunostaining, and microglia quantified in whole brain slices via the spots module in Bitplane Imaris 9. In PLX5622-treated mice, microglia were reduced by 68% (2002+/-615 spots drug diet vs 6303+/-306 spots control, difference 4301 +/- 1171). As a correlate, the concentration of PLX5622 extracted from the plasma in 75% acetonitrile was quantified using LC-ms/ms by comparison with a PLX5622 standard curve prepared in control mouse plasma. The mean calculated PLX5622 plasma concentration was 1619+/-26 ng/mL.
Although the reduction of microglia did reach 68%, this level is inadequate for studies aiming to infer a role for microglia in their absence. Even while using the strategy of ad libitum feeding of drug-loaded diet, some additional formulation is required. New methods easily accessible to life science investigators at low cost are now being investigated.

Disclosures: **B.L. West:** A. Employment/Salary (full or part-time):; Cytoscient. **A. Marimuthu:** A. Employment/Salary (full or part-time):; Cytoscient. **S. Yeung:** A. Employment/Salary (full or part-time):; Research Diets, Inc. **M. Pellizzon:** A. Employment/Salary (full or part-time):; Research Diets, Inc. **K.N. Green:** A. Employment/Salary (full or part-time):; University of California, Irvine. F. Consulting Fees (e.g., advisory boards); Ashvatta Therapeutics.

Poster

P944. Drug Delivery and Formulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P944.05

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: CSIR-SRF; Grant Number: 09/140(0176)/2019-EMR-I.
IBRO International Travel Grant

Title: Supersaturable lipid based nanoformulation of atazanavir: a strategy towards eradication of viral sanctuaries in the Brain.

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Abstract: Central nervous system (CNS) serves as an immunological privileged sanctuary site for HIV-1 virus that is capable of reactivating the infection.The injury resulting from HIV infection to the brain may leads to milder form of cognitive impairment and dementia in severe cases. Commercially available anti-HIV drugs, although effective in minimizing the viral load, cannot eradicate the virus significantly from the body. The failure of many highly active antiretroviral therapy (HAART) agents to block the accumulation of viral reservoirs is due to the low penetration across the blood-brain barrier (BBB). Therefore, formulating a novel approach foraugmenting the delivery of anti-HIV drugs are required for the treatment of neuro-AIDS. The current investigation entails the systematic development of Atazanavir Sulfate (ATZ) loaded supersaturable lipid formulation prepared with long chain triglycerides for enhanced oral bioavailability and CNS targeting. For this purpose, the optimized supersaturable lipidic formulation (F1) consists of Corn Oil: Oleic acid (1:1) (10% w/w), Propylene Glycol (40% w/w) and Tween 80 (50% w/w) was formulated by employing D-optimal mixture design. Two different radiolabeled formulations (F1 formulation and ATZ suspension) each containing 7 mg/Kg atazanavir were administered by an oral route to adult Wistar rats of either sex (n=6). The *in-vivo* biodistribution studies show significantly higher concentration ($P < 0.05$) of atazanavir in brain administered with F1 formulation, which was also evident by confocal laser scanning microscopy with respect to suspension group. Collectively, these findings suggest that lipid-based nanoformulation may enhance brain delivery of the potent and frequently used antiretroviral drug atazanavir.

Disclosures: S. Sethi: None. **V. Rana:** None.

Poster

P944. Drug Delivery and Formulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P944.06

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: NIH Grant DA045609
Center for Compulsive Behavior and Addiction, Rush University

Title: Vaporized Methamphetamine: A Novel Noninvasive Approach to Study Methamphetamine Abuse

Authors: *T. C. NAPIER[1], P. CZYZEWSKI[2], A. L. PERSONS[3];
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Abstract: Prevalence of nonmedical methamphetamine (meth) use is increasing in many countries. Personal vaporizers (electronic delivery devices, e-cigarettes), are a popular intrapulmonary delivery method for nicotine products, and these have been adapted to deliver illicit drugs, including meth. Powdered meth mixed into vape liquid provides bioavailability that is similar to nicotine (Peace et al; Office of Justice Programs' National Criminal Justice Reference Service, Doc #252921, May 2019). Though clearly important, the consequences of vaporized meth have received little scientific attention. To fill this knowledge gap and provide a model for future studies, we characterized vaporized meth HCl in young adult male Sprague-Dawley rats using off-the-shelf atomizer/tank cartridges secured in sealed e-Vape^{TM} chambers (La Jolla Alcohol Research, Inc). We tested concentrations of meth ranging from 50-200mg per 1mL of various combinations of a propylene glycol (PG)/vegetable glycerin (VG) vehicle. We also tested non-contingent schedules ranging from 3-5s vapor delivery every 2-5min for 45-60min sessions, for total meth delivery time of 60–150s. We determined that 100mg meth per 1mL of 30%/70% PG/VG vehicle delivered in 3s pulses/2min for 60min was optimal. Using this protocol, we tested vaporized meth and vaporized PG/VG vehicle (4 rats/treatment). After 60min, rats were removed from the e-Vape chamber, and motor activity was quantified in automated activity boxes for 60min. The vape protocol was repeated with vehicle only in a second cohort, and these rats were administered an intraperitoneal (ip) injection of meth (1.25mg/kg) or saline (1mL/kg) 30min before collecting locomotor data (4 rats/treatment). Both vaporized and ip meth robustly elevated motor function (e.g., locomotion and stereotypy increased 3-fold) and the hypermotility was sustained for the entire session (two-way ANOVA; $p<0.05$). A Bonferroni's multiple comparisons *post hoc* test revealed an effect of meth but no difference between the meth groups. To determine absorption of vaporized and ip meth, the experiment was repeated, and serum was collected 40-45min after vapor termination or 30min after ip injection. Blinded serum samples were analyzed by mass spectrometry, verified to a meth internal control. Serum concentrations were similar for vaporized and ip meth (68±17 and 50±6ng/mL, respectively; $p>0.05$). These results reveal that vaporized meth (1) is a feasible noninvasive means to deliver meth in rats, (2) is absorbed and detected in the bloodstream, and (3) alters motor function comparable to that of moderate meth doses injected ip.

Disclosures: T.C. Napier: None. **P. Czyzewski:** None. **A.L. Persons:** None.

Poster

P944. Drug Delivery and Formulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P944.07

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: Biodistribution of Spherical Nucleic Acids in the Mouse CNS Following ICV, ICM and IT Delivery

Authors: *L. MOORE, L. P. KELLY, M. SCHROFF, B. R. ANDERSON, G. T. CORBETT; Exicure, CHICAGO, IL

Abstract: Spherical nucleic acids (SNA^{TM}) are nanostructures comprised of densely packed oligonucleotides radially arranged around a nanoparticle core. Compared to their linear counterparts, SNA show increased cellular uptake and persistence in the central nervous system (CNS). In this study, we sought to assess the biodistribution and cell-type specific uptake of fluorescently labelled SNA in the mouse CNS following three routes of administration (RoA). We synthesized bi-fluorescent SNA harboring BODIPY-labeled DOPC cores and Cy5-labeled oligonucleotides (SNA-BODIPY-Cy5). Single injections of SNA-BODIPY-Cy5 were delivered to eight-week-old male NMRI mice via: i) intracerebroventricular (ICV), (ii) intra-cisterna magna (ICM), or (iii) intrathecal (IT) RoA. Two hours or 14 days after dosing, brains and spinal cords were processed for bone decalcification and tissue clearing prior to 3-dimensional light sheet microscopy. Strong BODIPY signals were observed in the CNS at 2 hours post-injection that were essentially absent at 14 days post-injection, while Cy5 levels were prominent throughout the CNS at both time points. These results suggest: i) rapid distribution of the SNA structure throughout the brain and spinal cord after delivery to cerebrospinal fluid and ii) SNA components are differentially metabolized. Fourteen days post-injection, strongest brain and spinal cord Cy5 levels were observed after ICV and IT delivery, respectively, in accordance with their points of injection. Particularly in ICV and IT injected animals, prominent Cy5 signals could be detected throughout all cortical and subcortical regions of the brain, including brainstem nuclei, cerebellum, thalamic and hypothalamic nuclei, hippocampus, and striated cortical layers. High Cy5 signals were detected throughout all sections of the spinal cord in IT-injected animals, including prominent distribution into dorsal root ganglia. Co-labeling of CNS tissues with cell-type specific markers revealed SNA uptake by all prominent CNS cell subtypes, including neurons, astrocytes and microglia. In conclusion, SNA distribute widely throughout the CNS following local administration to CSF and are taken up by all major CNS cell subtypes. These findings support the continued development of SNA therapeutics for neurological disorders of the brain and spinal cord.

Disclosures: L. Moore: None. **L.P. Kelly:** None. **M. Schroff:** None. **B.R. Anderson:** None. **G.T. Corbett:** None.

Poster

P944. Drug Delivery and Formulation

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Program #/Poster #: P944.08

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: KAKENHI 16H06532,16K21734,16H06524

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Health Sciences Research Grants from the Ministry of Health, Labour, and Welfare, Japan, Grant H27-KAGAKU-IPPAN- 007

Title: Quantitative detection of the effect on neuronal responses in tri-synapses with the acute application of bisphenol-A-related substances on hippocampal slices with voltage-sensitive dye (VSD)

Authors: Y. TOMINAGA[1], M. TAKETOSHI[1], K. TANEMURA[3], ***T. TOMINAGA**[1,2];
[1]Inst. of Neurosci., [2]Kagawa Sch. of Pharmaceut. Sci., Tokushima Bunri Univ., Sanuki, Japan;
[3]Lab. of Animal Reproduction and Development, Grad. Sch. of Agr. Sci., Tohoku Univ., Sendai, Japan

Abstract: Environmental chemical exposure and drug administration can cause central nervous system (CNS) toxicity. Especially, vulnerability to exposure during early developmental stages is concerned to raise the risk for various neuropsychiatric disorders, including Asperger spectrum syndrome. The neural circuit-level assay is important to fill in a gap between the cellular assay using primary or cell lines and in vivo assay. However, effects on neural circuit activity are challenging to detect with conventional electrophysiological methods. We developed a voltage-sensitive dye (VSD; Di-4-ANEPPS) imaging system to detect subtle neural activity modification in the neural circuits quantitatively on a large scale. Here, we used the hippocampal slice preparation as a representative neural circuit and tested acute effects of the environmental chemical bisphenol-A (BPA) and its related substances BBMTBP and MBMTBP (5 ppm in perfusate). We tested the hippocampal tri-synaptic responses by applying electrical stimulation to Schaffer collateral to CA1, mossy fiber to CA3, and perforant path to DG.The three different regions' responses in the hippocampus were accumulated across five to eight different slices. We assessed effects on excitatory and inhibitory networks using a GABA-A receptor blocker (SR95331) to the system (+Gz). Firstly, we tested with adult mice (eight weeks old). We evaluated the VSD signals with the amplitude of early response (ER, peak values within 40 ms from the stimulation), which corresponds to excitatory post-synaptic potential (EPSP) and action potentials (AP). We used selective ROI to evaluate the response corresponds to CA1, CA3, and DG regions. BBM and MBM reduced CA1-ER in the normal solution (-Gz) and CA3-EPSP components in +Gz. We also evaluated the late phase (40-500 ms) of the response as an integral of the response (LR). In LR, CA1 and CA3 were reduced by BBM in -Gz and -Gz. We also used young (two-week-old) mice to assess the vulnerability of circuit activity. To our surprise, the young group was more-or-less resistant to those chemicals, while still BBM and MBM showed significant suppression of CA3-EPSP components +Gz. In LR, CA1 responses were affected by BBM and MBM. The overall result indicates higher risks with BBMTBP and MBMTBP than with BPA.

Disclosures: Y. Tominaga: None. **M. Taketoshi:** None. **K. Tanemura:** None. **T. Tominaga:** None.

Poster

P944. Drug Delivery and Formulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P944.09

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: Advances in Gene Therapy: Convection enhanced delivery for Parenchymal Brain Administration guided by real time MRI

Authors: *E. LARSON[1], J. STARE[1], K. WONG[1], M. S. MAGHEZZI[1], C. LECRUX-LEBLOND[1], M. V. ACCARDI[1], S. AUTHIER[1,2];
[1]Charles River Labs., Laval, QC, Canada; [2]Fac. of Vet. Med., Univ. of Montreal, Montreal, QC, Canada

Abstract: Advances in Gene Therapy: Convection enhanced delivery for Parenchymal Brain Administration guided by real time MRI Erik A. Larson[1], Jerneja Stare[1], Karen Wong[1], M. Said Maghezzi[1], Clotilde Lecrux-Leblond[1], Michael V. Accardi[1], Simon Authier[1, 21]Charles River Laboratories, Laval, QC, Canada[2]Faculty of Veterinary Medicine, University of Montreal, QC, CanadaConvection-enhanced delivery (CED) has emerged to become a broadly used methodology for targeted delivery of cell and gene therapies directly into various brain regions. To ensure the highest translational value, the same methods are used in non-clinical models to assess safety and efficacy of cell and gene therapies in development. Considering this, we recently implemented real-time intraoperative magnetic resonance imaging (MRI) and a stereotactic navigation system to deliver novel neurological therapeutics in a non-human primate (NHP) model. A gadolinium-containing solution, infused into the targeted brain structure at rates of 1-5 µL/min, was used as a contrast agent which allowed for real-time MRI scanning confirmation of dose site accuracy and dose diffusion monitoring. Real-time MRI scanning was used to assess distribution relative to the targeted brain structures. Further post-hoc analysis using custom bilateral masks outlining target subcortical structures was used to calculate the estimated coverage of the structures. A sequential dosing paradigm was used in the preclinical model to cover structures bilaterally given anatomical constraints. During recovery from surgery, animals typically presented mild and expected procedure-related clinical signs (e.g. mild tremors) which generally resolved within four days. Histopathological assessments also identified the expected neuropathological changes indicative that surgical procedures were well tolerated. Since the use of NHP models is particularly important for testing the delivery of cell and gene therapies, this approach provides a clinically relevant preclinical methodology for targeted brain CED with high structural accuracy.

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Poster

P944. Drug Delivery and Formulation

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Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P944.10

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: CONACYT Grant 316317

Title: Gold nanoparticles produce transient reactive gliosis in the adult brain

Authors: *E. LIRA-DÍAZ[1,2], T. CAMPOS-ORDONES[1,2], D. ZARATE-LOPEZ[1,2], N. IBARRA-CASTANEDA[2], M. GONZALEZ-PEDROZA[3], R. MORALES-LUCKIE[3], O. GONZALEZ-PEREZ[2];
[1]Physiological Sci. PhD Program, Sch. of Medicine/University of Colima, Colima, Mexico; [2]Sch. of Psycology/University of Colima, Colima, Mexico; [3]Sustainable Chem. Res. Center/UAEMex-UNAM, Toluca, Mexico

Abstract: Background: Gold nanoparticles (GNPs) have unique physical and chemical properties that allow them to function as a drug-delivery system for several tissues: skin, eye, liver, and others. However, information about the biological response of brain tissue against GNPs is limited. Astrocytes and microglia cells are the first line of defense against brain insults and proper indicators of the level of brain damage. Objective: To evaluate the astrocytic and microglia response after an intracerebral injection (striatum) of polyethylene-glycol-coupled GNPs (PEGylated GNPs). Methods: Male CD1 mice (P60) were divided in three groups: Gold nanoparticles (GNPs), control-vehicle and control-sham. The GNPs received 100 nl of PEGylated GNPs (85×10^6 nanoparticles/nl). The control-vehicle group received 100 nl of 0.9% NaCl solution whereas control-sham group were submitted to craniotomy without meningeal disruption. All animals were sacrificed by transcardial perfusion at 3, 7, 14, 30 and 90 after administration. We evaluated the cellular response of astrocytes and microglia at all time-points after intracerebral injection by cell counting and morphologic changes in a stereological grid. Results: Astrocytic response shows an increase in their populations and changes in their cytoplasmic transformation at day 3 and decrease at day 90. Microglia response are less prominent and significant changes were found only at day 7 and 14. Conclusion: Our findings indicated that the glial response was transient and mainly circumscribed to the injection site.

Disclosures: E. Lira-Díaz: None. **T. Campos-Ordones:** None. **D. Zarate-Lopez:** None. **N. Ibarra-Castaneda:** None. **M. Gonzalez-Pedroza:** None. **R. Morales-Luckie:** None. **O. Gonzalez-Perez:** None.

Poster

P949. Computational Modelling of Synaptic Networks: Oscillations and Synchrony

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P949.01

Topic: B.09. Network interactions

Support: Grant-in-Aid for Scientific Research (A)

Title: A Whole Brain Synchronicity Neural Computing Model -Part 1

Authors: *T. MAEDA[1,2], N. NISHIO[1];
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Abstract: We propose a computational model of the brain that achieves identity of consciousness across the brain. In order for the brain to retain information as memory, it is believed that synapses must record temporally continuous signals in a temporally and spatially invariant state. In this paper, we propose a computational model of the brain that enables the brain to store information in a temporally and spatially invariant state. Just as synchronous circuits are necessary for large-scale electronic circuits, synchronization of the entire brain is necessary to ensure memory identity. We propose an Eigenfunction Synchronicity Model that utilizes standing waves as a physical phenomenon to achieve this synchronization. In this model, the neural field is regarded as an oscillatory field, as shown in the model of Wilson-Cowen et al. The nodes of the standing wave are the smallest units of memory. The formation of standing waves is defined as the recall of the whole memory and the formation of consciousness. The nodes of a standing wave are the roots of the eigenfunction that serves as its envelope waveform. This root can form a memory that satisfies the identity as nominal scale of 1DOF constraint in the orthogonal complementary space representation. What is the advantage of describing memory with standing waves if organisms use the same principle? This model embraces the principle of the generation of representations in the brain. Representations in the human brain are mappings that are associated with external events, and are information that can be recognized and stored as time-invariant relationships. The mechanism of this model, in which memories are traced in the brain through the "time-invariant and spatially-invariant" phenomenon of standing wave nodes, suggests that the principle of wave transmission is employed in brain information processing. In the wave transfer field, the boundary value of the closed domain and the internal and external states can be treated as mirror images via the Kirchhoff-Helmholtz integral equation. This means that the internal and external states are mapped to each other across the boundary surface, and if the closed domain corresponds to a human being, the boundary value corresponds to sensory-motor signals, the external state corresponds to external phenomena, and the internal state corresponds to representations in the brain. We can define a representation here as a map that extracts the time-invariance of external events. In the formation of these

representations in the brain, it is an essential mechanism that the mapping can be acquired as unsupervised learning only on the condition that the nodes constitute a time-invariant zero point.

Disclosures: T. Maeda: None. **N. Nishio:** None.

Poster

P949. Computational Modelling of Synaptic Networks: Oscillations and Synchrony

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P949.02

Topic: B.09. Network interactions

Support: Grant-in-Aid for Scientific Research (A)

Title: Two Types of Nerve Fields Realize Recall of Associative Memory on the Whole Brain-A Whole Brain Synchronicity Neural Computing Model Part 2-

Authors: *N. NISHIO[1], T. MAEDA[3,2];
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Abstract: In the first report, we showed that synchronization of the entire brain is necessary to ensure the memory identity on the whole brain. The synchronization of the entire brain is ensured by the eigenfunctions of the spatial direction of standing waves that are established synchronously on the whole brain. In the second paper, we first argue that the gray matter and white matter of the cerebral cortex works as wave fields in which standing waves of different frequencies are established. Based on the nerve field model proposed by Wilson-Cowan et al. VanRullen et al. suggested that the white matter works as a wave field. On the other hand, the gray matter also works as a wave field, as in the Wilson-Cowan model. In other words, these wave fields support the formation of standing waves throughout the brain across cortical regions. Neurons in the gray matter have no plasticity and can reproduce the same standing waves when given the same boundary conditions. This allows for a reproducible mapping of states of external phenomena to internal states of the brain. On the other hand, the axons of excitatory neurons passing through the white matter are plastic and learnable, and thus play a role in learning the relationship between internal states of the brain. Associative memory, as a fundamental property of human memory, requires whole memories to be recalled from partial memories. How can this associative memory be realized in a uniform wave field? The eigenfunctions in the spatial direction of the standing wave that satisfy the boundary condition are established in the section between the two edges set as the boundary condition. Any node of the standing wave can be set as a new boundary condition. Therefore, a new standing wave will also be established simultaneously in the subsection sandwiched between any two nodes of the standing wave. This

corresponds to partial memory for whole memory. This is also the basic principle that expresses the associative recall of partial memory from whole memory. In addition, it supports the possibility that when partial memories are recalled, whole memories which are associated with them can also be recalled. In the first paper, I argued that sensory-motor signals are the boundary values that map external phenomena to human beings as the closed domain. In the second paper, we will focus on the eigenfunctions of the spatial direction of standing waves established in the closed region as wave field. The eigenfunction has spatial orientation. This makes it possible to distinguish between signal propagations from each modality, although multiple modalities are propagating signals to the association areas of the cerebral cortex.

Disclosures: N. Nishio: None. **T. Maeda:** None.

Poster

P949. Computational Modelling of Synaptic Networks: Oscillations and Synchrony

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P949.03

Topic: B.09. Network interactions

Support: JSPS KAKENHI 20K12004

Title: Weak inhibition synchronizes neuronal populations with spontaneous firing by disengaging from sodium currents

Authors: *T. SHINOZAKI;
NICT CiNet, Suita, Japan

Abstract: Inhibitory inputs are considered to facilitate synchronization of neuronal populations (Hasenstaub et al., 2005; Sohal et al., 2009), and the mechanism of this synchronization is understood to be caused by anchoring the membrane potential to the reversal potential of the inhibitory synapse. On the other hand, neuronal populations in vivo are usually in a spontaneous firing state, and hyperpolarization to the reversal potential of inhibitory synapses requires enormously strong inhibition, which may be unnatural also in terms of efficiency. In this study, we report a synchronization phenomenon caused by weak inhibitory inputs in a neural population with spontaneous firing. Although this phenomenon has been confirmed in our previous computational neuroscience studies (Shinozaki et al., 2007, 2010), in this report, we have clarified the detailed mechanism by analysis using the Fokker Planck equation. In a neural population that does not cause spontaneous firing, the membrane potential forms a certain distribution by balancing the external noise with the leak current. On the other hand, at the membrane potential near the firing threshold, where spontaneous firing occurs, the depolarizing sodium current partially cancels out the leak current, and attenuates the force that suppresses the diffusion of the membrane potential, resulting in a desynchronized state. Thus, under conditions

of spontaneous firing, the membrane potential distribution becomes relatively desynchronized, and hyperpolarization induced by weak inhibitory input causes a disengagement from the desynchronized state, resulting in synchronization. These results were analyzed in models using exponential integrate-and-fire (EIF) (Fourcaud-Trocmé et al., 2003) neurons and validated by numerical simulations. Furthermore, it was confirmed that models using leaky integrate-and-fire (LIF) neurons, which do not have dynamics corresponding to sodium channels, does not exhibit such synchronization phenomena. Since inhibitory inputs are thought to be closely related to the mechanisms of attention in the brain (Mitchell et al., 2007; Synder et al., 2016), our results may contribute to the understanding of the neural mechanisms of attention and oscillation in the brain and to the realization of more energy-efficient neuromorphic information processing.

Disclosures: T. Shinozaki: None.

Poster

P949. Computational Modelling of Synaptic Networks: Oscillations and Synchrony

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P949.04

Topic: B.09. Network interactions

Title: Inhibitory network model captures asymmetry in the transient dynamics of hippocampal ripple oscillations

Authors: *N. SCHIEFERSTEIN[1,2], T. SCHWALGER[3,2], B. LINDNER[1,2], R. KEMPTER[1,2,4]; [1]Humboldt-Universität zu Berlin, Berlin, Germany; [2]Bernstein Ctr. for Computat. Neurosci., Berlin, Germany; [3]Technische Univ. Berlin, Berlin, Germany; [4]Einstein Ctr. for Neurosciences, Berlin, Germany

Abstract: Hippocampal ripple oscillations have been implicated in important cognitive functions such as memory consolidation (Buzsáki, 1989). Yet ripple generation remains obscure, and several mechanisms were proposed, relying on excitation and/or inhibition as the main pacemaker(s). Inhibition-first models can be subdivided into perturbation-based (Malerba et al., 2016) and bifurcation-based models (Donoso et al., 2018, Brunel, Wang, 2003). While all ripple models produce oscillations in the ripple-band (140-220 Hz), only the bifurcation-based inhibitory model reproduces intra-ripple frequency accommodation (IFA) — an asymmetry in the instantaneous network frequency in CA1 in response to transient, symmetric, sharp wave-like stimulation (Donoso et al., 2018, Ponomarenko et al., 2004).
Here we mechanistically explain IFA in bifurcation-based inhibitory ripple models, using numerical simulations and a theoretical mean-field approach.
We use a spiking network of leaky-integrate-and-fire units that are fully connected via delayed inhibitory pulse-coupling. All units receive independent white noise and the same excitatory drive, mimicking the CA3 Schaffer collateral input to CA1. For increasing excitatory drive this

network undergoes a bifurcation from a stationary to an oscillatory regime (Brunel, Hakim, 1999). To quantify IFA, we a) approximate the highly non-linear oscillation dynamics for constant drive beyond the bifurcation and b) show how the response to transient drive relates to those cyclo-stationary dynamics. Thus, we take the frozen-noise limit and approximate the density of membrane potentials in the mean-field (i.e. the solution of the associated Fokker-Planck equation) as a Gaussian with time-dependent mean. In this framework we can analytically approximate the frequency and amplitude of the network oscillation as a function of excitatory drive. We show that for a wide parameter regime this ansatz provides a good approximation of the cyclo-stationary dynamics beyond the bifurcation.
We demonstrate that IFA is an inherent feature of this model class that results from a speed-dependent hysteresis effect in the amplitude of the oscillatory mean membrane potential, given transient, sharp wave-like drive. We predict that ripples triggered by optogenetic stimulation of CA1 PV^+ basket cells in vitro show *less* IFA if the stimulus ramps up/down more *slowly*.
The present work thus provides new insights into the transient dynamics of oscillations in inhibitory networks that can contribute to model selection and the unravelling of the true generating mechanism of hippocampal ripples.

Disclosures: N. Schieferstein: None. **T. Schwalger:** None. **B. Lindner:** None. **R. Kempter:** None.

Poster

P949. Computational Modelling of Synaptic Networks: Oscillations and Synchrony

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P949.05

Topic: B.09. Network interactions

Support: MH122023

Title: Generation of sharp wave ripples in the CA1 Hippocampus: a role for chandelier interneurons

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Abstract: Sharp wave ripples (SWRs) are the most synchronous large-scale oscillatory activity in neural circuits. Predominantly investigated in the CA1 hippocampus, several circuit models have been proposed for their generation: 1) feedback inhibition 2) reciprocal inhibition 3) feedback and reciprocal inhibition and 4) axo-axonic coupling between pyramidal cells. The first

three models require extrinsic inputs from CA3. However, CA3 population bursts often do not correlate with ripples in the CA1 of the hippocampus. Secondly, isolated CA1 slices have also been shown to generate SWRs, indicating CA1 contains local circuit motifs to generate ripples. In the 4th model, electrical coupling between PN axons and stochastic discharge are proposed to generate SWRs. But intracellular events and pharmacological properties of SWRs suggest neuronal discharge in SWRs are controlled by synaptic inputs. PN discharge in CA1 SWRs are preceded by GABAergic inputs and enhanced by axonal $GABA_A$ receptors. Recently, GABAergic axo-axonic chandelier interneurons (INs) have been reported to initiate SWRs in the amygdala. Using a large-scale CA1 network model, we present a conceptual microcircuit organization that includes chandelier INs to pace local feedback and feedforward circuits to generate SWRs. This model provides an intrinsic circuit and synaptic mechanisms for SWRs generation in the CA1 and reconciles discrepancies in previous models. This model predicts distinctive role for axo-axonic and axo-somatic GABAergic neurons in the generation of SWRs.

Disclosures: G. Glickert: None. **B. Latimer:** None. **M. Bhagavathi Perumal:** None. **P. Sah:** None. **S.S. Nair:** None.

Poster

P949. Computational Modelling of Synaptic Networks: Oscillations and Synchrony

Location: Virtual Only

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Program #/Poster #: P949.06

Topic: B.09. Network interactions

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NIH PO50NS091856 (MS)
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NIH DP2MH104119 (SA)
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Title: Spatially heterogeneous cholinergic release generates theta-gamma coupling

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Abstract: Theta and gamma rhythms and their cross-frequency coupling contribute to attention and learning. While forebrain acetylcholine (ACh) signaling has been linked to theta-gamma coupling, an underlying mechanism has not been identified. Recent studies indicate that

cholinergic signaling is both temporally and spatially constrained, in contrast to the traditional notion of slow, spatially homogeneous, and diffuse neuromodulation. We show that spatially constrained cholinergic neuromodulation can generate theta-modulated gamma rhythms. Using biophysically-based excitatory-inhibitory (E-I) neural network models, the effects of ACh on neural excitability are simulated by varying the conductance of a muscarinic receptor-regulated K+ current. In E-I networks with local excitatory connectivity and global inhibitory connectivity, theta-gamma-coupled firing patterns emerge when ACh modulated network regions are spatially heterogeneous. Stable gamma activity occurs within regions of high ACh signaling, while theta or mixed theta-gamma activity occurs at the peripheries. Robust theta-gamma coupled activity occurs due to alternation at theta frequency of gamma activity between different high-ACh regions. These results are the first to indicate a causal role for spatially heterogenous ACh signaling in the generation of localized theta-gamma rhythmicity. Additional results provide novel insights into mechanisms by which ACh signaling supports attentional processing of sensory information.

Disclosures: Y. Yang: None. **H. Gritton:** None. **M. Sarter:** None. **S.J. Aton:** None. **V. Booth:** None. **M.R. Zochowski:** None.

Poster

P949. Computational Modelling of Synaptic Networks: Oscillations and Synchrony

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Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P949.07

Topic: B.09. Network interactions

Support: ERC 716643 FLEXNEURO

Title: When, not if: Oscillations and active dendrites support fast spiking computations

Authors: *T. S. J. BURGER, M. E. RULE, T. O'LEARY;
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Abstract: Oscillations are found throughout the brain (Buszaki et al., 2004).On a cellular level, they control the spike timing of neurons (Jacobs et al., 2007), and on the behavioural scale they appear to define windows of perceptual awareness (Chakravarthi & VanRullen, 2012).Do oscillations merely correlate with these important variables, or are they used for functional purposes?We present a biophysically motivated model network that can implement a computation very quickly and reliably with a low number of spikes because the computation is orchestrated by oscillations.Endogenous oscillations synchronise spiking activity in the network, which is crucial for the quick integration of related inputs.With this synchrony in place the network can use the all-or-nothing nature of spikes to compute quickly.To make computations flexible the phase of oscillation should be separated from the decision of a neuron to spike.We

demonstrate that this can be achieved with an excitable dendrite.An excitable dendrite equips the neuron with a short memory for integrating inputs, and a dendritic threshold supports binary computations.Meanwhile the soma, influenced by the oscillation, decides when (not if) to spike.In doing so the oscillation defines time windows for the integration of inputs.This decouples the integration from the oscillation; the oscillation orchestrates, but does not change, the computation.We show that by separating the integration from the mechanism controlling the firing time of the soma, a network with these neurons can display synchrony and reliably perform a binary computation at the same time: it acts as a clocked binary circuit.The advantages are efficiency (a low number of neurons and spikes is required), speed, and robustness (to noise and parameter uncertainty).

References:
Buzsaki, G. Neuronal Oscillations in Cortical Networks. *Science* **304**, 1926–1929 (2004).
Jacobs, J., Kahana, M. J., Ekstrom, A. D. & Fried, I. Brain Oscillations Control Timing of SingleNeuron Activity in Humans. *J. Neurosci.* **27**, 3839–3844 (2007).
Chakravarthi, R. & VanRullen, R. Conscious updating is a rhythmic process. *PNAS* **109**, 10599–10604 (2012).

Disclosures: T.S.J. Burger: None. **M.E. Rule:** None. **T. O'Leary:** None.

Poster

P949. Computational Modelling of Synaptic Networks: Oscillations and Synchrony

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:30 AM - 9:30 AM

Program #/Poster #: P949.08

Topic: B.09. Network interactions

Title: Network model provides insights into entorhinal cortex mechanisms of theta generation

Authors: *I. GUERREIRO[1], Z. GU[2], J. L. YAKEL[2], B. S. GUTKIN[1,3];
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Abstract: Hippocampal theta oscillations are prominent 4-12 Hz rhythmic activities in the hippocampal field potential of all mammals studied to date. They have been linked to spatial and episodic memory formation. After decades of research, the origins of the hippocampal theta rhythm remain elusive. In particular, it is not clear how theta oscillations are generated as a result of the interactions among the three essential brain subregions - the septum, hippocampus and entorhinal cortex (EC).
Our recent experimental studies indicate that the EC may be the generator of theta rhythm in a septo-entorhinal-hippocampal brain slice coculture preparation. Not only does the EC leads the hippocampus in the timing of theta rhythm activities, but all hippocampal sub-regions are

synchronized, suggesting that they respond to a common rhythmic input from the EC. However, it is important to note that the EC itself requires hippocampal feedback to maintain the theta rhythm (Zhenglin and Yakel, 2017). In this work, we propose a circuit model of the EC to study the intrinsic properties of the EC that allow for external excitatory inputs to drive the system into an oscillatory regime. We use Izhikevich's two-dimensional QIF neuron model (Izhikevich, 2003) to describe the three major classes of neurons observed in the EC: stellate cells (S), pyramidal cells (E), and fast-spiking interneurons (I). We then take advantage of recently developed exact reduction methods to get a simplified, exact description of the three neural populations. Next, to study the contributions of the neural populations in the generation of the theta, we use a machine learning approach (Gonçalves et al., 2020) to infer the space of connectivity parameters that give rise to theta rhythmic activity in the EC network model. We find that while a subnetwork of S and E-cells is capable to robustly generate synchronized theta oscillations, the I-cells facilitate the generation of low-frequency theta rhythm. The I and E coupling effectively modulates the oscillatory frequency while the S and E intercoupling controls the magnitude of the oscillations. Thereby our modeling results point out complementary roles of the excitatory and inhibitory entorhinal subcircuits in theta rhythmic generation.

Disclosures: I. Guerreiro: None. **Z. Gu:** None. **J.L. Yakel:** None. **B.S. Gutkin:** None.

Poster

P949. Computational Modelling of Synaptic Networks: Oscillations and Synchrony

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Program #/Poster #: P949.09

Topic: B.09. Network interactions

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Title: Inhibitory neuron participation in simulated M1 neuronal avalanches

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Abstract: Accumulating experimental evidence is consistent with the hypothesis that signal processing in the cerebral cortex is maintained near criticality. Proper balance must be maintained between excitation and inhibition to achieve synchronized activity among cortical neurons spanning a broad range of population sizes and durations. As previously presented, our data-driven detailed simulated mouse primary motor cortex (M1), when stimulated by a brief small unstructured current, produced self-organized and self-sustained neural dynamics with spikes clustered by size and duration distributed in log-log plot slopes around -1.5 and -2.0.We

analyzed avalanche durations and identified 4 population spiking patterns that combined to make up each individual avalanche: irregular pattern (exclusively in IT5B and IT6) and patterns with dominant frequencies -- delta (~1 Hz), beta (~22 Hz), and gamma (~40 Hz). We then clustered avalanches containing distinct combinations of these spiking patterns which gave us 4 main avalanche types: Irregular, Fragment, Beta, and Delta+ (note capital letters). Irregular avalanches are composed of only the irregular spiking pattern. Fragment avalanches are brief pieces of frequency-dominated avalanches. Beta, Delta+ contained the respective spiking patterns; the '+' indicating that many Delta+ included nested gamma, beta, irregular spiking patterns.Most Fragment avalanches included inhibitory neuron activity (95.0%+/-0.02%) compared to Irregular 0%; Beta: 100%; Delta+ 100%. The most active Fragment avalanche inhibitory neurons were somatostatin, neurons particularly in layer 2/3, included in 69.9% (SOM2/3) of the Fragment avalanches. Layers 5B and 6 somatostatin neurons were also active with 32.6% (SOM5B) and 35.4% (SOM6) respectively. Layer 5A somatostatin neurons were least active with 15.0% (SOM5A) included in Fragment avalanches. In contrast, a small number of parvalbumin neurons were active in Fragment avalanches, and only in layer 5, with 11.6% (PV5B) active in layer 5B, 6.4% in layer 5A, and 0.0% in the other layers (PV2/3 and PV6).

Disclosures: D. Doherty: None. **S. Dura-Bernal:** None. **W. Lytton:** None.

Poster

P018. Stem Cells: Technical Development

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P018.01

Topic: A.03. Stem Cells and Reprogramming

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Title: A tunable cell culture system to recapitulate endogenous neural stem cell behavior

Authors: *N. SOHONI[1], N. ZHANG[3], L. PENG[2], T. C. OUGHOURLIAN[2], A. WEINREICH[2], J. LEVI[2], J. SHEPPARD[2], M. A. BONAGUIDI[2];
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Abstract: Adult neural stem cells (NSC) were first isolated and characterized using cell culture about 30 years ago. However, recent advances in lineage tracing have elucidated that NSC

properties in vitro do not mimic their native behavior *in vivo*. We have developed a simple culture approach to recapitulate endogenous NSC fate choices. We determined that traditionally used growth factor concentrations promote NSC symmetric divisions while suppressing neuronal fate, and amplify glial-committed progenitors. Additionally, we have shown that high growth factor conditions increase the percentage of transit amplifying cells *in vitro*, a trend which is also replicated with mice treated with similar growth factor conditions *in vivo*. Instead, low growth factor application during an 'expansion phase' balances NSC symmetric, neurogenic, and astrogenic fate choices akin to multipotential NSC behavior *in vivo*. Our study provides an updated system to rapidly study new cell generation, neural circuit formation, and underlying mechanistic regulation.

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Poster

P018. Stem Cells: Technical Development

Location: Virtual Only

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Program #/Poster #: P018.02

Topic: A.03. Stem Cells and Reprogramming

Support: T32 Stem Cell Training Grant T32NS082174
UCI Institute for Clinical & Translational Science UL1 TR001414
UCI MSTP T32GM008620

Title: Blood vessel formation in a 3D scaffold is promoted by human neural stem cell-secreted material

Authors: *B. GUTIERREZ[1], L. A. FLANAGAN[2];
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Abstract: Human neural stem/progenitor cells (hNSPCs) have the potential to widen the currently narrow therapeutic time window for stroke as hNSPC transplantation several days post-stroke improves functional recovery in rodent stroke models. Transplanted hNSPCs increase angiogenesis and neovascularization in the peri-infarct region and this is linked to functional recovery. However, the mechanisms by which hNSPCs stimulate an increase in vessel formation after transplantation are not fully understood. Our lab modeled the hNSPC-induced increase in

vessel formation *in vitro* by seeding hNSPCs and human endothelial progenitor cells (hEPCs) in a 3D scaffold composed of salmon fibrinogen, laminin, and hyaluronic acid that mimics brain tissue properties. Using our *in vitro* neurovascular model, we tested our hypothesis that hNSPC-secreted material plays a role in the stimulation of vessel formation. Our previous RNA-Seq data showed that hNSPCs express high levels of angiogenic growth factors, such as VEGF, PDGF, and TGF-B1, that could be secreted by hNSPCs to increase vessel formation. In order to determine the effect of hNSPC-secreted material on vessel formation, hEPCs were seeded in 3D scaffolds alone, with hNSPCs, or with hNSPC-conditioned media (CM). We found hNSPC-CM is sufficient to induce an increase in vessel length, area, and branch points, showing that hNSPC secreted material and not contact stimulates vessel formation. CM contains soluble factors and extracellular vesicles (EVs) secreted by hNSPCs. Inhibiting hNSPC exosome production and secretion diminished the effect of hNSPC-CM on vessel formation. In conclusion, material secreted by hNSPCs can recapitulate the increase in vessel formation induced by hNSPCs and secreted EVs, such as exosomes, may play a role in promoting vessel formation. HNSPC secreted material could be beneficial therapeutically since it could be used to treat stroke several days after the initial insult and avoids limitations of cell therapy.

Disclosures: B. Gutierrez: None. **L.A. Flanagan:** None.

Poster

P018. Stem Cells: Technical Development

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Program #/Poster #: P018.03

Topic: A.03. Stem Cells and Reprogramming

Title: Homeobox Identity of Neural Stem Cells is Maintained Outside of a Developmental Context

Authors: *W. MCINTYRE[1,2], M. KHAZAEI[2], M. G. FEHLINGS[1,2];
[1]Inst. of Med. Sci., Univ. of Toronto, Toronto, ON, Canada; [2]Krembil Res. Inst., Toronto, ON, Canada

Abstract: ***Background***: Neural Stem Cell (NSC) therapy is a promising regenerative strategy to treat central nervous system (CNS) injury because NSCs replace the diverse cell niche that reflects a pre-injured state. However, an NSCs ability to integrate with spared tissue in a transplant environment has been suboptimal, which has halted NSC therapies from advancing into the clinic. The discordant identity of NSC grafts to their respective region of the CNS is perhaps responsible for their limited regenerative success. Supporting this, spinal-derived NSC grafts transplanted into the injured spinal cord promotes the greatest extent of regeneration when compared with brain-derived NSC grafts. Interestingly, an unexplored explanation for this may be conferred through the expression of regional-identifying transcription factors: Homeobox

(*Hox*) genes. During development, region-specific *Hox* promotes the localization of cells in the brain & spinal cord, and ensures appropriate neuronal connectivity to promote motor and sensory functions. Thus, if *Hox* genes are maintained in NSCs cultured for transplantation, this may suggest their developmental role is recapitulated during optimal regenerative attempts. ***Hypothesis***: We hypothesize that (b) brain and (s) spinal cord-derived NSCs will retain region-specific *Hox* expression following isolation, expansion, and differentiation *in vitro*. ***Methods:*** bNSCs and sNSCs (N=9), with distinct *Hox* expression *in vivo*, were dissected from the subventricular zone of the brain and central canal of the spinal cord, respectively. Subsequently, NSC colonies were expanded for several passages and differentiated *in vitro*. Gene and protein *Hox* expression was evaluated using RT-qPCR and Western blot, respectively. Each NSC's differentiation profile were assessed using immunocytochemistry. ***Results:*** Following both expansion and differentiation *in vitro*, both bNSCs and sNSCs retained region specific *Hox* expression. bNSCs maintained a greater proportion of brain-specific markers compared to sNSCs (*Otx2*, *FoxG1*, *Emx2*), whereas bNSCs did not express any spinal cord-specific markers (*Hox4-9*). Notably, each NSC line exhibited tripotency by producing neurons, oligodendrocytes, and astrocytes, a hallmark of NSC colonies. ***Conclusions:*** NSCs derived from developmental *in vivo* niches retain their developmental characteristics *in vitro*. This suggests that successful NPC transplants in experimental therapies may be due to the maintenance of *Hox* expression. Future work will involve evaluating the maintenance of NSC *Hox* identity in experimental CNS injury.

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Poster

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Program #/Poster #: P018.04

Topic: A.03. Stem Cells and Reprogramming

Support: JP Grant 16K10715

Title: Stat3 induces hypoxic preconditioning against oxidative stress in neural stem cells

Authors: *H. KINOUCHI, **N. FUKUDA**, T. WAKAI, T. TATEOKA, H. YOSHIOKA; Univ. of Yamanashi, Fac of Med., Chuo, Japan

Abstract: Transplantation of neural stem cells (NSCs) has been proposed as a promising therapeutic strategy in stroke; however, oxidative stress in ischemic host brains remarkably reduces the number of surviving transplanted cells. Hypoxic preconditioning (HP) is one of the approaches to improve the ability of NSCs to survive in harsh environments, but the mechanisms of such phenomenon are obscure. Activation of signal transducer and activator of transcription 3 (STAT3) via Janus kinase 2 (JAK2) induces cell differentiation and regulates proliferation in

NSCs, and also plays a pivotal role in hypoxia/ischemia-induced preconditioning in various type of cells. Accordingly, STAT3 may contribute to the beneficial effects of HP in NSCs, however, its role is not well elucidated. In this study, we investigated the role of STAT3 and the representative downstream signals such as vascular endothelial growth factor (VEGF) and cyclin D1 in hypoxic preconditioned NSCs. For the induction of HP, NSCs isolated from neonatal C57BL/6 mice were exposed to 5% hypoxia for 24 hours. HP significantly decreased cell injury of NSCs, and enhanced the viability against H_2O_2-mediated lethal oxidative stress. Western blot analysis revealed that the phosphorylation of JAK2 and STAT3, the activated forms of these proteins, was significantly increased after HP. Moreover, HP increased the concentration of VEGF in the culture media and the expression of Cyclin D1 in the cells. The administration of STAT3 inhibitor significantly suppressed the activation of STAT3 and the upregulation of the downstream signals, resulting in the diminishment of the efficacy of HP in oxidative injury. Our results suggest that the activation of STAT3 could induce HP in NSCs against oxidative stress, which might be due to protective function of autocrine/paracrine VEGF and cell proliferative effects of cyclin D1. Activation of this pathway could be a treatment strategy to improve the efficacy of cell therapy in stroke.

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Poster

P018. Stem Cells: Technical Development

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Program #/Poster #: P018.05

Topic: A.03. Stem Cells and Reprogramming

Support: NSF Grant 2024797
Stem Cell Research Fund
Honors Program, Iowa State University

Title: Development of a microfluidic ‘brain-on-chip’ device as an in vitro platform for 3D neural system development and analysis

Authors: *C. FONDER[1], R. YANG[2], T. SYLVESTER[1], S. PENG[1], L. QUE[2], D. SAKAGUCHI[1];
[1]Dept. of Genetics, Development, and Cell Biol., [2]Dept. of Electrical and Computer Engin., Iowa State Univ., Ames, IA

Abstract: In recent years, organ-on-chip models have become increasingly valuable due to their ability to provide a microenvironment platform for three-dimensional (3D) tissue development, which can mimic the interactions of in vivo cells and tissues. Brain-on-Chip models allow for

mimicking neural tissue that is not possible using conventional two-dimensional cell culture techniques which can facilitate study of various neurological disorders, such as Alzheimer's disease, or be used as a tool for drug screening. In our exploratory in vitro study, rat adult hippocampal progenitor cells (AHPCs) were cultured as dissociated cells and non-adherent neurospheres within a novel microfluidic device to determine if these devices allowed for the 3D culturing of the progenitor neural stem cells. The polydimethylsiloxane (PDMS) microfluidic device was fabricated by soft-lithography. Each 2 x 2 cm chip consisted of three independent "S"-shaped microarrays containing 15 microwells for cell culture. The AHPCs were cultured within the microfluidic devices for up to 12 days in vitro (DIV) in either conditioned culture media and/or differentiation media. The formation of neurospheres and their growth was analyzed by evaluating neurosphere size over the 12-day growth period by quantifying the average neurosphere diameter and perimeter distance of neurospheres within the microfluidic devices from 0 to 12 DIV. At 3, 7, and 11 DIV, the AHPCs were fixed and stained with propidium iodide (PI) to evaluate cell viability within the microfluidic devices, with results showing high cell viability at all timepoints for each media condition. At 7, 10, and 12 DIV, AHPCs were fixed and immunolabeled to evaluate proliferation (Ki-67) and cell differentiation looking at cell-type specific markers for neurons (TuJ1 and MAP2ab), oligodendrocytes (RIP), and astrocytes (GFAP) within the microfluidic devices for each timepoint. These results showed that AHPC neurospheres can be successfully generated and propagated within these novel microfluidic devices with continuing cell proliferation (Ki-67 expression) and neurosphere growth overtime for both media conditions. Immunolabeling with the cell-type specific markers indicated differentiation of multiple neural cell types within the growing neurospheres. Fabrication of microfluidic devices can be used to develop preliminary Brain-on-Chip models which may be useful to evaluate neurological disorders and act as drug screening platforms.

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Poster

P018. Stem Cells: Technical Development

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Program #/Poster #: P018.06

Topic: A.03. Stem Cells and Reprogramming

Support: R01 NS090617-05

Title: Biocompatibility Of Large Area Electronics-Grade 2D Materials With Stem Cells And Motor Neurons

Authors: *R. T. JAYANTH[1], N. WHITE[2], D. KIREEV[3], M. SERNA[3], E. OKOGBUE[5], Y. JUNG[6], S. SAKIYAMA-ELBERT[2], D. AKINWANDE[4];

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Abstract: Two-dimensional (2D) materials have the potential to become the functional elements of future bio-neuro-electronic interfaces. So far, studies directed towards understanding their biocompatibility have mostly utilized exfoliated flakes and suspensions resulting in large uncertainty and discrepancies in their cytotoxicity measures. In this work, as a first step before incorporating them in interface devices, we have performed a comprehensive experimental study of *in vitro* cell-viability of large-scale sheets of electronics-grade 2D materials that include platinum diselenide ($PtSe_2$), platinum ditelluride ($PtTe_2$), molybdenum disulfide (MoS_2), and graphene. These materials were selected because of their superior electronic and mechanical properties, and their atomically thin layered structure which facilitates efficient surface chemical functionalization. We cultured embryonic stem cells (ESCs) and ESC-derived motor neurons (MNs) on the surface of these large-area 2D sheets and quantified cell-viability using the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay and the Lactate Dehydrogenase (LDH) assay. Live-dead fluorescence imaging was performed to qualitatively understand the trends in cellular growth and proliferation. The results suggest that $PtSe_2$ displays cell-viability values on par with those of the controls, graphene, and MoS_2. Contrarily, $PtTe_2$ displays the lowest cell-viability value amongst all the 2D materials when observed through the lens of the MTT assay but displays cell-viability similar to that of the controls when observed through the lens of the LDH assay. We provide additional confirmatory tests to validate $PtTe_2$'s anomalous and area-dependent biocompatibility behavior. We believe the results from these *in vitro* biocompatibility experiments can help large-area 2D materials become an alternative for future cell-culture scaffolds and bio-neuro-electronic implants.

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Poster

P018. Stem Cells: Technical Development

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Program #/Poster #: P018.07

Topic: A.03. Stem Cells and Reprogramming

Support: TL1TR001880

Title: Emergence of structured neural activity in human brain organoids with maturation

Authors: *K. G. MENSAH-BROWN[1], J. T. LIM[1], C. D. ADAM[1], D. JGAMADZE[1], G.-L. MING[2], H. SONG[2], J. A. WOLF[1], H.-C. I. CHEN[1];
[1]Neurosurg., [2]Neurosci., Univ. of Pennsylvania, Philadelphia, PA

Abstract: Background: Brain organoids are 3D, stem cell-derived neural tissues that recapitulate neurodevelopment. Recently, they have been used to model Zika virus-induced microcephaly and GBM. Additional clinical applications could include modeling other relevant brain disorders or as structured substrate to repair brain circuitry. However, before we can levy their full translational potential, a deeper understanding of their intrinsic neural activity at the single cell and network level is essential. Here, we present our preliminary analysis of this activity in human forebrain organoids.
Methods: Organoids were generated from hiPSC lines derived from healthy volunteers. Linear microelectrode probes were employed to record extracellular activity from DIV77, 100, and 130 organoids. Extracellular recordings were collected at baseline and following the addition of MK-801 and NBQX. The recordings were processed with offline spike sorting followed by spatiotemporal analysis to characterize baseline unit and network characteristics across age groups.
Results: Nine organoids were recorded successfully (n=3 per time point). A significant difference in unit number was seen ($p < 0.05$). Post hoc comparisons using the Tukey HSD test showed significantly more units at DIV130 (51.67±14.15) than DIV100 (17.67±7.57) and 77 (16.33±14.98). A significant difference in baseline firing rate was observed ($p < 0.05$) with a higher baseline firing rate at DIV130 (0.83Hz ± 0.17) and DIV100 (0.86±0.40) than DIV77 (0.31Hz±0.06). A significant difference in fold response of firing rates to MK-801 and NBQX was seen ($p < 0.05$) with greater fold decreases in firing rate at DIV130 (-24.17%) and DIV100 (-36.81%) compared to DIV77(+1.2%). Global synchrony did not differ among age groups. However, there was a significant difference in the size of statistically significant synchronous unit clusters with larger clusters occurring at DIV100 than DIV77 and 130. Preliminary network analysis through spike time tiling coefficients (STTC) showed the emergence of functional connectivity across age groups. However, at these early time points, network topology showed no modularity or small-world features.
Conclusion: Consistent with findings from 2D cortical cultures, our data reflects increasing excitatory network activity with maturation. Furthermore, the reduction in the average size of clusters alongside the emergence of network connectivity could corroborate evidence of smaller, more efficient cell assemblies and increasingly complex neural activity. Additional work in this area will confer a better sense of how to rationally utilize brain organoids for translational purposes.

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Poster

P019. Regeneration: PNS

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Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P019.01

Topic: A.04. Transplantation and Regeneration

Support: JSPS 19H03850
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Title: Cell extract from adipose tissue-derived stem cells promotes peripheral nerve regeneration

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Abstract: Background: Peripheral nerves are well known to regenerate after injury, but this can induce neuroma, which results in intractable pain. The healing process receives enhancing or inhibitory signals from Brain-derived neurotrophic factor, VEGF, and semaphorin 3A. These factors mediate Schwann cells (SCs), vascular tissue, and neurons, which suggests that regeneration of injured nerves requires the contribution of these factors. However, it is difficult to control these factors simultaneously during the regenerative process. Adipose tissue-derived stem cells (ADSCs) have the potential to regenerate several tissues. A cell extract (CE) from ADSCs (CE-ADSCs) promotes functional recovery of salivary glands. Thus, CE-ADSCs may be applied to the treatment of injured peripheral nerves. In this study we examined the effect of CE-ADSCs on SCs and neurons in vitro and immunohistologically investigated the effects on axonal regeneration and localization of SCs in vivo.Animals/Methods: Adipose tissue was obtained from male C57B6/J mice and treated with collagenase. The stromal vascular fraction was cultured to establish ADSCs. CE was prepared by repeated freezing and thawing of ADSCs and centrifugation. CE-ADSCs was applied to SCs or PC12D cells, and the SC proliferation ability and protrusion elongation of PC12D cells were examined.Immunohistochemistry: Male SD rats (6 weeks old) were divided into three groups: nerve exposure surgery only (sham group), saline administration (control group; CG), and CE-ADSC administration (CE group; CEG). The inferior alveolar nerve (IAN) in the rat was transected. Seven days after the transection, fixation was performed by perfusion. The IAN was cut into a sagittal section. Fluorescence immunohistochemical staining was performed using primary antibodies against Protein gene product 9.5, Pan-neurofilament (pNF), and Myelin protein zero (MPZ), and secondary antibodies (Alexa Fluor® 488 and 594).Results: CE-ADSCs increased the number of SCs and PC12D cells with protrusion elongation. Immunohistology showed nerve fibers sprouting in the radiating direction and randomly extending in the distal site from the proximal stump in the control group, whereas regeneration of nerve fibers was observed in the CEG. Double staining for pNF and MPZ showed that fusiform myelin accompanied the axon at the proximal site, but that there was a lack of the axon that accompanied myelin at the distal site.Conclusion: CE-ADSCs might promote peripheral nerve regeneration by inducing proliferation of SCs and enhancing axon regeneration.

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Poster

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Topic: A.04. Transplantation and Regeneration

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Title: Using single-cell transcriptomics to explore the transcriptomic variance underlying the different axon regeneration ability between sensory neuron subtypes

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Abstract: Dorsal root ganglion (DRG) neurons are pseudo-bipolar neurons with two axonal branches, a peripheral branch innervating their target organ, and a central branch relaying somatosensory information into the central nervous system. The peripheral branch can readily regenerate upon peripheral axotomy, largely due to the activation of regenerative transcription programs. Thus, sensory neurons in the DRG provide a perfect model system to study the mechanisms underlying axon regeneration at the transcription level, and many studies have been built on this basis. However, as a highly heterogeneous neuronal population, sensory neurons in the DRG are comprised of many anatomically and functionally different subtypes that are likely to respond differently to peripheral axotomy. In addition, it has been previously reported that various subtypes possess different axon regeneration ability. Thus, to understand the transcriptomic difference underlying the different axon regeneration ability between various subtypes, we performed single-cell RNA-seq on DRG sensory neurons from mice receiving sham or sciatic nerve transection, by combining a sensory neuron specific GFP reporter mouse line, and the single-cell gene expression technique (10x genomics). After removal of low-quality cells and doublets, we identified 13 neuronal clusters from a total of 17,690 DRG neurons that can be categorized into non-peptidergic nociceptors (NP), peptidergic nociceptors (PEP), neurofilaments (NF), and C-fiber low-threshold mechanoreceptors (cLTMR). No significant

change of identified clusters or novel clusters were observed within 24 hours after injury. Using 297 genes known to be upregulated in the DRG by peripheral axotomy, we calculated a regeneration score for each neuron to evaluate the axon regeneration potential of the 13 identified neuronal clusters. The analysis revealed that one of the NP clusters had the lowest average regeneration score, suggesting that it may have the least ability in axon regeneration compared with other clusters. In addition, some clusters from NP, NF and cLTMR showed bimodal in the distribution of the score, indicating neurons of the same subtype may also have rather different axon regeneration ability.

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Poster

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Topic: A.04. Transplantation and Regeneration

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Title: Caldendrin is a negative regulator of neurite growth in dorsal root ganglion neurons

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Abstract: Caldendrin is a calmodulin-like Ca^{2+} binding protein that is expressed primarily in neurons and regulates multiple effectors including Ca_v1 L-type Ca^{2+} channels. Here, we tested the hypothesis that caldendrin regulates Ca_v1-dependent pathways that repress neurite growth in dorsal root ganglion neurons (DRGNs). By immunofluorescence, caldendrin was localized in medium- and large- diameter DRGNs. Consistent with an inhibitory effect of caldendrin on neurite growth, neurite initiation was enhanced, and the rate of neurite growth was faster, in dissociated DRGNs and DRGN explants, respectively, from caldendrin knockout (cald-KO) mice compared to those from wild type (WT) mice. In an *in vitro* axotomy assay, cald-KO DRGNs grew longer neurites via a mechanism that was more sensitive to inhibitors of transcription as compared to WT DRGNs. Strong depolarization, which normally represses neurite growth through activation of Ca_v1 channels, had no effect on neurite growth in DRGN

cultures from female cald-KO mice. Remarkably, DRGNs from cald-KO males were no different from those of WT males in terms of depolarization-dependent neurite growth repression. We conclude that caldendrin opposes neurite regeneration and growth, and this involves coupling of Ca_v1 channels to growth-inhibitory pathways in DRGNs of females but not males. Our findings suggest that cald-KO mice represent an ideal model in which to interrogate the transcriptional pathways controlling neurite regeneration and the mechanisms that distinguish these pathways in males and females.

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Poster

P019. Regeneration: PNS

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Program #/Poster #: P019.04

Topic: A.04. Transplantation and Regeneration

Support: Dr. Miriam and Sheldon G. Adelson Medical Research Foundation

Title: Loss of the axon initial segment inhibits axon regeneration

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Abstract: Peripheral nervous system (PNS) injury can be caused by a variety of traumas and has symptoms ranging from temporary numbness and mild pain to complete loss of feeling and motor control. Therapeutic targets for these injuries will require a comprehensive understanding of the mechanisms of regeneration. In neurons, the axon initial segment (AIS) is essential to generate the action potential and to maintain neuronal polarity. AnkyrinG (AnkG) is the master organizer of the AIS and functions to cluster voltage-gated sodium and potassium channels, as well as cell adhesion molecules. When AnkG is lost, the AIS is dismantled, leading to protein mistrafficking between somatodendritic and axonal domains. There are many examples of AIS disruption in injury and disease. However, the role of the AIS in axon regeneration is not well understood. We tested if the AIS is necessary for axon regeneration. We compared axon regeneration in control and AIS-deficient mice. AIS-deficient mice were generated by removing AnkG from motor neurons using *ChAT-Cre; AnkG$^{F/F}$* mice. We induced PNS injury at postnatal day 21 using a sciatic nerve crush model, then evaluated axon innervation of the lumbrical muscles in the foot. We used immunofluorescence microscopy and recorded compound muscle action potentials to determine if structural and functional recovery occurred after injury. Our data show that axon regeneration is significantly inhibited in the *ChAT-Cre; AnkG$^{F/F}$* mice compared to control mice. At 14 days post crush (DPC) there was 66% and 3% reinnervation of the NMJ in

control and *ChAT-Cre; AnkG$^{F/F}$* mice, respectively. Synapse function fully recovered by 21 DPC in control mice, but synapse function never completely recovered even by 35 DPC in *ChAT-Cre; AnkG$^{F/F}$* mice. Future work will focus on determining why loss of the AIS inhibits axon regeneration. Since the AIS has been shown to maintain polarity in neurons, we will combine mRNA and proteomic approaches to determine if trafficking is disrupted in regenerating axons that lack an AIS. These data will enable us to better understand the role of the AIS in PNS axon regeneration.

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Poster

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Topic: A.04. Transplantation and Regeneration

Support: The Health and Medical Research Fund (HMRF), Food and Health Bureau, Hong Kong Special Administrative Region Government (Ref. No.: 07181356)
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Title: Sustained axon regeneration and functional restoration induced by two FDA-approved bioactive small molecules

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Abstract: Complete motor functional recovery is virtually non-existent in patients with proximal peripheral nerve injury (PNI), largely due to the slow rate of axon regeneration leading to chronic target denervation after PNI. In central nervous system (CNS), most of the injured axons failed to regenerate due to the poor intrinsic regenerative capacity of adult neurons. The presence of growth-inhibitory molecules at the lesion site further limited the extent of axonal regrowth after CNS injuries. This remained the major challenge for scientists to identify effective drugs to enhance the intrinsic regenerative capacity and facilitate considerable axon regeneration after nerve injuries. Our pilot study demonstrated that *Lycium barbarum* polysaccharides (LBP), the major bioactive constituent in wolfberry, markedly enhanced the intrinsic growth capacity of axotomized peripheral neurons and accelerated *in vivo* sensory and motor functional recovery after severe PNI. To delineate the molecular mechanisms underlying the growth-promoting effects of LBP, we performed weighted gene co-expression network analysis (WGCNA) on the

microarrays of DRGs from LBP-treated mice. Our WGCNA indicated that LBP treatment induced up-regulation of key regeneration-associated genes and inhibited the expression of growth-inhibitory molecules. Subsequent *in silico* screening using LBP-induced key gene signatures identified two FDA-approved bioactive small molecules that recapitulated the growth-promoting effects of LBP. These two small molecules not only accelerated *in vivo* axonal regrowth after PNI, but also induced sustained axon regeneration after optic nerve crush (ONC) injury. Some of the injured axons can regenerate the entire length of optic nerve and reached optic chiasm 4 weeks after injury. To test if such extent of axonal regrowth was sufficient to elicit neural activity after ONC, we performed local field potential (LFP) recordings at superior colliculus (SC) upon optogenetic activation of injured retinal ganglion cells 6 weeks after ONC. While we failed to detect eye-evoked LFPs in vehicle-treated mice, mice treated with one of these two small molecules could elicit reliable eye-evoked LFP responses at SC 6 weeks after ONC. More remarkably, mice treated with this small molecule could partially restore the pupillary light reflex to constrict the pupil upon light stimulation to the injured eye. Taken together, our study highlighted the therapeutic potential of LBP and these two FDA-approved small molecules for functional recovery in patients with nervous system injuries. Further studies are required to optimize the dosage regimen for maximal clinical outcomes.

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Poster

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Topic: A.04. Transplantation and Regeneration

Support: NIH Grant HL093056
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Title: Cspg sulfation prevents sympathetic nerve regeneration following myocardial infarction

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Abstract: Sympathetic denervation of the heart following Myocardial Infarction (MI), which is sustained by chondroitin sulfate proteoglycans (CSPGs) in the cardiac scar, predicts risk of sudden cardiac death. Previous work demonstrated that blocking CSPG signaling restored

sympathetic axon outgrowth into the cardiac scar, decreasing arrhythmia susceptibility. Growth inhibition by CSPGs is thought to depend on the sulfation status of the glycosaminoglycans (GAGs) attached to the core protein. In the central nervous system, tandem sulfation of CSPGs at the 4th (4S) and 6th (6S) positions of n-acetyl-galactosamine is thought to be a primary inhibitor of axon outgrowth. It is not known if CSPG sulfation prevents sympathetic nerve regeneration. Our data suggest that CSPG sulfation prevents sympathetic axon outgrowth just as it alters regeneration in the CNS. Sympathetic neurite outgrowth across purified CSPGs is restored *in vitro* by removing 4S-CSPGs with the enzyme Arylsulfatase-B (ARSB). Neurite outgrowth across CSPGs was reduced to 69% of control levels while pre-treatment of CSPGs with increasing amounts of ARSB restored outgrowth to control levels in a dose dependent manner. Additionally, we co-cultured cardiac scar tissue with sympathetic ganglia *ex vivo* and found that removing 4S-CSPGs with ARSB restored axon outgrowth to control levels. Axon outgrowth towards infarcted myocardium was reduced to 66% of control (healthy myocardium) levels while treatment of infarcted myocardium with ARSB restored axon outgrowth to 97% of control levels. We examined levels of the enzymes responsible for adding and removing sulfation to CSPG-GAGs by western blot to determine if they were altered in the left ventricle after MI. We found that CHST11 (4-sulfotransferase) was downregulated, CHST15 (4S dependent 6-sulfotransferase) was upregulated, and ARSB (4-sulfatase) was downregulated after MI. This change was preceded by increased expression of Galectin 3, a key regulator of the inflammatory fibrotic response after ischemic injury. Increased CHST15 combined with decreased ARSB suggest a mechanism for production and maintenance of sulfated CSPGs in the cardiac scar.

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Poster

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Title: Transcriptomic changes toward brain remodeling in response to silk-based wearable bioreactor induced hindlimb regeneration in a non-regenerative animal model system

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Abstract: Advances in regenerative medicine and biomedical engineering are yielding tools with which the limbs of non-regenerative models can be regrown and repatterned following experimental amputations. The prospect of restoring form and function by initiating autopoietic tissue programs without genetic micromanagement is becoming a realistic possibility with significant clinical implications. However, as technologies rapidly develop, one significant set of challenges that must be addressed are the short- and long-term neuroplastic responses to injury and regeneration including neuropathic pain and the phantom limb phenomenon. Our silk-based wearable bioreactor - the Biodome - can successfully deliver time-released, multi-drug treatments to regrow and repattern limbs in *X. laevis* with new skin, muscle, bone, vasculature, and notable regrowth of peripheral nerve fibers and restoration of efferent neuromuscular connections. Indeed, sensorimotor responses in regenerated limbs were indistinguishable from non-amputated wild types, indicating complete regain of function. These results also include an evaluation of brain reorganization. Here, we present next generation sequencing (NGS) data from the brains of animals extracted 11hr, 24hr, and 7d post-amputation, comparing frogs treated with a Biodome intervention to animals receiving amputation only. We identified significant enrichment of genes associated with the reorganization of the brain's extracellular matrix (ECM), cell-cell interactions, porphyrin metabolism, and heme biosynthesis - particularly after 24hrs and 7d. These changes within the central nervous system reflect similar changes that we originally identified within the regenerating blastema proximal to the amputation site. A parallel immune signaling pathway revealed by transcriptomic analysis may represent a mechanism linking peripheral tissue repatterning and central nervous system plasticity. Together, these findings suggest the long-range transfer of information from local events in the distal hindlimb to the brain. In addition to revealing the systemic propagation of regenerative state information through the body, our data suggest the hypothesis that induced brain remodeling soon after intervention may functionally support long-term limb regeneration in non-regenerative systems.

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Poster

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Topic: A.04. Transplantation and Regeneration

Support: JSPS 19H03850

JSPS 19K10261

Title: Effects of cell extract from adipose-derived stem cells on peripheral nerve regeneration

Authors: *Y. IMAI[1], N. KISHIMOTO[1], Y. KOYAMA[1], J. TANUMA[2], K. TAKEUCHI[4], T. MAEDA[3], K. SEO[1];
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Abstract: *BACKGROUND*: Adipose-derived stem cells (ADSCs) have the potential to trans differentiate into other tissue types, such as nervous system or myocardial tissue. Cell extract produced from ADSCs (CE-ADSC) promotes functional recovery of the salivary gland, suggesting that some factors in CE-ADSC can induce tissue regeneration. This study aimed to investigate whether CE-ADSC has a regenerative effect on peripheral nerve tissue using incubated nerve cells. *METHODS*: ADSCs were isolated from adipose tissue from 8- to 15-week-old C57BL/6J mice, and CE-ADSC was then extracted by the repeated freezing and thawing of ADSCs. To evaluate the regenerative effects of CE-ADSC on nerve tissue, two experiments were performed. First, changes in proliferative capacity (using the MTT assay) and glial fibrillary acidic protein (GFAP) expression levels (using enzyme-linked immunosorbent assay) were compared before and after CE-ADSC application in immortalized rat Schwann cells (Cosmo Bio Ltd, Tokyo). In addition, the effects of CE-ADSC on rat dorsal root ganglion (DRG) neurons were evaluated by calculating the number of neurite-bearing cells (neurons with neurites that were longer than the cell body diameter) and by measuring the lengths of the neurites. Second; CE-ADSC that had been heated to 95℃ or treated with proteinase K was applied to the Schwann cells and DRG neurons, and the same changes in proliferative capacity and the number of neurite-bearing cells were measured. Statistical analysis was performed using *t*-tests or non-repeated measures analysis of variance (ANOVA), and the $p < 0.05$ was considered significant. *RESULTS*: (1) CE-ADSC increased proliferation (non-repeated measures ANOVA, $p < 0.01$) and GFAP expression (*t*-test, $p < 0.01$) in Schwann cells. (2) CE-ADSC increased the number of neurite-bearing cells (non-repeated measures ANOVA, $p < 0.01$) and the lengths of neurites (*t*-test, $p < 0.05$) in DRG neurons. (3) Heat or proteinase K treatment decreased the total amount of protein in CE-ADSCs; this inhibited both the proliferation of Schwann cells and the number of neurite-bearing cells (non-repeated measures ANOVA, $p < 0.05$). *CONCLSIONS*: Some proteins in CE-ADSC might have a regenerative effect on peripheral nervous cells. (This study was supported by JSPS 19H03850, 19K10261)

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Poster

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Program #/Poster #: P019.09

Topic: A.04. Transplantation and Regeneration

Title: Strategies to Optimize Functional Recovery in Distal Nerve Transfers

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Abstract: Strategies to optimize functional recovery in distal nerve transfers
Background: Distal nerve transfers (DNTs) are a popular reconstructive technique for peripheral nerve injuries. In this surgery, the distal motor targets of an injured nerve are reinnervated by a redundant branch of a nearby functioning nerve. As DNTs require the donor nerve to innervate new targets, synaptic plasticity is integral to success. Exercise therapy improves plasticity and enhances regeneration following a nerve injury, but its effects have never been evaluated in DNTs. We have recently shown that conditioning electrical stimulation (CES) significantly improves recovery following DNT. In this study we compare the effects of preoperative CES and postoperative exercise therapy on regeneration, reinnervation, and recovery following a lower limb DNT.**Methods**: Sprague-Dawley rats were divided into four cohorts i) CES, ii) exercise training, iii) CES and exercise training (CES-ExT), and iv) DNT. The common fibular nerve of all animals was crushed. Seven days later, CES was delivered to the tibial nerve. Fourteen days post-injury, a DNT was performed by coapting a tibial nerve branch to the common fibular nerve. Post-operatively, animals were run daily on a treadmill for either four ('short-term') or thirteen ('long-term') weeks. Outcomes included axonal extension, gait analysis, and skilled locomotion testing.**Results**: Animals treated with CES-ExT had significantly greater regeneration and functional recovery. The length of axon extension in CES-ExT nerves was significantly longer than other cohorts ($p<0.05$). These animals had significantly improvements on the horizontal ladder test and earlier recovery of dorsiflexion when compared to other cohorts. Restoration of a symmetrical gait was significantly improved ($p<0.01$) among CES-ExT treated animals, with increased weight-bearing and increased contact-time of the affected limb compared to the other groups ($p<0.01$). No significant difference was identified between short- and long-term exercise training.**Conclusions**: The combination of CES-ExT enhances regeneration and functional recovery following DNT. Both electrical stimulation and exercise training are clinically feasible, therefore, this perioperative regimen may be a strategy to significantly improve functional recovery for patients undergoing a DNT.

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Poster

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Dr. Miriam and Sheldon G. Adelson Medical Research Foundation

Title: Injury to the peripheral axon of dorsal root ganglia neurons alters the centrally projecting axonal transcriptome

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Abstract: The sensory neurons in dorsal root ganglia (DRG) are pseudounipolar, projecting one axon into the periphery and another into the spinal cord. Injury to the peripheral axon results in a robust regeneration response, whereas injury to the central axon does not. It is well established that initiation of an intrinsic growth program following injury to the peripheral nerve contributes to regeneration of the peripheral axon. Our lab has shown that mRNAs encoding growth-associated proteins, like growth-associated protein 43 (GAP-43), increase in the peripheral axons after injury, and the local translation of these mRNAs in the axon supports nerve regeneration. The possibility that peripheral nerve injury might also alter mRNA transport into centrally projecting sensory axons has not been tested. Alterations in transport and translation of mRNAs in centrally projecting sensory axons could alter their plasticity or function in the CNS, possibly contributing to pain signaling. We initially screened central dorsal roots for candidate mRNAs known to be increased in peripheral axons after axotomy. By RTddPCR using extruded axoplasm and FISH/IF, we found that *Gap-43* and *galanin* mRNAs are significantly increased in dorsal root axoplasm 7 days after sciatic nerve crush. Surprisingly, the increase in *galanin* mRNA is more substantial in the dorsal root than it is in the sciatic nerve. This initial screen raised the possibility that mRNA transport into centrally projecting DRG axons can be regulated differently than transport into the peripherally projecting axons. To systematically test this possibility, we performed RNA-Seq analyses of dorsal root axoplasm, sciatic nerve axoplasm, and DRG under naïve conditions and after sciatic nerve crush. We found 34 differentially expressed genes in the central axons and 1,223 in the peripheral axons after crush injury. 8 were seen in both the central and peripheral axons, while 16 were unique to the central axon and 1,087 unique to the peripheral axon. These results indicate that the transcriptome of the centrally projecting DRG axons is indeed altered by peripheral nerve injury, and mRNAs can be differentially directed to the periphery or to the central nervous system.

Disclosures: T. Smith: None. **J. Marryat:** None. **Q. Wang:** None. **R. Kawaguchi:** None. **J. Twiss:** None.

Poster

P019. Regeneration: PNS

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P019.11

Topic: A.04. Transplantation and Regeneration

Support: Dr. Miriam and Sheldon G. Aldelson Medical Research Foundation
NIH R01 NS117821

Title: Pura drives axonal localization of Calreticulin mRNA through conserved UTR motifs

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Abstract: Neurons are highly polarized cells with widely separated and specialized subcellular domains that are essential for function of the nervous system. Axons provide long-range connections in the nervous system and axons can extend from a few millimeters to a few meters depending on size of the organism. Axons locally synthesize proteins autonomous from their cell body that support growth, survival, and function of axons. Localization of mRNAs into axons is determined by proteins binding to sequence elements within the mRNA, and these are often present in the untranslated regions (UTRs) of the RNAs. Local translation of Calreticulin mRNA (Calr) in injured axons was reported to support initiation of regeneration. We have previously shown that Calr utilizes conserved regions in its 3' UTR to localize to axons. The 5' UTR of Calr is necessary for local translational regulation within axon but this regulation requires presence of a 3'UTR localization motif raising the possibility for common protein mediators binding to both UTRs, but the RNA binding proteins (RBPs) that mediate this post-transcriptional regulation of neuronal Calr mRNA are not known. We performed RNA affinity mass spectrometry (RAMS) analyses using the conserved Calr 5' and 3'UTR regions as baits to identify axonal proteins interacting with these motifs. 42 proteins were identified as high specificity interactors for Calr's 5' and/or 3'UTR motifs. Of these, only PurA and Ncl bound to both the functional 5' and 3'UTR Calr motifs. Using biotinylated RNA, PurA validated as precipitating with both the 5' and 3' UTR Calr motifs. RIP analyses for PurA protein immunoprecipitants, showed that endogenous Calr binds to these RBPs in sciatic nerve axoplasm. By high resolution single molecule FISH/IF, PurA colocalizes with Calr mRNA in axons of cultured rat DRG neurons. Depletion of PurA from these cultures decreased axonal Calr mRNA levels. Together, these data indicate that PurA is needed for the axonal localization and translation of the mRNA encoding the axon injury response protein Calreticulin.

Disclosures: M. Zdradzinski: None. **S.J. Lee:** None. **J. Oses-Prieto:** None. **A. Burlingame:** None. **J. Twiss:** None.

Poster

P019. Regeneration: PNS

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Program #/Poster #: P019.12

Topic: A.04. Transplantation and Regeneration

Support: Dr. Miriam and Sheldon G. Aldelson Medical Research Foundation

Title: Loss of the lipid phosphatase Fig4 results in decreased axonal mRNA transport and mitochondrial dysfunction

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Abstract: FIG4 mutations are causative for the neuropathic condition Charcot-Marie-Tooth Disease 4J [CMT4J]. Fig4 (also called Sac3) is a 5'-phosphoinositide phosphatase in the PI(3,5)P2 regulatory complex/PAS complex that plays an important role in endosome-to-lysosome maturation and vesicle trafficking. Considering the roles of endosomes/lysosomes in axonal transport, we performed proteomics on PNS axoplasm from $Fig4^{fl/fl}$ [WT] vs. $Fig4^{fl/fl}$ x Syn1-Cre [$Fig4^{-/-}$] mice deficient for Fig4 in neurons. These data show significant increase and decrease of proteins in $Fig4^{-/-}$ axons. Pathways disrupted by Ingenuity Pathway Analysis [IPA] included Ca^{2+} homeostasis and mitochondrial electron transport chain complex. IPA from transcriptomics data obtained from primary DRG neurons similarly pointed to disrupted oxidative phosphorylation following Fig4 depletion. Consistent with these findings, there is a marked loss in mitochondrial membrane potential in the axons of $Fig4^{-/-}$ neurons, with an exacerbated mitochondrial depolarization upon treatment with 2-deoxy-glucose to attenuate glycolysis. Recent reports show co-transport of axonal RNAs with endosomes/lysosomes. Axons from $Fig4^{-/-}$ neurons show a significant reduction of several nuclear-encoded mitochondrial mRNAs, both in culture and sciatic nerve in vivo. Taken together, these data point to a previously undetected disruption in axonal mRNA transport with the CMT4J-associated loss of Fig4. A decline in axonal mRNA levels of nuclear encoded mitochondrial proteins likely disrupting mitochondrial function. Given the importance of mitochondria in axonal health and survival through ATP production and Ca^{2+}-buffering, this raises the possibility that disruptions in endosome/lysosome-linked axonal mRNA transport contribute to CMT4J pathogenesis.

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Poster

P019. Regeneration: PNS

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Program #/Poster #: P019.13

Topic: A.05. Axon and Dendrite Development

Support: NIH Grant R01 NS050674
NIH Grant T32 AG000222

Title: The local translatome within central axons of dorsal root ganglia neurons

Authors: *E. SILAGI, M. PAZYRA-MURPHY, O. TASDEMIR-YILMAZ, R. SEGAL; Dana Faber Cancer Inst., Boston, MA

Abstract: Dorsal root ganglia (DRG) neurons are first-order neurons in the somatosensory pathway that communicate noxious and innocuous stimuli from the periphery to the central nervous system. DRG neurons are pseudo-unipolar, with one axonal branch that innervates peripheral tissues and another that synapses in the spinal cord and brainstem. Thus, DRG neurons provide an ideal system for understanding how spatially regulated mRNA localization and translation enable distinct subcellular protein compositions and functions in peripheral and central axons. We investigated local mRNA translation in the soma and axons of DRG sensory neurons from P0-P3 aged mice. In these mice, Nav1.8-Cre drives selective expression of a GFP-tagged ribosomal subunit, L10a-GFP, in many DRG sensory neuronal subtypes. Tagged ribosomes can be visualized in the DRG cell soma, in central axons in the spinal cord, and in peripheral nerves. Using these animals, we carried out Translating Ribosomal Affinity Purification (TRAP) followed by RNA sequencing of central axon and DRG cell bodies. Specifically, GFP-tagged ribosomes were purified from the lysates using anti-GFP antibody and the ribosomal-associated RNAs from DRG soma and central axon were isolated for sequencing (n=4; ~20 mice/set). Negative controls were prepared from L10a-GFP mice without Cre expression. Principal component analysis of GFP+ samples confirmed that central axon and DRG samples clustered separately yet have little variance within sample groups. The dataset identified 265 genes in the translatome of central axons and 1630 in the DRG; importantly, the soma and central axon included genes characteristic of multiple distinct DRG subtypes. In the central axon, significantly enriched Gene Ontology Biological Processes related to sensory perception and pain; meanwhile a significantly enriched Kegg Pathway was Neuroactive-ligand Receptor Interaction, containing genes associated with post-synaptic serotonin receptors. In contrast, the enriched biological processes in the DRG soma are related to microtubule transport,

RNA processing, and DNA damage. These data suggest that the DRG central axons may constitute a critical location for modulation of pain and somatosensory input by the serotonergic system. Further studies are ongoing to explore the role of local translation and the serotonergic pathway for modulating pain sensation.

Disclosures: E. Silagi: None. **M. Pazyra-Murphy:** None. **O. Tasdemir-Yilmaz:** None. **R. Segal:** None.

Poster

P028. Synaptic Structure

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Program #/Poster #: P028.01

Topic: B.06. Synaptic Transmission

Support: NSF 2014862

Title: Direct connections between the spine apparatus and the synapse are characteristic of large spine and are more frequent after associative learning

Authors: *E. PARRISH-MULLIKEN[1], L. OSTROFF[2];
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Abstract: The spine apparatus is a specialization of the smooth endoplasmic reticulum that is found in approximately 20% of all dendritic spines and is particularly associated with large, mature spines. The spine apparatus structure consists of membranous cisterns stacked between dense actin plates and it is involved in activity-dependent cytoskeletal changes of spine actin. These structural changes may be responsible for the undefined role the spine apparatus plays in synaptic plasticity and its association with calcium storage and synaptopodin. Additionally, the spine apparatus has been shown to form an actin-based physical connection to the post-synaptic density. However, detailed studies of its structural variability and distribution in the spine population are lacking, and whether every spine apparatus contacts the synapse is unknown. Synaptic plasticity in the lateral amygdala (LA) mediates aversive Pavlovian conditioning, and the spine apparatus is found more frequently in LA spines after this type of learning. Here, we investigated the structure of the spine apparatus and its physical connection to the synapse during the long-term memory phase of Pavlovian conditioning. Using serial section transmission electron microscopy, we examined reconstructions of the spine apparatus in 544 spines in the rat LA, and found that overall, the spine apparatus contacted the synapse only 18.9% of the time. The number of plates in the spine apparatus ranged from two to eight, and a connection to the synapse was associated with both more plates and a larger synapse. The spine apparatus was more likely to contact the synapse in the conditioned group, particularly if polyribosomes were

also present in the spine. Our results demonstrate that the connection between the spine apparatus and the synapse does not occur randomly, but instead is associated with synapse size and spine apparatus structure. These findings give further evidence that the spine apparatus is involved in synaptic plasticity and structurally plays a role through direct contact with the post-synaptic density.

Disclosures: E. Parrish-Mulliken: None. **L. Ostroff:** None.

Poster

P028. Synaptic Structure

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Topic: B.06. Synaptic Transmission

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Title: Nanoscale rules governing the organization of glutamate receptors in spine synapses are subunit specific

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Abstract: Dendritic spines are important sites for synaptic transmission and plasticity, and changes in their size and shape are thought to be structural correlates for learning and memory. Within the spine, the post synaptic density (PSD) is home to many proteins including MAGUK PSD-95 and glutamate receptors α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid receptor (AMPAR) and N-methyl-D-aspartate receptor (NMDAR). PSD-95 is organized in nanoscale puncta that scale in number, but not size as spine size increases. However, whether other postsynaptic proteins such as AMPAR and NMDARs share a similar organization is not known. Heterotetrameric glutamate receptors are essential for the development, function, and plasticity of spine synapses but how they are organized to achieve this is not known. Here we show that the nanoscale organization of glutamate receptors containing specific subunits define distinct subsynaptic features. Glutamate receptors containing GluA2 or GluN1 subunits establish nanomodular elements precisely positioned relative to Synaptogamin-1 positive presynaptic release sites that scale with spine size. Glutamate receptors containing GluA1 or GluN2B specify features that exhibit flexibility: GluA1-subunit containing AMPARs are found in larger spines,

while GluN2B-subunit containing NMDARs are enriched in the smallest spines with neither following a strict modular organization. Given that the precise positioning of distinct classes of glutamate receptors is linked to diverse events including cell death and synaptic plasticity, this unexpectedly robust synaptic nanoarchitecture provides a resilient system, where nanopositioned glutamate receptor heterotetramers define specific subsynaptic regions of individual spine synapses.

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Poster

P028. Synaptic Structure

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ANR NEUC-0001

Title: Calcium depletion versus replenishment conditions of the endoplasmic reticulum in dendritic spines are controlled by the nanoscale molecular architecture

Authors: *K. BASNAYAKE[1], D. MAZAUD[2], L. KUSHNIREVA[3], A. BEMELMANS[4], N. ROUACH[2], E. KORKOTIAN[5], D. HOLCMAN[6];
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Abstract: Dendritic spines are components of neuronal synapses receiving and transforming synaptic inputs into a succession of biochemical events regulated by calcium signaling. The spine apparatus (SA), an extension of smooth endoplasmic reticulum participates in slow and fast calcium transients inside spines.We previously showed that the fastest calcium ions arriving at ryanodine receptors (RyRs) present mostly at the base of the spines trigger calcium-induced calcium release events that deplete the SA calcium ion reservoir rapidly. However, the next signaling cycle requires refilling of these calcium stores, and how spines achieve such refilling without triggering calcium release remains unclear. In the present study, we employ a

combination of computational modeling & simulations along with calcium imaging and STED super-resolution microscopy to address this enigma. Our main goal is to determine the conditions under which SA-store refilling without triggering depletion through RyRs is possible via the store-operated calcium entry pathway involving ORAI1 channels. First, diffusion modeling revealed that a small amplitude and a slow timescale for the calcium ion influx is required not to trigger RyRs. We confirmed this prediction through calcium imaging of cultured hippocampal neurons (prepared as Korkotian & Segal 2000) under channel blockages letting only the ORAI1 calcium influxes into spines. We measured slow calcium transients with timescales in the order of 2s, contrasting with fast NMDA & AMPA currents that last only a few tens of milliseconds. Second, using diffusion simulations, we found that in spine heads, ORAI1 channels that allow calcium entry from the plasma membrane should be closely colocalized with SERCA pumps that refill the SA. We confirmed this organization with STED imaging of immunostained hippocampal tissues (prepared similar to Basnayake et al. 2019) and found that the average co-localization distance between SERCA and ORAI1 puncta in the spine heads containing SA have very short distances of a few tens of nanometers (106±4nm, n=378), while this distance is more than doubled (210±8 nm, n=290) at the bases of spines. Thereby, we delineated how spine's nanoscale organization determines efficiently the separation between SA replenishment and depletion conditions, preventing the simultaneous activation of both pathways. Finally, we conclude that the spine's nanoscale organization at the receptor level could underlie the differences of calcium dynamics during the induction of synaptic long-term potentiation versus depression, particularly at the region of the spine base.

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Poster

P028. Synaptic Structure

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P028.04

Topic: B.06. Synaptic Transmission

Support: Grant from Shinshu University

Title: New structure model for postsynaptic density, which contains tubulin as a key component.

Authors: *T. SUZUKI[1], N. TERADA[1], S. HIGASHIYAMA[2], K. KAMETANI[3], Y. SHIRAI[1], M. HONDA[4], T. KAI[4], W. LI[5], K. TABUCHI[1];
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Abstract: Structural changes in postsynaptic density (PSD) are important mechanisms for maintaining synaptic plasticity. The molecular mechanism underlying changes in PSD structure (PSD remodeling) is currently unknown. A complete understanding of the structure of PSD is indispensable to fully elucidate the molecular mechanisms of spine and PSD dynamics during the expression of synaptic plasticity. Currently, it is tacitly believed that PSD is formed as an assembly of various kinds of scaffold/adaptor proteins (Scaffold/adaptor assembly model). Based on our recent study on the purified PSD lattice, we propose a new structure model for PSD. In our initial study on this theme, we proposed "PSD lattice-based dynamic nanocolumn" model for the molecular architecture of excitatory PSD, in which the scaffold protein model and the PSD lattice model are combined (Suzuki et al., 2017, J Neurochemistry, 144: 390-497). Our latest model for PSD structure, which we provide in the meeting, is an expanded version of the above mentioned model with the addition of the concept of PSD lattice backbone structure. We found a PSD lattice backbone structure, which is constructed mainly with non-microtubule tubulin and almost free from typical PSD scaffold/adaptor proteins. In the new model, non-microtubule tubulin may play an important role as a platform to which PSD scaffold/adaptor proteins and various PSD-functioning molecules become associated while synapses mature and reorganize. Proteins associating with the PSD lattice include various proteins related with versatile functions at the postsynaptic site as well as postsynaptic scaffold/adaptor proteins. Postembedding immuno-gold EM using anti-tubulin antibody supports the role of tubulin as an essential component for PSD structure by demonstrating the presence in and around PSD in the mouse brain. Forebrains of rats (male, adult) were used for purification of the synaptic fraction.

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Poster

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Program #/Poster #: P028.05

Topic: B.06. Synaptic Transmission

Support: NIH Grant EY012141

Title: 3d sted super-resolution imaging of the ground squirrel cone photoreceptor synapse

Authors: E. A. SCHWARTZ[1], V. BINDOKAS[2], J. SHI[3], ***S. DEVRIES**[4];
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Abstract: The mammalian cone photoreceptor synapse plays a crucial role in daylight vision by faithfully transmitting signals associated with fine spatial and color discrimination. EM studies

reveal a compact, ~3 µm diameter concave synaptic surface that is supplied by ~20 invaginating ribbon release sites and juxtaposed with hundreds of discrete bipolar cell dendrites. Both geometry and electrophysiological responses suggest that transmitter diffusion plays an important role in shaping the responses of the bipolar cell types. While EM studies can reveal the detailed physical structure of the cone synapse, they are less informative about the localizations of cleft components that can interact with diffusing transmitter including receptors and transporters. To address this gap in our understanding at the nanoscale level, we have devised a 3D-STED microscopic technique that allows us to visualize up to 5 proteins at a time at high resolution. To obtain images, bipolar cells in some retinas were sparsely labeled with GFP by prior intravitreal injection of AAV2. Ground squirrel retinas were removed from eyecups and bathed in cold glyoxal fixative for 2 days. Both untreated and GFP-expressing retinas were labeled with primary antibodies to neuronal markers (including GFP) and synaptic proteins. Species-specific secondary antibodies were labeled in-house with Abberior and Biotium fluorophores emitting in the range of 488-680 nm. Tissue was embedded in melamine, cover slipped, and imaged with a Leica SP8 Scanning Confocal/STED Microscope. Images were processed by denoising followed by deconvolution and surface-fitting with custom routines. We found that both Off cb1a and On cb7a bipolar cells selectively contacted the center of the cone terminal. Other bipolar cell types showed less positional selectivity. Small clusters of EAAT5 transporter (70±2 per terminal) were distributed throughout the concavity and did not align with the contacts of either basally contacting Off bipolar cells (cb3a/b type) or GluK1 receptor densities. Additionally, while GluR4 receptors where localized in tight clusters close to ribbons and their anchoring protein bassoon, some On bipolar mGluR6 and Off bipolar GluK1 receptors were found in nearly equal densities both in apposition to the cone terminal and along the dendritic shafts beneath the cone terminal. Our data suggests an unexpected bullseye organization of contacts at the cone synapse, a widespread but discrete localization of EAAT5 glutamate transporters, and differences in the dendritic localization characteristics of AMPA (GluR4), kainate (GluK1), and metabotropic (mGluR6) glutamate receptors.

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Poster

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Topic: B.06. Synaptic Transmission

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Title: Differential nanoscale organization of excitatory synapses onto excitatory vs inhibitory neurons

Authors: *A. D. LEVY, P. A. DHARMASRI, T. A. BLANPIED;
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Abstract: The strength of excitatory synaptic transmission relies on the precise, nanometer-scale organization of synaptic proteins. Within single synapses, presynaptic release proteins and postsynaptic receptors and scaffolds concentrate in ~80 nm sub-regions of higher protein density, termed nanoclusters (NCs). Pre and postsynaptic NC alignment across the synaptic cleft results in stronger action potential-mediated excitatory postsynaptic currents, demonstrating a role for nanostructure in regulating synaptic strength. One context in which this may be particularly important is dictating excitatory drive onto diverse cell types in a circuit. For example, in the hippocampus, excitatory synapses are made onto both dendritic spines of excitatory principal neurons as well as dendrites of parvalbumin (PV)-expressing fast-spiking interneurons. These cellular and circuit contexts require different excitatory drive - compared to principal neurons, excitatory drive onto PV neurons must be stronger to overcome the electrotonic limitations of a non-compartmentalized synapse and sustain fast spiking, enabling PV neurons to deliver rapid and precise inhibition. Despite these different requirements, excitatory synapses onto both cell types consist largely of the same widely conserved group of proteins. Therefore, we reasoned synapses on these two cell types might differentially utilize nanoscale organization to tune the strength of excitation. We used immunocytochemistry and a combination of confocal and super-resolution microscopy to compare organization of common scaffold proteins at these synapses. We find that PV neurons have larger presynaptic Munc13 and postsynaptic PSD95 puncta, consistent with stronger synapses. However, while the larger Munc13 puncta contained more Munc13 per area, PSD95 did not, suggesting a possible nanostructural difference at these two cell types. Indeed, Munc13 and PSD95 at PV cells were organized into more NCs (Munc13: 1.85x, PSD95: 1.35x) that were larger (Munc13: 1.7x, PSD95: 1.3x) than their excitatory cell counterparts. Analysis and modeling of protein localization autocorrelation, which measures nanoscale heterogeneity of protein distribution, indicates both Munc13 and PSD95 are more concentrated into nanoclusters at PV cells. These results suggest that while excitatory synapses onto PV neurons are both larger and have more protein, the nanostructure is also optimized in such a way that predicts larger responses to action potential-evoked release. More broadly, we conclude that specialized aspects of nanostructural organization may contribute to functional diversity of excitatory synapses.

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Poster

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Topic: B.06. Synaptic Transmission

Support: DFG LU 1819/2-1 grant
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Max Planck Society

Title: Large molecular assemblies are involved in synaptic transmission

Authors: *C. PAPANTONIOU[1], C. IMIG[2], A. MARTINEZ-SANCHEZ[1], U. LAUGKS[1], Z. KOCHOVSKI[1], N. BROSE[2], W. BAUMEISTER[1], B. H. COOPER[2], V. LUCIC[1];
[1]Mol. Structural Biol., Max Planck Inst. of Biochem., Martinsried, Germany; [2]Mol. Neurobio., Max Planck Inst. of Exptl. Med., Goettingen, Germany

Abstract: Neurotransmitter release is mediated by a complex and highly regulated molecular machinery. We imaged isolated synapses by cryo-electron tomography (cryo-ET), a method that utilizes faithful sample preservation and direct 3D imaging of fully hydrated, unlabeled biological material at single nanometer resolution. Our quantitative structural characterization of the presynaptic terminal showed that Munc13 and SNAP25, proteins essential for synaptic vesicle priming for release and fusion, organize synaptic vesicles at the plasma membrane. Namely, these two proteins are necessary for two different types of tethers that position synaptic vesicles with a single nanometer precision. Furthermore, Munc13 and SNAP25 reorganize both tethers and short molecular bridges that interlink synaptic vesicles (termed connectors) in a coordinated fashion. This implies that synaptic vesicle priming and further steps towards the fusion involve extended molecular assemblies comprising multiple tethers and connectors that structurally organize synaptic vesicles on the nanoscale. In a separate set of cryo-ET experiments, we showed that molecular trans-synaptic assemblies link synaptic vesicles and tethers postsynaptic ionotropic glutamate receptors. Together, our data leads to a model where synaptic transmission is governed by large molecular assemblies of variable architecture that may involve multiple weak interactions.

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Poster

P028. Synaptic Structure

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Program #/Poster #: P028.08

Topic: B.06. Synaptic Transmission

Title: Correlative STED and Calcium Imaging of Quantal Events in Dendritic Spines

Authors: *H. JAFRI[1], M. B. DALVA[2];
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Abstract: Dendritic spines are the primary site of glutamatergic transmission in the brain. Spines are thought to be the site of plasticity in the brain, and changes in spine size are associated with synaptic and structural plasticity. Moreover, spine size is associated with increased synaptic strength and increased size of the postsynaptic density. The synapse within a spine is thought to consist of a single presynaptic release site, and contain vesicles, the active zone (AZ), precisely opposed to the postsynaptic density (PSD), containing glutamate receptors and scaffolding proteins. Super-resolution microscopy has revealed that key pre- and postsynaptic molecules, such as Synpatophysin-1, VGlut1, Bassoon, and PSD95, form nanopuncta that scale in number, but not size, with respect to spine area. These data suggest that both pre- and postsynaptic proteins may form modular structures termed “nanomodules” or “nanocolumns”. While 60% of spines contain a single nanomodule, larger spines contain two or more pairs of pre- and postsynaptic protein modules. However, it is not known how or whether these clusters of proteins relate to synaptic function. The relationship between the nanoscale organization of the synapse and presynaptic function in single dendritic spine synapses is not well understood. To explore this further, we developed an approach to visualize spontaneous events at spines with known numbers of nanomodules. Cortical neurons were transfected with FingR PSD-95-mturquoise2 (CFP) to label endogenous PSD-95 in spines, and GCaMP7f to visualize calcium signaling. The number of nanomodules in spines was determined with super resolution, Stimulated Emission Depletion (STED), nanoscopy of FingR PSD-95. In the presence of blockers of neuronal activity, AMPARs, calcium channels, and calcium stores, spontaneous calcium events were visualized within spines with identified numbers of PSD-95 nanomodules using GCaMP7f. Using line- and field-scanning, we quantified the kinetics of mini-synaptic calcium transients (mSCTs) in dendritic spines with one nanomodule of PSD95. The smallest, one quanta, events were significantly above noise and exhibited rise and decay kinetics expected of GCaMP7f. The frequency of mSCTs within single spines was 10 to 100 times less than the reported frequency of mEPSPs obtained by whole-cell patch clamp. The distribution of peak amplitudes of mSCTs was observed to fit a Poisson distribution exhibiting evenly spaced bins of increasing amplitudes, suggesting the events seen were quantal and multivesicular. Taken together our data challenge the ideas that central synapses undergo univesicular release.

Disclosures: H. Jafri: None. **M.B. Dalva:** None.

Poster

P028. Synaptic Structure

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P028.09

Topic: B.06. Synaptic Transmission

Support: MH081060
MH070727
MH66198

Title: Probing spontaneous and evoked neurotransmission using fluorescent neurotransmitter sensors

Authors: *C. WANG[1], N. L. CHANADAY[1], L. M. MONTEGGIA[3], E. T. KAVALALI[2]; [2]Vanderbilt Brain Inst., [1]Vanderbilt Univ., Nashville, TN; [3]Vanderbilt Brain Inst., Nashville, TN

Abstract: Synaptic transmission forms the basis of neural communication. Although previously thought to be similar in structure and function, recent work has demonstrated that spontaneous and evoked neurotransmission have unique molecular mechanisms and activate distinct downstream signaling pathways. However, their relative organization at the synaptic level remains incompletely understood. Further understanding of this organization can help provide insight into the fundamental structure of synaptic transmission. Here, we show that iGluSnFR, a fluorescent probe to visualize glutamate release, can be used as an imaging tool to visualize glutamatergic activity at the single synapse level within primary hippocampal cultures. Our results suggest that evoked release sites are likely clustered within single synapses, while spontaneous release sites are more mobile and more diffusely spread across the synapse. Collectively, our results demonstrate iGluSnFR imaging can be used to probe the organization of distinct forms of neurotransmission at the synapse, creating a broader framework to understand basic principles of neurotransmission.

Disclosures: C. Wang: None. **N.L. Chanaday:** None. **L.M. Monteggia:** None. **E.T. Kavalali:** None.

Poster

P028. Synaptic Structure

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Program #/Poster #: P028.10

Topic: B.06. Synaptic Transmission

Support: NIH Grant R37 MH080046
NIH Grant F31 MH117920

Title: Ionotropic glutamate receptors coordinate trans-synaptic scaffold nanostructure

Authors: *P. A. DHARMASRI, A. D. LEVY, M. ANDERSON, T. A. BLANPIED;
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Abstract: Information processing in the brain relies on the dynamic and precise organization of molecular ensembles at the excitatory synapse. Proteins subserving the mechanisms of synaptic transmission are heterogeneously organized on the nanometer scale. Presynaptic proteins such as Munc13 and RIM1 are found in ~80 nm diameter subsynaptic regions of high local density. These presynaptic *nanoclusters* (NCs) are aligned across the synapse with NCs of proteins such as PSD-95 and ionotropic glutamate receptors (iGluRs), forming a *trans-synaptic nanocolumn*. Our lab established that this nanocolumn is the preferential site of action potential evoked release and that dispersal of AMPARs from the nanocolumn to the remainder of the synapse specifically reduces the strength of the response to evoked release. This highlights how the nanocolumn is critical for synaptic transmission and underscores the importance of knowing what generates and regulates this alignment. We reasoned that a mechanism underlying trans-synaptic alignment would involve a protein that could act as either a structural or functional marker of successful transmission. Intriguingly, the iGluRs themselves embody both aspects. First, either AMPARs or NMDARs could trigger downstream mechanisms that sample receptor activation and direct receptor positioning. Second, often underappreciated, both AMPARs and NMDARs have an extended extracellular structure with distal N-terminal domains that could nucleate direct or indirect interaction with presynaptic proteins. iGluRs could also organize scaffold NCs through multivalent interactions, either through NMDAR c-tail or AMPAR TARP binding. Previous studies show that receptor structure, particularly that of the AMPAR subunit GluA2 and its N-terminal domain, influences synaptogenesis and presynaptic maturation. Thus, we hypothesized that the iGluRs align the trans-synaptic nanocolumn. To test this, we assayed nanoscale organization of Munc13, RIM1, and PSD95 in a total iGluR knockout mouse model as well as in individual knockouts of the GluA2 AMPAR subunit and NMDARs in rat primary hippocampal culture. Here we report that loss of iGluRs, even just GluA2, results in changes to pre- and postsynaptic nanoscale organization and strongly disrupted alignment. On-going experiments seek to elucidate the structural or functional influence of iGluRs. The organization of iGluRs at the synapse is typically thought to be downstream of pre-existing scaffold structure. This study instead presents an unorthodox view of AMPA and NMDA receptors: that they actively shape the structure of synapses and thus may dictate their own activation.

Disclosures: P.A. Dharmasri: None. **A.D. Levy:** None. **M. Anderson:** None. **T.A. Blanpied:** None.

Poster

P052. Rett Syndrome I

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Program #/Poster #: P052.01

Topic: A.07. Developmental Disorders

Support: NIH Grant P50HD103537
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Title: Safety and efficacy of genetic MECP2 supplementation in the R294X mouse model of Rett syndrome

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Abstract: Rett syndrome (RTT) is an X-linked neurodevelopmental disorder caused primarily by mutations in the *MECP2* gene, encoding transcriptional modulator methyl-CpG-binding protein 2. While no disease-modifying therapies are currently available for RTT, several proposed strategies, such as gene therapy, aim to restore levels of functional MeCP2. An important consideration for these strategies is the tolerable threshold of MeCP2, since *MECP2* gene duplication and subsequent gain of MeCP2 function causes a related neurodevelopmental disorder in people (*MECP2* Duplication Syndrome, or MDS). Furthermore, determining the tolerable threshold of MeCP2 for partial loss-of-function mutations could elicit mutation-specific safety profiles of MeCP2 supplementation. We addressed these questions by genetically modelling MeCP2 supplementation in the context of the partial loss-of-function R294X allele. To do so, we utilized a mouse model of MDS with a human *MECP2* transgene on an autosome (TG). We conducted a broad behavioral battery separately in male and female cohorts to evaluate rescue of RTT-like phenotypes and assess behaviors indicative of excess MeCP2. We found that male and female R294X mice share several RTT-like phenotypes, including impaired locomotor activity, breathing dysfunction, and decreased motor coordination. Additionally, male and female MDS model mice displayed opposing phenotypes, including enhanced motor coordination and enhanced fear memory. In males, RTT-specific phenotypes were rescued and bidirectionally dosage-sensitive phenotypes were normalized with genetic MeCP2 supplementation. However, female R294X TG mice demonstrated behavioral evidence of excess MeCP2 in motor coordination and forced gait assays, suggesting that the underlying motor circuitry is particularly sensitive to MeCP2 dosage in females. Quantitative Western blotting of brain tissue determined that R294X TG female mice express full-length MeCP2 at approximately 1.5x that of WT female mice. This is likely due to X-chromosome inactivation and may explain the sex specificity of these MDS-like behaviors. Collectively, these results suggest that MeCP2 supplementation is safe and effective in males and largely so in females. However, careful consideration of risk for adverse motor effects may be warranted for girls and women with RTT. While mutation type may not have impacted the therapeutic window of MeCP2 supplementation

in this study, mutation type does significantly impact clinical severity in people. Future work will investigate the mechanism of an understudied subset of mutations at the C-terminus of *MECP2*, which cause relatively mild disease in people.

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Poster

P052. Rett Syndrome I

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Program #/Poster #: P052.02

Topic: A.07. Developmental Disorders

Support: The Rett Syndrome Research Trust Heart Grant
Battelle Research Exchange Fellowship
Nationwide Chidlren's Hospital Research Exchange Trainee Association Fellowship

Title: Directly Converted Rett Syndrome Patient Astrocytes Show Mutation Related Gene Expression and Metabolic Profiles

Authors: *S. POWERS[1], K. KINLEY[1], C. N. DENNYS[1], R. RODRIGO[1], X. ZHANG[1], F. ROUSSEL[1], S. B. LIKHITE[1], Y. RODRIGUEZ[1], S. SINHA RAY[1], M. DUNNING[2], P. R. HEATH[2], L. FERRAIUOLO[2], K. C. MEYER[1];
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Abstract: Confidential: Rett syndrome (RTT) is an X linked neurodevelopmental disorder affecting approximately 1 in 10,000 girls. Patients exhibit a loss of established developmental milestones and progressive loss of motor function. Cellular pathology is characterized by a global compaction of neural somas with shortened and fewer neurites. Most RTT cases result from loss of function mutations in methyl-CpG binding protein 2 (MeCP2), an important epigenetic reader and transcription factor. MeCP2 functions by silencing/enhancing genes by interaction with various cofactors, but its mechanisms are poorly understood. Clinically, variation in MeCP2 mutations lead to differential disease severity. To further investigate the role of MeCP2 mutations on pathology, we developed a novel *in vitro* modeling system utilizing induced neural progenitor cells (iNPCs) directly converted from RTT patient fibroblasts. Using this system, we examined the morphology of neurons derived from RTT patients. We also examined the non-cell autonomous role of astrocytes in RTT through a co-culture assay with mouse neurons via automated imaging/analysis. We found that RTT neurons and astrocytes have a mutation dependent effect on neuronal morphology. To investigate the molecular mechanisms

underlying morphological differences, we performed microarray gene expression analysis, and metabolic (seahorse) screening on our patient derived cells. We found that RTT phenotypes had a basis in substantially different gene expression profiles and mutation specific differences in metabolic profiles. Biomarkers for RTT phenotypes *in vitro* identified in these screenings have been confirmed in RTT patient brain tissue and independently by our collaborators in the Gogliotti/Niswender laboratories. Our work offers new perspectives on mutation specific Rett syndrome phenotypes and their connection with mutation dependent changes in metabolism, and gene expression.

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Poster

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Program #/Poster #: P052.03

Topic: A.07. Developmental Disorders

Support: HD088832
HD064817

Title: Mitochondrial ROS-induced stress is atherapeutic target for Rett syndrome

Authors: *J. DI LUCENTE[1], K. BORKOWSKI[3], J. NEWMAN[3], H. WULFF[2], G. CORTOPASSI[4], L.-W. JIN[1], I. MAEZAWA[1];
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Abstract: Rett syndrome (RTT), a devastating neurodevelopmental disorder, is largely caused by loss-of-function mutations in the X-linked gene encoding MeCP2. MeCP2 is a “reader” of the epigenetic codes consisting of DNA methylation marks, the absence of which results in altered gene expression. However, the mutant-driven toxic mechanisms causing RTT remain unclear. In our studies of RTT microglia, we found that MeCP2 deficiency caused enhanced mitochondrial (mt) reactive oxygen species (mtROS) production and abnormal mt function, prompting us to hypothesize that MeCP2 controls coordinated expression of bioenergetic genes as part of the recently recognized “mitonuclear signaling pathways" to maintains mt integrity, including mt

stress response (MSR). We found enhanced mtROS level, impaired OXPHOS complex I and complex IV activities, and decreased mt DNA copy number in the brain of *Mecp2*$^{tm1.1Bird/+}$ (*Mecp2-KO*) model of RTT. Furthermore, the MSR to mtROS was impaired as evidenced by reductions of ATF4 and Nrf2, two transcription factors critical for orchestrating MSR. To test the translational significance of mtROS, we employed the "mCAT" approach, in which catalase, usually not expressed in mt, was engineered to target mt to decompose overproduced H_2O_2. mCAT expression extended the lifespan of *Mecp2-KO* mice from 6-8 weeks to up to 42 weeks, and recovered the body weight gain, phenotype score, and multiple measures of respiration, neuronal plasticity, behavioral and motor performance, neuronal morphology, and mitochondrial function to almost wild-type levels. Integrated transcriptomics and metabolomics analyses demonstrated a mechanism consistent with ATF4- and Nrf2-orchestrated MSR being the main target of mCAT. To translate our findings to clinical utility, we screened a library of 1,600 FDA-approved drugs based on their "mitoactive" properties and identified dyclonine as the most promising candidate. Daily intraperitoneal injection of dyclonine at 10mg/kg to *Mecp2-KO* mice activated ATF4 and Nrf2, and showed a therapeutic efficacy parallel to mCAT expression. Together, our data provide proof-of-principle that mtROS-induced mt stress is a therapeutic target and that the FDA-approved drugs able to enhance MSR such as dyclonine are promising clinical candidates for RTT.

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Poster

P052. Rett Syndrome I

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Program #/Poster #: P052.04

Topic: A.07. Developmental Disorders

Support: IRSF Grant 3609

Title: Reversal of neurological defects by painless Nerve Growth Factor in a mouse model of Rett syndrome

Authors: A. TIBERI[1], G. BORGONOVO[1], G. TESTA[1], M. CALVELLO[1], ***S. CAPSONI**[1,2], A. CATTANEO[1,3];
[1]Scuola Normale Superiore, Pisa, Italy; [2]Univ. of Ferrara, Ferrara, Italy; [3]European Brain Res. Inst., Rome, Italy

Abstract: Rett syndrome (RTT) is a severe neurological disorder affecting 1 in 10,000 females. At the moment, there is no pharmacological treatment able to prevent or revert this neurodevelopmental disorder. We decided to investigate the efficacy of a neuroprotective

approach to counteract the neuronal loss occurring in RTT. We focussed on the neurotrophin Nerve Growth Factor (NGF) because of i) its reduced levels in RTT brain tissue , ii) of its direct neurotrophic actions on basal forebrain cholinergic neurons, that are affected in the brain of RTT patients and iii) of its broader indirect neuroprotective actions mediated by microglia cells. Despite a strong rationale, the clinical application of NGF in RTT, as well as in other neurological diseases, has been hampered by the fact that NGF cannot cross the blood brain barrier and has a strong pro-nociceptive activity. To overcome these limits, (i) we have developed painless NGF (hNGFp), a variant of NGF with a tenfold reduced pain-sensitizing activity; (ii) we demonstrated that it can be effectively delivered to the brain using an intranasal route of administration and that (iii) hNGFp have a potent neuroprotective activity, also on neurons that do not express NGF receptors, that is carried out through microglia and astrocytes. This is particularly relevant for Rett syndrome, since it has been recently suggested that conditioned medium from Mecp2 null microglia decreases neuronal branching and the number of synapses. Here, we report data obtained from two separate treatments with hNGFp performed in heterozygous $Mecp2^{+/-}$ female mice (RTT mice). The first treatment started when hindlimb clasping appeared and lasted 1 month. The second treatment started at 2 months of age in absence of any apparent sign of deficits and lasted until we had to humane sacrifice the mice due to breathing difficulties. We found that the administration of hNGFp enhanced the survival of RTT mice with 93% of hNGFp treated RTT mice surviving against 40% of PBS treated mice after 24 weeks of treatment. The neurological phenotype was also improved since we found that in both groups of treatment there was a statistically significant reduction of hindlimb clasping, immobility, gait and breathing deficits. These findings were accompanied by an improvement of brain weight. At the cellular level, we found that hNGFp restores the ramified, resting morphology of microglia and reduces the asthenic phenotype of astrocytes. Data on neuronal and inflammatory markers will also be reported. In conclusion, our study shows that hNGFp is able to ameliorate RTT-like neurological defects in Mecp2 null mice, putting forward a promising therapeutic agent for this disease.

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Poster

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Topic: A.07. Developmental Disorders

Support: AMED Brain/MINDS Grant JP21dm0207001
NPO Rett Syndrome Supporting Organization Grant

Title: Generation and analysis of MECP2 mutant marmosets

Authors: *N. KISHI[1,2], K. SATO[3], J. HATA[1,4], M. OKUNO[1], T. ITOU[1], J. OKAHARA[1,3], H. J. OKANO[1,4], E. SASAKI[1,3], H. OKANO[1,2];
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Abstract: Rett syndrome is an X-linked neurodevelopmental disorder presenting almost exclusively in girls, with a prevalence rate of one in 10,000 to 15,000. After Down's syndrome, Rett syndrome is considered the second most common genetic cause of cognitive disability in girls. Children with Rett syndrome develop relatively normally for 6-18 months, after which they undergo a period of rapid regression, with loss of purposeful hand use, deceleration of head growth, and onset of repetitive, autistic behaviors. Mutations of the MECP2 gene on the X chromosome are found in over 95% of cases of classic Rett syndrome, and MECP2 has been implicated in several other neurodevelopmental disorders, including autism, childhood schizophrenia, and X-linked cognitive disability. Although it is evident that phenotypes of Mecp2 mutant mice and symptoms of Rett syndrome are attributable to lack of the Mecp2 gene in the CNS, there is little neuropathological understanding of the abnormalities in the CNS of Mecp2-null mice as well as of patients with Rett syndrome. Although rodent models have been used for these studies, these models do not faithfully recapitulate human neurodevelopmental processes. The establishment of nonhuman primate (NHP) models that are similar to humans in many aspects, such as brain structure and higher brain functions, is very important to understand the pathogenesis of Rett syndrome. To add to our understanding of pathological mechanisms of Rett syndrome, we created MECP2 mutant marmosets suitable for research on Rett syndrome, using genome editing technology, and have analyzed the NPH models. MRI imaging shows that the brain size of MECP2 +/- marmoset was smaller than wild-type ones by approximately 15% at 24 months of age, and less active than wild-type ones in the daytime. Furthermore, we also generated MECP2-null marmosets using CRISPR/Cas9, which have smaller brains and are less active even at early developmental stages. Those results indicate that those MECP2 mutant marmosets recapitulate symptoms of Rett syndrome, and this new primate model will potentially contribute to the development of future therapeutic strategies for patients with Rett syndrome.

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Poster

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Topic: A.07. Developmental Disorders

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The Association Française du Syndrome de Rett
INSERM Transfert

Title: Contribution of the adipocyte hormone leptin in the pathogenesis of Rett syndrome

Authors: *Y. BELAIDOUNI[1], D. DIABIRA[1], J.-C. GRAZIANO[2], C. MENUET[1], S. APPLEYARD[3], G. WAYMAN[3], J.-L. GAIARSA[1];
[1]INMED-INSERM, [2]CRCM-INSERM, Aix-Marseille Univ., Marseille, France; [3]Integrative Physiol. and Neurosci., Washington State Univ., Washington, WA

Abstract: Rett syndrome (RTT) is a non-inherited chromosome X-linked neurodevelopmental disorder characterized by a loss of acquired speech, hand stereotypies and gait abnormalities which appear following a period of apparently normal postnatal development. Other frequent symptoms include breathing difficulties, seizures, scoliosis and growth retardation. This syndrome is caused by mutations in the X-linked chromosome gene *Methyl-CpG-binding protein 2* (*MECP2*) encoding a transcriptional regulator. RTT accounts for up to 10% of severe intellectual disability of genetic origin in women. Currently there are no cures to treat this disease. In spite of different causative mutated gene and biological pathways affected, epidemiological and animal studies revealed abnormally elevated circulating levels of the adipocyte hormone leptin in patients with RTT and *Mecp2*-deficient mice. Besides its canonical role in the control of satiety and energy expenditure, leptin acts on many brain areas, and in doing so modulates cognitive functions, anxiety, breathing and more. Consequently, altered levels of leptin may contribute to the RTT-associated neuronal network dysfunctions. In the present study, we show that serum leptin levels are elevated in early (postnatal day (P) 30) and late symptomatic (P60) male *Mecp2-null* mice (*Mecp2$^{tm1\text{-}1}$ bird*, a well-established rodent model of RTT). We also show that daily subcutaneous injections of a validated leptin antagonist (Peg-SMLA, a competitive inhibitor of leptin BBB transport), during 10 days from P40, prevent the body weight loss, restore the hippocampal excitatory/inhibitory (E/I) balance (electrophysiological recordings on slices), slows down the progression of breathing dysfunctions (plethysmography) in the symptomatic male *Mecp2-null* mice. Conversely, leptin-treatment of wild type mice mimics some of the phenotypic manifestations observed in *Mecp2-null* mice, i.e. decreased GABAergic activity, increased hippocampal E/I balance, and increased number of apneas. These data provide further insights on the possible contribution of leptin in RTT pathogenesis and offer therapeutic perspective to alleviate RTT symptoms.

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Poster

P052. Rett Syndrome I

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Topic: A.07. Developmental Disorders

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Title: Modeling from single cell electrophysiology to neuronal network interactions in human induced pluripotent stem cell derived Rett Syndrome and isogenic controls

Authors: *K. S. PRADEEPAN[1,3], G. BENIGNO[2,4], W. ZHANG[5], R. S. MOK[6,7], M. W. SALTER[5], J. ELLIS[6,7], J. C. MARTINEZ-TRUJILLO[1,3,4], L. E. MULLER[2,4,3];
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Abstract: Rett syndrome (RTT) is a rare neurodevelopmental disorder that produces a distinct period of normal infancy followed by neurological regression. RTT is caused by a single heterozygous loss-of-function mutation in the gene methyl-CpG-binding protein 2 (*MECP2*) found on the X-chromosome - affecting almost exclusively females. Electrophysiological studies in mouse cortical slices, hippocampal cultures, and human induced pluripotent stem cell (hiPSC) models of RTT have shown that MECP2 mutant neurons exhibit decreased excitatory neuronal activity and synaptic connectivity. While these effects have been extensively studied at the level of single neurons, how these changes translate to network-level activity patterns remains unknown. Here we combine analysis of multielectrode array (MEA) recordings of hiPSC networks (Ellis Lab, SickKids, Toronto), single-cell intracellular recordings of hiPSC neurons (Salter Lab, SickKids, Toronto), and computational modeling to understand the network-level mechanism of RTT. By developing a tightly constrained computational modeling framework from single-neurons and networks, we find that an interplay between fast excitatory currents at the network level and slow adaptation currents at the single-neuron level can underlie changes in population spiking activity in hiPSC RTT (MECP2-null) networks.
hiPSC-derived networks exhibited bursting patterns that went from sparse firing to asynchronous bursting to synchronous bursting across 6 weeks of development. RTT networks exhibited significantly slower frequencies of network bursts compared to isogenic controls. Using data from intracellular recordings in vitro, we developed a single-neuron model to capture the subthreshold membrane potential and spiking dynamics following standard current injection protocols. We next implemented our single neuron models into a spiking network model of the excitatory hiPSC populations. The spiking network model revealed that increased adaptation currents at the single-neuron level can explain the changes in network burst frequency at the network level. These preliminary computational results implicate an interplay of single-neuron channel properties and network-level activity in changes observed in MEA recordings of RTT networks. These results further implicate channels involved in adaptation as downstream targets of MECP2, and we are currently designing new computational and experimental approaches to

create multi-scale validations of these predictions. [J.E., J.M.T., and L.M. co-supervised this work]

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Poster

P075. Human Imaging

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Topic: A.09. Adolescent Development

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NIH MH115357 , MH096773 (D.A.F), MH122066 , MH121276 , MH124567
Lynne and Andrew Redleaf Foundation

Title: Failure to reproduce a resting-state language laterality analysis in the ABCD data

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Abstract: The lateralization of language function to the left hemisphere (LH) is a conceptually robust and reproducibly measurable pattern of brain organization, and may be a marker of language development that can be revealed through task-based functional MRI (tfMRI) using comprehension and production tasks. For example, Szaflarski and colleagues (2006) described a general increase in LH lateralization between 5 years and early adulthood using tfMRI. But, under 5 years, tfMRI is challenging due to attention and motion issues, requiring that resting-state fMRI (rsfMRI) scans be used to fill this gap. Methods for calculating lateralization using rsfMRI data are much less well-defined than tfMRI methods. One proposed method for calculating lateralization with rsfMRI data is to subtract mirrored connectivity maps based on the right-hemisphere (RH) Broca and Wernicke homotopes from connectivity maps calculated from the canonical LH areas (Zhu et al., 2014). The authors reported two regions significantly more associated with LH Broca than RH; as well as six regions significantly more associated with

either LH or RH Wernicke using the NYU Test-ReTest sample (n=25; age: 20y).
Here, we attempted to reproduce Zhu and colleagues' findings using a modern surface-based, HCP-style pipeline and the Adolescent Brain and Cognitive Development (ABCD) study (age: 9-10y), as an intermediate step toward performing such an analysis in the Baby Connectome Project (age: 0-5y). We used the ABCD rsfMRI data (n=2000) with 10 min of low-motion data per scan, using Zhu and colleagues' Broca and Wernicke regions and parcels from the Gordon et al. (2016) and Glasser et al. (2016) atlases. We compare and contrast findings between region sets and present this as a case study in the advantages of newer acquisition protocols and processing methods and limitations in reproductions of older data.
We were unable to reproduce any previously reported lateralized regions. While Zhu et al. reported peak difference |Z| values in excess of 5, we found much smaller regions of lateralization near Broca and Wernicke, with lower peak values in the ABCD subsample. We will refine the most promising source regions from these exploratory analyses to replicate in a confirmatory ABCD subsample. Recent findings (Marek et al., 2020; Feczko et al., 2021) suggest this lack of replication might be a consequence of the small sample size and the limited usable data (4.5 min) in Zhu et al. (2014) compared to ABCD. It could also be because the ABCD participants are much younger than the Zhu et al. participants, which will need to be studied further. This study highlights the importance of revisiting prior results with new imaging standards.

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Poster

P075. Human Imaging

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Program #/Poster #: P075.02

Topic: A.09. Adolescent Development

Support: U.S. National Science Foundation, Grant/Award Number: 11519520
Brain and Creativity Institute Research Fund
University of Southern California Rossier School of Education
USC Provost

Title: Community violence exposure during mid-adolescence is related to greater grey matter volume reduction in urban youth

Authors: *K. HILLIARD, M. STONE, X.-F. YANG, M. IMMORDINO-YANG;
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Abstract: Community violence exposure is a form of chronic stress and a significant public health issue for urban youth. It has been linked with poorer cognitive outcomes (Delaney-Black et al., 2002; Sharkey, 2010) and increased risk for psychiatric illnesses (Fowler et al., 2009). Adolescence is a period of dramatic brain maturation and intense psychosocial development, characterized by a heightened sensitivity to social influences (Albert, Chein, & Steinberg, 2003), and is likely especially susceptible to social-relational stressors. However, research examining the effect of stress on brain maturation focuses mainly on early life adversities or on clinical populations (Hart & Rubia, 2012), and the few studies examining the impact of community violence exposure on healthy adolescents are cross-sectional in nature (Butler et al., 2018; Saxbe et al., 2018). It remains unclear whether community violence exposure uniquely impacts brain development during the mid-adolescent period. Here, fifty-five adolescents (mean age 15.9 years at wave 1, SD= 1.1) from mid to low-SES families living in moderate- to high-crime neighborhoods in Los Angeles participated in two waves of structural neuroimaging scans. All were healthy non-delinquent youth, and not the direct victims or perpetrators of violence. Using a self-report survey (Margolin et al., 2009), participants reported their lifetime cumulative community violence exposure at wave 1 and the additional violence they experienced over the two-year interval at wave 2 (proximal exposure). Structural neuroimaging data were processed independently for each wave using voxel-based morphometry (CAT 12 and SPM 12; http://www.fil.ion.ucl.ac.uk/spm). We found that participants with greater proximal violence exposure during the two-year period showed greater reduction in grey matter volume in the anterior and middle cingulate cortex and the right ventral anterior insula, regions broadly involved in salience detection and autonomic regulation (Bush, Luu, & Posner, 2000; Devinsky, Morrell, & Vogt, 1995; Seely et al., 2007). The patterns were consistent with our previous cross-sectional findings (Butler et al., 2018) and with patterns seen in PTSD (Kühn & Gallinat, 2013) and combat-exposed military populations (Butler et al., 2017). Results controlled for lifetime cumulative exposure, thereby isolating the effect of proximal violence exposure during this period. These findings highlight the unique vulnerability to environmental stressors during mid-adolescence even in healthy youth, and the urgent need for intervention and prevention to protect adolescents from the detrimental effects.

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Poster

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Topic: A.09. Adolescent Development

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Title: Intra-network connectivity in the cingulo-opercular network predicts variability in appraisals of emotional ambiguity in childhood development

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Abstract: Emotionally ambiguous signals, like a surprised expression, could be perceived as either positive (unexpected birthday party) or negative (witnessing an accident). In the absence of a clarifying context, people differ in their appraisals of these signals as having a more positive or a more negative meaning (i.e., valence bias). Valence bias is a stable, trait-like measure related to internalizing symptomology, highlighting the need to elucidate its underlying neurobiological mechanisms. Extant work suggests the initial response to ambiguity is negative, and that positivity relies on regulatory processes. Brain regions implicated in task control, like those of the cingulo-opercular network (CON), show greater activation during the resolution of emotional ambiguity in adults. Though it remains unclear whether these regions serve a regulatory role in childhood, and if resting-state connectivity is a useful predictor of individual differences in valence bias. Here, we use support vector regression (SVR) to assess this possibility that subtle differences in multivariate patterns in CON connectivity are related to individual variability in valence bias. This study includes 72 children (ages 6-17; 35 male) from Lincoln, Nebraska. Subjects completed a valence bias task, followed by a scanning session approximately one week later to collect resting-state data (15-20 mins). The resulting data were processed in a standard pipeline, including global signal regression, motion scrubbing, a bandpass filter, and spatial smoothing. Then, functional connectivity was computed for 264 whole-brain ROIs in each subject. Using SVR, intra-network connectivity in the CON (14 nodes, producing 91 edges or features) significantly predicted valence bias (R^2 = .14), even after regressing out age-related variance prior to model fitting (R^2 = .18). However, a larger network (default mode; 58 nodes, 1653 features) unrelated to task control did not (R^2 = .01). “Networks” of 14 random nodes - the same size as CON - also failed to significantly predict valence bias. We show that intra-network connectivity of the CON significantly predicts individual differences in valence bias during childhood. Together with evidence of the role of the CON in adults, these results suggest a putative regulatory role for this network in supporting positivity towards emotional ambiguity. Thus, CON intra-network connectivity holds promise as a diagnostic tool for identifying those with greater risk of developing internalizing symptomology.

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Poster

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Topic: A.09. Adolescent Development

Support: FONCICYT/37/2016, I000/667/2016
Medical Research Council United Kingdom - CONACyT Research Partnerships, Call 2015, Newton Fund

Title: Resting State MR networks modulated by BMI

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Abstract: Introduction: Several studies have suggested a direct association between human cognition and functional/structural connectivity patterns in the human brain; the latter, obtained through Resting State Functional Magnetic Resonance Imaging measurements (RS-fMRI). On top of this, a certain degree of cognitive impairment has been found in people who were obese during their infancy. The physiological mechanisms for these findings are still not clear.
Aim: This work was aimed at studying the RS-fMRI networks of a cohort of infants, divided into three different groups depending on their Body Mass Index (BMI). All this, to find differential brain region recruitment and interactions within-BMI groups.
Methodology: A cohort comprised of 142 children with a mean age of: 8.31 1.18 years, were classified into three groups according to BMI: normal weight, overweight and obese. RS-fMRI measurements were performed on a SIEMENS Skyra 3 Tesla scanner, with a 32-channel head coil, using a 3D Fast GE T_2* sequence. Data was latter preprocessed, analyzed and quantified with different softwares: SPM, GIFT and GRETNA. Through them we were able to obtain grouped RS networks of different kinds for each BMI group of children. Also, a series of different parameters from graphic analysis were obtained.
Results: 15 different RS-fMRI networks were consistently obtained for each BMI group. Larger volumes/surfaces were found when comparing Obese vs. Normal weight in the following networks: executive control, left and right memory networks. When comparing Normal weight vs. Obese larger volumes/surfaces were found in frontal cortex, ventral stream and bilateral frontoparietal networks.
Discussion and Conclusion: These results show differences in brain areas recruited between groups as well as a different physiological connectivity when addressing various cognitive functions. This is the first study of this type in a Mexican population and could shed some light on the reasons for the cognitive impairment found in obese children. In this work in progress, correlation of the different graphic analysis parameters from RS networks with: anthropometric measurements, IQ scores, depression scores, cognitive scores and blood and urine measurements, will be made. This way we might be able not just to see changes in RS networks, but to quantify

and find the physiological underlayer for the different cognition functionality dependance with BMI.

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Poster

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Program #/Poster #: P075.05

Topic: A.09. Adolescent Development

Title: Frontolimbic white matter pathways link prenatal cannabis exposure to child anxiety symptoms

Authors: *J. M. EVANSKI, B. BORG, A. MORALES, M. M. FARAJ, H. A. MARUSAK; Wayne State Univ., Detroit, MI

Abstract: Some pregnant women use cannabis to assuage symptoms of morning sickness or anxiety, and some cannabis-using women may have not known they were pregnant. However, research shows that approximately 1/3 of cannabis constituents, such as delta-9-tetrahydrocannabinol (THC) undergoes cross-placental transfer upon use by a pregnant woman and may therefore disrupt fetal neurodevelopment. In particular, THC may disrupt the endocannabinoid system, which plays a key role in neurodevelopment across the lifespan. Indeed, endocannabinoids and cannabinoid receptors are expressed in the fetal brain, including white matter, as early as 5 weeks into gestation. Prenatal cannabis exposure is associated with cognitive, motor, and social deficits that last into the adulthood of offspring. Here, we use data from the ongoing large-scale NIH Adolescent Brain Cognitive Development (ABCD) study to examine the impact of prenatal cannabis use on childhood anxiety and the integrity of frontolimbic white matter pathways. We focused on frontolimbic pathways that are known to be susceptible to cannabis use in adolescents and adults, and because variation in endocannabinoid signaling is to shown to be important for modulating frontolimbic development. This study reports on diffusion tensor imaging (DTI) data collected from 10,570 youth ($M \pm SD = 9.92 \pm 0.62$ years; 48% female; 55.1% White, 13.2% Black, 19.8% Hispanic, 1.9% Asian, 10.1% Other). Prenatal cannabis use was measured via parent retrospective report and anxiety was measured using the Child Behavior Checklist. Fractional anisotropy (FA) was estimated for frontolimbic white matter tracts. In this sample, 3.9% of parents (n = 410) reported using cannabis prior to knowledge of pregnancy, and 1.1% (n = 119) reported using after knowledge. Any prenatal cannabis exposure was associated with higher anxiety symptoms and lower FA of the left anterior thalamic radiations, left cingulum bundle, and right and left fornix. Cannabis

exposure before (but not after) knowledge of pregnancy was associated with lower FA of the corpus callosum. FA of the fornix was also positively associated with anxiety symptoms, and FA in the left fornix mediated the association between prenatal cannabis exposure and anxiety symptoms. These data add to the growing body of evidence linking prenatal cannabis exposure to altered neurodevelopment. These findings indicate that variation in frontolimbic white matter pathways may explain the link between prenatal cannabis exposure and elevated risk of mental health outcomes in offspring, and should be used to encourage women to abstain from cannabis use during pregnancy or pre-conception.

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Poster

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Program #/Poster #: P075.06

Topic: A.09. Adolescent Development

Support: Korea MSIT 0670-20210004
Brain Korea A0462-20210100

Title: Brain and Genetic Correlates of Psychotic Symptoms in Children

Authors: *K. KIM[1], K. KIM[1], H.-H. WANG[1], S.-Y. MOON[2], Y. JOO[4], J. CHA[3];
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Abstract: Childhood psychotic-like experiences are linked to a higher risk of diagnosis of a psychotic disorder during adulthood (Karcher, et al., 2018)., first psychotic episodes typically begin in adolescence or early adulthood (Arciniegas, 2015; Bozzatello, et al., 2019). Therefore, it is crucial to detect psychotic risk in early childhood. Here, we investigated brain and genetic underpinnings of the prodromal psychotic symptoms. We used the structural MRI of 5,629 children ages 9 to 10 years old from the Adolescent Brain Cognitive Development study. As a measure of psychotic symptoms, we used a prodromal questionnaire-brief child version (PQ-BC) including total (0 to 21) and distress score (0 to 126). Firstly, we examined the association between psychotic symptoms and brain structure (i.e., volume). The multiple linear analysis with demographic covariates (e.g., sex, age, race) showed that whole brain volume was negatively associated with total and distress score (total; standardized beta(β)=-0.040, P=0.020, distress; β=-0.046, P=0.008). To investigate the regional effect on the scores, we analyzed segmented brain regions. Right inferior parietal lobule (rIPL), right superior frontal gyrus (rSFG), right rostral middle frontal gyrus (rRMFG) have the largest effect on total score (rlPL; β=-0.704,

PFDR<0.001, rSFG; β=-0.621, PFDR=0.014, rRMFG; β=-0.565, PFDR=0.014). Interestingly, some brain regions showed a positive association with total score: right frontal superior gyrus (rFSG), right superior temporal sulcus (rSTS), angular gyrus (AG) (rFSG; β=0.466, PFDR=0.014, rSTS; β=0.417, PFDR=0.001, AG; β=0.358, PFDR=0.001). Besides, this relationship is consistent in the distress score. Secondly, we assessed genome-wide polygenic scores (GPSs) associated with psychosis using PRSice (Euesden, Lewis, and O'Reilly, 2015): schizophrenia (SCZ), autism spectrum disorders (ASD), major depressive disorder (MDD), and bipolar disorder (BIP) (Pourcain et al., 2017; Schulze et al., 2014). The multiple linear regression showed GPS of SCZ was not correlated with total and distress scores (βtotal=0.001, βdist=-0.000). Likewise, there is no correlation of ASD and BIP (ASD: βtotal=0.035, βdist=0.023; BIP: βtotal=-0.041, βdist=-0.034). However, PRS of MDD shows the correlation with total and distress scores (βtotal=0.052, PFDR(total)=0.031, βdist=0.066, PFDR(dist)=0.002). In summary, this study reveals some brain areas can be positively correlated with the symptoms. In the aspect of genetic factor, our finding shows a higher genetic risk for MDD is related to psychotic prodrome-related scores in prepubertal children.

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Poster

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Topic: A.09. Adolescent Development

Support: MOST 109-2410-H-152 -008 -MY2

Title: Neural correlates of semantic processing in Chinese-English bilingual children using functional Near Infrared Spectroscopy (fNIRS) imaging

Authors: *L.-Y. FAN[1], T.-L. CHOU[2], H.-C. CHEN[3], S.-Y. CHEN[4];
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Abstract: Bilingual experiences may change children's neural circuitry for learning to read. Previous neuroimaging studies of language acquisition in monolingual populations have revealed increased activation in the middle temporal gyrus (MTG) and inferior frontal gyrus (IFG) associated with semantic processing as a function of skill and development. The goal of this study is to investigate whether the lexical brain areas differ in meaning processing between Chinese monolingual and Chinese-English bilingual first grader children. Two groups of children were recruited: 20 Chinese monolingual and 20 Chinese-English bilingual first graders. Monolingual children were asked to perform an auditory semantic judgment task in Chinese,

while bilingual children were asked to perform an auditory semantic judgment task in both Chinese and English. During the fNIRS imaging task, children heard two words and were asked to indicate if word pairs were related in meaning. The task included two levels of difficulty requiring either high-association (easy) or low-association (difficult). Behavioral results showed that both monolinguals and bilinguals had poor accuracy in the low-association condition than the high-association condition. In addition, bilinguals had more accurate and faster reaction time in the unrelated condition than the low-association and high-association conditions during the English semantic task. Yet, this difficulty effect was only present in English of the bilinguals but not in Chinese. In fNIRS results, our imaging findings revealed that monolinguals showed increased neural activity in the left frontal, temporal, and parietal regions relative to bilinguals. For bilingual children, Chinese semantic association produced greater activation in the right inferior frontal gyrus, whereas English semantic association produced greater activation in the left inferior frontal gyrus. These findings suggest a neuro-cognitive model of language and literacy acquisition in Chinese-English bilingual first grader children. Moreover, bilingual children might develop different patterns of neural activity for each of their languages.

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Poster

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Topic: A.09. Adolescent Development

Support: NIMH Grant R01MH116147
NIMH Grant F32MH122057

Title: White matter sex differences in late childhood assessed with single- and multi-shell diffusion-weighted MRI metrics

Authors: *K. E. LAWRENCE[1], Z. ABARYAN[2], E. LALTOO[1], J. T. MCCRACKEN[3], P. M. THOMPSON[4];
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Abstract: A comprehensive characterization of sex differences in the brain is critical for improving our understanding of sex effects in health and disease. However, we still lack a complete understanding of such differences in developmental populations. Here we examined sex effects on white matter microstructure in a large-scale, community-based sample of youth

using advanced diffusion-weighted MRI measures. Study participants consisted of 9- to 10-year old children from the ABCD study (discovery/replication cohort: 3,979/3,975 youth, 47.8/48.6% female). White matter microstructure was quantified in 21 bilateral regions of interest (ROIs) across the brain using the advanced multi-shell diffusion model restriction spectrum imaging (RSI) and the traditional single-shell model diffusion tensor imaging (DTI). White matter metrics derived from RSI included intracellular anisotropic, isotropic, and total diffusion. Measures calculated using DTI included fractional anisotropy (FA), and mean, axial, and radial diffusivity (MD, AD, RD). Results were considered significant and replicable if they survived FDR correction across ROIs in the discovery cohort (q<0.05) and exhibited p<0.05 in the replication cohort. We found significant and replicable sex differences in RSI and DTI metrics for every white matter ROI examined (|std. betas|=0.06-0.48). Girls typically exhibited greater intracellular anisotropic, isotropic, and total diffusion relative to boys, suggesting that girls may exhibit greater neurite density and white matter cellularity on average. Boys generally displayed greater MD, AD, and RD across ROIs than girls on average, with higher or lower FA observed depending on the ROI. Findings were largely similar when examining the left and right hemispheres separately, as well as when adjusting for intracranial volume, head motion, pubertal status and dimensional internalizing or externalizing problems. Our results expand on previous conflicting DTI studies in developmental populations and, together with prior work in older adults, suggest that patterns of sex differences evolve over the lifespan. In sum, these findings robustly demonstrate sex effects on white matter in youth and provide an important foundation for understanding sex differences in health and disease.

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Poster

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Program #/Poster #: P075.09

Topic: A.09. Adolescent Development

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Title: Profound detrimental effects of obesity on the developing connectome in early adolescence

Authors: S. J. BROOKS[1], S. M. PARKS[2], *C. STAMOULIS[3];
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Abstract: Over 20% of adolescents in the US are obese, a highly alarming statistic given detrimental effects of unhealthy weight on physical, mental and cognitive health. In particular, there is growing evidence that obesity can lead to 'miswiring' of neural circuits, resulting in cognitive and behavioral deficits. During adolescence, the brain undergoes significant changes and is vulnerable to stressors and risk factors, including unhealthy weight, which may adversely impact the maturation of neural circuits supporting critical high-level functions such as decision-making and executive control, as well as mental health. To date, the effects of obesity on developing functional circuits during adolescence remain elusive. To address this significant gap in knowledge, using resting-state fMRI from 5566 early adolescents (median age = 120 months, inter-quartile range = 13.0 months; 52.1% females) from the Adolescent Brain Cognitive Development (ABCD) cohort, this study investigated the impact of obesity on the topology of developing functional brain networks. Children were included based on normal structural MRI, no history of ADHD or bipolar disorder, which may independently affect network connectivity, and quality of fMRI data. Using established age- and sex-specific thresholds for obesity (95th BMI percentile), n = 680 children (12.2% of the cohort) were identified as obese. Given overall low connectivity of brain networks at rest, for each participant the fMRI run with the lowest median connectivity was analyzed. Connectivity was estimated as the pairwise peak fMRI signal cross-correlation. Topological properties included network efficiency, clustering, modularity, robustness and stability. Statistical models were developed to compare these properties in obese versus non-obese children, adjusting for age, sex, race, ethnicity, family income and percent of censored frames in fMRI. Bilateral attention, somatomotor, default-mode, temporo-parietal, fronto-parietal control, limbic and reward networks as well as the thalamus had significantly lower median connectivity, efficiency, global clustering, robustness and stability ($p<0.001$ for all properties) in obese children. Together, these networks support cognitive processes spanning multiple domains, including sensorimotor function, memory, executive function, decision-making, the reward system and emotional processing. These results suggest that severely unhealthy weight may have profound detrimental effects on maturating neural circuits in adolescence, with widespread negative implications for cognitive function, and deficits that may persist across the lifespan.

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Poster

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Program #/Poster #: P075.10

Topic: A.09. Adolescent Development

Support: This work was accomplished with support from the National Institute on Alcohol Abuse and Alcoholism (Y1AA-3009)

Title: Body mass index influences the relationship between socioeconomic status and neurodevelopmental outcomes in children

Authors: *E. DENNIS[1], P. MANZA[1], N. VOLKOW[1,2];
[1]Natl. Inst. on Alcohol Abuse and Alcoholism, Bethesda, MD; [2]Natl. Inst. on Drug Abuse, Bethesda, MD

Abstract: Low socioeconomic status (SES) in childhood is associated with deficits in executive function and changes in cortical morphology. Furthermore, rates of childhood obesity are greater among low SES children and childhood obesity is also associated with cortical alterations and impaired neurocognition, specifically in the domain of executive function. To investigate the influence of BMI on the relationships between SES and both neurocognition and brain morphology, we used data from the Adolescent Brain Cognitive Development (ABCD) study, a multi-site, longitudinal study following over 11,000 9-10-year olds for 10 years throughout development, to construct multiple linear regression models and conduct mediation analyses. In a sample of 8,919 subjects (mean age = 9.91 ± 0.621 years, 52.6% male), SES as measured by household income, highest level of parental education, and area deprivation index, was associated with lower BMI, greater total and prefrontal cortical volume, and better performance on assessments of executive function (The Flanker Task, the List Sorting Working Memory Test, and the Dimensional Change Card Sort Task, measuring cognitive control, working memory, and cognitive flexibility respectively). Mediation analysis indicated that BMI had a significant indirect effect on associations between area deprivation and both total and prefrontal cortical volumes, as well as on the associations between area deprivation and composite neurocognitive scores, which were driven by performance on tasks of working memory and cognitive flexibility, but not cognitive control. The findings of this exploratory research suggest that BMI should be considered in future studies investigating the relationship between low SES and poor neurodevelopmental outcomes.

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Poster

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Topic: A.09. Adolescent Development

Support: Internal Funding from AWRI

Title: Exogenous Gonadal Hormone Treatment Reduces Connectivity between Prefrontal Cortex and Ventral Striatum at Rest in Transgender Adolescents

Authors: R. FRENCH[1], S. F. LIEBOWITZ[2], L. NAHATA[1], J. F. STRANG[3], W. I. MATTSON[1], *E. E. NELSON[1];
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Abstract: Adolescence is characterized by enhanced reward responsiveness and reductions in inhibitory control resulting in a shift toward increased levels of motivated behavior and impulsivity. Previous research has linked these behavioral changes to the surge in gonadal hormones at puberty. Testosterone and to a lesser extent estradiol have been shown to increase activation in the ventral striatum and may also alter functional connectivity between the ventral striatum and other brain regions – particularly the prefrontal cortex. While most previous work in this area has focused on alterations in reward system as a function of changes in endogenous hormone levels at puberty as dissociated from age. The current study focused on the effects of exogenously administered gonadal hormones, testosterone and estradiol, to transgender adolescents undergoing endocrine treatment for gender dysphoria.
One hundred and two participants between 13 and 22 years of age from gender development clinics in two treatment facilities underwent a 6-10 min resting state scan on a 3 Tesla MRI. Thirty-three of 68 trans-masculine (assigned female at birth) were undergoing exogenous testosterone treatment and twenty-two of 34 trans-feminine (assigned male at birth) participants were undergoing treatment with estradiol. Some of the untreated groups were undergoing a variety of other hormonal treatments including both puberty suppression with GnRHa and spironolactone to attenuate endogenous adrogen exposure. Whole brain connectivity maps were generated for each participant from seeds placed in the ventral tegmental area (VTA) and the Nucleus Accumbens (NAcc). A second level omnibus analysis included gonadal hormone treatment (GAH) , assigned sex at birth (AS), and their interaction as predictor variables; and age and site as covariates. This analysis indicated a significant region of reduced coupling in the ventromedial prefrontal cortex in both the VTA and NAcc seed maps as a function of hormone treatment (collapsed across both testosterone and estradiol). These results indicate that gonadal hormone treatment may act to reduce connectivity between prefrontal cortex and reward regions and is consistent with similar effects with endogenous hormones at puberty. Several trend regions in which hormone treatment by sex at birth were also found. Future planned analyses examining sub-sets of the untreated groups may help further elucidate these findings.

Disclosures: R. French: None. **S.F. Liebowitz:** None. **L. Nahata:** None. **J.F. Strang:** None. **W.I. Mattson:** None. **E.E. Nelson:** None.

Poster

P075. Human Imaging

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P075.12

Topic: A.09. Adolescent Development

Support: NIH Grant K22-MH109558

Title: Reduced cortical volume of the default mode network in adolescents with generalized anxiety disorder

Authors: *J. BASHFORD-LARGO[1,2], R. ZHANG[1], A. MATHUR[1], J. ELOWSKY[1], A. SCHWARTZ[1], M. DOBBERTIN[1], R. R. BLAIR[1], K. BLAIR[1], S. BAJAJ[1];
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Abstract: ABSTRACT
Background: Widespread structural alterations have been shown to be implicated in individuals with Generalized Anxiety Disorder (GAD). However, there have been inconsistent findings in cortical volume (CV) differences. Also, most structural neuroimaging studies looking at GAD used a region-based approach with relatively small sample sizes and have typically not focused on adolescents with GAD. We believe this is the first study to look at CV in adolescents with GAD using a network-based approach. The goal of the current study was to focus on three different brain networks (Limbic, Frontoparietal, and Default Mode Network [DMN]) in adolescents with GAD.**Method:** The study involved 81 adolescents with GAD and 112 typically developing (TD) comparison individuals matched on age (15.98 and 15.63 respectively), sex (42F/39M and 45F/67M), and IQ (101.90 and 103.94 respective means). Participants underwent structural MRI to look at CV and sub-cortical volume differences.**Results:** We found that adolescents with GAD showed significantly lower CV within the DMN compared to TD individuals. Within the DMN, CV was particularly reduced in the participants with GAD within prefrontal and temporal regions. Moreover, within the participants with GAD, severity of symptomatology was negatively associated with CV within the DMN (particularly within the prefrontal region of the DMN). Finally, participants with GAD showed significantly reduced hippocampal volume relative to TD participants.**Conclusion:** The current findings suggest structural alterations in adolescents with GAD, which are associated with severity of GAD in patients.

Disclosures: J. Bashford-Largo: None. **R. Zhang:** None. **A. Mathur:** None. **J. Elowsky:** None. **A. Schwartz:** None. **M. Dobbertin:** None. **R.R. Blair:** None. **K. Blair:** None. **S. Bajaj:** None.

Poster

P075. Human Imaging

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Program #/Poster #: P075.13

Topic: A.09. Adolescent Development

Support: Conacyt
Newton Fund UK 15/0178

Title: In vivo cerebral connectometry study based on the diffusion tensor: obese vs normal weight children

Authors: H. RAMIREZ[1], P. DIES-SUAREZ[2], B. DE CELIS[4], E. BARRAGAN[3], P. SO[5], *S. HIDALGO-TOBON[6];
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Abstract: Alterations in brain white matter relating to obesity have been poorly studied in childhood populations. Diffusion Tensor Imaging (DTI) provides information on the degree and directionality of water diffusion in tissues and can provide information regarding brain microstructure during brain development. Connectometry analysis is based on the central hypothesis that the associations between diffusion values and study variables suggests a common fiber pathway [1]. The cohort comprised 142 children with a mean age of (8.31 ± 1.18) years, who were classified into three by groups according to their Body Mass Index (BMI): normal weight, overweight and obese, in accordance with the World International Organization guidelines. Blood levels of low-density lipoproteins (LDL) and high-density lipoproteins (HDL) were recorded. DTI were performed on a SIEMENS Skyra 3 Tesla scanner, with a 32-channel head coil, using a 3D EPI diffusion-weighted sequence with bipolar gradients, 128 diffusion sampling directions, TE = 106ms, TR = 3200ms, flip angle 90º, a b-value of 1000 s/mm^2. DSI Studio software was used for the connectometry analysis, only the brain regions with white matter tracts that met the condition FDR < 0.05 were accepted, resulting in 16 white matter zones that were statistically significant. Variables, BMI and LDL, correlated with lower Axial Diffusivity (AD), Mean Diffusivity (MD), Radial Diffusivity (RD) and Quantified Anisotropy (QA) values, but higher Fractional Anisotropy (FA) and Generalized Fractional Anisotropy (GFA) values. The associations for HDL were generally the opposite to those for BMI and LDL. These results could indicate an increase in fatty acids in the extracellular space to increase the anisotropy of white matter axons [2]. Fatty acid accumulation could in turn, trigger an autoimmune response that leads to a low-grade neuroinflammation that decreases the anisotropy of white matter axons [3]. Interestingly, different results between adult populations and child populations, mainly in FA, have been observed [4,5], which could imply that obesity affects

developing brains differently compared to matured adult brains.

References

[1] Yeh, F. C., *et al*. Neuroimage, 125, 162-171, 2016.[2] Pakiet, A., *et al*, 16(1), 1-10, 2019.[3] Lumeng, C. N., & Saltiel, A. R.The Journal of clinical investigation, 121(6), 2111-2117, 2011.[4] Xiawei Ou, *et al*. Journal of MagneticResonance Imaging, 42(5):1205{1213, 2015.[5] Timothy D. Verstynen, *et al*. Psychosomatic medicine, 74(7):682, 2012.

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Poster

P075. Human Imaging

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Program #/Poster #: P075.14

Topic: A.09. Adolescent Development

Support: NINDS R01NS055903

Title: Neuropsychiatric traits and their relationship with striatal volume in children and adolescents from Huntington's Disease families

Authors: *E. E. REASONER[1], E. VAN DER PLAS[1], H. M. AL-KAYLANI[1], D. R. LANGBEHN[1], A. L. CONRAD[2], J. SCHULTZ[1], E. A. EPPING[1], V. A. MAGNOTTA[3], P. C. NOPOULOS[1];
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Abstract: Neuropsychiatric traits, broadly defined as executive dysfunction and psychiatric symptoms, are a prominent feature of Huntington's Disease (HD). Little is known about the development of these traits. Given previous evidence of atypical striatal development in mutant huntingtin (*mHTT)* carriers, we hypothesized that neuropsychiatric symptoms would be associated with striatum development. Using data from the Kids-HD study, we aimed to compare neuropsychiatric symptoms between child and adolescent carriers of *mHTT* and peers who did not inherit *mHTT*. We also sought to determine relationships between neuropsychiatric traits and striatal development.

Participants between 6-18 years old were recruited from families affected by HD. Following an accelerated longitudinal design, the sample included 59 gene-expanded (GE) individuals and 91 gene-non-expanded (GNE) individuals who provided 91 and 145 observations, respectively. The Pediatric Behavior Scale (PBS) and the Behavior Rating Inventory of Executive Function (BRIEF) were administered to assesses neuropsychiatric traits. Striatal volumes were extracted from 3T neuro-anatomical images. Multivariable linear regression models were conducted to

evaluate the impact of group, age, and striatal volume on neuropsychiatric symptoms. We found that depression/anxiety was higher in the GNE group compared to the GE group (Estimate = 4.26, $t(130) = 2.37$, $p = 0.019$). Additionally, age-dependent striatal volume (age*striatum interaction) predicted depression/anxiety scores independent of *mHTT* status (Estimate = 0.429, 95% CI 0.15:0.71, $p = 0.0029$). Increased depression was associated with smaller striatal volume in younger children; however, the inverse pattern was observed in adolescents (i.e., increased depression associated with larger striatal volume). When considered in the context of previous findings, these results suggest that differences in striatal development between GNE and GE may be associated with reduced risk of depression or anxiety in child and adolescent carriers of mHTT.

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Poster

P093. Voltage-Gated Sodium Channels

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P093.01

Topic: B.04. Ion Channels

Title: Identifying novel and selective sodium channel modulators from Theraphosidae venoms

Authors: E. M. DUNCAN[1], D. MCCULLOUGH[2], S. C. C. GRACE[1,3], S. BAKER[2], *J. P. O. KAMMONEN[1], P. GRANT[2], S. A. TRIM[2], V. LAZARI[1];
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Abstract: Voltage-gated sodium (Na_V) channels have been extensively studied as targets for therapeutic indications such as pain, epilepsy and paralysis, where modulating these channels could be beneficial by altering action potential firing. However, small molecule interventions have been largely unsuccessful in the clinic, primarily due to challenges in developing compounds with high subtype selectivity to minimise off-target effects. Many animal venoms have evolved as Na_V channel modulators in order to subdue prey and defend against predators. Several toxin peptides are highly potent and specific to individual Na_V subtypes, and as such have become widely-used tools in research to elucidate structure and function of the channels. These venom peptides are also inherently stable due to their biology. Increasingly, toxins have been used in the development of novel therapeutics as peptide frameworks for new bioactive molecules or targeting scaffolds for drug conjugates. However very few have moved to the clinic and thus new approaches are needed. Ion channels are notoriously challenging to study in drug

discovery, due to long and laborious manual patch clamp approaches. Recent advances in automated patch clamp technologies have allowed high quantities of meaningful data to be generated quickly. We have successfully collected, fractionated and standardised venoms from 17 Theraphosidae species to produce a Targeted Venom Discovery Array (T-VDA). This T-VDA, containing 960 theraphosid venom fractions, was screened in duplicate against $Na_V1.4$ and $Na_V1.7$ channels on the Sophion Qube high-throughput automated patch clamp platform in 384 well format. We identified 33 fractions, which modulated the activity of one or both channels, thus generating a hit rate of 3.4% across both targets with the majority modulating $Na_V1.7$. Here we present our findings and discuss the advantages and challenges of using automated patch clamp technology to identify novel hits using a venom peptide library.

Disclosures: **E.M. Duncan:** A. Employment/Salary (full or part-time):; Charles River. **D. McCullough:** A. Employment/Salary (full or part-time):; Venomtech. **S.C.C. Grace:** A. Employment/Salary (full or part-time):; Charles River. **S. Baker:** A. Employment/Salary (full or part-time):; Venomtech. **J.P.O. Kammonen:** A. Employment/Salary (full or part-time):; Charles River. **P. Grant:** A. Employment/Salary (full or part-time):; Venomtech. **S.A. Trim:** A. Employment/Salary (full or part-time):; Venomtech. **V. Lazari:** A. Employment/Salary (full or part-time):; Charles River.

Poster

P093. Voltage-Gated Sodium Channels

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P093.02

Topic: B.04. Ion Channels

Support: Israel Science Foundation 1454/17

Title: Functional analysis of the developmental delay SCN8A G1625R mutation reveals prominent persistent currents

Authors: *S. QUINN[1], M. BRUSEL[1], H. BASSAN[2], R. BEN-SHALOM[3], M. RUBINSTEIN[1]; [1]Goldschleger eye research institute, Dept. of human molecular genetics and biochemistry, Tel Aviv university, Tel Aviv, Israel; [2]Pediatric neurology and development center, Shamir medical center, Be'er Ya'akov, Israel; [3]Ctr. for Integrative neuroscience, Kavli institute for fundamental neuroscience, Univ. of California, San Francisco, CA

Abstract: The voltage-gated sodium channel $Na_V1.6$, encoded by the gene *SCN8A,* plays an important role in neuronal firing. Mutations in the *SCN8A* gene result in a range of neurodevelopmental impairments, including developmental and epileptic encephalopathies, autism spectrum disorders, developmental delay without seizures, and movement disorders. Functional testing revealed that gain of function mutations in the *SCN8A* gene are associated

with severe epilepsy, while loss of function mutations are associated with intellectual disability with or without seizures. We performed a functional analysis of a $Na_V1.6$ mutation, G1625R, identified in a 5-year-old patient with significant global developmental delay, mild epilepsy, and quadriparetic cerebral palsy. Voltage-clamp experiments in whole-cell configuration were performed on transfected mouse neuroblastoma cells, revealing mixed gain and loss of function effects. While the maximal current amplitudes of the $Na_V1.6^{G1625R}$ currents were smaller compared to WT channels ($Na_V1.6^{WT}$ at -10 mV: -313±51.72 pA/pF, n=12; $Na_V1.6^{G1625R}$: -138.6±38.3 pA/pF, n=15, p=0.01), persistent currents were markedly enhanced ($Na_V1.6^{WT}$: 3±0.68%; $Na_V1.6^{G1625R}$: 27.23±3.9%, p<0.0001), leading to a slightly higher total current that passes through the $Na_V1.6^{G1625R}$ channel over a longer period. Additionally, $Na_V1.6^{G1625R}$ channels showed a depolarizing shift in the activation and inactivation curves. The effect of different anti-epileptic drugs was tested (Carbamazepine, Lacosamide, and Cenobamate) with the main goal of inhibiting the persistent current and shifting the inactivation to hyperpolarized potentials. Carbamazepine shifted the inactivation curve toward hyperpolarized potentials, partially correcting the gating properties, but had no effect on the persistent currents, which remained prominent in $Na_V1.6^{G1625R}$. We further investigated the effect of the mutation on neuronal firing by generating an ion channel model with biophysical properties of $Na_V1.6^{G1625R}$ .We incorporated it into a numerical model of an excitatory neuron, in order to determine the overall contribution of the mixed loss and gain of function characteristics of the mutation to the pathophysiology of this channelopathy. Together, our analysis demonstrated complex gain and loss of function properties in the *SCN8A* mutation $Na_V1.6^{G1625R}$, which are not corrected by Carbamazepine but might be corrected with a drug that will have a more specific effect on the persistent current.

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Poster

P093. Voltage-Gated Sodium Channels

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Program #/Poster #: P093.03

Topic: B.04. Ion Channels

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Title: Phenotype and neurophysiology of Scn2a^{K1422E}mice reflect complex Na$_V$1.2 sodium channel dysfunction related to DEKA > DEEA mutation of the selectivity filter.

Authors: *D. M. ECHEVARRIA-COOPER[1,3], N. A. HAWKINS[1], S. M. MISRA[1,2,4], C. H. THOMPSON[1], A. D. NELSON[5], A. M. LIPKIN[5,6], A. L. GEORGE[1,3], K. J. BENDER[5], J. A. KEARNEY[1,3];
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Abstract: Genetic variants in *SCN2A*, encoding the Na$_V$1.2 voltage-gated sodium channel, have been described in a range of neurodevelopmental disorders. Genotype-phenotype relationships in *SCN2A* disorders can be investigated using animal models of unique variants. The variant *SCN2A*-p.K1422E is associated with infant-onset developmental delay, epilepsy, and autism spectrum disorder. Compared to wild type (WT) Na$_V$1.2 channels, K1422E channels exhibit novel calcium permeability, reduced current density, and tetrodotoxin (TTX) resistance. To investigate the pathogenic effects of this variant, we generated a mouse model using CRISPR/Cas9 genome editing. To characterize cellular effects of this variant, we performed voltage clamp recordings in dissociated hippocampal neurons, as well as current clamp recordings and 2-photon calcium imaging in neocortical slice preparations. Dissociated neurons from *Scn2a^{K1422E}* heterozygous mice (*Scn2a$^{E/+}$*) exhibited a novel TTX-resistant current with a reversal potential of 32.7 mV compared to 38.5 mV observed in WT mice. Measurements in *Scn2a$^{E/+}$* neocortical slices showed altered action potential waveforms and ectopic calcium transients during the rising phase of the action potential at the axon initial segment, demonstrating complex effects on channel function. To assess how this complex channel phenotype might contribute to seizure susceptibility, juvenile mice were implanted with EEG headmounts and monitored for ≥ 96 hours. *Scn2a$^{E/+}$* mice exhibited rare (< 1 per week) spontaneous localized seizures during sleep without observable behavioral changes. Similar events were not observed in WT mice. Due to the rarity of these seizures, we used flurothyl to induce a stereotyped progression to generalized tonic-clonic seizures (GTCS) in juvenile mice. While latency to myoclonic jerk was not affected, *Scn2a$^{E/+}$* mice took longer to reach GTCS compared to wild type (WT) controls. This indicates resistance to seizure spreading in *Scn2a$^{E/+}$* mice, consistent with the localized seizures observed using EEG. We also observed a reproducible non-unimodal distribution of GTCS latencies in *Scn2a$^{E/+}$* females compared to WT. Neurobehavioral phenotypes of *Scn2a$^{E/+}$* mice were assessed in a series of assays between 6-12 weeks of age. *Scn2a$^{E/+}$* mice exhibited reduced anxiety-like behavior and alterations in olfactory-guided social behavior. Overall, *Scn2a^{K1422E}* mice exhibit phenotypes that are similar yet distinct from other *Scn2a* models, consistent with complex effects of K1422E on Na$_V$1.2 function. This underscores how *Scn2a^{K1422E}* mice can serve as a platform to investigate phenotypic complexity of *SCN2A* disorders.

Disclosures: D.M. Echevarria-Cooper: None. **N.A. Hawkins:** None. **S.M. Misra:** None. **C.H. Thompson:** None. **A.D. Nelson:** None. **A.M. Lipkin:** None. **A.L. George:** B. Contracted

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Poster

P093. Voltage-Gated Sodium Channels

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Program #/Poster #: P093.04

Topic: B.04. Ion Channels

Support: FamilieSCN2A Foundation
NSF GRFP DGE-1842166
Showalter Research Trust
NIH R01NS117585 and R01NS123154

Title: The reversibility of social deficits in adult Scn2a gene-trap knockout mice

Authors: *M. EATON[1,4], J. ZHANG[1], X. CHEN[1], J. WU[2], Y. YANG[3];
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Abstract: Recent genetic studies have discovered variants in voltage-gated sodium channel $Na_V1.2$ (encoded by gene *SCN2A*) as the leading cause of monogenic autism spectrum disorder (ASD), as well as other neurodevelopmental disorders. While an estimated 400 or more new cases with *SCN2A*-associated disorders are diagnosed each year, no curable treatments are available. In the early stage of brain development, $Na_V1.2$ is suggested to be the main voltage-gated sodium channel expressed in the axon initial segment. Later in the development, $Na_V1.2$ is re-distributed to other parts of the neurons including proximal axon initial segment and dendrites, altogether contributing to the initiation, propagation, and backpropagation of action potentials. Homozygous *Scn2a*$^{-/-}$ knockout mice die perinatally. While heterozygous *Scn2a*$^{+/-}$ mice (with ~50% $Na_V1.2$ expression level) survive to adulthood, behavior abnormalities of these mice are minor to modest. To further study the relationship between the dysfunction of *SCN2A* and ASD, we have characterized a gene-trap knockout mouse model of *Scn2a* with about 75% reduction of *Scn2a* mRNA and protein expression. This mouse model has pronounced social deficits in multiple social behavior tests including empty versus novel mouse (three-chamber), fake toy versus novel mouse (three-chamber), sociability over 24 hours (home cage), stranger interaction,

and reciprocal interaction. Approaches to rescue abnormal behaviors are being explored, including the restoration of *Scn2a* expression in adult mice. Our data will demonstrate to what extent that restoration of *Scn2a* expression can correct behavioral abnormalities in adulthood. Our results will also reveal circuit mechanisms underlying abnormal social behaviors related to severe $Na_V1.2$ deficiency. Knowledge gained in our study will set a solid foundation to further determine other forms of interventions (gene therapy, small molecule drugs) using behavioral rescue as a direct, phenotypic readout.

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Poster

P093. Voltage-Gated Sodium Channels

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Topic: B.04. Ion Channels

Support: NIH R01NS117585
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Purdue Institute for Integrative Neuroscience (PIIN)
Ralph W. and Grace M. Showalter Research Trust
Purdue Big Idea Challenge 2.0
Purdue University Institute for Drug Discovery (PIDD)
Purdue Autism Research Center (PARC)

Title: Modeling SCN2A gain-and-loss of function mutations L1342P and C959X with CRISPR-edited human-induced pluripotent stem cell-derived neurons and cortical spheroids

Authors: *M. I. OLIVERO-ACOSTA[1], Z. QUE[1], T. NGUYEN[1], L. YUNIS[1], Y. YANG[2]; [2]MCMP, [1]Purdue Univ., West Lafayette, IN

Abstract: Human-induced pluripotent stem cell (hiPSC) models are valuable tools that enable the generation of monolayer and three-dimensional neuronal cultures that recapitulate features of neurological diseases. The *SCN2A* (Voltage-Gated Sodium Channel Alpha Subunit 2) gene encodes for sodium channel Nav1.2, a protein that plays a vital role in mediating action potentials in neurons. *SCN2A* pathogenic variants have been identified in individuals with epilepsy and autism spectrum disorders (ASD). These variants leading to abnormalities in sodium channel topology and function, possibly influencing neuronal activity patterns. Two examples of variants are L1342P (p. Leu1342Pro) and C959X (p. Cys959X). The gain-of-function (GoF) L1342P variant was identified from several patients with intractable seizures. While the loss-of-function (LoF) C959X variant was identified from a patient with ASD, developmental delay, and no previous history of epilepsy. Our recent work has demonstrated that

hiPSC-derived 2D-neuronal monolayers carrying the CRISPR-Cas9-edited L1342P-mutant channel display a marked hyperexcitability phenotype. This hyperexcitability is characterized by increased intrinsic excitability, enhanced synchronous network firing, and bursting (Que et al., 2021). However, the impact that the GoF L1342P mutation could have upon the development of neurons and organoids remains unexplored. On the other hand, we are interested in modeling the *de novo* LoF C959X, a protein-truncating variant. Previous studies have shown that when the C959X mutation is transfected in HEK293 (Human embryonic kidney 293 cells), the cells presented no detectable sodium current, indicating deficiencies in ion channel formation (Ben-Shalom et al., 2017). Nonetheless, it is unknown how the mutation may impact neuronal activity and development. In the present study, we used human-induced pluripotent stem cells to generate cortical neurons and 3D spheroids to identify and compare the neurobiological phenotypes arising from *SCN2A* gain-and-loss of function sodium channel variants. Techniques applied include patch-clamp recordings, microelectrode array (MEA) recordings, live-cell calcium imaging, and neurite reconstructions. Our results will elucidate how gain vs. loss-of-function Nav1.2 variants may affect neuronal activity and development of neurons distinctly, contributing to different disease phenotypes. Our work will also provide a highly physiologically relevant platform for testing novel therapeutical interventions, including small molecules, gene therapies, and antisense oligonucleotides.

Disclosures: M.I. Olivero-acosta: None. **Z. Que:** None. **T. Nguyen:** None. **L. Yunis:** None. **Y. Yang:** None.

Poster

P093. Voltage-Gated Sodium Channels

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Program #/Poster #: P093.06

Topic: B.04. Ion Channels

Support: Stanley Center for Psychiatric Research

Title: Discovery of $Na_V1.2$ Modulators for Treating SCN2A-Related Disorders

Authors: *E. LEBOIS[1], M. WEIWER[1,2], A. CAMPBELL[1,2], H.-R. WANG[1], S. CHOI[1], A. GHOSHAL[1], D. BAEZ[1], M. FITZGERALD[1], M. FLEISHMAN[1], Z. FU[1], K. PEREZ DE ARCE[1], J. SACHER[1,2], Q. XU[1], P. SPRATT[3], K. BENDER[3], G. FENG[1], D. LAL[1], Y.-L. ZHANG[1], Å. JÄGERVALL[4], J. PIHL[4], P. KARILA[4], E. SCOLNICK[1], J. PAN[1], F. WAGNER[1], J. COTTRELL[1];
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Abstract: Recent discoveries have shown that *SCN2A* genetic variants are causal for a number of neurodevelopmental disorders, including autism spectrum disorder (ASD), intellectual disability (ID), and infantile seizure disorders. *SCN2A* encodes the voltage-gated sodium channel $Na_V1.2$, with loss-of-function variants causing ASD and ID, and gain-of-function variants causing infantile seizure disorders. Given this genetic linkage to disease, we set out to discover small molecule positive modulators of $Na_V1.2$ for the treatment of ASD. We first established a novel primary high-throughput screening (HTS) fluorescence-based assay to allow the detection of voltage changes in $Na_V1.2$-expressing HEK cells in response to electrical stimulation. This physiologically relevant assay platform enabled us to complete an HTS of ~77,000 compounds, where we identified 378 $Na_V1.2$ activators and 1837 inhibitors. Follow up concentration-response assay yielded a number of potent and efficacious activators, and select compounds were followed up in a synaptic function model where a synaptically transmitted response to electrical stimulation was quantified in primary murine cortical cultures to confirm the compounds' effect. Subsequent electrophysiological characterization of one hit, BRD4032, revealed that it activates $Na_V1.2$ by changing both the voltage sensing mechanism and the inactivation kinetics of the channel, and that BRD4032 elicited marked widening of the action potential waveform in *Scn2a*$^{+/-}$ mouse brain slices. Finally, *in vivo* EEG studies showed significantly altered alpha, beta, and delta frequency bands in *Scn2a*$^{+/-}$ mice, indicating that these alterations may be useful translational biomarkers. Taken together, we have successfully implemented a robust screening strategy to identify $Na_V1.2$ modulators and are poised to follow up on the selectivity, mechanism of action, and *in vivo* effects of these compounds.

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Poster

P093. Voltage-Gated Sodium Channels

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Program #/Poster #: P093.07

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Title: The ASD-associated genes Ank2 and Scn2a converge to co-regulate dendritic excitability in neocortical pyramidal neurons

Authors: *A. NELSON[1], A. CATALFIO[2], J. PHILIPPE[2], L. MIN[2], R. CABALLERO-FLORAN[2], K. PERKINS[2], C. ELVIRA[2], A. SAHAGUN[1], S. SANDERS[1], K. BENDER[1], P. JENKINS[2];
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Abstract: Neuronal voltage-gated sodium channels (Na_Vs) play critical roles in action potential (AP) generation and propagation. *SCN2A*, which encodes the sodium channel $Na_V1.2$, has repeatedly emerged from whole-exome sequencing studies with some of the strongest association scores to autism spectrum disorder (ASD). In developing glutamatergic pyramidal neurons of the medial prefrontal cortex, $Na_V1.2$ is the only sodium channel subtype detected at the axon initial segment (AIS) and nodes of Ranvier. In mature neurons, $Na_V1.2$ is replaced by $Na_V1.6$ at the AIS and is relocalized to the dendrites. Here, $Na_V1.2$ promotes the backpropagation of APs from the AIS to distal dendritic arbors, an important feature of excitable dendrites that is essential for normal synaptic structure, function, and plasticity. While previous studies have shown Na_Vs are localized to the AIS by ankyrin-G (*ANK3*), the mechanisms that underlie $Na_V1.2$ localization within the dendritic membrane are unknown. We hypothesize that ankyrin-B, product of the high-confidence ASD gene *ANK2*, is the primary ankyrin that localizes $Na_V1.2$ to dendrites. Immunostaining cultured neocortical neurons at 19 days *in vitro* showed ankyrin-B is highly expressed throughout the dendrites, distinct from ankyrin-G at the AIS, which suggests that ankyrin-B is well-positioned to control the dendritic localization of $Na_V1.2$. To test this, we performed knockout-and-rescue experiments of ankyrin-B in mature cultured neocortical neurons. Knockout of ankyrin-B resulted in a significant reduction in $Na_V1.2$ levels at the dendritic membrane and rescue with wild-type ankyrin-B restored dendritic $Na_V1.2$ localization. Therefore, loss of ankyrin-B was predicted to have similar consequences on dendritic excitability as *Scn2a* loss. Consistent with this hypothesis, AP backpropagation into the distal dendrites of layer 5 pyramidal neurons was attenuated in acute brain slices from *Ank2* haploinsufficient mice, an effect we observed previously in *Scn2a* haploinsufficient conditions. Furthermore, we found subsequent deficits in excitatory postsynaptic function identical to those previously identified in *Scn2a* haploinsufficient mice. These findings highlight a novel dendritic role for ankyrin-B in the proper localization of $Na_V1.2$ and reveal convergent dendritic function for both *Scn2a* and *Ank2*, two of the most commonly disrupted genes in ASD.

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Poster

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Title: Severe deficiency of voltage-gated sodium channel $Na_V1.2$ elevates neuronal excitability in adult mice

Authors: *J. ZHANG, X. CHEN, M. EATON, J. WU, T. XIAO, Z. QUE, M. OLIVERO-ACOSTA, N. LANMAN, Y. YANG;
Purdue Univ., West Lafayette, IN

Abstract: $Na_V1.2$ channel, encoded by *Scn2a*, is a major voltage-gated sodium channel in the central nervous system (CNS) supporting action potential (AP) firing. $Na_V1.2$ is strongly expressed in the principal neurons of the cortico-striatal circuit, including pyramidal neurons in the medial prefrontal cortex (mPFC) and medium spiny neurons (MSNs) in the striatum. The current paradigm suggests that $Na_V1.2$ gain-of-function variants enhance neuronal excitability resulting in epilepsy, whereas $Na_V1.2$ loss-of-function or protein-truncating variants (collectively referred to as $Na_V1.2$ deficiency) impair neuronal excitability contributing to autism. This paradigm, however, does not explain why 20~30% of patients with $Na_V1.2$ deficiency still develop seizures. As hyperexcitability and hypersynchronization of neuronal firings are associated with seizure generation, it is thus intriguing how $Na_V1.2$ deficiency, predicted to reduce neuronal excitability, contributes to seizures and epilepsy. Here we report a counterintuitive finding that severe $Na_V1.2$ deficiency results in increased neuronal excitability, challenging the current dogma. Using a $Na_V1.2$-deficient mouse model, we show enhanced intrinsic excitabilities of principal neurons in the prefrontal cortex and striatum, brain regions known to be involved in *Scn2a*-related seizures. This increased excitability is autonomous, and is reversible by the genetic restoration of *Scn2a* expression in adult mice. RNA-sequencing reveals the downregulation of multiple potassium channels including $K_V1.1$. Correspondingly, K_V channel openers alleviate hyperexcitability of $Na_V1.2$-deficient neurons. This unexpected neuronal hyperexcitability revealed by our study may serve as a cellular basis underlying $Na_V1.2$ deficiency-related seizures.

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Poster

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Title: Microglial regulation of hyperexcitability in cortical neurons derived from human iPSCs carrying epilepsy-associated sodium channel Nav1.2 variant

Authors: *Z. QUE, M. OLIVERO-ACOSTA, J. ZHANG, J. WU, M. EATON, Y. YANG; Purdue Univ., West Lafayette, IN

Abstract: Rapid progress in the discovery of genetic variants from patients with epilepsy and autism highlights a leading role of *SCN2A* variants. Thus, understanding the pathophysiology related to SCN2A variants may help to advance the development of novel therapeutic interventions. Nav1.2, encoded by gene *SCN2A*, is predominantly expressed in the pyramidal excitatory neurons across the brain, and is responsible for action potential firing and propagation. *De novo* variants of Nav1.2 (e.g., L1342P) were discovered in patients with encephalopathy and seizures. Biophysical property of Nav1.2-L1342P variant was studied in a heterologous expression system, which revealed complex and puzzling characteristics with both gain and loss-of-function features. Therefore, how this variant perturbs neurons and shapes the overall neuronal excitability remains unclear. Using computational modeling, we suggested a gain-of-function phenotype in a single-compartmental pyramidal neuron model. To experimentally test our prediction, we used CRISPR genome editing and generated an L1342P variant and isogenic controls in reference human induced pluripotent stem cell (hiPSC) line. We found elevated action potential firings and altered firing patterns of cortical neurons derived from iPSCs carrying Nav1.2-L1342P variant. Using multi-electrode array recording, we observed a significant increase in neuronal network excitability in L1342P neuron culture. Importantly, we recapitulated features of the phenytoin-resistance effect observed in the clinical treatment. We further demonstrated that a Nav1.2 isoform-specific blocker (Ptx3) can efficiently alleviate the hyperexcitability of L1342P neurons. Neuronal hyperexcitability especially in the chemically induced seizures models can be regulated by microglia, which is an innate immune cell in the brain. However, how human microglia regulate the activity of human neurons carrying seizure-related mutations is not known. Thus, we continue our exploration to study how microglia behave in the presence of hyperexcitable neurons carrying Nav1.2-L1342P variant. Using microglia-neuron co-culture system, we studied changes of microglia calcium signaling in culture with L1342P neurons. We also studied how the microglia regulate neuronal activities

using MEA and patch-clamp recording. Understanding the relationship between hyperexcitable L1342P neurons and microglia would open the possibility of exploring the modulation of microglia as a novel approach to control neuronal hyperexcitability and eventually alleviate seizures.

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Poster

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Topic: B.04. Ion Channels

Title: Constructing a novel functional interactome to study the dynamic crosstalk between neuroinflammation and neural excitability

Authors: J. VON MORGENLAND[1], A. ALLAHVERDIAN[2], R. KNORR[1], *S. VENUGOPAL[3];
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Abstract: Maintaining neural excitability is a key function of the nervous system and is essential for normal behaviors. Ongoing perturbations such as aging and nervous diseases can cause homeostatic shifts in excitability through modifications in the palette of intrinsic and synaptic ion channel proteins (ICPs) in neurons. Here we hypothesize that neuroinflammatory cytokines associated with injury and disease are homeostatic modulators of ICPs and neural excitability. To test this, we conducted a systematic meta-analysis of experimental evidence for cytokine modulation of neuronal excitability. Our initial approach of database search using popular bioinformatics tools such as the STRING surprisingly did not reveal strong evidence for protein-protein interactions between cytokines and ICPs. However, a more detailed examination of the scientific literature on PUBMED uncovered a few yet convincing mechanistic studies which demonstrated cytokine-induced changes in not only ICP mRNA expression, but also functional changes in ion channel conductivity and, action potential threshold and frequencies. For example, the proinflammatory cytokine TNF-α; released by resident glial cells increased voltage-gated sodium channel mRNA, channel conductivity and action potential firing. Driven by these findings, we developed a novel *functional interaction score* (FIS) through which we quantitate the strength of evidence for pleiotropic actions of cytokines on ICPs not only at the molecular level, but also at the functional levels which lead to altered neural excitability. We then used the FIS to generate protein-protein interactomes of cytokines and ICPs to better understand their functional relationships. Our analysis revealed 1-to-N and M-to-1 associations

between cytokines and ICPs. We further demonstrate application of graph theory and dynamic model simulations to analyze the functional links between neuroinflammation and homeostatic changes in neural excitability. Our results also prompt experimental work to clarify cytokines effects on neurons at pre-symptomatic stages of neurodegenerative diseases for therapeutic neuromodulation.

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Poster

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Topic: B.04. Ion Channels

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Title: Regulation of surface expression of the hyperpolarization activated cyclic nucleotide gated channel 2 (HCN2) through SUMOylation and interaction with the auxiliary subunit TRIP8b.

Authors: *L. A. FORSTER[1], D. J. BARO[2,1];
[1]Neurosci. Inst., [2]Biol., Georgia State Univ., Atlanta, GA

Abstract: HCN2 mediates the subthreshold conductance, Ih, that influences intrinsic cellular properties such as membrane potential, pacemaker activity and temporal summation. HCN2 is widely expressed in both the CNS and PNS, and channelopathies that alter Ih amplitude contribute to disease states such as epilepsy, chronic pain, Parkinson's disease, and major depression. HCN2 surface expression can be tuned by SUMOylation at K669, or co-expression with the interacting protein TRIP8b. Enhancing HCN2 SUMOylation at K669 or interaction with TRIP8b isoform 1a-4 produces an increase in surface expression. We are investigating if a common mechanism is involved. Increased exocytosis or decreased internalization can enhance surface expression. To test if SUMO regulated internalization, HCN2 was heterologously expressed in HEK293 cells, and Ih maximal conductance (Gmax) was characterized with whole cell patch clamp. Inclusion of SUMO peptide in the patch pipette to enhance SUMOylation increased Ih Gmax by ~61% relative to control recordings that lacked SUMO (one way ANOVA, $F(3, 31) = 4.672$, $p=0.0083$; Tukey's, $p=0.0355$). This experiment was repeated, but the clathrin inhibitor Pitstop 2, was or was not included in the superfusate and pre-applied for 20min to block endocytosis. Pitstop 2 produced a significant ~80% increase in Ih Gmax relative to controls lacking any treatment (Tukey's, $p=0.0188$); Pitstop 2 + SUMO produced a significant

~71% increase in Ih Gmax relative to control (Tukey's, p=0.0247); the effect of Pitstop 2 was not significantly different compared to Pitstop 2 + SUMO (Tukey's, p= 0.9700). These data suggest that SUMOylation prevents internalization. Consistent with this interpretation, a biotin internalization assay showed 23% of HCN2 is internalized in HEK293 cells after 2.5 hours at 18°C, however enhancing SUMOylation reduced internalization to 6.3% (t-test, p= 0.0125). The experiment is being repeated with HCN2 K669R to determine if SUMOylation at K669 is mediating this effect. TRIP8b and SUMO may act to increase HCN2 surface expression through a common mechanism. Co-expression of TRIP8b with HCN2 in HEK293 cells increased Ih Gmax by 114% compared to HCN2 alone (one way ANOVA, F (3,32) = 12.56, p<0.0001; Tukey's, p<0.0001), no further increase was observed when SUMO + UBC9 were co-expressed with TRIP8b (Tukey's, p=0.7239). This suggests that TRIP8b and SUMO may act to increase HCN2 surface expression through the same pathway. Biotin internalization assays will examine if TRIP8b is preventing internalization of the channel.

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Poster

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Title: The role of $P2X_2$ receptor for acetylcholine synthesis pathway

Authors: *T. MARUYAMA, A. MANO, T. ISHII, Y. KAKINUMA, M. KANEDA; Physiol., Nippon Med. Sch., Tokyo, Japan

Abstract: Acetylcholine, an excitatory neurotransmitter, is synthesized from choline and acetyl-CoA by choline acetyltransferase (ChAT) in cholinergic neurons. Currently, as choline taken up through high-affinity choline transporters is thought to be the only source for acetylcholine synthesis, the activity of cholinergic neuron is thought to correlate with the activity of high-affinity choline transporter. We have previously reported that the mouse retinal cholinergic neurons with weak immunoreactivity for high-affinity choline transporter have choline-permeable cation channels formed by $P2X_2$ receptor. In this study, therefore, we investigated whether choline transported into cells through $P2X_2$ receptors is used for the acetylcholine synthesis in recombinant system by using immunostaining, immunoblotting and HPLC techniques. We expressed $P2X_2$ receptors in both HEK293T cells and HEK293T cells stably

expressing ChAT (ChAT-HEK293T cells). In both HEK293T cells and ChAT-HEK293T cells, intracellular acetylcholine levels were significantly increased when choline uptake through $P2X_2$ receptors was activated by ATP. Our results support the idea that $P2X_2$ receptors work as a novel pathway of choline transport for acetylcholine synthesis.

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Poster

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University of Kentucky Neuroscience Research Priority Area and College of Arts and Sciences Summer Research Fellowship to N.P.
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Title: The effect of calcium ions on mechanosensation and neuronal activity in proprioceptive neurons

Authors: *N. PARKER[1], H. TANNER[3], R. COOPER[2];
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Abstract: Proprioception of all animals is important in being able to have coordinated locomotion. Stretch activated channels (SACs) transduce the mechanical force into electrical signals in the sensory endings. The types of SACs vary among sensory neurons in animals as defined by pharmacological, physiological and molecular identification. The chordotonal organs within insects and crustaceans offer a unique ability to investigate proprioceptive function. The effects of the extracellular environment on neuronal activity as well as the function of associated SACs are easily accessible and viable in minimal saline for ease in experimentation. The effect of extracellular [Ca2+] on membrane properties which affect voltage-sensitivity of ion channels, threshold of action potentials and refractory periods can be readily addressed in the chordotonal organ in crab limbs. It is of interest to understand how low extracellular Ca2+ enhances neural activity considering the SACs in the sensory endings maybe Ca2+ channels and that all neural activity is blocked with Mn2+. It is suggested that axonal excitability might be affected independent from the SAC activity due to potential presence of calcium activated potassium channels (K(Ca)) in the axons. We are experimenting to separate the role of Ca2+ on the

function of the SACs and the excitability of the axons in the nerves associated with chordotonal organs. These experiments may aid in understanding the mechanisms of neuronal hyperexcitability in hypocalcemia within mammals.

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Poster

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Title: Hypoglycosylation of Piezo1: new pathological mechanisms and therapeutic target for neurological disorders in phosphomannomutase-2 deficiency?

Authors: A. EDO, J. CARRILLO-GARCÍA, *J. FERNANDEZ-FERNANDEZ;
Univ. Pompeu Fabra, Barcelona, Spain

Abstract: Congenital Disorders of Glycosylation (CDG) is a disease family caused by mutations in genes involved in the glycosylation pathways. Phosphomannomutase-2 (PMM2) catalyzes an early step in N-glycosylation, and its genetic variation is associated to PMM2-CDG. CDGs involve a wide range of clinical manifestations with severe neurological alterations. Stroke-like episodes (SLEs) are among the acute neurological complications that may occur in PMM2-CDG, which are characterized by confusional status, hemiparesis and epileptic seizures. Although their pathomechanisms are not fully understood, head trauma has been recently described as a potential trigger. The brain is a mechanosensitive organ capable of transducing mechanical forces into neurological responses, and mechanosensitive ion channels have been suggested to underlie the transduction of these forces. Although the molecular nature of these channels remains unknown, Piezo family channels are well positioned candidates. Interestingly, Piezo1 has been identified as an N-glycosylated protein.
Our goal was to study Piezo1 hypoglycosylation in cellular physiology and its possible pathophysiological relevance for SLEs in PMM2-CDG. We studied the effects of Piezo1 hypoglycosylation on its activity and/or biophysical properties in both heterologous expression system and primary cultures of murine cortical neurons. Our results showed that hypoglycosylation of wild-type (WT) human Piezo1 (hPiezo1) by using the glycosylation

blocker kifunensine induced an increase in the mechanical sensitivity in response to increasing pulses of negative pressure on cell-attached patches of HEK293 cells. Additionally, hypoglycosylated WT hPiezo1 showed slower inactivation kinetics. Interestingly, asparagine-to-glutamine hPiezo1 mutants N2293Q and N2330Q mimicked the effect of kifunensine on channel mechanical sensitivity and inactivation kinetics. N885Q mutant also showed slower inactivation kinetics but similar mechanical sensitivity when compared to the WT channel. Similarly, kifunensine- and swainsonine-induced hypoglycosylation of Piezo1 in murine cortical neurons resulted in an increased calcium influx at the soma in response to Piezo1 specific agonist Yoda1. However, immunostaining of Piezo1 revealed a reduced channel density in neurites upon treatment with glycosylation blockers kifunensine or swainsonine.
Based on these results, we suggest that hypoglycosylation of Piezo1 might contribute to the neurological symptoms found in PMM2-CDG patients, by favoring neuronal excitability in response to mechanical stimulation.

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Poster

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Program #/Poster #: P098.05

Topic: B.04. Ion Channels

Title: Trpv1-tubulin complex: a molecular target to understand chemotherapy-induced neuropathic pain

Authors: *S. MOHANTA[1,2], S. SAHA[1,2], C. GOSWAMI[1,2];
[1]Natl. Inst. of Sci. Educ. and Res., Bhubaneswar, India; [2]Homi Bhabha Natl. Inst., Mumbai, India

Abstract: <u>TRPV1-Tubulin complex: A molecular target to understand chemotherapy-induced neuropathic pain</u>Microtubule dynamics plays important function in stabilization and maturation of dendritic spines, and thereby affects overall neuronal circuit functions. Taxol and Vincristine are commonly used compounds that alter microtubule dynamics and these are also used as chemotherapeutic agents against different forms of Cancer. One major side effect of Taxol application in cancer patients is the development of severe peripheral neuropathy, which not only affects microtubule dynamics of peripheral neurons, but also other apparently unrelated functions such as mitochondrial functions and Ca^{2+}-signalling events. In last few years it has been suggested that TRP channels are involved in the development of microtubule-drug-induced Neuropathic pain regulation. In this context, cross talk between microtubule cytoskeleton with TRP channels especially with TRPV1 and TRPV4 at various level has been demonstrated. So far multiple groups have demonstrated the importance of TRPV1 in chemotherapy induced

neuropathic pain, both *in vitro* and *in vivo* experimental model systems. TRPV1, also known as the “Capsaicin receptor” is a nonselective cation channel found in a sub-set of peripheral neurons, often localizes in the nerve endings of nociceptors and is activated by different noxious stimuli such as high temperature, low pH and a plethora of endogenous as well as exogenous chemical stimuli. Taxol is known to increase the expression of TRPV1 in DRG neurons. TRPV1 seems to regulate microtubule dynamics and cytoskeletal elements in turn regulate proper localization and function of TRPV1, presenting a strong feedback mechanism. Hence detailed understanding of TRPV1-tubulin complex appears as a potential therapeutic target relevant for cancer, cancer pain, peripheral neuropathy and many other pathophysiological systems. Previously we reported the biochemical interaction between TRPV1 and tubulin and vertebrate-specific conservation of the same. In this work, we demonstrate that specific positively charged residues present within the C-terminus of TRPV1 are crucial for tubulin interaction. Substitution of these amino acids with neutral amino acids resulting in altered sensitivity to ligands, suggesting that the TRPV1-Tubulin complex is able to regulate channel function as well. We conclude that understanding the nature of interaction between TRPV1 and microtubules is highly relevant to unravel the involvement of ion channels in chemotherapy induced pain. These findings may have broader implication in human health and welfare.

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Poster

P098. Other Ion Channels

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Topic: B.04. Ion Channels

Title: Pharmacology of Transient receptor potential cation M8 (TRPM8) channels using different activation stimuli

Authors: I. RINKE-WEIß[1], A. HORVÁTH[1], N. BECKER[1], T. A. GOETZE[1], G. OKEYO[2], T. STRASSMAIER[2], R. HAEDO[2], N. FERTIG[1], ***A. OBERGRUSSBERGER**[1];
[1]Nanion Technologies GmbH, Munich, Germany; [2]Nanion Technologies Inc., Livingston, NJ

Abstract: Transient Receptor Potential (TRP) channels are widely distributed throughout the mammalian central and peripheral nervous systems. The TRP channel subfamily M (melastatin) member 8 (TRPM8) is a nonselective cation channel, proposed to be the predominant thermoreceptor for cellular and behavioral responses to cold stimuli. TRPM8 channels can be found in multiple organs and tissues, and are involved in regulation of various important processes such as inflammatory reactions, immunomodulatory effects, pain, cell proliferation, migration and apoptosis, and vascular muscle tension. Disorders ranging from migraine, dry eye disease and cancer, amongst others, have been attributed to TRPM8 rendering this channel a

promising target for drug discovery. Here, we studied the responses of TRPM8 expressed in CHO cells activated using different stimuli on automated patch clamp (APC) systems. TRPM8 is activated by chemical cooling agents including menthol and icilin or by temperatures lower than ~26 °C. Using a high throughput APC device, TRPM8 was activated by increasing concentrations of menthol with an EC_{50} of approximately 20 µM. In addition, TRPM8 was activated when the system was cooled to 18°C and further activated at 12°C. A small, lower throughput APC device was also used to activate TRPM8 repetitively using solution at 10°C. This was subsequently blocked by increasing concentrations of capsazepine applied at 10°C with an IC_{50} of 12.9 µM. External solution warmed to 35°C also blocked the TRPM8-mediated current. To this end, TRPM8 could be reproducibly activated by different stimuli, and blocked by pharmacological agents or heated solution. Therefore, the use of TRPM8 expressed in cell lines coupled with high throughput APC provides a promising tool for basic research and may have important implications in drug development for compounds acting on TRPM8.

Disclosures: I. Rinke-Weiß: A. Employment/Salary (full or part-time):; Nanion Technologies GmbH. **A. Horváth:** A. Employment/Salary (full or part-time):; Nanion Technologies GmbH. **N. Becker:** A. Employment/Salary (full or part-time):; Nanion Technologies GmbH. **T.A. Goetze:** A. Employment/Salary (full or part-time):; Nanion Technologies GmbH. **G. Okeyo:** A. Employment/Salary (full or part-time):; Nanion Technologies Inc. **T. Strassmaier:** A. Employment/Salary (full or part-time):; Nanion Technologies Inc. **R. Haedo:** A. Employment/Salary (full or part-time):; Nanion Technologies Inc. **N. Fertig:** A. Employment/Salary (full or part-time):; Nanion Technologies GmbH. **A. Obergrussberger:** A. Employment/Salary (full or part-time):; Nanion Technologies GmbH.

Poster

P098. Other Ion Channels

Location: Virtual Only

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Program #/Poster #: P098.07

Topic: B.04. Ion Channels

Support: NIGMS-RISE R25 GM061838
NSF-PIRE OISE-1545803
National Institute on Minority Health and Health Disparities 8G12-MD007600
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National Institute of General Medical Sciences GM084854
NEURO-ID R25NS080687

Title: Intermittent cocaine self-administration but not passive cocaine infusions reduces h-current in putative dopaminergic neurons of the ventral tegmental area

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Abstract: Intermittent Access (IntA) cocaine self-administration is a protocol suggested to better simulate human addictive behavior due to the intermittent pattern of drug administration. IntA is also known to produce incentive salience and psychomotor sensitization. Dopamine (DA) neurons display a prominent mixed cation current conductance known as the hyperpolarization-activated cyclic nucleotide current, or I_h, that contributes to neural processes such as resting membrane potential, firing frequency modulation, and synaptic integration. Previous results from our laboratory demonstrated that I_h amplitude is significantly reduced after cocaine sensitization. This I_h reduction resulted in an increased temporal summation, mean depolarization and EPSP amplitude, all factors related to an enhanced excitability state. Since the cocaine sensitization model involves noncontingent drug injections given by the experimenter it is crucial to determine if electrophysiological changes in VTA DA cells are present when drugs are self-administered (contingent). In the present study we explored if DA neurons intrinsic alterations are present after exposure to IntA. Our hypothesis is that VTA DA cells intrinsic neuroadaptations are dependent on associative processes acquired during the operant conditioning. Using the whole-cell patch-clamp technique in brain slices, we investigated the effects of 3 days of cocaine IntA, and passive cocaine infusions (yoked controls) on I_h amplitude, temporal summation, and neuronal excitability. Hyperpolarizing current injections followed by sudden depolarizing steps were used to analyze rebound excitation. Increasing depolarizing current injections were used to evaluate how neurons respond to a depolarizing stimulus. To analyze synaptic integration, a 33-Hz train of 5 αEPSCs ($\alpha = 5$ ms; Imax = 50) were injected into the soma of putative VTA DA neurons when clamped at -70 mV. Our results demonstrate that an IntA protocol, but not passive cocaine infusions, produces a significant I_h amplitude reduction ($P<0.05$). No differences in rebound action potentials (APs) where observed. A depolarizing current protocol showed a significant increase in the number of APs ($P<0.05$). Temporal summation was increased at depolarized potentials in the IntA group and Yoked controls ($P<0.0001$). These results suggests that I_h modulation and intrinsic activity regulation is dependent on associative learning to drug cues. The findings also suggest that enhanced synaptic integration could possibly be a cocaine-induced pharmacological effect.

Disclosures: C. Calo: None. **O. Velez-Acevedo:** None. **K.Y. Bosque Cordero:** None. **D. Consuegra Garcia:** None. **R. Vázquez-Torres:** None. **C.A. Jimenez-Rivera:** None.

Poster

P143. Epilepsy: Mechanisms and Networks

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P143.01

Topic: B.10. Epilepsy

Support: European Research Council (ERC) grant No 855109
INCR predilepsy project

Title: Absence seizure simulations in a full virtual brain model explain the transient loss of consciousness

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Abstract: Conscious perception results from an interplay between two processes interacting with each other: *awareness* and *wakefulness*. *Awareness* is supported by large scale cortico-cortical functional connectivity whereas *wakefulness* depends critically on thalamocortical connectivity[1]. We have recently developed a full virtual human brain computational model named COALIA able to simulate realistic EEG, as compared to real EEG recorded in human at two level of consciousness during wake and deep sleep[2].Typical absence seizures are characterized by a sudden and transient (10-15 sec) loss of conscious perceptions associated with an interruption of ongoing activities associated with a fixed gaze. At the same time, the EEG shows generalized spike-wave discharges (SWD) at 2.5-4 Hz at the level of scalp electrodes. In childhood absence epilepsy, these seizures can occur in children up to 200 times a day, which can lead to attention, cognition, memory and mood disorders and ultimately to learning disabilities at school. In this study we used COALIA to understand the neurobiological mechanisms of typical absence seizures and how these mechanisms are responsible for a transient loss of consciousness. Accounting for brain characteristics present in immature brain, and the human structural connectivity we were able to generate realistic absence seizures with our computational model. Results suggest that a focal initiation in the frontal cortex triggers a switch in the firing mode of the cognitive/associative nuclei of the thalamus from tonic to “UP and down” discharge mode due to and excessive feed forward cortico-thalamic inhibition. This thalamic bursting mode initiate spike-wave discharge onto the associative area of the cortex through an exaggerated thalamocortical feed forward inhibition (hyperexcitation of cortical SST GABAergic subtype onto pyramidal cells) that explain the large wave of the SWD. This large cortical inhibition transiently disrupts the cortico-cortical connectivity by suppression of the PV-PC loop. Simulated TMS-evoked potential confirmed a dramatic drop of the consciousness index during absence seizures.1: Decoding the circuitry of consciousness: From local microcircuits to brain-scale networks, 2020. PMID: 325375302: COALIA: A Computational Model of Human EEG for Consciousness Research. 2019 , PMID: 31798421

Disclosures: P. Benquet: None. **M. Yochum:** None. **M. Kuchenbuch:** None. **R. Nabbout:** None. **F. Wendling:** None.

Poster

P143. Epilepsy: Mechanisms and Networks

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Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P143.02

Topic: B.10. Epilepsy

Support: NIH/ NINDS grant number NS115049 awarded to Dr. Isgor

Title: Sleep-seizure electroencephalography associations predictive of severity and death risk in epilepsy

Authors: *A. TYULMENKOVA, G. GARCIA-MENDOZA, D. KEITH, C. ISGOR;
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Abstract: Sleep-seizure electroencephalography (EEG) associations may serve as biomarkers to track severity of upcoming seizures in order to monitor death risk using a new mouse model of adult-onset, spontaneous epilepsy. The mice overexpress brain derived neurotrophic factor (BDNF) in the forebrain under the CAM kinase II alpha promoter (TgBDNF). TgBDNF mice develop seizures at ~4 months of age which worsen with each episode. Seizures are elicited via tail lifts and cage agitation. Seizures become more severe as indicated by increased duration of postictal cortical activity suppression (i.e., EEG flat or EEG waves in delta frequency) associated with loss of posture/consciousness. In more advanced stages, the mice expire following long period of postictal generalized EEG suppression resulting in cardiorespiratory arrest. We surgically implanted subdural cortical electrodes on the mice skulls prior to the first seizure episode (3-channel EEG system, Pinnacle Technology, KS) to record seizure and sleep EEG for 7 weeks. The recordings targeted time periods of moderate to severe phases of epilepsy. We used age- and litter-matched wild-type mice as controls. We hypothesize that as epilepsy progresses, detectable alterations in sleep structure may indicate upcoming life-threatening seizures. Twenty-four hours after seizure induction, mice were connected to EEG apparatus for recording sleep in inactive phase. We analyzed early (first 1 hr) and late (last 1 hr) sleep separately to define the changes in rapid eye movement (REM) and slow-wave sleep (SWS) phases. Data was assessed by simple regression analyses between multiple seizure and sleep parameters. A shift in increased duration of SWS in late compared to early sleep was linked to a remarkable increase in postictal generalized cortical suppression duration. Shorter delay to onset of SWS in late phase (last 1 hr) of sleep was associated with increased total seizure duration. Number of REM bout counts in early sleep was negatively associated with duration of postictal generalized cortical suppression (delta frequency), ictal duration and ictal spectral power. Number of REM bout counts in late sleep was negatively associated with length of postictal generalized cortical suppression (delta frequency) and cumulative duration of spiking wave discharges observed in seizure recovery accompanied by behavioral freezing. Our data reveals that dynamic alterations in sleep-seizure EEG measures indicate epilepsy severity progression. Identifying critical sleep-

seizure EEG associations as epilepsy progresses may predict severity of upcoming seizures and, in turn may inform impact of such episodes on sleep architecture.

Disclosures: A. Tyulmenkova: None. **G. Garcia-Mendoza:** None. **D. Keith:** None. **C. Isgor:** None.

Poster

P143. Epilepsy: Mechanisms and Networks

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P143.03

Topic: B.10. Epilepsy

Support: NIH 1R21NS106434

Title: Interictal bursts in mouse model of acute seizure engage hippocampal pyramidal cells and resemble mini seizures

Authors: Y. CHEN[1], N. DRISCOLL[1], P. MULCAHEY[1], F. VITALE[1], *H. TAKANO[2,1]; [1]Univ. of Pennsylvania, Philadelphia, PA; [2]The Children's Hosp. of Philadelphia, Philadelphia, PA

Abstract: Interictal bursts, abnormal electrographic activity characterized by brief epileptiform discharges, are commonly observed in intracranial EEG recordings from both human patients and animal models. Despite the ubiquity of these bursts, the cellular nature of interictal activity and their relationship to seizures remain unclear. In this study, we aim to understand how electrographic activity and cellular recruitment during interictal bursts change over the course of acutely induced seizures. In the hippocampus of transgenic mice expressing calcium indicator gCaMP6 in pyramidal neurons, we implanted a transparent graphene-based microelectrode array (Gr MEA). This set-up allows us to simultaneously follow pyramidal cell recruitment in the dorsal hippocampus by two-photon microscopy and record electrographic signals from the surface of the hippocampus. To induce seizures, we applied a convulsant, 4-aminopyridine, to the contralateral cortical surface. We present two main findings: 1) During interictal events, pyramidal cell recruitment corresponds to periods of high gamma activity (80 - 250 Hz). Line length in the high gamma band, a feature which measures signal energy and complexity, predicts pyramidal cells recruitment during interictal activity. 2) Interictal bursts resemble mini seizures - metrics that are elevated during seizures, such as phase locked high gamma, LFP (4 to 30 Hz), and high gamma broadband power are transiently elevated during the onset of interictal activity. Taken together, these results suggest that interictal bursts manifest as mini seizures that are rapidly quashed. Excitatory principal cell activity is enhanced during the onset of the burst, a period where the interictal activity most closely resemble seizure onset. However, they are quickly silenced. By identifying the cells that are recruited during the onset of interictal bursts,

we can speculate about the mechanisms and cellular identity of interictal activity termination. Inhibitory interneurons are a likely candidate, and the Gr MEA enables us to elucidate cell type specific dynamics during interictal bursts.

Disclosures: Y. Chen: None. **N. Driscoll:** None. **P. Mulcahey:** None. **F. Vitale:** None. **H. Takano:** None.

Poster

P143. Epilepsy: Mechanisms and Networks

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P143.04

Topic: B.10. Epilepsy

Title: Analysis of network changes in the kindling model of temporal lobe epilepsy

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Abstract: Analysis of network changes in the kindling model of temporal lobe epilepsy
In this work we analyzed the firing rate (FR) of single neurons involved in the neural network during hippocampal rapid kindling (RK 3 days). Thirty male Wistar rats were implanted with a bipolar Macroelectrode (MacroE) for stimulation in the CA1 region of the right ventral hippocampus and also, eight Microelectrodes (MicroE) were implanted to record the electrical activity of the CA1 region of the right dorsal hippocampus. Two groups were created out of the animals that successfully completed the procedure: The RK group, consisting of 3 animals and the sham group with the remaining rat that was implanted but not stimulated. We proceeded according to the RK model stimulation parameters, and we analyzed the electrical signal. The analysis was performed in 57 single neurons in total, 47 in the RK group and 10 in the sham group. The FR was determined during the Basal (pre-stimulations) and After-Discharge (AD) periods. The analysis of the Basal and AD periods of kindled animals showed a progressive increase in the daily average of the FR along the protocol and a significant difference was found between day 3 and day 1. We also found that data dispersion increased as the stimulations progressed. On the other hand, the qualitative analysis of the FR evidenced a possible classification of the neurons, with some of them increasing their FR during the AD period -the excitatory type (35%)-, some decreasing it -inhibitory type (32%)-, and some not changing their FR at all, not directly involved in the epileptogenic network (33%). We demonstrated that the FR

is strongly affected by the epileptogenic activity associated with the RK. We believe these findings are relevant for a better understanding of kindling-associated network changes and might provide new targets for therapeutic strategies in patients with epilepsy.

Disclosures: **A. Nasimbera:** A. Employment/Salary (full or part-time):; CONICET. **S. Cordisco:** None. **M. Tibaldi:** None. **B. Gori:** None. **F. Nishida:** None. **L. Ledesma:** None. **S. Kochen:** A. Employment/Salary (full or part-time):; CONICET.

Poster

P143. Epilepsy: Mechanisms and Networks

Location: Virtual Only

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Program #/Poster #: P143.05

Topic: B.10. Epilepsy

Support: NIH Grant R01NS094211
NIH Grant R03NS103111
NIH Grant R01NS110012
NIH Grant 1T32NS09957801A1

Title: Automated prediction of epileptic activity in mouse models of epilepsy

Authors: *A. I. BECK[1], D. KUNDA[2], R. E. A. SANCHEZ[3], A. M. BARD[4], J. SKIBO[4], K. J. MILLEN[4], F. K. KALUME[4], H. O. DE LA IGLESIA[2];
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Abstract: Epilepsy is a neurological disorder characterized by the presence of excessively synchronized neural activity (seizures) and epileptiform discharge (interictal spikes). In addition, epileptic activity is modulated by circadian rhythms and the sleep-wake cycle. Because increased frequency of interictal spikes can be associated with seizure risk, real-time detection of interictal spikes could lead to seizure prediction and interventions to prevent seizures or their adverse outcomes. Using mouse models of epilepsy, we developed a machine-learning (ML) based method for autonomously detecting interictal spikes. Neural activity was chronically recorded in mice using two electrocorticography electrodes and one electromyography electrode. We used mouse models of Dravet syndrome (DS; heterozygous *Scn1a* gene deletion) and focal cortical dysplasia (FCD; PIK3CA gene mutation), along with corresponding wild-type controls. Data were normalized to a sampling rate of 400 Hz and min/max value of ±400 mV, and interictal spikes were manually scored in a subset of data. A total of 96 time- and frequency-based features were extracted in epochs of 2 seconds. The data were randomly under-sampled to ensure balanced classes and used to train a single-layer Perceptron-based ML model using L1 regularization to classify interictal spikes. The outcome was a binary classification; interictal

spike present/absent in the given epoch. The fully trained model had a mean test accuracy of 74.56 (SD=18.66%). When separated by mouse model, the accuracy was 75.39% for DS signals, 64.24% for FCD signals, and >95% for corresponding wild-type signals. The model achieved recall and precision values of 1.00 and 0.84 for epochs without interictal spikes, and recall and precision values of 0.06 and 0.75 for epochs with interictal spikes. False positive epochs tended to resemble manually labeled interictal spikes. Additionally, in DS signals, interictal spikes predicted by the model displayed circadian rhythm-modulated frequency in multi-day recordings, as well as increased interictal spike frequency and circadian rhythm disruption when transitioning between light-dark cycles. These results suggest our ML-based method captures epileptic activity in two mouse models of epilepsy with differing genetic etiology and presentation of epileptic activity. Because the model uses regularization aimed at reducing data dimensionality, this algorithm could be used to detect interictal spike frequency in real-time, and potentially forecast potential seizure onset.

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Poster

P143. Epilepsy: Mechanisms and Networks

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Program #/Poster #: P143.06

Topic: B.10. Epilepsy

Support: CURE Epilepsy Award
NIH Grant K08NS105929
NIH Grant R21NS122011

Title: Sleep is Fragmented and Reduced in a Mouse Model of Temporal Lobe Epilepsy

Authors: *N. P. PEDERSEN[1], L. M. AIANI[2], L. I. ROSENBERG[2], B. J. HARVEY[3]; [1]Neurol. and Biomed. Engin., [2]Neurol., [3]Neurol. and Program in Neurosci., Emory Univ., Atlanta, GA

Abstract: Sleep and epilepsy are inextricably and bidirectionally intertwined. Sleep disruption occurs in human epilepsy, and sleep loss can result in worsening seizures. Sleep-wake brain networks determine forebrain rhythms and excitability, but the role of these circuits in epilepsy is unknown. In people with epilepsy, sleep disorders, particularly insomnia, are worse. These sleep disorders may also contribute to other comorbidities of epilepsy. We implanted 47 mice with electrocorticogram, hippocampal, and EMG electrodes. Mice were recorded for a baseline week with synchronized video before experimental mice (n =37) received intraamygdala kainate injections, and control mice (n = 10) underwent intraamygdala saline injections. Mice were then

recorded for three weeks before perfusion for histological analysis. 642 seizures were recorded in 23 experimental mice. Experimental mice with seizures (n = 10, >= 19 seizures) had significantly more wake than control mice (10.4% increase ± 3.5 (SE), two way ANOVA, $p < 0.01$). Seizures were most common in the waking state, but proportionally seizures were most common in the post-ictal state ('clustering'), REM, then wakefulness, and NREM sleep. Sleep was also fragmented in experimental mice. These data provide a parallel of human sleep disruption in epilepsy. Studies now examine the mechanisms of sleep disruption, as well as manipulations of sleep-wake circuits to treat sleep disorders in epilepsy, ameliorate comorbidities, and a as novel candidate means to treat epilepsy and seizures.

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Poster

P143. Epilepsy: Mechanisms and Networks

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Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P143.07

Topic: B.10. Epilepsy

Support: KAKENHI 16H06532,16K21734,16H06524
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KAKENHI 21K06702,21H03532,20H04341,19H01142,19K12190
KAKENHI 21K15247

Title: In vitro assay for seizure liability using voltage-sensitive dye imaging (VSDI) in the mouse hippocampus

Authors: *Y. UTSUMI[1,2], M. TAKETOSHI[2], N. OHARA[3], M. MIWA[3], Y. TOMINAGA[2], T. TOMINAGA[1,2,4];
[1]Grad. Sch. of Pharmaceut. Sciences, Tokushima Bunri Univ., Sanuki, KAGAWA, Japan; [2]Inst. of Neuroscience, Tokushima Bunri Univ., Sanuki, KAGAWA, Japan; [3]Kagawa Sch. of Pharmaceut. Sciences, Tokushima Bunri Univ., Sunuki, KAGAWA, Japan; [4]Kagawa Sch. of Pharmaceut. Sciences, Tokushima Bunri Univ., Sanuki, KAGAWA, Japan

Abstract: The central nervous system (CNS) toxicity is a critical factor among possible adverse effects in clinical and non-clinical trials in drug development. Seizure and convulsions are frequent appearances of the CNS-toxicity and are the leading cause of drug development attrition. The existing state of seizure liability assessment relies on conventional behavioral batteries, such as Irwin's test and the functional observational battery (FOB), to detect seizure and convulsions susceptibility. Recently, several laboratories have developed in vitro electrophysiological assay system using hippocampal brain slices from various animal species

(rats, dogs, monkeys, and mini-pigs) for seizure liability assessment. Here, we report a novel in vitro assay system using voltage-sensitive dye (VSD) imaging in mice hippocampus. The VSD allows us to record the membrane potential change in the neurons entire membrane components under a field of view. That is, we can record action potential (AP) components and the excitatory and inhibitory postsynaptic potentials (EPSP and IPSP) even below the threshold. We prepared acute transverse hippocampal slices (350 µm thick) from the two age groups (4 - 7, and 8 - 10 weeks of female mice). Electrical stimulation was applied onto Schaffer collateral pathway from a stimulation electrode placed near the CA1/CA3 border.The responses were recorded every thirty seconds with an imaging system (MiCAM02, Brainvision Ltd.) and field potential recordings from stratum radiatum. Four convulsive compounds of picrotoxin (PiTX; 1, 10, and 100 µM), SR95331 (Gabazine; 0.1, 1, and 10 µM), 4-aminopyridine (4AP; 1, 40, and 100 µM), and pilocarpine (Pilo; 10, 30, and 100 µM) are tested. We perfused the drugs with cumulative ascending concentrations for 20 min per concentration. We designed the tests to include the stimulus-response relationships during perfusion. PiTX and gabazine ($GABA_A$ receptor antagonist) enhanced the duration of the response in the stratum radiatum (SR). An application of 4AP induced significant slow components in the SR. We found that the application of Pilo suppressed EPSP and thus action potential. The different effect on CA1 activity indicates that the system can evaluate drugs on different mechanisms in the neurons, thus examining CNS toxicity (seizure liability).

Disclosures: Y. Utsumi: None. **M. Taketoshi:** None. **N. Ohara:** None. **M. Miwa:** None. **Y. Tominaga:** None. **T. Tominaga:** None.

Poster

P196. Alzheimer's Disease: Genetics, Genomics, and Other Omics Approaches V

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P196.01

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Owens Family Foundation
Cure Alzheimer's Fund
NIH grant AG051085

Title: Tandem transcriptional and translational analysis of gene expression in mouse models of Alzheimer's disease

Authors: G. EASTMAN[1,2], E. R. SHARLOW[1], J. S. LAZO[1], J. R. SOTELO-SILVEIRA[2], *G. S. BLOOM[1];
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Abstract: Defining cellular mechanisms that drive Alzheimer's disease (AD) pathogenesis and progression is aided by studies describing how gene expression patterns change during pre-symptomatic AD and the ensuing periods of steadily declining cognition. The majority of relevant published studies have emphasized control of mRNA levels, but not translation regulation, leaving the ultimate results of gene expression relatively unexplored in the context of AD. To identify genes whose expression might be regulated at the transcriptional, and especially at the translational levels in AD, we analyzed gene expression in cerebral cortex of two AD model mouse strains, CVN (APP_{SwDI};$NOS2^{-/-}$) and Tg2576 (APP_{Sw}), and their companion wild type (WT) strains at 6 months of age by tandem RNA-Seq and Ribo-Seq (ribosome profiling). Identical starting pools of bulk RNA were used for RNA-seq and Ribo-seq. Differential gene expression analysis was performed at the transcriptional and translational levels separately, and also at the translational efficiency level. Regulated genes were functionally evaluated by gene ontology tools. Compared to WT mice, AD model mice had similar levels of transcriptional regulation, but displayed differences in translational regulation. A specific microglial signature associated with early stages of Aβ accumulation was up-regulated at both transcriptome and translatome levels in CVN mice. Although the two mice strains did not share many regulated genes, they showed common regulated pathways related to APP metabolism associated with neurotoxicity and neuroprotection. This work represents the first genome-wide study of brain translation regulation in animal models of AD, and provides evidence of a tight and early translational regulation of gene expression controlling the balance between neuroprotective and neurodegenerative processes in brain.

Disclosures: G. Eastman: None. **E.R. Sharlow:** None. **J.S. Lazo:** None. **J.R. Sotelo-Silveira:** None. **G.S. Bloom:** None.

Poster

P196. Alzheimer's Disease: Genetics, Genomics, and Other Omics Approaches V

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P196.02

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH R01AG067025
NIH R21CA237955
NIH U01MH116492

Title: Analysis of cell-type gene regulatory network characteristics in Alzheimer’s disease

Authors: *C. GUPTA, D. WANG;
Univ. of Wisconsin Madison, Madison, WI

Abstract: Alzheimer's Disease (AD) is a chronic neurodegenerative disorder with complex molecular mechanisms that can be highly cell-type specific. Gene regulatory networks, a key mechanism controlling gene expression of single cells, serve as powerful tools to explain the cellular heterogeneity underlying AD phenotypes. In this study, we analyzed gene regulatory networks depicting interactions between transcription factors, regulatory elements, and target genes for major cell types in the human brain: excitatory and inhibitory neurons, microglia, and oligodendrocytes. We identified genes that act as hubs in different cell types using various network centrality measures and found that several functional pathways showed centrality differences between healthy and AD conditions, such as synapse organization in neurons and neutrophil-mediated immunity in microglia. Because gene regulatory networks naturally form a hierarchy with downward information flow, we also estimated hierarchical scores of genes over each network. We found that genes related to cognition and glial cell fate commitment had the most extensive changes in the hierarchical scores in microglia and oligodendrocytes, respectively. In contrast, hierarchical changes in neuronal cells were relatively less extensive. We also estimated the level of co-regulation among genes and identified cell-type-specific modules consisting of known AD-risk genes. For example, one microglia module consists of 7 known AD-risk genes, including HS3ST1, KAT8, SORL1, TREM2, BIN1, ABI2, and ECHDC3. Furthermore, we show that the network-based features of known AD-risk genes can be used in a machine learning framework to predict new genes potentially involved in AD pathogenesis. These results suggest that our network analysis of cell-type gene regulatory networks can complement AD gene prioritization and functional interpretation. Finally, our analysis is open-source available as a general pipeline and has potential applications to other neurodegenerative diseases.

Disclosures: C. Gupta: None. **D. Wang:** None.

Poster

P196. Alzheimer's Disease: Genetics, Genomics, and Other Omics Approaches V

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P196.03

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant 1R01 AG065206-01
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Mathers Charitable Foundation

Title: Early changes in spine density and gene expression in cortex of APP/PS1 mice

Authors: *J. E. TOMORSKY, M. K. DREWS, K. R. LEBLANC, C. J. SHATZ;
Dept. of Biol., Stanford Univ., Stanford, CA

Abstract: Understanding the early pathogenesis of Alzheimer's Disease (AD) is an important step in designing treatments and preventing more advanced pathology and cognitive decline in the later stages of the disease. Early in the progression of AD, soluble forms of oligomeric amyloid beta (oAbeta) are thought to increase, leading to deficits in synaptic plasticity and synapse loss. We have previously shown that in an APP/PS1 mouse model of AD, ocular dominance plasticity is diminished as early as P30, long before plaque formation and overt cognitive defects are detectable. These early deficits in ocular dominance plasticity were rescued by knocking out the paired immunoglobulin-like receptor B (PirB), a high-affinity neuronal receptor for oAbeta. Here we show preliminary data suggesting that structural changes in synapse density might also occur at P30 in an APP/PS1 mouse model. Dendritic spine densities on L5 basal dendrites in visual cortex trended lower in APP/PS1 mice relative to healthy controls at P30. This trend was not observed when PirB was knocked out, suggesting that oAbeta may act through PirB to decrease spine density long before overt pathological or behavioral changes are detectable. We also identified several neuronal transcripts that were differentially expressed between APP/PS1 and WT mice at P30 using the RiboTag technique to isolate ribosome-bound mRNA in neurons followed by RNA-seq. Differentially expressed genes largely fell into Gene Ontology pathways relating to neuropeptide signaling, MAPK signaling, axon development, circadian rhythm, cell adhesion, memory, and apoptosis. Together these results indicate that the very early accumulation of oAbeta can trigger significant changes in neuronal gene expression which accompany disruptions in synaptic plasticity and possible synapse loss.

Disclosures: J.E. Tomorsky: None. **M.K. Drews:** None. **K.R. LeBlanc:** None. **C.J. Shatz:** None.

Poster

P196. Alzheimer's Disease: Genetics, Genomics, and Other Omics Approaches V

Location: Virtual Only

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Program #/Poster #: P196.04

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: 5R01AG056287-04
5R01AG057915-04
U19 AG065156-01

Title: Identifying differences in cellular and disease phenotypes across human Alzheimer's disease tissue by multiplexed ion beam imaging

Authors: *B. CANNON, K. VIJAYARAGAVAN, D. TEBAYKIN, D. MRDJEN, J. OLIVERIA, A. BARANSKI, N. GREEENWALD, M. ANGELO, T. MONTINE, S. BENDALL; Stanford Univ., Palo Alto, CA

Abstract: Identifying cell-based and regional phenotypes in human brain tissue is a necessary task to understand neurodegenerative pathologies in Alzheimer's disease. Using MIBI (multiplexed ion-beam imaging) we have simultaneously quantified tissue-level expression of 40 different targets encoding a deep set of neurological phenotypes with nanometer scale spatial resolution. To localize protein expression to specific cells or tissue restricted regions, we use a combination of single cell and single object segmentation methods. Applying this technique to multiple MIBI scans of archival human hippocampus cross-sections, we are able to identify the spatial distribution and expression heterogeneity of glial cell types and proteopathies in both cognitively healthy and diseased human brain. Using off-the-shelf and in-house developed computational strategies we are able to identify unique protein-disease relationships in neurons that may lead to additional work testing the functional significance of the descriptive work outlined here. We believe this will be a straight forward approach to discretize and select features for multiplexed imaging technologies as applied to human brain tissue in both degenerative and non-degenerative contexts.

Disclosures: B. Cannon: None. **K. Vijayaragavan:** None. **D. Tebaykin:** None. **D. Mrdjen:** None. **J. Oliveria:** None. **A. Baranski:** None. **N. Greeenwald:** None. **M. Angelo:** None. **T. Montine:** None. **S. Bendall:** None.

Poster

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Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH/NIA RF1 AG059778

Title: Hypothalamic RNAseq highlights lysosomal gene Laptm5 as a potential driver of cognitive and metabolic dysfunction in Alzheimer's disease

Authors: *M. DAI, A. DUNN, J. ZHANG, V. PHILIP, K. O'CONNELL, C. C. KACZOROWSKI;
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Abstract: Alzheimer's disease (AD) is the most common form of dementia that leads to progressive decrease in memory, cognitive function, and eventually ability to perform daily tasks. Cognitive dysfunction in AD is often accompanied (and preceded) by metabolic dysfunction, which may worsen neurological symptoms and quality of life in patients. It has been difficult to identify gene modifiers of AD that may only make a relatively small contribution to the overall disease risk. However, we recently developed a panel of AD mouse strains (AD-BXDs; Neuner, et al. 2019) that harbors the genetic and phenotypic heterogeneity similar to

those observed in human AD patients. The AD-BXDs are an ideal resource to identify genetic modifiers of late-onset AD that manifest through molecular, neurological and metabolic phenotypes. Using this model, we performed weighted gene co-expression network analysis (WGCNA) in hypothalamic bulk RNA sequencing data. We identified several gene modules whose expression levels significantly correlate with cognitive and metabolic traits. Gene ontology (GO) enrichment search showed that functions of these modules included immune response, neuronal myelination, cell metabolic process, etc. We identified *Laptm5* as the hub gene of the immune response module, and showed it has higher expression in aged and AD mice. Multiple previous studies have proposed *Laptm5* as a target associated with amyloid-beta load in both human and mouse data (Salih et al. 2019, Chang et al. 2017, Bonham et al. 2019, Patel et al. 2020). In conclusion, we hypothesize that *Laptm5* modifies vulnerability to both cognitive and noncognitive symptoms in AD.

Disclosures: M. Dai: None. **A. Dunn:** None. **J. Zhang:** None. **V. Philip:** None. **K. O'Connell:** None. **C.C. Kaczorowski:** None.

Poster

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Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant 3RF1AG059717-01S1

Title: Microglial INPP5D isoform expression in AD and AD-associated SNP contributions to allelic expression

Authors: *D. ZAJAC[1,2], J. SIMPSON[1,2], J. MORGANTI[2,3], S. ESTUS[1,2];
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Abstract: *INPP5D* (Phosphatidylinositol-3,4,5-trisphosphate 5-phosphatase 1), the gene encoding SHIP1, contains single nucleotide polymorphisms (SNPs) that are strongly associated with Alzheimer’s Disease (AD) risk. In the brain, *INPP5D* is expressed as several isoforms, mostly in microglia. Full-length SHIP1 is encoded by 27 exons, including an amino-terminal SH2 domain followed by the phosphatase domain. Truncated isoforms lacking the SH2 domain begin from internal transcription start sites. To better understand the function of *INPP5D* in the human brain, we investigated *INPP5D* isoform expression as a function of AD status, and AD-associated SNPs. Single cell RNAseq was performed on APP/PS1 mice to understand the relationship between *INPP5D* expression and microglial activation. The expression of microglial *INPP5D* isoforms was analyzed using quantitative polymerase chain reaction on RNA from AD

and non-AD anterior cingulate human brain samples. Samples were genotyped for the AD-associated SNPs rs35349669 and rs10933431 using TaqMan SNP kits (Thermo). Isoform expression results were analyzed as a function of microglial gene expression (ITGAM and AIF1), total *INPP5D* expression, AD status, and SNP status. In addition, reporter SNPs in the 5' UTR region were used to detect unequal allele expression in the two full-length isoforms of *INPP5D* as a function of AD SNP status. APP/PS1 mice showed an increased expression of disease-associated microglia compared to controls. *INPP5D* was expressed at equivalent levels in both homeostatic and disease-associated microglia. Expression of each *INPP5D* isoform was strongly correlated with microglial gene expression and showed an increase with AD neuropathology but did not show a significant association with AD SNPs. Sequencing of samples heterozygous for both AD SNPs provided preliminary evidence for unequal allele expression modulated by SNP status. Our next step is to quantify the unequal expression using next generation sequencing. In summary, *INPP5D* expression is uniform in both disease-associated and homeostatic microglia. Expression of *INPP5D* isoforms is increased with AD neuropathology. The mechanisms whereby AD genetics influence *INPP5D* expression is not clear, but the rs35349669 allele that increases AD risk may be associated with allelic expression.

Disclosures: D. Zajac: None. **J. Simpson:** None. **J. Morganti:** None. **S. Estus:** None.

Poster

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Harold and Margaret Southerland Alzheimer's Research Fund

Title: Heterogeneity of APOER2 isoforms specific to AD human brains

Authors: *C. GALLO[1,3], A. LABADORF[2], A. HO[3], U. BEFFERT[3];
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Abstract: Age and the ε4 allele of apolipoprotein E (APOE) are the two greatest risk factors for developing Late Onset Alzheimer's Disease (LOAD). APOE mediates its physiological effects in the brain by binding to cognate receptors like APOER2. Recent studies have discovered a novel link between aging, neurodegeneration and alternative splicing. Interestingly, APOER2 is highly alternatively spliced in neurons, and one alternative splicing event in APOER2 was identified

previously as altered in Alzheimer's disease (AD). Therefore, abnormal alternative splicing of receptors like APOER2 in aging and neurodegeneration may alter the interactome of the key AD risk factor APOE. Investigating how altered splicing of APOE receptors alters APOE physiological function highlights a new scientific paradigm to investigate the role of APOE in LOAD. Based on this rationale, we hypothesize that APOER2 isoform balance is progressively perturbed in AD contributing to synaptic and neuronal dysfunction. To examine APOER2 isoform distribution across the AD brain, we have utilized long-read single molecule RNA sequencing to generate novel APOER2 transcript maps from the hippocampus and the parietal cortex of three female control patients and three female Braak Stage IV LOAD patients. Our data indicates both common and unique isoforms amongst control and AD patients in the hippocampus and the parietal cortex and highlighted several isoforms of interest. Using in vitro cell based assays, we found hippocampal AD-specific APOER2 isoforms show unique functional biology compared to control-specific isoforms, indicating the combination of alternative splicing events in APOER2 is critical to defining APOER2 receptor biology. To ensure experimental rigor and analyze APOER2 splicing quantitatively, we will examine over 200 control and AD tissue samples in the hippocampus, prefrontal cortex, parietal cortex and control region cerebellum using qPCR. Our findings suggest a novel scientific model to view conferred APOE risk in AD through altered splicing of APOER2.

Disclosures: C. Gallo: None. **A. Labadorf:** None. **A. Ho:** None. **U. Beffert:** None.

Poster

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Topic: C.02. Alzheimer's Disease and Other Dementias

Support: F99NS120365
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R21AG068370

Title: Novel TREM2 isoforms highly expressed in human brain and their potential impact on Alzheimer's disease

Authors: *B. C. SHAW[1], H. C. SNIDER[2], D. J. ZAJAC[1], J. F. SIMPSON[1], S. ESTUS[3]; [1]Physiol., [2]Univ. of Kentucky, Lexington, KY; [3]Physiol., Sanders-Brown Ctr. Aging, Lexington, KY

Abstract: Multiple variants in *TREM2* which decrease protein function act as risk factors for Alzheimer's Disease (AD). In the brain, *TREM2* is primarily expressed in microglia. The five exons of the *TREM2* gene encode a cell surface receptor with exon 2 encoding the ligand binding

domain, while exon 4 encodes the transmembrane domain. In this study, we have found alternative splicing within *TREM2* mRNA from human brain tissue which results in skipping of exons 2, 3, and 4 (D2-, D3-, and D4-TREM2, respectively) with some isoforms having multiple exons skipped. We also show that these isoforms are not brain-specific as they are detected in multiple tissues. Quantitative PCR data indicate exon 2 is skipped at a frequency of approximately 11.4%. We are evaluating whether there is a functional genetic variant which increases exon 2 skipping. In transiently transfected HMC3 human microglial cells, we show using confocal microscopy that the novel *D2-TREM2* mRNA isoform is translated into protein and are currently investigating subcellular localization. Hence, we propose that these novel mRNA isoforms are likely translated to protein and represent alternative splicing-derived mimetics of the soluble TREM2 (sTREM2) and C-terminal fragment TREM2 which result from ADAM10 and gamma secretase proteolytic cleavage. Overall, modulating *TREM2* splicing represents a potential avenue for AD treatment strategies.

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Poster

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Garrison Institute on Aging, TTUHSC
Center for Translational Neuroscience and Therapeutics, TTUHSC

Title: All-trans retinoic acid deficiency in Alzheimer’s disease: evidence from human transcriptomics and a novel mouse model

Authors: J. ALMAGUER[1], B. R. KISBY[1,2], K. CASTENEDA[1], A. J. PASCULLO[1], R. BARNES[1], M. MANCZAK[1,3,4,5], J. D. BAILOO[6], I. PONOMAREV[1,5], ***J. J. LAWRENCE**[1,3,5]; [1]Pharmacol. and Neurosci., [2]Grad. Sch. of Biomed. Sci., [3]Garrison Inst. on Aging, [4]Neurol., [5]Ctr. for Translational Neurosci. and Therapeut., [6]Cell Biol. and Biochem., Texas Tech. Univ. Hlth. Sci. Ctr., Lubbock, TX

Abstract: Alzheimer’s disease (AD) features oxidative stress, transcriptional dysregulation, and hippocampal-dependent learning impairment. The antioxidant all-trans retinoic acid (ATRA), the active form of retinol, plays dual roles as a ROS scavenger and transcriptional regulator via its

function as an agonist at retinoic acid receptors (RARs). We hypothesized that reduced bioavailability of ATRA in the hippocampal dentate gyrus (DG) is associated with AD. To investigate whether ATRA deficiency occurs in the hippocampus of human AD, we performed a secondary analysis of hippocampal transcriptomic data from post-mortem AD and control brains (van Rooij et al. 2019). ATRA-responsive genes (ADLH1A3, CYP26A1, CYP26B1, RBP1, RBP4, NGRN, GAP43, and CALB1) were downregulated, but RAR co-repressors (NCOR1, PML, ZBTB16, and TNIP1) were upregulated. Moreover, consistent with antioxidant depletion, ROS sensors Nrf1/Nrf2 (NFE2L1/NFE2L2) were upregulated. In the J20 AD mouse model crossed with a somatostatin-CRE:tdTomato (SOM:tdT) reporter mouse, we tested the hypothesis that ATRA delays AD-related behavioral, circuit, and transcriptional abnormalities in the DG (3-9 mice per group, 4.8 ± 0.8 months of age, M/F sexes, 31 mice total). We administered ATRA (20 mg/kg) or vehicle (corn oil) IP 3x/week for 8 weeks, followed by Y-maze and open field maze (OFM) testing. Consistent with the hyperactivity phenotype of J20 AD mice, vehicle-treated (VT) mice traveled a greater distance compared to WT mice ($U = 5.00$, $p = 0.009$). By contrast, ATRA-treated (RT) AD and WT mice did not vary in their overall distance traveled ($U = 16.00$, $p = 0.727$). In the OFM, a similar pattern of results was observed. We then performed correlative histological and RNA-seq analyses on DG tissue. Through SOM:tdT expression, we found that DG SOM circuits were aberrantly targeted to the DG inner molecular layer in $J20^{+/-}$ AD mice. By contrast, ATRA treatment prevented formation of this aberrant SOM:tdT circuit in 6/6 males but not in 3/3 female mice. Finally, genes differentially expressed between genotypes and treatments were analyzed using Ingenuity Pathway Analysis. Pairwise comparison of VT WT and VT AD groups indicated dysregulation of Synaptogenesis, PKA, and Ca^{2+} Signaling pathways. ATRA treatment partly normalized these pathways. In summary, chronic treatment with ATRA normalized behavior, prevented formation of aberrant SOM:tdT circuits in the DG, and normalized 6/10 of the top dysregulated pathways. Thus, these human and preclinical studies support the hypothesis that hippocampal ATRA deficiency occurs in AD, suggesting that sustained hippocampal ATRA levels delay AD pathogenesis and progression.

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Poster

P196. Alzheimer's Disease: Genetics, Genomics, and Other Omics Approaches V

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Program #/Poster #: P196.10

Topic: C.02. Alzheimer"s Disease and Other Dementias

Title: Transcriptome-wide Involvement of the central LH receptor in the interaction between reproductive senescence and Alzheimer's disease development

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[1]Dept. of Pharmacol. and Therapeut., Univ. of Florida, Gainesville, FL; [2]Dept. of Biomed. Sci., [3]Dept. of Biol. Sci., Kent State Univ., Kent, OH

Abstract: Menopause is a knownrisk factor for Alzheimer's disease (AD) development in women. However, themechanisms underlying this increased risk remain unclear. Estrogen losshas been closely implicated in driving such risk. More recently, dataalso support the involvement of luteinizing hormone receptors in the regulationof cognition, plasticity, and AD-related cellular changes. However, themechanisms that underlie such changes are unknown. To address this question, wecarried out a transcriptomic analysis (RNAseq) in C57BL/6 and APP/PS1 miceunder reproductive intact or ovariectomized (OVX) status. A subset of theseanimals was administered an LH analog (hCG)intracerebroventricular (ICV), toaddress the mechanisms associated with central activation of the LH receptor.Animals had been characterized for cognitive function and pathology, both ofwhich demonstrated the benefits of LH receptor activation. Ourtranscriptomic analysis reveals important transcriptional changes that thehippocampus undergoes as a result of gonadal loss that can explain OVX-relatedcognitive deficits and acceleration of AD pathology. Interestingly thesechanges were genotype-dependent with different pathways enriched in C57BL/6 andAPP/PS1 mice. LH receptor activation in OVX APP/PS1 mice, which showed asignificant improvement in cognition and normalization of plaque pathology, thedifferentially expressed genes and the complete topology of the networkchanges, suggests that such effects maybe mediated through pathways associatedwith RNA homeostasis, cellular metabolism, and oxidative stress. Together ourdata further our understanding of mechanisms involved in cognitive loss andpathology regulation under gonadal loss and highlight a novel and directfunctional role of the central LH receptor in mediating AD-related cellularaspects.

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Poster

P206. Amyloid Beta: Changes in Synaptic Function

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Program #/Poster #: P206.01

Topic: B.02. Ligand-Gated Ion Channels

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Title: Nicotinic receptor modulation of early Alzheimer's disease in (mouse) hippocampus

Authors: *M. H. SABEC, U. MASKOS;
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Abstract: Current treatments for Alzheimer's disease (AD) commonly rely on elevating cholinergic function in the brain, however such interventions are symptomatic and do not prevent the disease's progression. β-amyloid, a peptide thought to underlie neural disruption reported in early AD development (Busche et al, 2008), can interact with the cholinergic system though nicotinic acetylcholine receptors (nAChRs). Previous studies from our group revealed that a genetic loss of β2 nAChR subunits could protect against memory impairment which is induced by β-amyloid over-expression (Lombardo et al, 2016). The presented work therefore investigates the influence of nAChR on β-amyloid induced synaptic and network dysfunction in the hippocampus, and how such modulation may affect the manifestation of cognitive deficits in early AD pathogenesis. We developed a model of localized β-amyloid over-expression by viral transduction of adult mouse dentate gyrus (DG). This local model is used to test the impact of genetic and pharmacological inactivation of nAChRs on memory function and neural activity through behavioural analysis, *in vivo* two-photon calcium imaging, and immunofluorescent techniques. An early-onset development of cognitive deficits and DG hyperactivity was observed in the localized AD model. Furthermore, whilst pharmacological β2 nAChR inhibition was able to protect against β-amyloid induced impairments, this was not associated with a normalization of the DG network activity. Thus, β2 nAChR inactivity is able to protect against β-amyloid induced cognitive dysfunction in a manner independent to changes in overall activity levels within the DG. Identification of the specific nicotinic-β-amyloid interactions underlying the reported protection will facilitate more targeted cholinergic therapies against early AD and may provide mechanistic insights essential for the development of preventative medicine.

Disclosures: M.H. Sabec: None. **U. Maskos:** None.

Poster

P206. Amyloid Beta: Changes in Synaptic Function

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Program #/Poster #: P206.02

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Abl1 is downstream effector of mAβ1-42 in its impact on spontaneous synaptic release

Authors: *M. KABIROVA[1], M. REICHENSTEIN[3], D. GOROBETS[1], A. PINHASOV[1], I. MICHAELEVSKI[2];
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Abstract: Amyloid plaques are one of the major hallmarks of Alzheimer's disease. While three structural species: monomers, oligomers and fibrils of amyloid beta have been identified (Kamenetz et al., 2003), oligomers and fibril were major target of the last decades investigations, providing less evidence about monomeric forms of amyloid beta (mAβ) and its function. In our research, we aimed to investigate the role of monomeric amyloid beta on spontaneous synaptic transmission. Our previous finding showed that 42 amino acids' long mAβ (mAβ42) markedly reduces spontaneous synaptic activity in cultured hippocampal neurons. Moreover, Abl kinases were shown to be downstream effectors of mAβ42 effect on spontaneous synaptic transmission (Reichenstein et al., 2021). Abelson tyrosine (Abl) kinase family consisting Abl1 (c-Abl) and Abl2 (Arg) isoforms, both are regulators of cell motility, morphology and adhesion (Colicelli, 2010). While the activity of c-Abl is crucial for proper neuronal development, it is relatively quiescent in healthy adult neurons. Activation of c-Abl in adult brain has been linked to human neurodegenerative diseases (Schlatterer et al., 2011). Here we report that Abl differentially regulates spontaneous synaptic release in excitatory and inhibitory neurons. Namely, DPH induced activation of Abl strongly impaired spontaneous transmission in excitatory neurons, while leading to enhancement of spontaneous inhibitory synaptic activity, presumably reducing E/I ratio. Interestingly, prolonged (24 h) incubation of hippocampal neurons in DPH led to reduction of both excitatory and inhibitory synapses. Surprisingly, Abl1 knock-down led to increasing of spontaneous release in both, excitatory and inhibitory neurons. Quantitative phosphoproteomic screening of Abl activity impact on synaptic machinery revealed 3813 phosphorylation sites, while further rigorous bioinformatic analysis of phosphomotifs revealed synaptotagmin 2 and complexin 2 to be the most possible candidates to mediate the effect of Abl signaling on neurotransmission. Based on our findings, we assume that Abl kinases signaling pathway could be activated by Aβ peptides, leading to protective effect from excessive excitatory activity via reduction of excitatory/inhibitory balance on the short-term time scale, while under pathological condition chronic mAβ exposure entails to synaptic disfunction/elimination.

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Poster

P206. Amyloid Beta: Changes in Synaptic Function

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Topic: C.02. Alzheimer's Disease and Other Dementias

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ISN

Title: An oxidative mechanism for cholinergic dysfunction in Alzheimer's disease

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Abstract: The neurotransmitter acetylcholine is synthesized by the enzyme choline acetyltransferase (ChAT), which is also a phenotypic marker of cholinergic neurons. Defective cholinergic transmission is involved in the onset and development of several CNS pathologies, including Alzheimer's disease (AD). Previous work by our group, using the avian retina as a CNS model, showed that ChAT activity in cultured neurons is markedly and specifically down-regulated by amyloid-β peptide oligomers (AβOs), diffusible toxins that accumulate in AD brains. This effect was shown to be linked to excitotoxicity, being likely caused by oxidative damage to the enzyme. ChAT inhibition by AβOs precedes any changes in cell viability or enzyme levels, and requires calcium influx and nitric oxide (NO) production to occur. Here, we expand these observations to a mammalian model, using cultured neurons of the rat septal region, and attempt to identify oxidative modifications involved in ChAT inhibition, in both animal models and in postmortem tissue from AD patients. Using S-nitrosothiol resin-assisted capture and thiol labeling, we show that cysteine modification is not central to the mechanism of inhibition. Tyrosine nitration, however, is induced in post-mortem hippocampal tissue from AD patients, and in cultures exposed to AβOs, correlating well with loss of ChAT activity. In immunoprecipitated samples, we show that nitrated ChAT is increased in the hippocampus of AD patients, as compared to controls. Results suggest a novel mechanism of cholinergic dysfunction that precedes neuronal death and may be relevant in early-stage AD pathology.

Disclosures: L.E. Santos: None. **S.T. Ferreira:** None. **F.G. de Mello:** None.

Poster

P206. Amyloid Beta: Changes in Synaptic Function

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Topic: C.02. Alzheimer's Disease and Other Dementias

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Title: The metabolic syndrome plus the injection of amyloid-β 25-35 peptide into the hippocampus modifies the expression of cholinergic markers, NGF and alters spatial memory.

Authors: *I. LIMON PEREZ DE LEON[1], O. REYES-CASTRO[2], F. LUNA-MORALES[3], A. PATRICIO[4];
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Abstract: The consumption of sweetened beverages has promoted the development of metabolic syndrome (MetS). MetS increases the risk of developing Alzheimer's disease (AD). In AD there is loss of cholinergic innervation in the septo-hippocampal pathway. Cholinergic neurons from the medial septum maintain acetylcholine levels with the choline acetyltransferase (ChAT) and acetylcholinesterase (AChE). On the other hand, nerve growth factor (NGF) maintains the cholinergic phenotype by being transported retrogradely to the soma of cholinergic neurons. The aim of this work was to evaluate the effect of MetS and intrahippocampal administration of the Aβ25-35 peptide on spatial memory, the expression of cholinergic enzymes, and NGF in rats. We used Wistar male rats (250-280 g) randomly assigned in the MetS and control group. MetS model was obtained by consuming a 20% sucrose solution while control group received drinking water, ad libitum for six months. All rats were yield to stereotaxic surgery to the injection of vehicle or Aβ25-35 [100µM] into CA1 subfield of Hippocampus, (AP: -4.0, L: ±2.3, P: -2.6). The groups were control+vehicle, MetS+vehicle, control+Aβ25-35 and MetS+Aβ25-35. On the 15th day post-surgery, experimental groups were tested for spatial learning in Morris Water Maze (MWM) and 5 days later, were memory tested. At the end of the spatial memory test in the MWM, the expression of ChAT, AChE and NGF in the hippocampus and medial septum was evaluated. The results indicate that all the experimental groups there were not modify spatial learning. In the memory test, the MetS + vehicle, control + Aβ25-35 and MetS + Aβ25-35 groups show damage in the spatial memory. Immunohistochemical analysis indicated that the MetS + vehicle, control + Aβ25-35 and MetS + Aβ25-35 groups significantly decrease ChAT expression in the medial septum, in the CA1, CA3 region and the dentate gyrus of the hippocampus. The expession of AChE increased in the medial septum of the MetS + Aβ25-35 group, as well as NGF in the regions of the septum-hippocampus pathway. Our findings suggest that the expression of ChAT and AChE in regions of the septohippocampal pathway are vulnerable to the presence of MetS, which is exacerbated by Aβ25-35 probably due to oxidative and pro-inflammatory mechanisms; however. These condiction damage the spatial memory. It is likely that MetS also stimulates defense mechanisms such as the release of NGF

Disclosures: I. Limon Perez De Leon: None. **O. Reyes-Castro:** None. **F. Luna-Morales:** None. **A. Patricio:** None.

Poster

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Title: Role of orexin signaling in beta-amyloid-induced cell death in SH-SY5Y cells

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Abstract: Alzheimer's disease (AD) is a degenerative disease which is accompanied by cognitive decline and progressive memory loss. It has been reported that orexin, a neuropeptide produced in hypothalamus, is responsible for regulation of sleep-awakening cycle and energy metabolism. However, roles of orexin in neurodegenerative diseases like AD has not been clearly examined as yet. In this study, we investigated role of orexin signaling in beta-amyloid (Aβ)-induced cell death in SH-SY5Y cells. Aβ induced cell deaths in a dose-dependent manner and increased the expression of orexin receptor. Pretreatment of orexin receptor 1 antagonist, SB334867 significantly inhibited Aβ-induced cell death but not orexin receptor 2 antagonist, TCS-OX2-29. SB334867 decreased apoptotic signaling induced by Aβ, including the ratio of Bax/Bcl-2 expression and cleavage of Caspase-3. In addition, Aβ increased phosphorylation of Akt/mTOR while pretreatment of SB334867 inhibited the phosphorylation of Akt/mTOR which had been induced by Aβ. Taken together, these findings suggest that orexin signaling may play a role in pathogenesis of AD and may be a potential therapeutic target for treatment of AD.

Disclosures: H. Bae: None. **B. Do:** None. **J. Jang:** None. **G. Park:** None.

Poster

P206. Amyloid Beta: Changes in Synaptic Function

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P206.06

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: SAGA-17-418878
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R03AG063162
2016-NIRG-397187_completed

Title: Protective effects of SSRI Citalopram against Ab-induced toxicities on dorsal raphe in vivo and in vitro AD models.

Authors: ***A. REDDY**[1], N. SAWANT[2], H. MORTON[2], L. E. BUNQUIN[2], H. P. REDDY[2,3,4,5,6];
[1]Texas Tech. Univ., LUBBOCK, TX; [2]Intrnl. Med., [3]Pharmacol. and Neurosciences, [4]Neurol.,

[5]Speech, Language and Hearing Sci. Departments, [6]Publ. Hlth. Department,, Texas Tech. Univ. Hlth. Sci. Ctr., Lubbock, TX

Abstract: Protective effects of SSRI Citalopram against Aβ-induced toxicities on dorsal raphe *in vivo* and *in vitro* AD models. Abstract: The purpose of our study is to understand the protective effects of selective serotonin reuptake inhibitor (SSRI), citalopram in Alzheimer's disease (AD). Depression is among the most common neuropsychiatric comorbidities in AD. Apart from its anti-depressive and anxiolytic effects, SSRI treatment also offers cellular modifications that improve synaptic transmission, neurogenesis and reduce AD pathologies, including amyloid burden, phosphorylated Tau and synaptic toxicity. Recent research also suggests besides canonical neurotransmission serotonin (5HT) increase intracellular energy metabolism and improve neuronal resilience. **Experiment design:** The current study was designed to explore the AD-related cellular changes and protective effects of SSRIs on serotonergic raphe neurons in the *in vitro* neurons and rodent raphe brain of AD. **Method:** Here we investigated the protective role of SSRI, Citalopram (Cit) *in vitro*, in rat serotonergic, medullary raphe neurons - RN46A-B14 and *in vivo*, in the dorsal raphe nucleus (DRN) region of transgenic mouse models of APP and Tau. We transfected RN46A-B14 neurons, with wild-type and mutant APP and Tau cDNAs for 24 hrs, then treated them with 20 uM Cit for 24 hrs. Harvested cells were analyzed for mRNA and protein levels of TPH2, BDNF, SERT and 5HTR1A, in addition to pTau and amyloid beta (Aβ). We also treated 10-month-old APP and Tau mice for a period of 8 weeks with Cit (20 μg/gm body weight). This was followed by cognitive behavior analyses and mRNA and protein levels assessments. **Results:** Cit treated mutant APP and Tau RN46A-B14 neurons showed reduced pTau levels while increased levels of TPH2, SERT, BDNF and 5HTR1A genes. Likewise, the Cit treated APP and Tau mice exhibited reduced cognitive decline and increased levels of serotonin related genes (TPH2, SERT and 5HTR1A) and reduced AD genes (APP and pTau) compared to the untreated ones. **Conclusion:** Taken together these findings suggest Cit could not only be a promising therapeutic drug for treating depression in AD but also mitigate AD pathologies and improve cognition, probably though increased serotonin production.

Disclosures: A. Reddy: None. **N. Sawant:** None. **H. Morton:** None. **L.E. Bunquin:** None. **H.P. Reddy:** None.

Poster

P206. Amyloid Beta: Changes in Synaptic Function

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Program #/Poster #: P206.07

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: FONDECYT 1161065

CARE-Chile-UC AFB170005
FONDEF D10E1077
CONICYT Doctoral Fellowship 21200584

Title: Abl1 promotes axon initial segment disassembly and tau missorting in response to amyloid-β fibrils and allosteric activation

Authors: *N. STUARDO, Á. CÁCERES, A. ÁLVAREZ;
Dept. of Cell. and Mol. Biol., Pontificia Univ. Católica De Chile, Santiago, Chile

Abstract: The axon initial segment (AIS) is an axonal subdomain that is critical for correct neuronal compartmentalization and action potential initiation. AIS disassembly is a known consequence of amyloid-β (Aβ) toxicity but the specific mechanisms that underlie this process in the context of Alzheimer's disease (AD) are not fully understood. The non-receptor tyrosine kinase Abl1 is a potential candidate in the mediation of AIS disassembly, as it becomes activated by Aβ aggregates, promoting several deleterious effects on normal neuronal cytoskeleton organization including tau hyperphosphorylation and loss of postsynaptic protein clusters. To evaluate a possible role for Abl1 in AIS disassembly we used neuronal cultures (14-16DIV) treated with Aβ fibrils, Abl kinase inhibitors and DPH, an allosteric activator of Abl kinases. Immunofluorescence and western blot analyses were performed to evaluate changes in AIS cytoskeletal organization and Abl1 signaling activation. We find that short Aβ fibril treatments (15 min to 1 hr) and DPH induce AIS disassembly—evaluated by loss of localized Ankyrin G staining—, and this was prevented by inhibition of Abl kinases. Similarly, Abl1-KO neurons do not suffer AIS disassembly in response to Aβ fibril insults, further reinforcing the notion that Abl1 could play a relevant role in the collapse of this structure. We find that Aβ fibril-mediated AIS disassembly is reversible, suggesting a mechanism that involves short-lived alterations of cytoskeleton arrangement or reversible post-translational modifications. We also evaluated changes in the subcellular distribution of tau, which have been associated to AIS alterations in AD. We find that Aβ fibrils and DPH increase PHF-1 antibody signal in the cell body, a sorting defect that has previously been associated to AIS disruption. Our results suggest that Abl1 could be a key player in the process of AIS disassembly and tau missorting in the context of AD. AIS collapse could be a key event in the amyloid cascade, as this structure mediates critical processes in normal neuronal function which have been found to be altered in AD.

Disclosures: N. Stuardo: None. **Á. Cáceres:** None. **A. Álvarez:** None.

Poster

P206. Amyloid Beta: Changes in Synaptic Function

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P206.08

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH R01NS105615

Title: Role of Fragile X Mental Retardation Protein in Amyloid Beta Induced Translational Suppression

Authors: *S. LIZARAZO, Y. YOOK, N.-P. TSAI;
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Abstract: Alzheimer's Disease (AD) is the most common cause of dementia where the accumulation of the peptide Amyloid-Beta (Aß) is the main theory linked to the pathogenesis of the disease. Fragile X Mental Retardation Protein (FMRP) is an RNA-binding protein that functions to suppress global translation. Because Aß is known to be a toxic peptide capable of inducing the Endoplasmic Reticulum (ER) stress response and because augmented translational suppression is one of the main molecular phenotypes in ER stress response, we ask whether and how FMRP is involved in Aß induced translational suppression. In this study, we observed that treatments of synthetic Aß peptides in culture primary mouse cortical neurons induce the expression of FMRP and FMRP-dependent ER stress response and subsequent translational suppression. We further characterized that two translational regulators, eukaryotic translation initiation factor 2α (eIF2α) and eukaryotic translation elongation factor 2 (eEF2), are involved in Aß induced translational suppression and are dysregulated in the absence of FMRP. Mechanistically we found that FMRP is responsible for regulating the levels of Protein Phosphatase 2 (PP2A) and Protein Phosphatase 1 (PP1) that differentially modulate the phosphorylation status and thereby the activity of eIF2α and eEF2 following treatment of Aß. Using a pharmacological approach to inhibit PP2A and PP1 with Okadaic Acid, we were able to rescue the impairment of eIF2α and eEF2 as well as the translational suppression in the absence of FMRP. These results suggest a novel function of FMRP in regulating translational suppression following Aß treatment by fine-tuning two distinct signaling pathways involved in global translation.

Disclosures: S. Lizarazo: None. **Y. Yook:** None. **N. Tsai:** None.

Poster

P206. Amyloid Beta: Changes in Synaptic Function

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P206.09

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: ZEN-02-643453

Title: Parp-1 activation is a key mediator of pathogenesis in familial alzheimer's disease model

Authors: *A. JHALDIYAL[1], T.-I. KAM[2], T. M. DAWSON[3], V. L. DAWSON[4]; [1]JOHNS HOPKINS UNIVERSITY, Baltimore, MD; [2]Inst. for Cell Engineering/ Dept. of Neurol., Johns Hopkins Univ. Sch. of Med., Ellicott City, MD; [3]Inst. for Cell Engin., Johns Hopkins Univ. Inst. for Cell Engin., Baltimore, MD; [4]Inst. Cell Engin., Johns Hopkins Univ. Sch. Med., Baltimore, MD

Abstract: Alzheimer's disease (AD) is the most common form of dementia. It affects about 5.7 million people within the United States. This disease is marked by occurrence of two major pathological proteins- Amyloid-beta plaques (A plaques) and Tau mediated Neurofibril tangles (NFT), wherein emergence of A plaques precede decades before NFT pathology. The clinical outcome of these protein aggregations is the progressive loss of memory further leading up to the inability to perform daily tasks. A major roadblock in the field is the lack of understanding of how amyloid pathology is connected to tau pathology. We report that Poly [ADP-ribose] polymerase 1 (PARP-1) activation is the missing link between these two pathologies. PARP-1, a DNA damage sensing enzyme, plays a major role in stress-mediated signaling. Our data in primary cortical neuron cultures show that oligomeric A (oA_{1-42}) treatment results in robust Poly [ADP-ribose]/PAR activation, concomitant with increased tau phosphorylation. We show that this PAR activation is upstream of tau phosphorylation and PARP-1 inhibitor decreases tau phosphorylation in these oA_{1-42} treated neurons. Additionally, we performed genetic deletion of PARP-1 in the 5xTg Familial AD mice model and found increased neuron survival and decreased gliosis. Taken together, our data show that PARP-1 plays a pivotal role in AD pathogenesis and is a very potent target for therapeutic intervention in this disease.

Disclosures: A. Jhaldiyal: None. **T. Kam:** None. **T.M. Dawson:** None. **V.L. Dawson:** None.

Poster

P206. Amyloid Beta: Changes in Synaptic Function

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P206.10

Topic: C.02. Alzheimer's Disease and Other Dementias

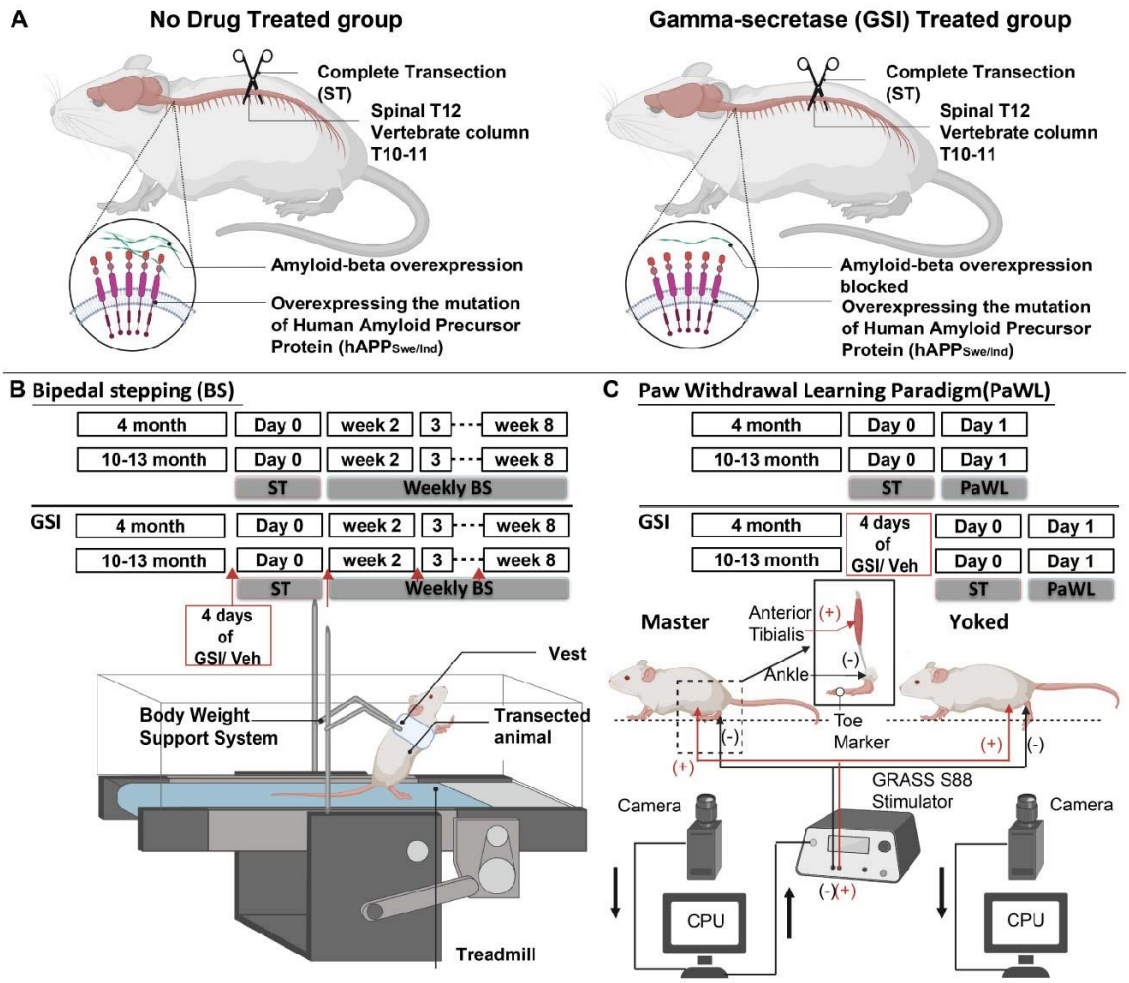
Support: Louis and Harold Price Foundation
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J. Yang Family Foundation

Title: Amyloid beta deposition in the lumbosacral spinal circuit impairs both the short-term and long-term neuroplasticity of locomotion

Authors: *R. HUANG[1], M. S. JOSEPH[2], H. ZHONG[1], R. ROY[1], J. CHANG[1], M. IGLESIAS[1], P. A. TRUONG[1], S. SHAHRESTANI[3], M. MADHAVAN[1], Y. ZHOU[1], K. LI[1], J. LEITER[4], E. KOO[5], R. EDGERTON[1], D. C. LU[6];

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Abstract: Sensorimotor perturbations are often reported in patients with Alzheimer's disease (AD) and attributed to cortical and not spinal dysfunction. This is due to the fact that one canonical biomarker of AD, amyloid-β (Aβ) plaques, are rarely observed in the spinal cord of AD patients. However, more evidence supporting spinal cord involvement in AD is emerging. Atrophy of the spinal cord, neurofibrillary pathology, and increased amyloid-beta precursor protein (APP) expression levels in the spinal cord have been identified in the spinal cord inAD patients and AD animal models. Furthermore, APP and soluble Aβ can impair neuronal functions even without forming the plaques. In this study, we investigated the contribution of spinal APP and its proteolytic products to locomotor functions by studying the lumbar locomotor circuit of J20 mice. To rule out the impact of the supraspinal structures, we transected the spinal cord completely at thoracic level 10 to cut off all connections between the lumbosacral spinal cord and all the rostral parts of the central nervous system (CNS). The short-term plasticity of the lumbosacral locomotor circuit was evaluated by an instrumental learning model, paw withdrawal paradigm (PaWL) and the long-term plasticity was investigated by the weekly bipedal treadmill stepping paradigm. Both 4mon and 13mon J20 mice showed significant decreases in the PaWL compared to their WT littermates ($p = 0.0075$). The spontaneous bipedal locomotion recovery was also inhibited in spinal J20 mice when compared to WT littermates ($p = 0.0031$). Notably, these impairments of lumbar circuit plasticity was reversed by the administration of a gamma-secretase inhibitor (Benzodiazepine-type LY-411575) in the spinal J20 mice, suggesting the generation of amyloid-beta in local spinal cord circuit contribute to the locomotor deficits.



Disclosures: R. Huang: None. **M.S. Joseph:** None. **H. Zhong:** None. **R. Roy:** None. **J. Chang:** None. **M. Iglesias:** None. **P.A. Truong:** None. **S. Shahrestani:** None. **M. Madhavan:** None. **Y. Zhou:** None. **K. Li:** None. **J. Leiter:** None. **E. Koo:** None. **R. Edgerton:** None. **D.C. Lu:** None.

Poster

P217. Influences of Amyloid / APP on Excitability

Location: Virtual Only

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Program #/Poster #: P217.01

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R03NS111493
NIH Grant F31AG069496

Title: Entorhinal-hippocampal desynchronization in a mouse model of Alzheimer's disease pathology

Authors: *L. M. VETERE, L. CHEN, Z. CHRISTENSON WICK, D. J. CAI, T. SHUMAN; Neurosci., Icahn Sch. of Med. at Mount Sinai, New York, NY

Abstract: Background: Alzheimer's disease (AD) is a devastating neurodegenerative disease characterized by memory loss and progressive age-related cognitive decline. While leading hypotheses have posited that accumulation of amyloid-beta (Aβ) and tau proteins drives neurodegeneration, reducing these proteins without preventing or reversing functional changes may be insufficient to alleviate memory impairments. Therefore, it is critical to understand the functional changes that occur in brain circuits required for memory. Impairments in hippocampal function are well established in AD, but it is unclear whether this is driven by local changes or abnormal inputs, such as those from medial entorhinal cortex (MEC).
Methods: To understand how AD pathology impacts communication within and between brain regions that are critical for memory, we used 3xTg mice, which express mutations in APP, presenilin, and tau. We performed acute *in vivo* silicon probe recordings to simultaneously record from 512 channels across hippocampus and MEC layers II and III. Head-fixed mice were trained to run on a virtual reality track to receive water rewards and recorded during active navigation. By recording two age groups, we were able to compare neural activity before and after the onset of memory impairments and examine how and when synchrony of individual cells and local field potentials breaks down.
Results: Our data suggest that synchrony between MEC and hippocampus is impaired in 3xTg mice compared to WT controls prior to the emergence of spatial memory impairments. In MEC, we found reduced theta power and altered phase locking of single units to ongoing theta oscillations. We also found reduced theta coherence between MEC and hippocampus. Interestingly, these effects were observed at 6 months of age, before the onset of spatial memory impairments. In the hippocampus, we found that theta power was largely intact at 6 months of age but significantly decreased at 8 months, when memory deficits are observed. These findings support the hypothesis that entorhinal dysfunction precedes and may contribute to later hippocampal changes. Possible factors driving early entorhinal dysfunction include hyperexcitability of MEC stellate cells or loss of MEC parvalbumin interneuron function.

Disclosures: L.M. Vetere: None. **L. Chen:** None. **Z. Christenson Wick:** None. **D.J. Cai:** None. **T. Shuman:** None.

Poster

P217. Influences of Amyloid / APP on Excitability

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P217.02

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH R01 MH113924

NIH T32 GM007356
NSF CAREER

Title: Differential impact of amyloid β pathology on population activity along dorsal-ventral axis of CA1 in APP/PS1 mice

Authors: *U. CHOCKANATHAN, K. PADMANABHAN;
Univ. of Rochester, Rochester, NY

Abstract: Alzheimer disease (AD) causes a range of cognitive and behavioral symptoms that often lead to loss of independence, decreased quality of life, and death. A major histopathological hallmark of AD is the amyloid beta (Aβ) plaque. The hippocampus (HPC), an important region for learning and memory, is particularly susceptible to amyloid pathology. Recent studies have documented significant anatomical and functional variations along the longitudinal hippocampal axis, with dorsal HPC implicated in episodic memory and spatial cognition and ventral HPC involved in affective processing and social cognition. As most animal studies of Aβ pathology have been performed in dorsal HPC, comparatively little is known about how Aβ affects ventral HPC, specifically how the molecular and cellular changes associated with Aβ affect the activity of neuronal networks and, in turn, behavior. To address this question, we made high density recordings of neuronal populations in the APP/PS1 mouse model of Aβ pathology. Using high-density silicon electrode arrays, we recorded large populations of neurons in dorsal CA1 (dCA1) and ventral CA1 (vCA1) in awake head-fixed mice as they ran in a virtual reality environment.
First, we found no differences in single-neuron activity, such as spatial information or mean firing rate, between the two subregions across APP/PS1 and control mice. However, we found that features of collective activity, such as pairwise correlations, were higher in dCA1 than vCA1 in control mice, but that this difference was abolished in APP/PS1 mice. These functional interactions were critical for the population-level encoding of position, as the collective spatial information of the population was higher in dCA1 than vCA1 in controls, but this difference was abolished in APP/PS1 mice. Our results point to two critical features of Aβ-associated disruptions in hippocampal activity: 1) Aβ may differentially affect the responses of single neurons as compared to network activity and 2) these effects vary along the longitudinal axis of the HPC. This suggests that the effects of Aβ are highly variable within a single brain region, which may, in turn, provide insight into the diversity of behavioral symptoms observed in AD.

Disclosures: U. Chockanathan: None. **K. Padmanabhan:** None.

Poster

P217. Influences of Amyloid / APP on Excitability

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P217.03

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: App modulates excitability through the axon initial segment and node of Ranvier with implications for Alzheimer's disease

Authors: *F. MA, H. AKOLKAR, R. BENOSMAN, K. HERRUP;
Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Alzheimer's disease (AD) is the most common cause of dementia of the elderly. Although rare, familial AD is most often caused by one of only three known disease genes one of which is *APP*(amyloid precursor protein). APP is the source of the Aβ peptide fragment, whose aggregation produces the characteristic amyloid plaques found in AD brains. Indeed, the amyloid cascade hypothesis - the most widely cited disease model - assumes Aβ is the driver of AD. Yet dozens of clinical trials based on Aβ removal have been unsuccessful, leading many to question the central role of the amyloid cascade. Curiously, the genetics of familial AD identify *APP*, not Aβ, yet over the years the normal function of the APP holoprotein itself has received only scant attention. In striving to fill this gap in our understanding we have found that APP functions in the brain as a damage response protein. Gene expression doubles within minutes of an excitotoxic stimulus and the excess APP protein dampens neuronal activity through its ability to regulate the length and position of the axon initial segment (AIS) and the node of Ranvier (NOR). APP protein is not only present in neurons. In both mouse and human there is a striking presence of APP in the myelin sheath. Primary cultures of oligodendrocytes confirm the colocalization of APP and MBP. We verified the APP-dependent changes in the AIS had electrophysiological consequences. We used high density multielectrode-arrays to demonstrate that APP-overexpression reduced the firing rate of neurons in the in vitro networks. Our results support the idea that the cognitive decline in Alzheimer's disease, while perhaps modulated by Aβ is caused primarily by the alterations in neuron firing potential and action potential propagation brought about by dysregulation of the APP holoprotein acting through the AIS and the NOR. Our study proposes an alternative explanation for Alzheimer's disease that might be called 'APP Cascade Hypothesis'.

Disclosures: F. Ma: None. **H. Akolkar:** None. **R. Benosman:** None. **K. Herrup:** None.

Poster

P217. Influences of Amyloid / APP on Excitability

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Program #/Poster #: P217.04

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH grant NS085171 (JC)
NIH grants AG065290 (JC)

Title: An age-related shift in the distribution of epileptiform spikes across sleep-wake states in a transgenic mouse model of Alzheimer's disease

Authors: *M. SILVA-PÉREZ, J. PARK, J. CHIN;
Baylor Col. of Med., Houston, TX

Abstract: In Alzheimer's disease (AD) patients and mice, there is an increased incidence of seizures and epileptiform activity, which may be underestimated due to the prevalence of nonconvulsive seizures, and the observation that seizures and epileptiform spikes primarily occur during sleep. In AD patients, epileptiform spikes are non-uniformly distributed across sleep-wake states and occur predominantly during nonrapid eye movement (NREM) sleep. The mechanisms that give rise to epileptiform spikes, and their preferential occurrence during NREM sleep, are not clear. Several mouse lines that express human amyloid precursor protein (APP) containing AD-linked mutations also exhibit epileptiform spikes primarily during sleep. In APP mice from Line Tg2576, epileptiform spikes occur only during rapid eye movement (REM) sleep at early stages of disease, but invade NREM sleep as disease and age progress. APP mice from Line J20 have been found to also primarily exhibit epileptiform spikes during REM sleep at early stages of disease. However, whether the occurrence of epileptiform spikes shifts to other brain states as disease and age progress in J20 mice is unclear. Determining if the occurrence of epileptiform spikes in specific sleep states is similar between patients and mice is necessary for assessing the extent to which insights gained from mouse models can be translated to the human disease. To investigate whether the distribution of epileptiform spikes across sleep-wake states changes with age in J20 APP mice, we performed video-EEG monitoring of mice at early and later stages of disease. Wakefulness, NREM sleep, and REM sleep were scored in Spike2 according to the EEG activity in the delta (0.5-4 Hz), theta (6-10 Hz), and high-frequency (100-300 Hz) ranges. Epileptiform spikes were identified by Spike2 using duration and amplitude criteria, manually verified, and spike number and rate were calculated. In young mice at early stages of disease, REM sleep exhibited a higher rate of epileptiform spikes than NREM sleep. Despite the proportionally low abundance of REM sleep, half of total daily epileptiform spikes occurred during REM sleep. At later stages of disease, the proportion of epileptiform spikes during NREM sleep increased, demonstrating a shift in prevalence over time. This shift from REM to NREM sleep indicates that the epileptiform activity exhibited by aged APP mice more closely reflects that observed in AD patients. These results also suggest that identifying the mechanisms in APP mice that give rise to epileptiform spikes, and underlie the shift from REM to NREM sleep, may provide novel insights into the basis of epileptiform activity in AD patients.

Disclosures: M. Silva-Pérez: None. **J. Park:** None. **J. Chin:** None.

Poster

P217. Influences of Amyloid / APP on Excitability

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Program #/Poster #: P217.05

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH R01 NS086965
NIH R01 NS085171

Title: Seizure-induced changes in sFRP3 expression contribute to dysregulation of adult hippocampal neurogenesis in a transgenic mouse model of Alzheimer's disease

Authors: *C.-H. FU[1], J. PARK[1], U. TOSI[2], X. ZHANG[3], Y. ZHENG[1], J. CHIN[1];
[1]Dept. of Neurosci., Baylor Col. of Med., Houston, TX; [2]Weill Cornell Med., New York, NY;
[3]Dept. of Neurol., Children's Hosp. of Philadelphia, Philadelphia, PA

Abstract: Alzheimer's disease (AD) is associated with hippocampal dysfunction, and one contributing factor may be the dysregulation of adult hippocampal neurogenesis. Neurogenesis in the adult dentate gyrus (DG) is important for cognition and mood, both of which are affected in AD. We previously showed, using a transgenic mouse model overexpressing mutant human amyloid precursor protein (APP), that seizure activity that begins early in disease progression aberrantly increases the proliferation of a finite pool of neural stem cells (NSCs) in the DG. Such increased proliferation was associated with accelerated depletion of the NSC pool and impaired spatial discrimination, a behavior that relies on adult-born neurons. These alterations were prevented by treatment with an anti-seizure drug. However, the specific molecular mechanisms that drive such seizure-induced activation and depletion of NSCs is unclear. Using RNA-sequencing, we queried which components of the Wnt signaling pathway, a pathway critically involved in inducing neurogenesis in both development and adulthood, were differentially expressed in the DG of APP mice and their nontransgenic (NTG) littermates. Surprisingly, few Wnt components were differentially expressed, of which secreted frizzled-related protein 3 (sFRP3) stood out. sFRP3 is an inhibitor of the Wnt signaling pathway, and thus acts as a "brake" on neurogenesis. It is normally constitutively expressed and secreted by mature DGCs. Notably, sFRP3 expression was decreased in APP mice, but was restored after treatment with an anti-seizure drug, suggesting that seizures drive suppression of sFRP3 in APP mice. In addition, we found that sFRP3 may be epigenetically regulated by seizure-induced transcription factors. We hypothesized that seizure activity in APP mice recruits such transcription factors to suppress sFRP3 expression, which then accelerates proliferation and depletion of the NSCs and thus impairs spatial discrimination. To test this hypothesis, we designed an adeno-associated virus (AAV) to express sFRP3 in DGCs of APP and NTG mice. We found that viral-mediated expression of sFRP3 does indeed decrease neurogenesis, and that long-term expression of sFRP3 improved spatial discrimination in APP mice. Our results suggest that sFRP3 may play a key role in the seizure-driven dysregulation of NSC proliferation and neurogenesis, and consequent cognitive impairment, in APP mice and perhaps also in AD.

Disclosures: C. Fu: None. **J. Park:** None. **U. Tosi:** None. **X. Zhang:** None. **Y. Zheng:** None. **J. Chin:** None.

Poster

P217. Influences of Amyloid / APP on Excitability

Location: Virtual Only

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Program #/Poster #: P217.06

Topic:

Support: NS116357
R03 NS111493
R21 DA049568
R01 MH120162
DP2 MH122399
Klingenstein-Simons Fellowship Award in Neuroscience
NARSAD Young Investigator Award

Title: Selective and progressive changes in excitability of MECII stellate cells in the 3xTg mouse model of Alzheimer's disease pathology

Authors: *L. CHEN, Z. CHRISTENSON WICK, L. VETERE, D. J. CAI, T. SHUMAN; Icahn Sch. of Med. at Mount Sinai, New York, NY

Abstract: Alzheimer's disease (AD) is a disorder characterized by progressive cognitive impairments. In mouse models of AD pathology many studies have found deficits in spatial memory and hippocampal spatial coding. Yet it remains unclear whether these deficits are due to local circuit changes or reflect changes in upstream inputs. The medial entorhinal cortex (MEC) is the primary input into the hippocampus and an early site of AD pathology. Specifically, MEC has two distinct layers, layer II (MECII) and III (MECIII), that project to hippocampal CA1 through distinct pathways. In order to understand how spatial coding deficits emerge in AD, we examined the cellular and circuit alterations in distinct MEC cell types before and after the onset of spatial memory impairments. In the 3xTg mouse model of AD pathology, we found that hippocampus-dependent spatial memory impairments on a novel object location task appeared between 6 and 8 months of age. We then used *in vitro* whole-cell electrophysiology to investigate whether distinct MEC cell types (MECII stellate cells, MECII pyramidal cells, and MECIII excitatory neurons) were altered before and after the onset of memory impairments. We found that MECII stellate cells in 3xTg mice had altered excitability as early as 3 months, demonstrated by a depolarized resting membrane potential (RMP). However, when we injected current steps the number of evoked action potentials (eAPs) remained comparable to controls, indicating a cellular compensation to reduce spiking. This compensation was lost by 10 months of age, as MECII stellate cells in 3xTg mice exhibited increased number of eAPs in addition to a depolarized RMP. Further, by quantifying the amplitude and frequency of spontaneous excitatory and inhibitory post synaptic currents (sEPSC and sIPSC, respectively), we have found a reduction in the sIPSC frequency and a significantly increased E/I ratio in MECII stellate cells in 10-month-old 3xTg mice. Importantly, all the cellular differences were observed only in

MECII stellate cells, while MECII pyramidal cells and MECIII cells remained normal in 3xTg mice at 10 months of age. Together, our data suggests that cellular excitability deficits in 3xTg mice emerge as early as 3 months of age and are restricted to stellate cells in MEC. These changes begin well before the onset of memory impairments and progress into severe cellular and synaptic deficits by 10 months of age in the 3xTg mouse model. We propose that early disruptions in MECII stellate cell excitability leads to local and downstream circuit changes and spatial memory impairments with the progression of AD pathology.

Disclosures: L. Chen: None. **Z. Christenson Wick:** None. **L. Vetere:** None. **D.J. Cai:** None. **T. Shuman:** None.

Poster

P217. Influences of Amyloid / APP on Excitability

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P217.07

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH NS085171
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Title: Chronic ΔFosB activity regulates key target genes in dentate granule cells and modulates neuronal excitability and recurrent seizures in APP mice

Authors: *G. STEPHENS[1], Y. ZHENG[1], J. PARK[1], A. L. EAGLE[2], J. YOU[1], C.-H. FU[1], S. CHOI[1], Y. LIU[3], A. ROBISON[2], J. CHIN[1];
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Abstract: Recurrent seizures are associated with cognitive deficits in many neurological disorders, including epilepsy and Alzheimer’s disease (AD). We reported that seizures induce expression of ΔFosB, a transcription factor with an unusually long half-life that epigenetically alters gene expression and persistently impairs cognition in mouse models of epilepsy or AD. Recently, we showed that ΔFosB binds to genes involved in key pathways that may regulate excitability in dentate granule (DG) cells in mice with recurrent seizures, suggesting that ΔFosB may play a neuroprotective role in conditions associated with excitotoxicity. Understanding which ΔFosB target gene pathways are most influential in moderating DG cell excitability and seizure activity may guide novel therapeutic strategies to enhance endogenous mechanisms of neuroprotection in AD and other conditions with recurrent seizure activity. To determine the impact and time-course of ΔFosB activity on DG excitability and seizure activity, we used viral strategies to overexpress or block ΔFosB activity in the dentate gyrus of amyloid precursor

protein (APP) transgenic mice, a model of AD that exhibits spontaneous recurrent seizures. Whereas short-term (3-4 week) blockade of ΔFosB activity did not affect seizure activity, long-term (3 months) inhibition of ΔFosB activity in DG cells exacerbated seizure activity and spatial memory deficits in APP mice. Conversely, overexpression of ΔFosB in DG cells reduced neuronal excitability measured in electrophysiological recordings in hippocampal slices from wild-type mice overexpressing ΔFosB in the dentate gyrus. Moreover, overexpression of ΔFosB in primary hippocampal neurons was neuroprotective in an NMDA toxicity assay. We performed whole-hippocampal ChIP-sequencing to identify ΔFosB target genes that may be involved in moderating DG cell excitability in APP mice, and used GOTerm clustering analyses to find that many ΔFosB target genes in APP mice are involved in excitability and neurotransmission, including numerous epilepsy-related genes. Our results indicate that the epigenetic regulation of gene expression resulting from chronic ΔFosB activity may help constrain neuronal excitability and reduce seizure occurrence.

Disclosures: G. Stephens: None. **Y. Zheng:** None. **J. Park:** None. **A.L. Eagle:** None. **J. You:** None. **C. Fu:** None. **S. Choi:** None. **Y. Liu:** None. **A. Robison:** None. **J. Chin:** None.

Poster

P217. Influences of Amyloid / APP on Excitability

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Program #/Poster #: P217.08

Topic: C.02. Alzheimer's Disease and Other Dementias

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Title: The mode of cooperative suppression on large conductance calcium-activated potassium channel by amyloid β oligomer and amyloid precursor protein

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Abstract: We have previously reported that intracellular amyloid β (Aβ) injection suppresses the large-conductance calcium-dependent potassium (BK) channel in cortical pyramidal cells from wild type (WT) mice (Yamamoto et al. 2011). In triple transgenic AD model (3xTg) mice, intraneuronal Aβ is genetically destined to accumulate, which suppresses BK channel. However, multiple lines of evidence have indicated that the amyloid precursor protein (APP) is also

causative in the toxicity attributed so far solely to Aβ in mouse AD models that overproduce APP. The possibility that not just the end product Aβ but APP per se might be harmful, still remains to be fully examined. This study investigated the mode of BK channel blockade mediated by not just amyloid β but also APP. Whole cell recordings were performed from the pyramidal neurons in frontal neocortical slices of WT or 3xTg mice. Spike properties were recorded under the intracellular injection of recombinant full length APP or various antibodies against APP or Aβ through patch pipette. An anti-Aβ-oligomer antibody (11A1), but not anti-monomer-Aβ-antibodies, was effective in restoring BK channel activity in 3xTg neurons. Other anti-Aβ-oligomer antibodies, A11 and 82E1, failed to recover BK channel activity. Antibodies against APP were also found effective. In WT neurons, APP as well as Aβ suppressed BK channels by itself. In cortical neurons from 3xTg mice that bear Homer1a knockout (4xTg mice), neither anti-APP antibodies nor 11A1, but only the 6E10 antibody that binds both APP and Aβ, rescued the BK channel activity. These results revealed a different synergy of Aβ and APP in WT and 3xTg neurons. Given that Homer1a was shown to disrupt Aβ-BK channel coupling both in WT and 3xTg neurons and Homer1a expression is activity dependent and 3xTg neurons are hyperexcitable, Homer1a is likely to be expressed sufficiently in 3xTg neurons, thereby mitigating the synergistic suppressive influence of APP and Aβ on BK channel. BK channel suppression still occur even both in intracellularly APP-injected WT neurons in the presence of 11A1, and in 3xTg neurons in the presence of Bri2, which disrupt the link between Aβ and APP, and the processing of APP to Aβ. In Aβ-injected WT neurons, BK channel suppression failed to be reversed by antibodies against APP. Considering that the APP C-terminus domain is known to bind to the Homer EVH domain, these observations suggest that long Homer and BK channel, together with APP and Aβ oligomer, may form a molecular complex that underlies synergistic BK channel suppression by APP and Aβ oligomer.

Disclosures: K. Yamamoto: None. **R. Yamamoto:** None. **N. Kato:** None.

Poster

P217. Influences of Amyloid / APP on Excitability

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Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P217.09

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Rapid calcium-dependent suppression of macroscopic homomeric Kv1.2 currents by amyloid beta (1-42) peptide: Implications for Alzheimer's disease

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Abstract: Amyloid beta peptides (Aβ) have long been implicated in the pathogenesis of Alzheimer's disease (AD). Past studies have shown that Aβ is involved in the disruption of intracellular calcium $[Ca^{2+}]_i$ homeostasis, synaptic communication, learning and memory mechanisms (e.g., hippocampal long term potentiation (LTP)), as well as neuronal death. The critical molecular targets and synapto- and neuro-toxic mechanisms for Aβ remain unclear however. Because voltage-dependent potassium channels, and several Kv1 family members specifically, are activated during an action potential, regulate Ca^{2+} influx in a depolarized cell, and can be neurotoxic when suppressed, they are important candidates in studies of Aβ targets and AD pathogenesis. Previous studies from our lab of Aβ effects on murine homomeric Kv1.1 channels (-expressed heterologously in *Xenopus* oocytes) found that bath-addition of 1 μM Aβ(1-42) produced ~50% suppression of macroscopic Kv1.1 currents within 30 m (measured at +20 mV). Little or no suppression was produced by equimolar reverse peptide Aβ(40-1) or solvent controls. Previous studies also found that the Aβ(1-42) suppression involved $[Ca^{2+}]_i$-dependent signaling pathways: chelation of $[Ca^{2+}]_i$ by 1,2-Bis(2-aminophenoxy)ethane-N,N,N',N'-tetraacetic acid tetrakis (acetoxymethyl ester) (BAPTA-AM), significantly attenuated the 30 min suppression value from ~50% suppression to 20%. Here we extend our observation of Aβ-suppression of Kv1-family channel currents to include (homomeric, apha-subunit) Kv1.2 channels, expressed in oocytes. Bath application of appropriately "aged" 1 μM Aβ(1-42) resulted in ~50% suppression of macroscopic Kv1.2 currents(± 5.4; n=7), after 30m., DMSO controls (± 3.46; n=10) showed little (~6% suppression). Pre-exposure of oocytes (± 6.16; n=6) to 10 μM BAPTA-AM reduced the Aβ(1-42) suppression level to ~17% at 30 m, significantly less than the 50% observed in the absence of BAPTA-AM (p =0.045).Kv1.1 and Kv1.2 play diverse functional roles in several neuronal compartments, especially in the regulation of pre- and post-synaptic membrane excitability. Kv1.2 subunits can also co-localize with over Kv1 subunits to form heteromeric channels. For example, Kv1.2 co-localizes with Kv1.1 in the nodes of Ranvier in myelinated axons, and in the brain, in particular, in axons and nerve terminals. We propose that Kv1 channels represent promising and novel targets for attempts to treat Aβ-driven pathology in AD.

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Poster

P217. Influences of Amyloid / APP on Excitability

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Program #/Poster #: P217.10

Topic: C.02. Alzheimer's Disease and Other Dementias

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Title: Identification of candidate key regulators of neural stem cell dynamics and neurogenesis in a transgenic mouse model of Alzheimer's disease

Authors: *Y. FURUTA, C.-H. FU, J. CHIN;
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Abstract: Neural stem cells (NSCs) ensure lifelong neurogenesis, and reduced neurogenesis is associated with cognitive decline in Alzheimer's disease (AD). Therefore, elucidation of molecular mechanisms that regulate NSCs, and therapeutic intervention to preserve the NSC pool, may be a novel strategy for AD. Using transgenic AD model mice (APP mice), our lab previously reported that early seizure activities that occur in AD affect gene regulations of mature granule cells and drive aberrant NSC division and neurogenesis, accelerating the depletion of a finite pool of NSCs in the dentate gyrus. However, the molecular mechanisms by which the seizures that occur in AD disrupt NSCs and neurogenesis dynamics are not clear. To gain some perspective about molecules that could affect NSC and neurogenesis dynamics in AD, we examined a dentate gyrus RNA-seq dataset from our lab which provided a list of differential expressed genes between APP mice and nontransgenic littermate controls. We conducted a literature research about the functions of highly up/downregulated genes from the list up to about 200 genes. Through this work, we found 13 candidate genes which may regulate NSCs and neurogenesis, and benchtop confirmed the expression level of the genes in the RNA-seq dataset. One interesting molecule among the candidates we identified is Follistatin, a secreted protein that antagonizes various TGF-β superfamily proteins. Previous reports found that Follistatin inhibits NSC division and neurogenesis after acute excitotoxic neurodegeneration in the hippocampus. In our study, we found that the expression of Follistatin is significantly altered in APP mice. Furthermore, our data suggest that Follistatin may be under the regulation of seizure activities in APP mice. Together, these data indicate that the regulation of Follistatin expression may be one of the mechanisms by which seizure activities drive aberrant NSC and neurogenesis dynamics in APP mice.

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Poster

P254. Dopamine and Non-Dopamine Pathways II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P254.01

Topic: C.03. Parkinson's Disease

Title: Measuring endogenous dopamine during approach-avoidance task

Authors: *H.-Y. LU[1], H. STEALEY[1], Y. ZHAO[1], E. CONTRERAS-HERNANDEZ[1], K. T. KISHIDA[2], S. R. SANTACRUZ[1];

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Abstract: Dopamine is a neurotransmitter that plays critical roles in neuromodulation, such as motivation, reward, motor control, and value-based decision making. However, the nature and the exact function of dopamine is not clearly understood even though it has been studied extensively. This can in part be attributed to the challenge of measuring dopamine in vivo during behavior. In order to overcome this challenge, we customized a probe made of carbon-fiber electrode for dopamine measurement using fast-scan cyclic voltammetry, which includes an outer guide electrode that serves as a reference electrode. We collected the non-background-subtracted cyclic voltammetry responses based on known concentrations of dopamine solutions across multiple probes. An elastic-net penalized regression model was trained to predict the dopamine concentrations in held-out samples and was used for *in vivo* experiments after validation. From our preliminary results, we built a regression model that can accurately predict the dopamine concentrations of held-out data. The in vivo measurements were collected from a non-human primate (rhesus macaque) during an approach-avoidance task. In this task, the subject chose between an appetitive (juice) and aversive (bitter solution) outcome in each trial. While measuring dopamine, we also recording neural spiking activity and local field potentials from frontostriatal circuitry. We found that dopamine was modulated by the two alternative outcomes, and that selective neurons within the circuitry encoded separately each alternative. This work will have important implications in understanding approach and avoidance behaviors, the balance of which is often impaired in the presence of disease.

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Poster

P254. Dopamine and Non-Dopamine Pathways II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P254.02

Topic: C.03. Parkinson's Disease

Support: CIHR grant MOP 136778

Title: Amyloid beta deposition and cognitive decline in Parkinson's disease: a study of the PPMI cohort

Authors: *A. MIHAESCU[1], M. MASELLIS[2], A. GRAFF-GUERRERO[3], M. VALLI[4], C. URIBE[4], R. GANDHI[4], M. DIEZ-CIRARDA[4], A. STRAFELLA[5];
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Abstract: The accumulation of amyloid beta (Aβ) in the brain has a complex and poorly understood impact on the progression of Parkinson's disease (PD) pathology. Increased Aβ burden has been associated with cognitive decline in PD, especially in PD with dementia. Some studies however have found no association between high Aβ burden and cognitive decline in PD patients, instead suggesting normal aging may be responsible for both Aβ accumulation and cognitive decline. To better understand the relationship between PD cognitive decline and Aβ deposition, our study focused on a homogeneous group of PD patients whose cognitive abilities were measured longitudinally for 2 years after their Aβ scan. Our study used PD patients and healthy control data from the Parkinson's Progression Marker Initiative (PPMI) cohort: a multi-center cohort of 25 *de novo* idiopathic PD patients and 30 healthy controls (HC). These participants underwent a [18F]florbetaben positron emission tomography scan which can measure the density of Aβ in vivo in the brain. The data was analyzed using IBM SPSS version 27 software with linear regression modeling, Pearson correlations, and hierarchical cluster analysis. A stepwise linear regression of the PD group revealed a strong adjusted R2 of 0.495 in a model explaining their Montreal Cognitive Assessment (MoCA) score 1-year post-scan using the standardized uptake value ratios from the 20 regions of interest (ROI) as independent variables. The ROIs used in this stepwise model were the left rectus, the left anterior cingulate cortex, and the right parietal cortex. We found the PD group's 20 ROIs formed three clusters, with increased Aβ burden in cluster two correlating more strongly with a poorer cognitive score, while Aβ burden in clusters one and three had a mixed effect upon MoCA score. The results suggest Aβ does have a moderate association with cognitive decline in PD, independent of age. We found Aβ accumulation in the brain has a different effect on PD cognition depending on which ROI is affected, with increased Aβ in the rectus gyrus having no detrimental effect on MoCA score 1-year post-scan. The inconsistencies in the literature regarding Aβ and PD cognitive decline may be due to the patchwork impact Aβ burden has on different brain regions.

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Poster

P254. Dopamine and Non-Dopamine Pathways II

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Topic: C.03. Parkinson's Disease

Support: 5T32NS086749-05
2T32NS086749-06

F32 DC 018431

Title: What determines selective vulnerability and circuit integration of olfactory bulb dopaminergic neurons?

Authors: *T. KUNKHYEN[1], A. A. LAUER[1], T. R. BRECHBILL[1], A. N. RANGEL[1], C. E. J. CHEETHAM[2];
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Abstract: One of the hallmarks of many neurogenerative diseases is the selective loss of specific sub-populations of neurons. Dopaminergic (DA) neurons in the mouse olfactory bulb (OB) are highly activity dependent. While 40% of DA neurons undergo cell death after a month of sensory input blockade by naris occlusion, the remaining 60% are resilient to this loss of sensory input. Furthermore, OB DA neurons are continuously generated in the subventricular zone throughout life, enabling them to fully repopulate a month after reversal of naris occlusion. This makes OB DA neurons an ideal model system in which to determine what underlies selective vulnerability in subset of DA neurons and how newborn DA neurons are functionally integrated to replace previously lost neurons. We have developed an experimental paradigm that enables rapid elimination followed by gradual restoration of sensory input to the OB. By performing chronic *in vivo* 2-photon imaging in DAT-cre;Ai9;Ai162 mice that express both a red fluorescent protein as a morphological marker and a green genetically encoded calcium indicator in DA neurons, we can track the survival and integration, as well as the odor response properties, of individual neurons over long periods of time (10 weeks). We found that loss of DA neurons was significantly elevated during the first week after ablation of OB sensory input compared to control mice (-9.5% vs. -0.94%, $P < 0.001$, Sidak's test after two-way ANOVA). In contrast, there was no effect on DA neuron loss 7-14 days after OSN ablation (-3.0% vs. -1.4%, $P = 0.29$). We are currently analyzing whether there are differences in the vulnerability of the previously described large embryonically-generated and small postnatally-generated DA neurons. We are also quantifying the rate of newborn DA neuron integration before and after ablation of OB sensory input. Finally, we are analyzing whether there are differences in the odor response characteristics between DA neurons that are vulnerable or resilient to sensory input ablation, and whether newly integrated DA neurons share odor response properties with DA neurons that had previously been lost. This work will provide new insight into what distinguishes vulnerable neurons from their neighbors, which may inform novel targeted therapeutic strategies for the treatment of neurodegenerative diseases.

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Poster

P254. Dopamine and Non-Dopamine Pathways II

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Topic: C.03. Parkinson's Disease

Support: NIH Grant T32GM136499
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Title: Sonic hedgehog signaling onto striatal cholinergic interneurons impinges on D2 signaling via the integrated stress response pathway to mediate L-dopa induced dyskinesia induction

Authors: *S. URIBE-CANO[1], K. LIU[2], M. HOLMES[2], L. MALAVE[3], A. H. KOTTMANN[1]; [1]Physiology, Pharmacol. and Neurosci., CUNY City Col. of New York, New York, NY; [2]CUNY Sch. of Med., City Col. of New York, New York, NY; [3]Dept. of Psychiatry, Columbia Univ., New York City, NY

Abstract: Dopamine neuron (DAN) loss, a hallmark sign of Parkinson's Disease (PD), is treated with L-Dopa therapy which attenuates bradykinesia and akinesia in patients. Unfortunately, prolonged treatment with L-Dopa leads to a debilitating side effect called L-Dopa induced dyskinesia (LID) in a majority of medicated PD patients. Cholinergic interneurons of the striatum (CIN), which are projection targets of DAN and undergo pathophysiological changes in response to DAN degeneration, have been increasingly implicated in LID. However, the mechanistic underpinnings of how L-Dopa-derived Dopamine (DA) signaling onto CIN interacts with the pathophysiological changes CIN undergo following DAN degeneration to produce LID remains poorly understood. CIN express both inhibitory D2 and facilitatory D5 receptors. In the work presented here, we probe the degree to which DA action via D2 on CIN following L-Dopa administration may be responsible for LID induction. Amazingly, selective ablation of the D2 receptor from CIN in 6-OHDA animals treated with L-Dopa attenuated LID and reduced levels of the LID cytochemical marker p-ERK among CIN. Unexpectedly however, quantification of the neuronal activity marker p-rpS6 in CIN suggested that D2 ablation leads to lower activity levels in CIN following L-Dopa administration. In parallel lines of study, we recently found that release of the signaling peptide sonic hedgehog (Shh) from DAN to CIN is crucial in the prevention of aberrant CIN physiology and LID induction. Prior work from our group has also demonstrated that Shh signaling onto CIN impinges on the Integrated Stress Response pathway (ISR), suggesting that changes in physiological cell stress among CIN following DAN degeneration are mediated by changes in Shh signaling. These findings are made relevant to D2 signaling on CIN given recent work by Helseth and colleagues (2021) demonstrating that attenuation of the ISR pathway in CIN can produce an inversion of D2 from acting as inhibitory to excitatory. Specifically, we hypothesize that altered ISR activity in CIN mediated by changes in Shh signaling can lead to disruptions of physiological D2 signaling and promote LID. Here, alongside our characterization of how D2 ablation from CIN prevents LID induction, we begin to investigate how Shh signaling impinges on the ISR pathway and impacts D2 signaling on CIN to prime LID formation following DAN degeneration in PD.

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Poster

P254. Dopamine and Non-Dopamine Pathways II

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University of Calgary Graduate Faculty Council Doctoral Scholarship

Title: Global remodelling of afferent and efferent projections of the A13 region following unilateral nigrostriatal degeneration using 6-hydroxydopamine

Authors: *L. H. KIM[1], T. CHOMIAK[2], M. A. TRAN[3], S. TAM[3], C. MCPHERSON[3], S. E. A. EATON[3], Y. OU[5], Z. H. T. KISS[4], P. J. WHELAN[3];
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Abstract: Parkinson's disease (PD) is a complex disease affecting many facets of movement and cognitive function. Gait abnormalities worsen over time and remain difficult to treat. We found previously that the A13 nucleus of rostromedial zona incerta (ZI) projects to brainstem locomotor regions, suggesting that a parallel dopaminergic pathway in addition to the nigrostriatal pathway exists. Dopaminergic cells within the ZI-hypothalamic region are preserved in the advanced PD brain despite a global remodelling of the dopaminergic system following nigrostriatal degeneration. To explore alternative targets to treat gait dysfunctions in PD, we targeted the A13 connectome. We used an established unilateral 6-OHDA PD model (N=8, male C57BL/6 mice 7-8 weeks of age). Sham mice (N=3) received a vehicle solution of the same volume. AAV8-CamKII-mCherry and AAVrg-CAG-GFP were microinjected at the ipsilesional A13 region. Brains were processed 5 weeks later using a modified iDISCO clearing method and imaged on a lightsheet microscope (LaVision Biotech UltraMicroscope). Brains with insufficient quality in labelling were excluded from analysis. Immunolabeled cells were manually counted, and fibres were segmented using Ilastik and ImageJ. WholeBrain software was used to register with the Allen Brain Atlas. Segmentations from any given brain region were normalized to the total amounts across all regions per mouse and analyzed using Spearman's cross-correlation matrices. Despite A13 cells being largely preserved, there was a remarkable change in both the afferent and efferent A13 connectome. The sham group showed strong interconnections with the cerebral cortex and A13 region. Although these cortical-A13 afferents and efferents were

downregulated most severely following nigrostriatal degeneration, afferent projection patterns from the isocortical (motor, sensory, visual, and prefrontal) areas remain anti-correlated with the rest of the neuraxis. In contrast, the efferent projection patterns show greater cross-correlation across the neuraxis. In a human PD patient brain with gait dysfunction we found substantial degeneration of the substantia nigra compacta with preservation of the A13 and the locus coeruleus. Our findings suggest that executive control of behavioural response, which could be modulated by descending ZI efferents, remains intact following nigrostriatal degeneration. This work provides a comprehensive insight into the plasticity in A13 connectome that could contribute to heterogeneity of progression and severity in PD.

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Poster

P254. Dopamine and Non-Dopamine Pathways II

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Topic: C.03. Parkinson's Disease

Support: University of California Riverside start-up package

Title: Pharmacological functional ultrasound imaging to characterize the effects of NMDA-antagonist MK-801 on blood perfusion in the mouse brain

Authors: *K. AGYEMAN[1], L. CROWN[2], I. HOANG[3], N. LAZKANI[5], C. LIU[3], V. N. CHRISTOPOULOS[6], D. J. LEE[4];
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Abstract: NMDA hypofunction has been proposed as a mechanism underlying the cognitive deficits observed in neuropsychiatric disorders such as schizophrenia. MK-801 is a potent and selective NMDA antagonist and has been used to model NMDA hypofunction in rodents. Despite its widespread use as a research model, little is known about the effects of MK-801 on blood perfusion in brain regions and circuits important to cognitive function. Pharmacological functional ultrasound (Pharmaco-fUS) is a relatively new technique that uses functional ultrasound imaging (fUSI) to quantify the effects of pharmacologically induced changes to blood perfusion with unprecedented spatiotemporal resolution (100 μm and up to 10 ms). In this study, we assess the pharmacodynamics of blood perfusion changes in the mouse hippocampus, thalamus and isocortex induced by MK-801. We acquired baseline cerebral perfusion recordings

for 5 minutes followed by an intraperitoneal injection of MK-801 (1 mg/kg) in a group of 14 isoflurane-anesthetized male C57BL/6 mice. Drug effects were compared to saline-vehicle injection in another group of 14 animals. By recording fUS activity for 40 min post injection, we found that MK-801 causes a significant reduction in blood perfusion in cortical and sub-cortical regions [MEAN +/- 95% Confidence Interval (CI) over the last 5min; hippocampus (-2.88 +/- 0.27 %, p = 2.22e-4), isocortex (-4.22 +/- 0.60 % p = 0.0047), and thalamus (-4.89 +/- 0.68% p = 0.0042)]. No significant changes of blood perfusion were found over time in the saline control animals [mean difference 0.357 % (p = 0.918)]. The greatest change in blood perfusion was observed in the thalamus, which exhibits a relatively linear decrease in blood perfusion over time following MK-801 injection [average rate, -0.114 +/- 6.0e-4 % per min, 95% CI]. Blood perfusion in the hippocampus also decreased [average rate, -0.066 +/- 6.0e-4 % per min, 95% CI] in a linear manner as well. Intriguingly, blood perfusion in the isocortex exhibited an immediate perfusion decrease [average rate, -0.624 +/- 0.012 % per min, 95% CI], restoring increase [average rate, 0.282 +/- 0.012 % per min, 95% CI], followed by decrease [average rate, -0.186 +/- 6.0e-3 % per min, 95% CI] in perfusion akin to that observed for the thalamus and hippocampus. Together, these preliminary results indicate that pharmacologic NMDA receptor antagonism has regionally distinct effects on blood perfusion. These insights may shed light on the dynamics of pharmacological models of NMDA hypofunction, providing further insight into models of altered brain function in neuropsychiatric disorders.

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Poster

P254. Dopamine and Non-Dopamine Pathways II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P254.08

Topic: C.03. Parkinson's Disease

Support: NIH NI AG065682

Title: Gpr139, a modulator of mu opioid receptor, is a target of sonic hedgehog signaling in cholinergic interneurons of the adult striatum and modulates cholinergic activity

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Abstract: Cholinergic interneurons (CIN) are key components of striatal circuits. CIN have been implicated in various processes including learning, aging, addiction, and Parkinson's disease (PD). The gold standard treatment for PD is dopamine (DA) substitution therapy with L-Dopa,

which aims to mitigate the functional consequences of dopamine cell loss. However, long-term treatment leads to the development of debilitating L-Dopa induced dyskinesia (LID). We have previously shown that activation of the GPCR Smoothened (Smo) on CIN attenuates L-Dopa induced dyskinesia (LID) in rodent and macaque models of PD (Malave et al., 2021). The predominant source of the physiological ligand for CIN expressed Smo, Sonic Hedgehog (Shh), are ventral midbrain DA neurons (DAN) (Gonzalez et al., 2012). To elucidate possible mechanism(s) by which loss of Shh signaling from DAN might mediate CIN pathophysiology in PD, we identified CIN-specific downstream transcriptional targets of Smo. We utilized translating ribosome affinity purification (TRAP), to isolate mRNA selectively from CINs for RNAseq in mice with conditional ablation of Shh from DAN and controls. The results from the RNAseq analysis identified the orphan G-coupled receptor GPR139 as a potential target for further investigation. Subsequent experiments showed that striatal GPR139 expression is reduced in mice with conditional ablation of Shh from DAN compared to controls. GPR139 expression in the wild-type striatum reveals a prominent low (medial) to high (lateral) expression gradient that matches the expression gradient of the transcriptional effector of Shh signaling, Gli3. GPR139 is a cell autonomous negative modulator of endo-opioid signaling acting downstream of mu-opioid receptor (MOR) activation by morphine and fentanyl. Previous studies have shown morphine decreases ACh release, while antagonists of MOR increase striatal ACh. Further, antagonists of MOR were previously found to attenuate LID in animal models of PD. Thus, we hypothesized that Shh signaling might attenuate LID by modulating the CIN response to MOR activation. We used fiber photometry with DA- and ACh- sensors to monitor *in vivo* extracellular levels of DA and ACh combined with localized micro-fluid injection of drugs in the striatum and detected increased striatal ACh levels in response to GPR139 agonist. To test if this effect is cell autonomous, we performed acute, Cre-dependent CIN CRISPR knock-down of GPR139. Using this novel paradigm, we will present pharmacological epistasis experiments that investigate the molecular details of the modulation of MOR signaling by Smo in CIN and the effect of GPR139 pharmacology on LID.

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Poster

P254. Dopamine and Non-Dopamine Pathways II

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Program #/Poster #: P254.09

Topic: C.03. Parkinson’s Disease

Support: NRF-2020R1A4A1018714
2021R1I1A1A01059678

Title: Associated white matter connectivity networks with the patterns of striatal dopamine depletion in Parkinson's disease

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Abstract: Recent studies suggest that the patterns of nigrostriatal dopaminergic denervation are associated with clinical heterogeneity in Parkinson's disease (PD), and the patterns have individuality and are consistent over the disease progression. In our previous study, we identified the four patterns of striatal dopamine depletion by the factor analysis based on the dopamine transporter (DAT) availability in each striatal subregion. The results suggest that the patterns could provide information on the long-term prognosis of early-stage PD patients, and could also be contributed as a predictive marker of motor complications or dementia, which reflects the heterogeneity in PD. In this regard, this study aims to investigate whether the striatal dopamine depletion pattern is associated with a white matter (WM) connectivity network in drug-naïve early-stage PD. A total of 240 patients (age = 70.6 ± 8.9; female, No. (%) = 125(52.1%)) with newly diagnosed PD who underwent ^{18}F-FP-CIT PET scans and diffusion tensor imaging at initial assessment were enrolled. Factor analysis-derived striatal dopamine loss patterns were estimated in each patient for four striatal sub-region factors (*caudate*, *more-affected sensorimotor striatum*, *less-affected sensorimotor striatum*, and *anterior putamen*). Here, a higher value of the factor indicates DAT availability of that factor is relatively preserved than other striatal sub-regions. Then, the structural networks associated with the composite scores of each striatal sub-region factor were identified. Lastly, Pearson's correlation analyses were performed to assess the relationship of cognitive domain with the composite scores of each striatal sub-region factor, and with the connectivity strength of identified subnetworks. The composite score of *caudate* was associated with strong structural connectivity in a subnetwork composed of the left caudate and left frontal gyri. Selective dopamine loss in the caudate was associated with strong connectivity in the structural subnetwork whose hub nodes were bilateral thalami and left insula that was connected to the anterior cingulum. Whereas there was no subnetwork correlated with other striatal sub-region factors. The composite score of *caudate* and the connectivity strength of positively correlated subnetwork with *caudate* showed a significantly positive association with the frontal/executive function domain. These findings suggest that different patterns of striatal dopamine depletion are closely associated with WM structural alterations, which may contribute to heterogeneous clinical features in individuals with PD, particularly in terms of cognitive function.

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Poster

P254. Dopamine and Non-Dopamine Pathways II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P254.10

Topic: C.03. Parkinson’s Disease

Title: In vitro models: Tools to advance Parkinson’s disease research

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Abstract: Parkinson's disease (PD) is a neurodegenerative disorder that predominantly affects the dopaminergic (DA) neurons of the brain and results in loss of motor function. Studying the underlying molecular mechanisms and subsequent drug development against Parkinson's disease is often challenging with primary human dopaminergic neurons not being accessible. Stem cells therefore serve as powerful tools in generating specific cell types *in vitro.* These differentiated stem cell models can then be used to study the disease in the right biological context. Here, we show human induced pluripotent stem cells (iPSCs) being differentiated into dopaminergic neurons which were then characterized using specific antibodies against cell specific markers like TH, DAT, LMX1B and FOXA2: TH is an enzyme which is involved in the conversion of L-tyrosine into dopamine precursor L-DOPA. DAT is a transmembrane transporter which is vital in maintaining the dopamine levels. LMX1B and FOXA2 are transcription factors that regulate specification and differentiation of DA neurons. Mutations on genes like PARK7, SNCA, LRRK2, MAPT, PINK1 and PARK2 have been found to be linked with Parkinson’s disease. Knockout models for these key genes were generated and were used to ascertain specificity of Invitrogen antibodies against these targets. Antibodies against these genes are incredibly useful in understanding disease progression and arrive at novel methods of treatment. Given the importance of PD, it is imperative that scientists are provided with the necessary resources or tools which will enable them to take their research forward. Recognizing this critical need, Invitrogen offers several specific and extensively validated antibodies for the key players of this disease. The data here shows that these antibodies were tested in the most relevant cell models and have been confirmed for specificity.

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Poster

P256. Alpha-Synuclein: Models I

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Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P256.01

Topic: C.03. Parkinson’s Disease

Support: NINDS R56NS117465

Title: Early synaptic dysfunctions associated with Parkinson Disease-relevant alpha-synuclein pathology

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Abstract: The main pathological hallmarks of Parkinson's Disease (PD) are loss of dopaminergic neurons in the substantia nigra pars compacta (SNpc) and accumulation of alpha-synuclein (α-syn) in intracellular, insoluble inclusions in cell bodies and axons, collectively termed Lewy pathology (LP). α-Syn normally localizes to membranes at the presynaptic terminal where its interaction with SNARE proteins has been implicated in synaptic transmission. Aggregated synuclein corrupts presynaptic endogenous α-syn to form pathologic aggregates. Experimentally, aspects of progressive α-syn pathology can be recapitulated in model systems by exogenous introduction of recombinant α-syn preformed fibrils (PFFs). Templated α-syn aggregation reduces spine density *in vitro* and *in vivo* hippocampal primary neurons in rodent models. This causes increased frequency of miniature excitatory postsynaptic currents (EPSCs) and presynaptic docked vesicles, suggesting enhanced presynaptic function. These observations suggest that one of the initial consequences of α-syn pathology may be dysfunctional synaptic transmission preceding neurodegeneration and DA terminal loss. However, research focused on identifying these early synaptic alterations *in vivo,* in circuits relevant to PD symptomatology, is lacking. Indeed, the role aggregated α-syn in the impairment of corticostriatal synaptic transmission and plasticity remains incompletely studied despite cumulative evidence that dysfunction of these synapses plays a key role in mediating the deficits associated with PD. We investigated synaptic transmission and plasticity at dorsolateral corticostriatal glutamatergic synapses onto spiny projection neurons, in adult male mice 8 weeks post intrastriatal injection of α-syn preformed fibrils (PFF) or monomer α-syn. Using whole cell patch clamp recordings, we found that compared to monomer, PFF-injected mice had increased basal synaptic transmission, enhanced spontaneous release of glutamate and increased presynaptic sensitivity to Ca2+, as indicated by increased paired pulse ratio. Despite the enhanced glutamatergic transmission, corticostriatal synapses in PFF injected animals were not able to express long-term synaptic potentiation (LTP) assessed in extracellular recordings, and LTD was decreased. Altogether, this data reveals an early deficit in glutamatergic corticostriatal synaptic transmission and plasticity induced by α-syn pathologies.

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Poster

P256. Alpha-Synuclein: Models I

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Topic: C.03. Parkinson's Disease

Support: NIH Grant NS038065
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Title: Integrated Stress Response in Alpha-Synucleinopathy

Authors: *C. FANG[1], D. JANGIR[1], P. GRIFFIN[1], R. KARIM[1], J. MEINTS[1], S. MORE[1], M. K. LEE[2];
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Abstract: Abnormalities in α-synuclein (αS) is directly linked to the pathogenesis of Parkinson's disease (PD) and related disorders called α-synucleinopathies. We showed that α-synucleinopathy in a αS transgenic model (TgA53T) and humans causes chronic endoplasmic reticulum stress (ERS)/unfolded protein response (UPR)/Integrated Stress Response (ISR). Treatment of TgA53T with salubrinal, a compound that inhibits dephosphorylation of eIF2α and attenuates ERS induced toxicity, can significantly delay onset of αS pathology and motor deficits. To further establish ERS/UPR/ISR as an important pathogenic factor in α-synucleinopathy, we examined if reducing phosphorylation of eIFα by the Protein kinase R-like ER Kinase (PERK), exacerbates α-synucleinopathy. We show that conditional deletion of PERK in neurons of TgA53T mice leads to significantly earlier onset of α-synucleinopathy, showing the pathologic importance of PERK-eIF2α pathway. While salubrinal inhibits both the protein phosphatase 1 regulatory subunit 15A (PPP1R15A, Gadd34) and PPP1R15B (CReP), studies show that inhibition of Gadd34 could be neuroprotective. Significantly, neither the pharmacological inhibition of Gadd34 or genetic loss Gadd34 function attenuated α-synucleinopathy in TgA53T model. Significantly, treatment of TgA5t model with an CReP inhibitor delays disease onset and attenuates pathology in TgA53T model. Our data suggest ISR components, particularly inhibition of CReP, is a therapeutic target for α-synucleinopathy.

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Poster

P256. Alpha-Synuclein: Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P256.03

Topic: C.03. Parkinson's Disease

Title: Fluorescence-activated nuclei sorting (FANS) and a novel combinatorial fluidic indexing to elucidate molecular pathways involved in the pathogenesis of Parkinson's disease.

Authors: *M. AVALLONE[1], J. PARDO[1], S. PALO[1], U. PFISTERER[2], P. BRATTÅS[2], J. RAJOVA[1], M. ÅKERBLOM[1], M. DAVIDSSON[1], T. BJORKLUND[1];
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Abstract: Parkinson's Disease (PD) is a neurodegenerative disorder characterized by loss of dopamine (DA) neurons of the Substantia Nigra (SN). There is a big body of evidence suggesting that alpha-synuclein (aSyn) plays a central role in the pathogenesis of PD. However, there is no molecularly precise, single cell in vivo transcriptomic studies with accurately defined aSyn overload. Here we describe a novel strategy and recent results achieved based on AAVs engineering and transgenic animals. The engineered MNM008 AAVs capsid, capable of retrograde transport in DA neurons, is injected in the striatum of TH-Cre rats and DAT-Cre mice. The vector carries uniquely barcoded aSyn genomes or barcoded-nonpathogenic tagBFP. Injecting in the striatum with Cre-recombinase restriction of the expression to DA neurons of the SN, we avoid possible injection-related inflammation and cell loss. The nuclei from the microdissected SN area are enriched for DA neurons using FANS. This is achieved thanks to the viral vector prep containing a Cre-inducible histone-linked GFP (H2B-GFP), which is only expressed in the population of interest. Using a novel combinatorial-indexing based single nuclei sequencing approach called Sci-Fi Seq we can pre-label all mRNA in each nucleus with an identifier for that specific sample. These samples are then pooled together and prepared for sequencing using 10X genomics droplet technology. Immunohistochemistry results showed that the virus injected in the striatum was retrogradely transported and expressed in DA neurons of the SNpc. The FANS enrichment strategy for our population of interest expressing the virus showed to be robust and consistent with an average of 2500 GFP positive (DA) nuclei retrieved per animal. Transcriptomic analysis on the GFP positive sorted nuclei revealed DA markers specific to the SN population, and the viral vector expression was restricted to this population, confirming the high selectivity of our approach. The combinatorial indexing and molecular barcoding will allow us to accurately measure the aSyn expression level in each DA neuron arranging them into disease stages in what we call pseudo-time. Moreover, this approach will identify genetic triggers and causative cascades of changes at the transcriptional level. From this study, we will further our knowledge on PD disease progression as well as possible new treatments.

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Poster

P256. Alpha-Synuclein: Models I

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Topic: C.03. Parkinson's Disease

Support: JP21dm0207001

Title: A transgenic marmoset as a novel nonhuman primate model of Parkinson disease and its preclinical states

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Abstract: Parkinson disease (PD) is an adult-onset, intractable neurodegenerative disease characterized byslowly progressive symptoms, including motor, sleep, and olfactory disturbances. Here, we describe transgenic marmoset lines carrying a pathogenic mutation in *SNCA*(A30P) as forming a nonhuman primate (NHP) model of PD. The animals exhibited diverse late-onset PD-like behaviors, including REM sleep disruptions and agait disturbance that was partly responsive to dopamine replacement. Multiple subcortical regions along with the dopaminergic system exhibited α-synuclein-related pathologies, and long-term loss of dopamine transporters and oligomerization of α-synuclein (but not Lewy-like inclusions) were observed in the prodromal phase. Analysis of several animals at different ages and stages of disease progression indicated that this model animal exhibited hallmark pathologic abnormalities at various phases of disease, including as α-synuclein oligomerization at a preclinical/prodromal phase and Lewy body-like α-synuclein inclusions at a later phase. This work documents the first transgenic NHP in which α-synuclein anomalies were demonstrated to be associated with the late-onset symptoms and diverse pathologiesof PD. We propose that this modelwill enable the identification of predictive biomarkers and the assessment of preemptive and disease-modifying therapiesfor PD.

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Poster

P256. Alpha-Synuclein: Models I

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Program #/Poster #: P256.05

Topic: C.03. Parkinson's Disease

Title: Longitudinal single-cell tracking to assess survival of human neurons: application to synucleinopathy

Authors: *J. CHOI[1], H. KII[2], J. NELSON[1], Y. YAMAZAKI[2], F. YANAGAWA[3], A. KITAJIMA[2], T. UOZUMI[2], Y. KIYOTA[2], D. DOSHI[1,4], H. SADLISH[1], K. RHODES[1,5], R. SCANNEVIN[1,6], C. CHUNG[1];
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Abstract: Developing proper phenotypic assays and analysis methods is an important step in the drug discovery process. Assays using iPSC-derived human neurons are emerging as powerful tools for the treatment of neurological disease. We have previously shown that the longitudinal single cell tracking enabled the detection of survival/death of neurons after overexpression of mutant alpha synuclein (A53T), a familial Parkinson's disease mutation. The reliance of this method on manual counting, however, renders the process labor intensive, time consuming and error prone. To overcome these hurdles, we developed algorithms for automated detection of neurons using BioStation CT live imaging system and CL-Quant software. With these algorithms, we successfully measured the risk of neuronal death caused by the overexpression of A53T alpha synuclein with similar accuracy and improved consistency compared to manual counting. It also provided an additional key readout of neuronal fitness: total neurite length and the number of nodes from which neurites were projected from the cell body. The algorithm was able to detect the beneficial effects of BDNF treatment in A53T alpha synuclein overexpressing neurons. These data show that the automated algorithm will improve the consistency of the neuronal health measurement and considerably shorten the assay time, making this assay amenable for small molecule screening.

Disclosures: **J. Choi:** A. Employment/Salary (full or part-time):; Yumanity Therapeutics. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Yumanity Therapeutics. **H. Kii:** A. Employment/Salary (full or part-time):; Nikon. **J. Nelson:** A. Employment/Salary (full or part-time):; Yumanity Therapeutics. E. Ownership Interest (stock, stock options, royalty, receipt of

intellectual property rights/patent holder, excluding diversified mutual funds); Yumanity Therapeutics. **Y. Yamazaki:** A. Employment/Salary (full or part-time):; Nikon. **F. Yanagawa:** A. Employment/Salary (full or part-time):; Nikon. **A. Kitajima:** A. Employment/Salary (full or part-time):; Nikon. **T. Uozumi:** A. Employment/Salary (full or part-time):; Nikon. **Y. Kiyota:** A. Employment/Salary (full or part-time):; Nikon. **D. Doshi:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Yumanity Therapeutics. **H. Sadlish:** A. Employment/Salary (full or part-time):; Yumanity Therapeutics. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Yumanity Therapeutics. **K. Rhodes:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Yumanity Therapeutics. **R. Scannevin:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Yumanity Therapeutics. **C. Chung:** A. Employment/Salary (full or part-time):; Yumanity Therapeutics. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Yumanity Therapeutics.

Poster

P256. Alpha-Synuclein: Models I

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Program #/Poster #: P256.06

Topic: C.03. Parkinson's Disease

Support: NIEHS R21 ES031211-01A1
NIGMS 1P20GM134974-01A1
NASA 80NSSC21K0273

Title: A novel genetic sensor and "Seek and Rescue" strategy to unravel the pathogenic role of environmental risk factor-driven neuronal DNA damage response in sporadic neurodegenerative disorders

Authors: *M. S. GRAMES[1], M. B. W. EL-SAADI[1], X. TIAN[1], K. M. KEYS[1], M. REN[1], C. E. HAACKER[1], E. L. KNOTT[1], H. M. LI[1], S. WANG[2], L. HARRISON[3], X.-H. LU[1]; [1]Pharmacology, Toxicology, and Neurosci., [2]Biochem. and Mol. Biol., [3]Mol. and Cell. Physiol., LSU Hlth. Sci. Ctr. Shreveport, Shreveport, LA

Abstract: Parkinson's disease (PD) is the second-most prevalent neurodegenerative disease, where the vast majority of incidences have no identifiable inheritance and occur in a sporadic form. Environmental risk factors (e.g., pesticide/herbicides, exposure to metals and solvents, virus infection, and head trauma) have been speculated as a trigger to the sporadic PD. To explore the causal and pathogenic evidence of environmental triggers, we developed a genetic

sensor exploiting the interaction of DNA virus with host cell DNA damage response (DDR) as an innate antiviral defense mechanism. We harnessed the recombinant Adeno-associated virus (rAAV) genome-processing machinery and the instability of a hypermutable repeat sequence to detect neuronal genomic instability and visualize neurodegeneration in post-mitotic differentiated neurons. We demonstrated the viral genetic probe is sensitive in detecting neurons with increased DNA damage response. To quantify DDR in vivo, we developed an efficient mutation reporter assays using the DNA Endonuclease T7E1, targeted next-generation sequencing, and capillary electrophoresis. Exposure to paraquat, a PD-associated environmental toxicant, heightened neuronal genomic instability and elicited neuropathology revealed by single-cell analysis. The increased neuronal genomic instability was also probed in a genetic alpha-synuclein (aSyn) transgenic mouse model infused with pre-formed fibrils. Based on the neuronal burden of synucleinopathy, we documented four pathological stages of Lewy body formation in vivo. We identified a possible "initiation stage," where aSyn aggregates colocalized with DNA damage foci inside the nucleus without cytoplasmic Lewy body formation. A similar pathology was also observed in the caudate nucleus of patients with Lewy body dementia, suggesting a pathogenic causal role of DNA damage response in aSyn aggregation/propagation and proteotoxicity. To determine the causal link and to explore the potential for disease-modifying therapies, we developed a novel genetic strategy to "Seek-and-Rescue" neurons with DNA damage non-invasively via the systemic introduction of a genetic sensor and actuator of DDR. Dose responses for various amounts of vector genomes of the genetic sensor were determined. A genetic actuator was implemented via a Cre-dependable conditional shRNA to knockdown ATM, a major orchestrator of the DDR in the hope to slow down the progression of neurodegeneration. These studies can pave the way for new paradigms to determine the causal and pathogenic roles of DDR and exposure to environmental toxicants in the progression of dementia and identify therapeutic targets.

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Poster

P256. Alpha-Synuclein: Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P256.07

Topic: C.03. Parkinson's Disease

Title: Impaired striatal plasticity and dendritic spine loss and remodeling in the premotor stage of an animal model of progressive dopaminergic degeneration

Authors: *M. RODRIGUEZ-OROZ[1], L. MERINO-GALAN[2], M. ZAMARBIDE[3], A. BELLOSO-IGUERATEGUI[4], B. GAGO[5], D. DUMITRIU[6], Y. GROSSMAN[7], A. QUIROGA-VARELA[8];
[1]CIMA-Clínica Univ. de Navarra-IdiSNA, Pamplona/Iruna, Spain; [2]CIMA-University of the Basque Country, Pamplona/Iruna, Spain; [3]CIMA-IdiSNA, Pamplona, Spain; [4]CIMA, Pamplona, Spain; [5]Inst. de Investigación Biomédica de Málaga, Univ. de Málaga, Málaga, Spain; [6]New York State Psychiatric Institute, Columbia Univ., New York, NY; [7]Duke Univ., North Carolina, NC; [8]Neurosci., CIMA-IdiSNA-Girona Biomed. Res. Inst. (IDIBGI), Salt, Spain

Abstract: Parkinson's disease (PD) is characterized by a progressive loss of dopaminergic neurons in the substantia nigra compacta (SNc) and accumulation of α-synuclein (α-syn), which underlies complex functional and structural changes in striatal spiny projection neurons (SPNs). Although a reduction in SPNs dendritic spine density has been reported in either postmortem studies of advanced PD patients or animal models of parkinsonism, it is unknown if these alterations occur since the onset of the nigrostriatal dopaminergic degeneration. Thus, our aim was to study the temporal sequence of functional and structural changes in striatal dendritic spines in the premotor stage of parkinsonism. For that purpose, rats were inoculated in the SNc with an adeno-associated viral vector coding for A53T mutated human α-syn (hα-syn) and evaluated at 72h, 1, 2 and 4 weeks post-inoculation (p.i.). Synaptic plasticity by chemical stimulation of LTP (cLTP) in isolated striatal synaptosomes and measurement of striatal dopamine by HPLC were assessed. The analysis of the dendritic arborization and the density and morphology of SPNs dendritic spines was performed by microinjection and high-resolution confocal microscopy. The ultrastructure of the dendritic spines and the presence of the spine apparatus (SA) were analysed by electron microscopy. The hα-syn group showed an inhibition of cLTP ($p<0.001$) and a decrease in dopamine content ($p<0.01$) since 72h p.i. even before the presence of hα-syn in the striatum and significant dopaminergic neurodegeneration. These functional alterations are followed by a dendritic spine remodelling, as observed by a significant loss of thin spines ($p<0.05$) at 4w p.i., along with an increase in the head volume of mushroom spines ($p<0.01$). In addition, we observed a decrease in the proportion of spines with SA since the 1st w p.i. in hα-syn group ($p<0.05$), along with an increase in SA_{area}/$SpineHead_{area}$ at 4w p.i ($p<0.01$). These structural changes occur before the development of parkinsonian motor signs and could represent a compensatory mechanism to enhance the function of existing spines, balancing the observed decrease in spine turnover. Thus, our results indicate dysfunctional dopaminergic neurotransmission by impaired striatal synaptic plasticity since very early time points, leading to dendritic spine remodelling and ultrastructural changes indicative of increased protein synthesis, before the manifestation of motor impairment.

Disclosures: M. Rodriguez-Oroz: None. **L. Merino-Galan:** None. **M. Zamarbide:** None. **A. Belloso-Iguerategui:** None. **B. Gago:** None. **D. Dumitriu:** None. **Y. Grossman:** None. **A. Quiroga-Varela:** None.

Poster

P256. Alpha-Synuclein: Models I

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Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P256.08

Topic: C.03. Parkinson's Disease

Support: CONNECT H2020 – FET ProAct-01-2018

Title: Connecting neural networks: a new model to study alpha-synuclein pathology and spreading in Parkinson disease

Authors: *G. GOMEZ-GIRO, D. FRANGENBERG, G. ROBERTSON, J. SCHWAMBORN; Luxembourg Ctr. for Systems Biomedicine (University of Luxembourg), Belvaux, Luxembourg

Abstract: Parkinson Disease (PD) is the second most common neurodegenerative disorder affecting around 1-2 % of the population above 65 years. The number of people with PD is expected to double by 2030. The primary neuropathology appears to be the chronological accumulation of alpha-synuclein (in Lewy bodies and neurites) in various parts of the brain and nervous system. The staging system of the PD brain pathology suggests that the protein aggregates are first seen in the posterior part of the brainstem. From there, the pathology takes an ascending course, reaching different nuclei in the pons and then the midbrain, affecting the dopaminergic neurons in the *substantia nigra* (SN). Furthermore, the influential hypothesis of Braak (2006) suggests that these aggregates might find their origin in the gut and are transported retrogradely via the enteric nervous system (ENS) towards the central nervous system (CNS). *In vitro*, there is growing evidence of transneuronal transport of different forms of alpha-synuclein, including potentially toxic oligomeric protein. In this project, we are aiming to model the complex interaction between the peripheral nervous system and the CNS through the incorporation of both cell types in compartmentalized devices, building on nervous-system-on-chip (NoC) technology. To represent the CNS, we use human induced pluripotent stem cell (hiPSC)-derived midbrain/hindbrain
organoids (MOs), and as a peripheral nerve contribution, we generate hiPSC-derived enteric neurons. Using PD as a showcase, we derive both cellular types from controls and a PD patient line that carries a triplication on the alpha-synuclein gene (3xSNCA). Up until this point in the project, using a specific protocol for extracting soluble and insoluble protein fractions, we succeeded in finding an increase of insoluble α-synuclein in the PD organoids compared to the controls (N=3). Additionally, we applied high content imaging and in-house built algorithms to detect alpha-synuclein and of its phosphorylated form (pS129), associated with aggregation and consequent toxicity. Considering the results obtained thus far, we can conclude that the successful development of our model in this project will enable, for the first time, to study a simple nervous system connection *in vitro*, instead of isolated tissues. Moreover, it might reveal new PD signatures in the context of alpha-synuclein pathology and spreading, providing a strong and innovative basis for the next generation *in vitro* on-chip disease models, which ultimately will reduce the need for pre-clinical animal studies and increase the efficacy of drug development.

Disclosures: G. Gomez-Giro: None. **D. Frangenberg:** None. **G. Robertson:** None. **J. Schwamborn:** None.

Poster

P256. Alpha-Synuclein: Models I

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Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P256.09

Topic: C.03. Parkinson's Disease

Support: National Institute of Health [NIH P51OD011106]
Parkinson's Foundation [PF-APDA-SFW-1922]
Trewartha Senior Honors Thesis Research Award from the University of Wisconsin - Madison

Title: Alpha-synuclein and Tau are abundantly expressed in the appendix of humans and the ceca of common marmosets and rhesus macaques.

Authors: *A. D. ZINNEN[1], J. VICHICH[1], J. M. METZGER[1], J. C. GAMBARDELLA[1,2], V. BONDARENKO[1], H. A. SIMMONS[1], M. E. EMBORG[1,2,3];
[1]Wisconsin Natl. Primate Res. Ctr., [2]Cell. and Mol. Pathology Grad. Program, [3]Dept. of Med. Physics, Univ. of Wisconsin - Madison, Madison, WI

Abstract: α-Synuclein (α-syn) proteinopathy in the neurons of the enteric nervous system is proposed to have a critical role in Parkinson's disease (PD) onset and progression. Interestingly, the human appendix harbors abundant α-syn, and appendectomy has been linked to decrease risk and delayed onset of PD, suggesting that the appendix may influence PD pathology. Common marmosets and rhesus macaques lack a distinct appendix (a narrow closed-end appendage with a distinct change in diameter at the junction with the cecum), yet the cecal microanatomy of these monkeys is similar to the human appendix. Sections of human appendix (n=2) and ceca from common marmosets (n=4) and rhesus macaques (n=3) were evaluated to shed light on the microanatomy and the expression of PD-related proteins. Analysis confirmed that the human appendix and marmoset and rhesus ceca present thick walls comprised of serosa, muscularis externa, submucosa, and mucosa plus abundant lymphoid tissue. Expression of α-syn and tau in the cecum/appendix was present within myenteric ganglia and along nerve fibers of the muscularis externa and mucosa in all species. In the myenteric ganglia α-syn, p-α-syn, tau and p-tau immunoreactivities (ir) were not significantly different across species. The percent area above threshold of α-syn-ir and tau-ir in the muscularis externa and mucosa were greater in the human appendix than in the nonhuman primate cecum (α-syn-ir $p<0.0001$; tau-ir $p=0.0006$). Overall, this study provides evidence that the common marmoset and rhesus macaque ceca are remarkably similar to the human appendix as they share comparable microanatomy and dense immunoreactivity of α-syn, p-α-syn, tau and p-tau.

Disclosures: A.D. Zinnen: None. **J. Vichich:** None. **J.M. Metzger:** None. **J.C. Gambardella:** None. **V. Bondarenko:** None. **H.A. Simmons:** None. **M.E. Emborg:** None.

Poster

P256. Alpha-Synuclein: Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P256.10

Topic: C.03. Parkinson's Disease

Support: 1F30DK124987-01

Title: Altered cholinergic neurotransmission in the ENS of mThy1-hSNCA mice

Authors: *E. ZIEHL[1], N. YELLESWARAPU[2], J. J. GALLIGAN[3];
[1]Michigan State Univ. Neurosci. Program, East Lansing, MI; [2]Neurosci., [3]Neruoscience Program, Michigan State Univ., East Lansing, MI

Abstract: α-synuclein (α-Syn), a presynaptic terminal protein, is the pathological biomarker in Parkinons's disease (PD) causing cell death of dopaminergic neurons and resulting in motor deficits. Many patients with Parkinson's disease also have several gastrointestinal (GI) symptoms, the most common of which is constipation. As α-Syn is also found within the nervous system of the gut, the enteric nervous system (ENS), we hypothesize that overexpression of α-Syn within the ENS disrupts neurotransmission and results in colonic dysmotility. We used the mThy1-hSNCA, which expresses four-fold human α-Syn, and wild-type (WT) littermate mice to measure food intake, whole gut transit time, fecal pellet output, and transmural electrical stimulation on longitudinal smooth muscle contractions in the duodenum, ileum, and proximal and distal colon in the isometric tension isolated organ baths. We found that male mThy1-hSNCA mice have significantly decreased distal colon longitudinal muscle contractions compared to WT mice whereas female mThy1-hSNCA mice have significantly decreased duodenal longitudinal muscle contractions compared to WT mice. As contractions in the smooth muscle of the gut are primarily mediated by cholinergic neurotransmission, we predict that an overexpression of α-syn is altering cholinergic neurotransmission. This work supports our previous findings showing that α-Syn plays an important role in modulating enteric neurotransmission.

Disclosures: E. Ziehl: None. **N. Yelleswarapu:** None. **J.J. Galligan:** None.

Poster

P296. Cellular Stress and Death Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P296.01

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH K01 MH112983
NIH R01 NS112171

Title: Implications of heat shock protein family A member 6 (HSPA6) in neurological diseases based on in vivo analyses of a novel transgenic mouse line

Authors: *C. HOLT[1], S. CHENNAREDDY[2], S. GONZALEZ[1], R. G. GOGLIOTTI[1];
[1]Loyola Univ., Chicago, IL; [2]Vanderbilt Univ., Nashville, TN

Abstract: The heat shock protein 70 (HSP70) family is a multigene group of proteins that are highly conserved and many of its members are well studied in their role in neurological disorders. Increased expression of the *heat shock protein family A member 6* (*HSPA6*) gene is a conserved finding in autopsy samples from patients with Alzheimer's disease, Parkinson's disease, idiopathic autism, and Rett syndrome; however, unlike many of its HSP70 family members, *HSPA6* is not expressed in common model organisms. As a result, its contribution to these common neurological disorders remains unknown. While highly homologous to other HSP70 family members, *in vitro* data suggests that HSPA6 plays a functionally unique role in shuttling RNA-polymerase (Pol) II between nuclear speckles and sites of active transcription during times of cellular stress and recovery. In this context, HSPA6 may control the rate of transcriptional recovery to insult; a neuroprotective mechanism that is presently absent from most disease models. To address this critical knowledge gap, we have generated the first HSPA6 transgenic mouse (*HSPA6-TG*). Our novel HSPA6-TG mouse contains the human HSPA6 locus and the entire 3kb endogenous promoter. The entire construct is flanked by Lox-P sites, rendering it "conditional-off" in the presence of Cre-recombinase. Here we will report our progress establishing a baseline characterization of the HSPA6-TG mouse, including its temporal and spatial expression pattern, responsiveness to hyperthermia, and capacity to be induced by the HSP90-inhibitor 17-Dimethylaminoethylamino-17-demethoxygeldanamycin (17-DMAG).

Disclosures: C. Holt: None. **S. Chennareddy:** None. **S. Gonzalez:** None. **R.G. Gogliotti:** None.

Poster

P296. Cellular Stress and Death Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P296.02

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Title: The role of ferroptosis in microglia during neurodegeneration

Authors: S. RYAN[1], M. ZELIC[1], L. CHEN[2], L. GUO[2], C. LI[2], F. PONTARELLI[1], E. TEEPLE[2], M. SADEGHI[2], H. YINGNAN[2], D. KUMAR[2], M. ZHANG[2], A. BLAZIER[1], J. GANS[2], B. ZHANG[2], D. OFENGEIM[1], *T. HAMMOND[2];
[1]Rare and Neurologic Dis. Therapeut. Area, [2]Sanofi, Framingham, MA

Abstract: Ferroptosis is an iron-dependent form of regulated cell death. This pathway, as well as iron dysregulation, has been implicated in multiple neurodegenerative disorders, including Parkinson's disease (PD). Mutations in iron handling can lead to a rare set of disorders known as Neurodegeneration with brain iron accumulation. While neuron susceptibility has been well established, the role of ferroptosis in microglia during disease is not well understood. However, iron-loaded microglia have been found in affected brain regions of many disorders, including Multiple Sclerosis, Amyotrophic Lateral Sclerosis, and Alzheimer's Disease. We determined that microglia are susceptible to ferroptosis, which can be rescued by standard ferroptosis inhibitors. In a tri-culture of human iPSC-derived neurons, astrocytes, and microglia, the microglia were the most sensitive to ferroptosis at the transcriptomic, protein, and functional level. Single-cell RNA sequencing (RNA seq) revealed a distinct ferroptotic signature in the microglia which correlated with marked increases in Ferritin at the protein level and increases in IL-8 and IL-1β production. These ferroptotic signatures were rescued with pharmacological inhibitors. A genome wide CRISPR screen revealed potential regulators of ferroptosis in microglia associated with necroptotic pathways and G protein-coupled protein receptor signaling. Transcriptional analysis of PD patient blood samples revealed changes in iron, mitochondrial, and IL-8 signaling, and single cell RNA seq of PD patient brain tissue revealed a ferroptotic signature in microglia. This work places microglial ferroptosis in the context of disease, reveals a potential role for this pathway in driving neuroinflammation, and suggests that further research into therapeutics that mitigate ferroptotic stress in not just neurons but also microglia, may be of interest in treating neurodegenerative disorders.

Disclosures: S. Ryan: A. Employment/Salary (full or part-time):; Sanofi. **M. Zelic:** A. Employment/Salary (full or part-time):; Sanofi. **L. Chen:** A. Employment/Salary (full or part-time):; Sanofi. **L. Guo:** A. Employment/Salary (full or part-time):; Sanofi. **C. Li:** A. Employment/Salary (full or part-time):; Sanofi. **F. Pontarelli:** A. Employment/Salary (full or part-time):; Sanofi. **E. Teeple:** A. Employment/Salary (full or part-time):; Sanofi. **M. Sadeghi:** A. Employment/Salary (full or part-time):; Sanofi. **H. Yingnan:** A. Employment/Salary (full or part-time):; Sanofi. **D. Kumar:** A. Employment/Salary (full or part-time):; Sanofi. **M. Zhang:** A. Employment/Salary (full or part-time):; Sanofi. **A. Blazier:** A. Employment/Salary (full or part-time):; Sanofi. **J. Gans:** A. Employment/Salary (full or part-time):; Sanofi. **B. Zhang:** A. Employment/Salary (full or part-time):; Sanofi. **D. Ofengeim:** A. Employment/Salary (full or part-time):; Sanofi. **T. Hammond:** A. Employment/Salary (full or part-time):; Sanofi.

Poster

P296. Cellular Stress and Death Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P296.03

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: MH109382

Title: Dolutegravir-mediated integrated stress response activation induces neurotoxicity

Authors: *N. ALBOLOUSHI[1], C. AKAY-ESPINOZA[2], K. JORDAN-SCIUTTO[2];
[1]Doctor of Sci. in Dent. Candidate, Sch. of Dent. Med., [2]Dept. of Basic and Translational Sciences, Sch. of Dent. Med., Univ. of Pennsylvania, Philadelphia, PA

Abstract: *Keywords:* Human immunodeficiency virus; antiretroviral drug; integrated stress response; dolutegravir; neuron *Title:* Dolutegravir-mediated integrated stress response activation induces neurotoxicity *Background:* The incidence of human immunodeficiency virus (HIV)-associated dementia has been decreasing with the advent of antiretroviral therapy (ART) due to efficient systemic viral control; however, the prevalence remains high, partially due to increase life expectancy. Interestingly, about 50% of the HIV-infected patients have minor cognitive impairment, which led us to hypothesize that antiretroviral drugs (ARVs) induce integrated stress response (ISR), leading to neuronal dysfunction and damage, contributing to the changing clinical picture seen in HIV-positive individuals. *Methods:* We treated primary rat cortical neurons with the commonly prescribed integrase inhibitor dolutegravir (DTG) in therapeutically relevant doses and assessed whether ISR activation led to neuronal death. *Results:* We found that DTG led to neuronal death after 24 hours of treatment, as determined by MAP2 loss. In addition, we found that ISR was activated, as detected by the increased phosphorylation of eIF2alpha and ATF4 expression. Our data also suggest that pretreatment with the trans-ISRIB, which prevents eIF2alpha phosphorylation, might inhibit DTG-mediated neuronal death. *Conclusion:* Overall, understanding the underlying mechanisms of toxicity will lead to the development of new strategies to develop adjunctive therapies to complement ART while providing crucial information about potential neurotoxic side effects in the development of new ARVs.

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Poster

P296. Cellular Stress and Death Mechanisms

Location: Virtual Only

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Program #/Poster #: P296.04

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant 1R15NS112920-01A1

Department of Defense Grant W911NF2010275

Title: Inducible hypoxia tolerance is associated with downregulation of gene expression involved in Ca^{2+} homeostasis

Authors: *A. ALVAREZ[1], J. M. SANTIN[2];
[1]Univ. of North Carolina Greensboro, Greensboro, NC; [2]Univ. of North Carolina At Greensboro, Greensboro, NC

Abstract: Under hypoxic/ischemic conditions the brain fails to regulate ion homeostasis. Ion dysregulation in such pathologies leads to high levels of Ca^{2+} entering the cytosol, triggering neuronal dysfunction and cell death (termed "excitotoxicity). In health, various Ca^{2+} transporters and buffering mechanisms finely regulate Ca^{2+} entry and removal from the cytosol, while others buffer it by sequestering Ca^{2+} within organelles such as the endoplasmic reticulum (ER) or the mitochondria. Most vertebrates exposed to anoxic (no oxygen) conditions perish within minutes, and the model we use, the American Bullfrog (*Lithobates Catesbeianus*) is no different; after a few minutes of energy deprivation, rhythmic output from the brainstem respiratory network fails within a matter of minutes. However, after hibernation, when subjected to O_2 deprivation, the circuit becomes robustly tolerant and maintains steady rhythmic output for up to 4 hours, making a within-species model for inducible hypoxia tolerance. We hypothesized that one factor involves a coordinated modification of Ca^{2+} regulators such that transporters involved in removing and buffering Ca^{2+} are upregulated, while those that import it into the cytosol are downregulated, ultimately preventing Ca^{2+} overload. To test this hypothesis, we identified 16 Ca^{2+} pumps, exchangers, and channels in the bullfrog genome and designed quantitative PCR assays to determine if mRNA gene expression of each regulator correlated its role in an "anti-excitotoxic" phenotype (n=7 control, n=7 hibernation/hypoxia-tolerant). Surprisingly, we observed decreases in mRNA expression for 12/16 genes involved in Ca^{2+} homeostasis. The only downregulated targets consistent with protecting neurons from Ca^{2+} overload involved the IP_3 receptor and 4 non-selective cation channels (TRP channels). Therefore, our study suggests hypoxia tolerance corresponds with altered gene expression for Ca^{2+} homeostasis, specifically in plasma membrane and ER ion channels. Future studies are required to determine if these processes contribute to a large improvement in neuronal function during anoxia.

Disclosures: A. Alvarez: None. **J.M. Santin:** None.

Poster

P296. Cellular Stress and Death Mechanisms

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Program #/Poster #: P296.05

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant R03 AG063162
Alzheimer's Association, SAGA Grant
Garrison Family Foundation
McNair Scholars Program
Undergraduate Research Scholars Program at Texas Tech University

Title: Brain metabolic dysfunction in in vitro neurons

Authors: *L. BOLENG, N. SAWANT, S. KSHIRSAGAR, L. E. BUNQUIN, H. P. REDDY, A. P. REDDY;
Texas Tech. Univ., Lubbock, TX

Abstract: Metabolic dysfunction resulting from obesity and diabetes-induced inflammation is a significant risk factor in Alzheimer's disease (AD) and other dementias. Brain metabolic dysfunction affects multiple physiological pathways in the systemic and central nervous systems, adding to the complexity of AD and Alzheimer's disease related dementias (ADRD). The long-term effect of defective energy homeostasis is loss of synaptic transmission, hypometabolism, and neuronal death, and metabolic dysfunction indirectly affects neural resilience by increasing mitochondrial stress. Our research aims to investigate metabolic dysfunction in neurons that primarily contribute to dementia and comorbidities of obesity and depression. Metabolic dysfunction of serotonin in the neurons of three distinct brain areas contribute to depression, diabetes, and dementia—obesity: hypothalamus, depression: raphe, and dementia: hippocampus. The immortalized embryonic neuronal cell lines of the mouse-specific hypothalamus (mHypo), raphe (RN46A-B14), and hippocampus (HT22) express dopaminergic, serotonergic, and cholinergic neuronal characteristics, respectively, and have neurotransmitter expression. The aim of this research is to induce metabolic dysfunction by using high glucose and palmitic acid then treating with citalopram, a selective serotonin reuptake inhibitor, to assess improved energy homeostasis and cellular resilience. We propose that if treated with palmitic acid and high glucose, mHypo, RN46A-B14, and HT22 immortalized embryonic neurons will show a metabolic dysfunction phenotype, and citalopram treatment will improve cellular resilience. Cells were treated with 100 μM palmitic acid, 4% glucose, and citalopram for 24 hours. Control and treated cells were harvested and analyzed for mRNA levels of genes representing the preadipocyte to adipocyte transitions. We used qRTPCR based low-density array to analyze genes related to mitochondrial dysfunction, immune and cytokine function, serotonin pathways, synaptic signaling, and development. Immunocytochemistry and immunoblotting were conducted on adiponectin, IL6, and TNFα, PI3K, and Glut4. Results confirm that the immortalized mouse neurons show metabolic dysfunction and cellular susceptibility to mitochondrial stress. Together, these findings suggest that the use of multiple drug targets can combat comorbid diseases.

Disclosures: L. Boleng: None. **N. Sawant:** None. **S. Kshirsagar:** None. **L.E. Bunquin:** None. **H.P. Reddy:** None. **A.P. Reddy:** None.

Poster

P296. Cellular Stress and Death Mechanisms

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Program #/Poster #: P296.06

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant EY30747

Title: Differential expression of endocannabinoid signaling proteins in response to oxidative stress in human neural progenitor cell-derived neurons

Authors: R. S. DUNCAN, ***P. KOULEN**;
Univ. of Missouri Kansas City, Kansas City, MO

Abstract: OBJECTIVE: Determine which neuroprotective endocannabinoid signaling proteins are expressed by differentiated neural precursor cells (NPC) and how oxidative stress affects expression levels of these proteins.
RATIONALE: Rodent primary neurons and cancer cell lines have been used extensively to study neurodegeneration and identify potential neuroprotectants, including endocannabinoids. NPC-derived neurons are potentially an effective replacement for these traditional cell culture models to use in neuroprotection studies, but it is not clear whether they express proteins involved in neuroprotective endocannabinoid signaling.
HYPOTHESIS: We hypothesized that NPC-derived neurons express critical endocannabinoid signaling proteins implicated in neuroprotection and that their expression levels are altered in response to oxidative stress.
METHODS: Human NPC (MilliporeSigma, Burlington, MA) were grown in neural stem cell maintenance medium (MilliporeSigma) differentiated by withdrawing basic fibroblast growth factor (Gibco, Frederick, MD) and epidermal growth factor (Gibco) from the media for 7 - 10 days leading to cells that ceased dividing, exhibited well-defined neurites and displayed high levels of neural cell adhesion molecule (NCAM) and microtubule associated protein 2 (MAP2) immunoreactivity (IR). We exposed these differentiated neuronal cells to sub-lethal oxidative stress (addition of 2 or 10μM tert-butyl hydroperoxide to the culture medium for 24 hours). IR for endocannabinoid signaling proteins, cannabinoid receptor 1 (CB1), cannabinoid receptor 2 (CB2), fatty acid amide hydrolase (FAAH), *N*-acylethanolamine acid amidase (NAAA) and *N*-acyl-phosphatidylethanolamine-hydrolyzing phospholipase D (NAPE-PLD), was determined with immunocytochemistry and confocal microscopy imaging.
RESULTS: NPC-derived neuronal cells displayed IR for CB1, CB2, NAPE-PLD, FAAH and NAAA. Exposure to oxidative stress led to an increase in NAAA IR and a decrease in CB1, NAPE-PLD and FAAH IR, while oxidative stress had no effect on CB2 IR.
CONCLUSIONS: Critical endocannabinoid signaling proteins involved in neuroprotective signaling are expressed in NPC-derived neuronal cells, and their expression levels are modified differentially in response to oxidative stress.

Disclosures: R.S. Duncan: None. **P. Koulen:** None.

Poster

P296. Cellular Stress and Death Mechanisms

Location: Virtual Only

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Program #/Poster #: P296.07

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: the Brain Research Program of the National Research Foundation (NRF) funded by the Korean government (MSIT) (No. 2017M3C7A1048040)

Title: Asthma, Allergic Rhinitis, Nonsuicidal Self-Injury, and Suicide Attempts

Authors: *E. JO, G. KIM, J.-W. HUR;
Sch. of Psychology, Korea Univ., Seoul, Korea, Republic of

Abstract: A growing body of literature has shown that asthma and allergic rhinitis are associated with mental health problems such as suicidality and self-harm. However, to our knowledge, there are no studies to date that have examined the link between nonsuicidal self-injury (NSSI) and such diseases. In this study, we aimed to investigate whether and to what extent the diagnosis of asthma or allergic rhinitis was linked to the NSSI and suicide attempts. A total of 696 participants (age range = 19-29) enrolled in the study, including 317 in the NSSI with suicide attempts (SA) group, 240 in the NSSI without SA group, and 139 in the control group. We compared a medical history of asthma and allergic rhinitis, NSSI acts, suicide attempts, and demographic characteristics, including gender and socioeconomic status, among the groups. The associations between dysregulated immune systems, NSSI, and suicide attempts were estimated through odds ratios from multinomial logistic regression. Results showed that the NSSI without SA group had a significant association with a diagnosis of allergic rhinitis even after adjusting demographic variables. The findings also suggest that individuals diagnosed with asthma, allergic rhinitis, or asthma with allergic rhinitis had a greater risk of co-occurrence of NSSI and SA, respectively. However, only the effect of asthma with allergic rhinitis on NSSI with SA remained significant after adjustment. This study provides novel evidence for the relationship between NSSI and the disturbance in inflammatory conditions, including asthma and allergic rhinitis. Our findings also indicate a need for a closer examination of the history of inflammatory diseases required for individuals at high risk of self-harm.

Disclosures: E. Jo: None. **G. Kim:** None. **J. Hur:** None.

Poster

P296. Cellular Stress and Death Mechanisms

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Program #/Poster #: P296.08

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: National NeuroAIDS Tissue Consortium (NNTC) and CHARTER Neurobiobank
National Institute of Mental Health grant MH096625
National Institute of Neurological Disorders and Stroke NS105584

Title: Radiation and temozolomide induces tunneling nanotubes formation in glioblastoma tumors that promote tumor survival and adaptation to treatment

Authors: S. VALDEBENITO[1], K. P. L. BHAT[2], *E. EUGENIN[1];
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Abstract: Glioblastoma (GBM) is an aggressive and heterogeneous type of primary brain cancer with a poor prognosis (5 years of median survival) despite the advances in treatments. The only suggested treatment involves surgical resection, with subsequent radiation and temozolomide (TMZ) regiment, which results in an incremental increase in 5% survival. TMZ is a DNA alkylating, which triggers the mismatch repair system leading to a double-stranded break of the genome resulting in apoptosis. TNTs are minimally expressed under physiological conditions and only induced under cancer conditions, providing a unique therapeutic target to reduce the devastating consequences of GBM. Our preliminary data indicate that GBM tumors are highly communicated via TNTs, resulting in the spread of organelles and soluble factors between communicated cells. We detected that compromised mitochondria with deletions and mutations in the cytochrome B gene can be transferred. The transfer of mitochondria with mutated DNA enables non-tumor cells to become adapted to the tumor environment. Here we use primary glioma stem cells to study the transfer of mitochondria and tumor adaptation metabolism, using a co-culture model to examine TNT mediated transfer. We perform live cells imaging using mitotracker, immunofluorescence, Western blot, and Seahorse. Our results showed that the transfer of these mitochondria via TNTs enables cells to survive tumor growth, provide a better adaptation to environmental metabolic changes, and resist radiation and TMZ treatment. In conclusion, our data indicate that diffusion of key metabolites by TNT enables treatment-sensitive tumor cells to survive treatment and adapt non-tumor cells to the tumor microenvironment. Thus, blocking TNT communication induced by the tumor and treatment can provide an alternative mechanism to treat glioblastoma cells.

Disclosures: S. Valdebenito: None. **K.P.L. Bhat:** None. **E. Eugenin:** None.

Poster

P296. Cellular Stress and Death Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P296.09

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: CONACYT 287959

Title: Expression of pro-apoptotic proteins and Bcl-2 by exposure to ozone in rat´s cerebellum, frontal cortex, hippocampus and striate

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Abstract: Ozone (O_3) is one of the major air pollutants around the world, it is created by a photochemical reaction of volatile organic compounds, nitrogen oxides, and carbon monoxide that accumulates in the troposphere. According to the world health organization, O_3 concentrations above 100 µg/m3 (510 parts per million) with an exposure of 8 hours can be dangerous for humans causing moderate to severe effects on health. Several studies about the exposure to O_3 have been made in animal models, concluding to an increase in oxidative stress, resulting in lipoperoxidation, inflammation, metabolic disturbances, alteration in cell signaling, and the possible initiation of cell death in vulnerable areas such as the olfactory bulb, cerebellum, striatum, hippocampus and frontal cortex which are not fully described. The inflammatory response can represent an initial factor of cell death since a high concentration of TNF-α can activate caspase 3, which is one of the main effectors to bind substrates such as immune molecules that causes cell death. Apoptosis is a programmed cell death, which activates caspases leading to DNA fragmentation, forming and phagocyting apoptotic bodies with or without an inflammatory response. Apoptosis onset can occur through 2 pathways: intrinsic (mitochondrial) and extrinsic (death receptors), where multiple proteins interfere. Therefore, we aim to evaluate the participation of caspase 3 and caspase 8, Bcl-2, cytochrome C, VDA-C, TUNEL, and BAX proteins, in the initial damage resulting from ozone exposure in rat´s cerebellum, frontal cortex, hippocampus, and striatum. We used twenty male Wistar rats, who were randomized, in two groups, control and experimental equally divided. The experimental group was subjected to a hermetic ozone chamber for 12 hours provided with 1 ppm of O_3. Subsequently, they were immediately sacrificed to perform immunohistochemistry analysis. Compared to the control group, we found a decrease in Bcl-2, an anti-apoptotic protein, and an increase in TUNEL, caspase 3, 8, and BAX who trigger the cell death, this indicates that the two pathways (extrinsic and intrinsic) were activated. We also found a decrease in VDA-C and cytochrome C, this could be explained due to the release of cytochrome C. Also, cytochrome C is independent of its oxidation state and it becomes reduced in the cytosol. O3 induces cell death

through the extrinsic and intrinsic apoptotic pathways, this could explain some manifestations in the rats such as motor alteration, lack of memory, sleep disturbances, among others.

Disclosures: P. Rodríguez Quintero: None. **W. Moreno Roman:** None. **M. Rubio Osornio:** None. **R. Luna:** None. **L. Hernández López:** None. **J. Brito Brito:** None. **C. Rubio:** None.

Poster

P296. Cellular Stress and Death Mechanisms

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Program #/Poster #: P296.10

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

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NEI R01 EY022349
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Potoscnak Family-CSC Research Fund

Title: Neuronal Nrf2 modulates glaucoma pathogenesis onset and severity

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Abstract: NRF2, a transcription factor that modulates a cell's response to reactive oxygen species (ROS), has been implicated as a therapeutic target in models of neurodegeneration including glaucoma. We used the well-characterized microbead occlusion model of glaucoma (MOM) to elevate intraocular pressure (IOP). This model causes retinal ganglion cells (RGC) degeneration and death over time. We have previously demonstrated that there is an increase in ROS at 1- and 2- weeks post-IOP elevation in this model. We also detected an NRF2-mediated endogenous antioxidant response at 2-weeks post-IOP elevation. Notably, detectable pathology in the MOM does not occur until 4-weeks later. The goal of this study was to determine if NRF2 activation occurred within the RGCs. We intravitreally injected $Nrf2^{fl/fl}$ mice with an adeno-associated vector driving Cre from the mouse RGC-specific gamma synuclein (mSncg) promoter (AAV2/2.mSncg.Cre.IRES.TdTomato). We then elevated the IOP and assessed the endogenous antioxidant response at 2-weeks and pathology at 5-weeks post-IOP elevation. The mice with knock-down of Nrf2 in the RGCs had a greater increase in nitrotyrosine labeling in the RGC layer, a marker for ROS, compared to mice injected with the control vector, AAV2/2.eGFP. The

retinas from these mice had a 33% decrease in levels of PRDX6 and a 35% decrease in levels of SOD3 as compared to the AAV2/2.eGFP controls. These are two examples of the many antioxidant enzymes regulated by NRF2-dependent activation of the antioxidant response element (ARE). At 5 weeks post-IOP elevation, the RGC-specific Nrf2 knock-down mice had a 37% decrease in their photopic negative response (PhNR) amplitude and a 26% decrease in the visual evoked potential (VEP) amplitude in comparison to AAV2/2.eGFP injected controls. The PhNR measures RGC function and the VEP measures activity from the visual cortex *in vivo.* In a separate group of C57Bl/6 mice, we overexpressed Nrf2 in ganglion cell layer neurons by intravitreal injection of AAV2/2.CMV.Nrf2. Nrf2 overexpression resulted in lower superoxide levels, preservation of the PhNR amplitude, and decreased axon degeneration. Together, these data suggest that the endogenous Nrf2/ARE-mediated protective response is driven, at least in part, by RGC localized NRF2. Further, RGC-directed Nrf2 over-expression is a viable therapeutic approach for the treatment of glaucoma that should be explored further.

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Poster

P296. Cellular Stress and Death Mechanisms

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Program #/Poster #: P296.11

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: CONACYT Mexico Master's Degree Scholarship

Title: Obese Zucker rats express dendritic retraction in neurons of the hippocampal trisynaptic circuit and deficits in spatial memory

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Abstract: Obesity is a chronic disease characterized by excessive fat accumulation and is related to the appearance of other chronic diseases such as hypertension and type II diabetes mellitus. In recent years, it has been suggested that obesity may cause brain damage, and this relationship is being studied using animal models. The aim of the present study was to analyze the spatial memory capacity and the neuronal morphology of the hippocampal trisynaptic circuit in obese Zucker rats. We use male the Zucker rat that is an animal model of genetic obesity caused by a mutation in the leptin receptor. Five month-old animals were grouped into 2 sets of 16 rats each (8 male rats of the Long Evans strain as the control group and 8 male rats of the Zucker strain as the experimental group). We monitored the weekly body weight and morphometric parameters

of the rats and determined the glucose, cholesterol, and triglyceride serum concentrations. The first set of animals was subjected to a learning and spatial memory test in a Morris water maze to measure the following variables: escape latency, average speed, maximum speed, number of entries to the white quadrant, time spent in the white quadrant, latency to the first crossing to the white quadrant, and distance traveled. The second set of organisms was used to evaluate the morphology of CA1, CA3, and entorhinal cortex pyramidal neurons, as well as granular neurons of the dentate gyrus, where we obtained the parameters of dendritic arborization, length by branching order, and total length and density of dendritic spines using Golgi-Cox staining and Sholl analysis. Our results showed that Zucker rats exhibited clear signs of an obese phenotype and metabolic dysregulations related to increases in glucose, triglyceride, and cholesterol concentrations. In addition, deficiencies were observed in the learning test on the first two days of testing and a decrease in swimming speed. Furthermore, during the memory phase, the obese rats showed a lower number of entries to the quadrant containing the platform and traveled a shorter distance in the same quadrant. With respect to the neuronal morphology, we observed a decrease in dendritic arborization and dendritic length of hippocampal pyramidal neurons and granular neurons of the dentate gyrus, as well as a loss of dendritic spines in all neurons. These results indicate that obesity and its comorbidities cause dendritic retraction in the neurons that compose the trisynaptic circuit of the hippocampus. In conclusion, our work shows that there is an evident alteration in the neuroanatomy of the neurons that make up the trisynaptic circuit of the hippocampus, causing deficits in spatial memory capacity.

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Poster

P296. Cellular Stress and Death Mechanisms

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Title: Lead-induced hippocampal changes in BDNF and TrkB gene expression and neuronal survival

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Abstract: Lead, a non-biodegradable metal, is one of the most ubiquitous persistent toxicants present in the environment. Previous studies showed that the hippocampus was an important area where gene expression changes occur due to lead (Pb) exposure. These changes most pronounced during childhood and may negatively modify important neuronal pathways in the hippocampus, cortex, and thalamus implicated in synaptic plasticity, cell survival and expression. The brain-derived neurotrophic factor (BDNF), along with its receptor TrkB (tyrosine kinase) is a protein known for promoting the development and survival of nerve cells within the hippocampus by playing a role in the growth, differentiation, and maintenance of these cells. This is a known target of Pb contamination in CNS. In this study we address the transgenerational effect of a range of consumption; 0 or 27 ppm (low-dose mice equivalent concentration) to parents and 0 ppm or 27 ppm to offspring, of Pb on hippocampal neuronal survival and brain expression of relevant proteins in adolescent female and male mice. We hypothesize that changes in BDNF and TrkB gene expression are key in determining known changes in neuronal survival due to physiologically relevant concentrations of Pb exposure specifically in the hippocampus. Results have shown that lead exposure at 27 ppm during two weeks in adult mice decreases both BDNF and TrkB expression in the hippocampus. These results correlate with neuronal survival rates. Furthermore, mice exposed in utero also showed significant changes in both survival and BDNF/TrkB expression. This study will contribute to our understanding of the effects of physiologically relevant concentrations of Pd-exposure to the hippocampus regions in both adults and in early adolescence, after in-utero exposure.

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Poster

P296. Cellular Stress and Death Mechanisms

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Program #/Poster #: P296.13

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: R21NS081182
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U01NS103883

Title: Stau1 modulation of p53-mediated apoptosis

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Abstract: STAU1 is an RNA-binding protein that becomes highly overabundant in multiple neurological diseases, including amyotrophic lateral sclerosis (ALS), Parkinson's disease, Alzheimer's disease and spinocerebellar ataxia type 2 (SCA2) and in response to numerous pharmacological stressors [1, 2, 3]. Increased levels of STAU1 form cytoplasmic aggregates, impair the progression of autophagy, induce ER stress with activation of the integrated stress response (ISR) and cause apoptosis [1, 2]. The transcription factor p53 has a central role in cellular stress responses, including induction of apoptosis. In ALS, elevated levels of p53 are found in motor neurons in regions that are selectively vulnerable to the disease, and p53 has been shown to drive neurodegeneration caused by mutations in C9ORF72 associated with ALS. The objective of this study was to determine if STAU1 functions as a modulator of p53-mediated apoptosis and if targeting STAU1 could prevent apoptosis triggered by C9ORF72 mutations. We found that STAU1 modulates the p53 pathway at multiple levels. Elevation of STAU1 induced p53-mediated apoptosis and enhanced apoptosis triggered by various p53 activators, evidenced as increased levels of cleaved caspase 3 and of p53-upregulated modulator of apoptosis (PUMA) in HEK293 cells. Conversely, STAU1 knockdown prevented p53-dependent apoptosis triggered by a wide variety of stressors. STAU1 knockdown increased antiapoptotic factors and key DNA repair proteins. Additionally, RNA-seq of cells after STAU1 knockdown and hallmark pathway analysis showed p53 and apoptosis pathways were negatively enriched. To test whether STAU1 can prevent p53-dependent apoptosis triggered by C9ORF72 mutation, we knocked down STAU1 in human fibroblasts derived from a patient with C9ORF72 expansion. We found that this resulted in decreased levels of PUMA and cleaved caspase 3. Our results suggest that STAU1 may be a novel target to prevent neurodegeneration in ALS and other neurological diseases where p53 has a role in neuronal death.
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Poster

P296. Cellular Stress and Death Mechanisms

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Program #/Poster #: P296.14

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Title: Oxidative stress induces Progranulin expression to control neurite outgrowth in HT22 cells

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Abstract: Progranulin (PGRN) is a multifunctional growth factor abundantly expressed in central nervous system. When heterozygous mutations occur in the *Pgrn* gene, it will cause

PGRN haploinsufficiency. PGRN haploinsufficiency has been reported for the association with frontotemporal lobar degeneration (FTLD), but it's mechanism(s) has not been clarified, so it is crucial to understand the physiological and pathophysiological roles of PGRN. Moreover, although PGRN expression is regulated by various stressors (e.g., oxidative stressors), its regulatory mechanism(s) has not been completely clarified. In this study, we have investigated physiological roles of the oxidative-stress induced PGRN expression in HT22 murine hippocampal cells. In addition, we also analyzed the molecular mechanism(s) of how the oxidative stressors control PGRN expression. First, we investigated the oxidative stress-dependent response of HT22 cells (e.g., neurite length, number, and cell body size) and found that lengths and numbers of neurites are significantly increased by oxidative stress. *Pgrn* knockdown (KD) experiments suggested that at least partially of these changes were mediated by oxidative stress-induced PGRN expression. Interestingly, the addition of exogenous recombinant PGRN did not alter the length and number of neurites, which suggested that the increase in neurite length and numbers induced by oxidative stress was mainly mediated by the intracellular PGRN, not by the secreted PGRN. We then investigated signaling pathway(s) responsible for PGRN induction by oxidative stress in HT22 cells. We observed the activation of Erk1/2, JNK and p38 MAP kinase by adding oxidative stressors, and SB203580, a potent and selective inhibitor of p38 MAP kinase, attenuated PGRN induction by oxidative stress. On the other hand, significant effects of U0126 (an inhibitor of MEK1 and MEK2) and SP600125 (an inhibitor of JNK) were not observed. Overall, these results suggested that oxidative stress on HT22 cells induce PGRN via p38 MAP kinase pathway, thereby controlling neurite outgrowth of the cells. Importantly, the intracellular PGRN accumulation by oxidative stress appears to be crucial for neurite outgrowth at least in HT22 cells. Further studies will reveal the physiological differences between the intracellular PGRN and secreted PGRN, which may be an useful information to understand the progression of FTLD.

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Poster

P296. Cellular Stress and Death Mechanisms

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Program #/Poster #: P296.15

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NSF Grant 1818140

Title: C. elegans as tool to study chronic stress effects

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Abstract: Chronic stress is one of the main risk factors for depression and other neuropsychiatric diseases. Long exposure to chronic stress results in changes in brain gene expression that are deleterious for organisms. Due its relevance to human health, our goal is to investigate the molecular pathways disrupted by chronic stress to understand how this leads to such diseases. Using rodent models of chronic stress, we found that stress alters neuronal protein GPM6A. GPM6A participates in neuronal differentiation and morphology establishment and human *GPM6A* has been linked to schizophrenia, bipolar disorder, claustrophobia and suicide patients. This links GPM6A to the stress phenomenon and depression. Nevertheless, there is a gap between the cellular GPM6A functions and its role in systemic stress response. To fill this gap, we use the nematode *Caenorhabditis elegans* as model due to shared features between nematode and mammal nervous system and because of the genetic tools available. *C. elegans* exhibits a GPM6A ortholog, the neuronal membrane glycoprotein 1 (NMGP-1), thus, here we used *C. elegans* as a simpler model to study NMGP-1 participation in stress response. First, worms expressing GFP under the *nmgp-1* promoter indicated us that *nmgp-1* expresses in sensory amphid and phasmid neurons and in the egg-laying apparatus. Second, we have characterized NMGP-1 functions using RNAi knockdown and two non-null, hypomorphic mutant alleles. Analysis of dsRNA (*nmgp-1)*-treated or mutant alleles showed an increased recovering time from the stress-resistant dauer stage and a reduced egg-laying rate with respect to control worms. In addition, defects in egg-laying induced egg retention (bag of worms) in *nmgp-1*-deficient worms. Also, worms lacking NMGP-1 showed a normal response to the attractant diacetyl, but an altered repulsive response to SDS. Moreover, morphologically, *nmgp-1*(RNAi) worms showed alterations on ASJ chemosensory neurons located at the nerve ring, responsible of dauer exit. Altogether these results suggest that NMGP-1 is involved in the stress response in *C. elegans*. To move forward, we will present and discuss a battery of experiments to score stress response: Temperature acute and chronic exposure, oxidative and osmotic stress. The characterization of stress response in worms lacking *nmgp-1* will allow us to deepen on the stress molecular bases and mental diseases.

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Poster

P302. Neuroinflammation: Animal Models II

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Program #/Poster #: P302.01

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: General Research Fund (17123217)

Title: Investigating the significant roles of PKR on systemic inflammation triggered neuroinflammation, tau phosphorylation and cognitive dysfunctions

Authors: *W. Y. CHENG[1], Y. S. HO[3], R. C. C. CHANG[1,2];
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Abstract: Increasing lines of evidence have shown that systemic inflammation may contribute to neuroinflammation and accelerate the progression of neurodegenerative diseases. Double-stranded RNA-dependent protein kinase (PKR) is one of the key signaling molecules regulating immune responses by regulating macrophage activation, different inflammatory pathways, and inflammasome formation. In this study, we aim at investigating whether PKR can be a pharmacological target to prevent both systemic inflammation-triggered neuroinflammation, tau phosphorylation and cognitive dysfunctions. Laparotomy, a non-bacterial endotoxin mouse model, is adopted to address the systemic inflammation triggered by surgery.
We have shown that laparotomy induced systemic inflammation and neuroinflammation in male C57BL/6J mice. At 4 h after laparotomy, a significant increase in interleukin-1β (IL-1β) and MCP-1 was detected in the liver of the laparotomy group compared with the control group. The expression of IL-1β was also significantly upregulated in the hippocampus after laparotomy. On POD 1, no significant differences in cytokine expression were observed in the liver. This suggests that the induced systemic inflammation is transient. Meanwhile, the level of IL-1β remained high in the hippocampus after surgery. Second, on POD 14, for the laparotomy group, a significant increase in tau protein phosphorylation (AT8 and pS416) was observed in the frontal cortex.By immunohistochemical analysis and flow cytometry, an increased number of CD68-eGFP-positive cells was observed in the laparotomy group compared with the control, implying macrophage infiltration or an increased number of activated microglia.Moreover, cognitive impairment was observed after laparotomy in the form of reduced recognition memory with lower discrimination index in the NOR test. By genetic knockout of PKR in mice, it potently downregulated the increased IL-1 expression induced by laparotomy in both the liver and frontal cortex. Meanwhile, a significant reduction in tau protein phosphorylation (AT8 and pS416) was observed in frontal cortex of PKR-KO mice. Furthermore, recognition memory remained intact in PKR-KO mice after laparotomy.
This study reveals that genetic knockout of PKR in mice could attenuate laparotomy induced systemic inflammation, neuroinflammation, abnormal tau phosphorylation and cognitive impairment.Further studies will be performed to examine the effect of knockout of PKR in macrophages and cholinergic neurons on systemic inflammation, neuroinflammationand cognitive functions.
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Disclosures: W.Y. Cheng: None. **Y.S. Ho:** None. **R.C.C. Chang:** None.

Poster

P302. Neuroinflammation: Animal Models II

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Support: National Multiple Sclerosis Society RG 4587-A-1
National Science Foundation 1648822
National Eye Institute RO1EY025687
National Eye Institute R01EY032342
Cole Eye Institute Vision Science Training Program 5T32EY024236

Title: Axonal injury in central nervous system white matter precedes retrograde neuronal degeneration during experimental autoimmune encephalomyelitis

Authors: *G. M. MEY[1], K. S. EVONUK[1], L. M. WOLFE[1], M. CHAPPELL[1], R. SINGH[2], J. C. BATOKI[2], M. YU[2], B. ANAND-APTE[2], N. S. PEACHEY[2], T. M. DESILVA[1];
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Abstract: Background: Multiple sclerosis (MS) is a chronic demyelinating disease of the central nervous system (CNS) that eventually results in permanent neuronal loss. Immunomodulatory therapies can reduce relapses, but there are no effective indicators or treatments for the degenerative phase of MS. Evidence of visual system impairments have been shown in MS, presenting a unique system to monitor disease. The pathology underlying visual assessments and how it relates to pathology in the rest of the CNS, however, remains ill-defined.
Objectives: The purpose of this study was to investigate the spatiotemporal relationship between demyelination/axonal injury and neurodegeneration in the visual system and spinal cord during experimental autoimmune encephalomyelitis (EAE).
Methods: EAE was induced in adult male C57BL6J mice using MOG_{35-55} peptide. Histopathological analyses were coupled with visual assessments including optical coherence tomography (OCT) to track retinal thickness and visually evoked potentials (VEP) to test the neuronal response to visual stimuli. Assessments were performed at 15, 35, and 60 days post-EAE induction (dpi) compared to naïve age-matched controls. OCT images and VEP recordings were collected at baseline, 15 dpi, and 35 dpi.
Results: Our data show that $CD3^+$ T cells robustly infiltrate both the spinal cord and optic nerve white matter at 15 dpi. Concurrently, demyelination and axonal loss and reactive gliosis were observed in both of these regions and sustained at 35 dpi. Loss of $NeuN^+$ neurons in the ventral horn of the spinal cord and $Brn3a^+$ retinal ganglion cells (RGCs) in the retina, however, was not observed until 35 dpi. No neuronal loss was observed in the ventral posterolateral nucleus (VPL) nor in the lateral geniculate nucleus (dLGN) of the thalamus at any time point during EAE, but synaptic loss was observed in the dLGN beginning at 15 dpi. Consistent with RGC loss, a

significant reduction in retinal nerve fiber layer thickness shown by OCT was observed at 35 dpi. Functional visual deficits began at 15 dpi with delayed VEPs, followed by a decrease in VEP amplitude at 35 dpi.
Conclusions: Our data suggest a retrograde loss of neuronal cell bodies following demyelination and injury of their respective axons. Neuronal loss was not observed anterograde to axonal injury, further supporting a prominent retrograde neurodegeneration. Our results also indicate that the visual system presents a similar time course of EAE pathology to that in the spinal cord. Along with OCT imaging showing loss of RGCs in chronic EAE, our data suggest that the visual system may be an important marker of neurodegeneration in the CNS.

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Poster

P302. Neuroinflammation: Animal Models II

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Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

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VA Merit Award I01CX001923-01 awarded to S. Chatterjee

Title: TRPV4 deletion increases neuroinflammation and neurodegeneration in murine model of chronic multi-symptom illness-associated with gulf war veterans

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Abstract: Chronic multi-symptom Illness (CMI) is a condition affecting war veterans like Gulf War (GW) veterans and elderly. Neurological symptoms, memory loss and cognitive dysfunction are among the major symptoms reported in CMI. However, the pathophysiology of CMI remains elusive. Our previous studies in GW mouse models indicated increased neuroinflammation and neurodegeneration. Recently, it was reported that transient receptor potential vanilloid 4 (TRPV4) an osmo-mechano sensitive cation channel regulates the blood brain barrier (BBB) and is associated with neuroinflammation. Our lab showed previously that TRPV4 deletion increased

CYP2E1 mediated toxicity. In this study we exposed GW chemicals pyridostigmine bromide (2mg/kg) and permethrin (200mg/kg) in adult C57BL/6J (n=6) and TRPV4 knockout (TRPV4KO) (n=6) mice for 15 days to study if deletion of TRPV4 modulated GW chemical exposure-associated brain pathology. Compromised BBB was observed by a decreased expression of tight junction protein Claudin 5 and increase in matrix metalloproteinase 9 in GW chemical exposed TRPVKO mice over wild type mice. GW chemical exposure significantly increased the expression of proinflammatory cytokine interleukin 1β and 6 in the frontal cortex of TRPV4KO mice compared to wild type mice. Increased expression of translocator protein (TSPO), a mitochondrial membrane protein also indicated neuroinflammation with microglial activation in TRPV4KO mice exposed to GW chemicals. Furthermore, increased expression of cytochrome P4502E1 and lipid peroxidation stable adduct 4-hydoxynonenal indicated heightened oxidative stress in GW chemical exposed TRPV4KO mice. Administration of GW chemicals increased neurodegeneration in TRPV4KO mice as seen by an increase in Tau level and decreased expression of brain-derived neurotrophic factor. Hence, deletion of TRPV4 exacerbated GW chemical associated neuroinflammation and neurodegeneration suggesting TRPV4 as hitherto underappreciated therapeutic target for CMI.

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Poster

P302. Neuroinflammation: Animal Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P302.04

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: the Basic Science Research Program (2019R1I1A2A01060115) through the National research Foundation of Korea (NRF) funded by the Ministry of Science, ICT & Future Planning
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Title: Effect of GR 82334, neurokinin 1 (NK1) receptor antagonist, on the induction of osteoarthritic pain in rat models with MIA-induced osteoarthritis (OA)

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Abstract: Osteoarthritis (OA) is a degenerative disease leading to functional deterioration in joint and surrounding tissue and inducing chronic pain of joint which make patients uncomfortable in their life. Substance P, one of neuropeptides, induces secretion of inflammation mediated-cytokines followed by vasodilation and edema in tissue and transmit pain signal to PNS and CNS in pathologic environment of OA. The aim of this study is to investigate the role of NK1 receptor on the development of chronic pain following OA, using GR 82334, the NK1 receptor antagonist, in rat with MIA-induced OA. OA was induced by injecting monosodium iodoacetate (MIA; 4 mg/50 µl) into the right knee joint of rats. GR 82334 10 µM diluted with 30 µl of saline were injected by single intra-articular injection with 30 gauge insulin syringe, to animals of pre administration group 30 minutes before MIA injection and to animals of post administration group 1 day after MIA injection. Any administration was not performed to the MIA group after MIA-induced OA. Paw withdrawal threshold (PWT), knee joint diameter, knee bending test, and weight bearing test were performed before and after MIA injection. Histological change of knee joint was evaluated by safranin O fast green (SOFG) staining and immunohistochemistry was used to quantify the expression of SP and calcitonin gene related-peptide (CGRP) in L3-5 of spinal cord. Knee bending score and knee joint diameter in the both pre- and post-GR 82334 group were smaller than those of MIA group at 3 days after MIA injection. At 10 and 14 days after MIA injection, PWT in the post-GR 82334 group was significantly higher compared to the pre-GR 82334 and MIA group. Also, the expression of SP and CGRP at spinal cord significantly less in post administration group than pre administration and MIA group at day 14. There were no significant differences in values of PWT among three groups at 28 days after MIA injection. The present data demonstrated that NK1 receptor antagonist is effective for inhibiting inflammation and induction of pain following OA in MIA-induced OA rat models. These results suggest that SP-NK1 signal might contribute in pathophysiology of OA from acute inflammation to central sensitization inducing chronic pain and be a key target for fundamental therapeutic approach to treat OA.

Disclosures: M. Kwon: None. **H. Yoo:** None. **T. Kim:** None. **D. Nam:** None. **J. Kim:** None.

Poster

P302. Neuroinflammation: Animal Models II

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Program #/Poster #: P302.05

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: Tisch MS Research Center of New York (Private Funds)

Title: Antibodies in primary progressive multiple sclerosis CSF impair remyelination of lysolecithin-induced lesions

Authors: N. M. CELESTIN, *J. K. WONG, S. A. SADIQ;
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Abstract: Multiple sclerosis (MS) is characterized by inflammatory demyelination, astrogliosis and axonal loss in the CNS. Most patients present with relapsing-remitting MS (RRMS) then eventually enter a phase of disease progression termed secondary progressive MS (SPMS). However, approximately 10-15% of patients diagnosed with primary progressive multiple sclerosis (PPMS) experience unremitting disease progression from disease onset. This progressive clinical course of PPMS suggests that remyelination is impaired in contrast to relapsing-remitting forms of the disease. We previously reported that intrathecal injection of cerebrospinal fluid (CSF) from PPMS patients, but not RRMS or SPMS patients, delayed spontaneous remyelination in lysolecithin-induced lesions. However, the PPMS CSF factors contributing to impaired remyelination have yet to be identified. Here, we investigated whether antibodies present in PPMS CSF are responsible for delayed remyelination of lysolecithin-induced lesions. CSF obtained from untreated PPMS patients was passed through a tangential flow filtration system to remove components larger than 100 kDa (including antibodies). Adult C57BL/6 female mice underwent a laminectomy at cervical level 5 (C5) and 1µl of 1% lysolecithin was injected into the dorsal column. At 5 days post lysolecithin injection, mice received 3µl injections into the C5 subarachnoid space of either: 1) CSF from PPMS patients, 2) filtered PPMS CSF, or 3) PPMS or RRMS recombinant antibodies (rAbs) produced after FACS sorting B-cells in CSF then DNA sequencing IgG variable regions. Mice were perfused 12 days after lysolecithin injection. Spinal cords were post-fixed overnight in 4% paraformaldehyde, cryoprotected in 30% sucrose, then cryosectioned for histological analyses. Luxol fast blue staining revealed significantly larger lesion volumes in PPMS CSF-injected mice compared to mice injected with filtered PPMS CSF, suggesting that the factor in PPMS CSF responsible for impeding remyelination is larger than 100 kDa. Preliminary data suggest that mice injected with PPMS rAbs also have larger volumes of demyelination than mice injected with RRMS rAbs, implicating a role of antibodies in PPMS CSF in delaying remyelination. Together, these data suggest therapeutic strategies that reduce or remove CSF antibodies may ameliorate disease progression in PPMS.

Disclosures: N.M. Celestin: None. **J.K. Wong:** None. **S.A. Sadiq:** None.

Poster

P302. Neuroinflammation: Animal Models II

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Program #/Poster #: P302.06

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIAID Grant AI097299-01

NINDS Grant NS104351

Title: The Role of Sympathetic Neuronal Pathways in Regulating HSV1 and HSV2 Genital Infection

Authors: *G. A. MOORE, S. LEE, A. A. N. ABBOTT, A. M. IVES, A. S. BERTKE; Virginia Tech., Blacksburg, VA

Abstract: Herpes simplex virus 1 (HSV1) and HSV2 are known to establish latency and reactivate in sensory neurons after orofacial or genital infection. However, the viruses are also found in autonomic ganglia, including both sympathetic and parasympathetic branches of the autonomic nervous system (ANS) in humans and in animal models. HSV1 shows a preference for sympathetic pathways and HSV2 shows preference for parasympathetic pathways, suggesting that autonomic pathways may provide an alternative reservoir for reactivating virus that may contribute to different recurrence frequencies of HSV1 and HSV2. To determine the contribution of sympathetic pathways to acute genital disease and recurrence frequency, guinea pigs were treated with 6-hydroxydopamine (6-OHDA) prior to infection to ablate sympathetic neuronal axons. Chemical ablation of sympathetic axons using 6-OHDA allows selective removal of sympathetic neurons, making them unavailable for infection and the establishment of latency, resulting in significantly reduced severity and neurological involvement during acute disease for HSV1, but not HSV2. Animals treated with 6-OHDA prior to infection also had reduced clinical recurrences by nearly 75% for HSV1 and by 50% for HSV2 ($p \leq 0.001$). Furthermore, viral shedding in genital secretions was significantly reduced, as determined through the analysis of daily vaginal swabs. Thus, sympathetic neurons and pathways play a significant role in acute disease severity and neurological involvement of HSV1, but not HSV2. Additionally, sympathetic pathways are responsible for a significant portion of HSV1 and HSV2 recurrences, with a greater impact on HSV1 than HSV2.

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Poster

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Program #/Poster #: P302.07

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NASA grant #NNX15AJ20H

Title: Understanding the Impact of Chronic, Low-Dose GCR Particles on Behavior and Systemic Inflammation in Mice

Authors: *C. LAFRENIER[1], P. FRIEND[1], B. GLEESON[1], P. NOLTE[1], F. RULISON[2], P. DEYOUNG[2], P. RIVERA[1];
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Abstract: A major component of NASA's 2018 strategic plan was to send astronauts to and beyond our lunar orbit within the next couple of decades. A potential risk to mission success is an astronauts' exposure to galactic cosmic radiation (GCR), a mixture of low-dose, high-charge ion particles (HZE). However, studies examining the impact of low-energy GCR particles on long-term mission success are scarce. Therefore, our first study examined the effects of chronic, low-dose proton radiation on behavior and systemic inflammation in mice. We tested the hypothesis that chronic low-dose proton radiation negatively impacts mouse behavior by causing lasting systemic inflammation. Mice were divided into three treatment groups: Irradiation (IRR), No-Irradiation/Constraint (No-IRR), and Home Cage (HC). The irradiation group of mice received approximately 0.17mGy/day of proton radiation on 13 non-consecutive days over a period of 50 days. Ten weeks after irradiation, all mice underwent anxiety- and depressive-like behavioral tests. Then, mice were euthanized and brain, liver, and spleen samples were extracted for molecular analysis. Results suggest that mice irradiated with a cumulative dose of ~2.5 mGy of protons exhibit an increase in overall activity and compulsive-like behavior. Our secondary study examined the impact of chronic low-dose neutrons on sociability and cognitive learning. Mice were irradiated at a dose of 0.005 mGy 33 times over 70 days, resulting in an expected cumulative dose of approximately 0.165 mGy. We expect to see a differential effect of neutron radiation on sociability behaviors and cognitive spatial learning in a sex-dependent manner. We suspect that these behavioral differences will be the result of systemic inflammation. Taken together, proton and neutron chronic low dose radiation may negatively impact long-term mission success to Mars.

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Poster

P302. Neuroinflammation: Animal Models II

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Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P302.08

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Title: In vivo neuroinflammation modelling by acute, sub-chronic and chronic LPS-administration, and in transgenic APP, APPxPS1, TAU and APPxTAU mouse models of Alzheimer's disease

Authors: *A. TANGHE, **T. CORNELISSEN**, W. DEJONCKHEERE, S. CARMANS;
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Abstract: Compelling evidence indicates that the etiology of Alzheimer's disease (AD) is multifactorial, with neuroinflammation, and its capacity to exacerbate Aβ and Tau pathology, taking a center stage in the AD pathogenesis (see e.g., Onyango *et al.*, 2021). Albeit none of the currently available AD animal models are a faithful reproduction of the human disease, they have been and will remain essential tools to improve our understanding of AD biology and to evaluate specific therapeutic approaches, including the role of neuroinflammation and the potential of anti-inflammatory therapies (see e.g., Saito & Saido, 2018). In mice, systemic inflammation and neuroinflammation ensuing peripheral or central injections with the pro-inflammatory component lipopolysaccharide (LPS), have shown to evoke an elevation in Aβ levels, in Tau hyperphosphorylation and tangle formation, and in neuronal cell death, finally resulting in cognitive impairment (Zhao *et al.*, 2019; Batista *et al.*, 2019). Encouraged by the robust yet transient effects obtained in acute and sub-chronic intraperitoneal LPS-administration regimens, we set out to develop a chronic LPS-induced neuroinflammation mouse model both in wild type and Tau-transgenic mice, with read-outs of AT8-responsive Tau and CD68-responsive phagocytic microglia. Optimization of the LPS-concentration, the number of administrations and the time of sacrifice after the last dosing, provided insights in the transient or sustained character of the effects, assessed via the abovementioned read-outs as well as via TNF alpha-levels in serum and a qPCR-based panel of inflammasome markers. We compared the data of the acute, sub-chronic and chronic LPS-induced neuroinflammation models with the neuroinflammation hallmarks in a series of related AD transgenic models (APP-London, APP-London x PS1, TauP301S, and APP-London x TauP301S) and discuss the discriminative and complementary value of the altogether of these models for the assessment of therapeutic interventions. In the APP-London x TauP301S transgenic, we confirmed the previously postulated temporal development and spatial co-localization of amyloid, Tau and neuroinflammation pathology. As a next step, we are extending the above set of models and concomitant neuroinflammation read-outs to a second-generation APP knock-in model. Humanizing animal models at the level of microglia, to bridge the species barrier upfront, is another exciting avenue that we are exploring. This will open a whole new array of questions that can be more credibly posed already in preclinical stage.

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Poster

P302. Neuroinflammation: Animal Models II

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Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: PICT 2016-3642

01/Q666 (20020130100666BA; Universidad de Buenos Aires Ciencia y Tecnología [UBACYT]) from University of Buenos Aires (to O.D.U.)

Title: Expression of ASIC1 channels throughout the pain pathway in the CNS in the formalin acute pain mouse model

Authors: M. N. GOBETTO, N. E. CONTRERAS, L. C. SALINAS CASTELLANOS, **O. D. UCHITEL**, *C. WEISSMANN;
IFIBYNE-CONICET, Buenos Aires, Argentina

Abstract: Hindpaw injection of formalin in rodents is used to assess acute intense, short-lasting (minutes to tens of minutes) persistent pain. The response to formalin is biphasic, with an initial response in the first minutes, and a later response around 40min after the injection with an increase in licking of the paw. The initial response is thought to be contributed by inflammatory, peripheral mechanisms, while the latter is linked to central mechanisms. This model has been especially useful to analyze the effect of drugs at either phase. Different channels have been shown to be involved in these responses according to specific drugs used. Acid-sensing ion channels (ASICs) regulate synaptic activities and play important roles in neurodegenerative diseases as well as in pain conditions. Psalmotoxin-1 (Pctx-1), a toxin that inhibits ASIC1a-constituted channels, as well as antisense ASIC1a RNA, injected in mice before the formalin test, have been previously shown to affect both phases (Mazzuca et al. 2007). However, whether ASIC1 protein levels are affected remains controversial, as most work focused on mRNA levels as protein levels proved difficult to detect. We decided to analyze ASIC1a protein levels at different levels in the pain pathway centrally, in regions in which ASIC1a channels are enriched (spinal cord, SC, and the anterior cingulate cortex, ACC, of the brain). In addition, we subdivided the areas into different lumbar (L) segments (SC at L3, L4 and L5 levels) as well as ipsilateral and contralateral to the injection for ACC-to avoid the dilution of any contributing effect- in formalin and PBS (control) injected mice. Our results showed the biphasic behavior of mice to formalin. This behavior was accompanied by an increase in ASIC1 levels at the ipsilateral ACC compared to the contralateral ACC and greater than in ACC of animals injected with PBS. At the spinal cord, the effect of ASIC1 levels follows a gradient stronger at L3 levels and decreasing towards L5 in formalin-injected mice. This work highlights the important role ASIC1 channels play in pain and the potential role of pharmacological therapies aiming at this channel.

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Poster

P302. Neuroinflammation: Animal Models II

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Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P302.10

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Title: An experimental animal model of disseminated cerebral aspergillosis

Authors: *B. N. SULLIVAN[1], C. GUILLORY[2], M. A. BAGGETT[2], M. JONES[3], C. STEELE[3]; [1]Neurosci. Grad. Program, [2]Neurosci., [3]Microbiology & Immunol., Tulane Univ., New Orleans, LA

Abstract: Invasive pulmonary aspergillosis (IPA), a leading cause of death in immunosuppressed patients, is an infection most often attributed to the opportunistic fungi *Aspergillus fumigatus* (AF). The most common site of primary infection with this invasive disease is the lungs, and the most prevalent site of secondary infection following hematological dissemination is the central nervous system (CNS). Dissemination of AF to the CNS, termed cerebral aspergillosis (CA), occurs in up to 40% of immunocompromised patients with IPA and has a mortality rate approaching 90%. While the experimental murine model of IPA and the resulting immune response have been extensively characterized, the current models available for CA are inconsistent and physiologically irrelevant. This lack of relevant models results in a considerable gap in knowledge for the neurological pathogenicity of infection. To address this gap, we infected wild-type male and female C57BL/6 immunocompetent, or immunosuppressed (via high-dose corticosteroid) mice with AF intranasally. Following immunosuppression and infection, mice were monitored for severe weight loss (>20%), neurological symptoms (e.g., altered-gate, severe head-tilt, and/or spinning in cage), and mortality daily over 8 to 10 days. Brains were assessed for fungal burden at the time of death, study end, or at a specific time point (3 days post-infection (PI)). Predictably, primary pulmonary infection occurred in immunocompetent and immunosuppressed mice. However, encouragingly, significant weight loss, neurological symptoms, and fungal burden in the brain were found exclusively in immunosuppressed mice, occurring in as little as 3 days PI. Further, in the immunosuppressed infected group, we observed a significant correlation between weight loss and neurological symptoms indicating the successful induction of severe disease. Interestingly, like immunosuppressed humans with IPA, cerebral dissemination, defined by detection of AF in the brain, was observed in approximately 50% of immunosuppressed mice. In conclusion, we have developed a well-defined, physiologically relevant, model of disseminated CA in immunosuppressed mice with primary pulmonary infection. This model will serve to advance understanding of the disease mechanism, identify immunopathogenic processes, and aid in defining the neuroinflammatory response to CA.

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Poster

P317. Pre-Clinical Therapeutic Strategies

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Program #/Poster #: P317.01

Topic: C.10. Brain Injury and Trauma

Support: R01CA194924
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Title: Circadian influences on chemotherapy efficacy in a mouse model of brain metastases of breast cancer

Authors: *W. H. WALKER, II[1], S. A. SPROWLS[2], J. R. BUMGARNER[1], J. A. LIU[1], O. H. MELÉNDEZ-FERNÁNDEZ[1], J. C. WALTON[1], P. R. LOCKMAN[2], A. C. DEVRIES[3], R. J. NELSON[1];
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Abstract: Chemotherapy is more effective in the treatment of peripheral tumors than brain metastases, likely reflecting the reduced ability of chemotherapy to cross the blood brain barrier (BBB) and/or blood tumor barrier (BTB) at efficacious concentrations. Recent studies demonstrate circadian regulation of the BBB. Thus, we sought to determine whether BBB/BTB permeability to chemotherapeutic agents displays circadian regulation. We predicted that optimally timed chemotherapy would increase anti-tumor efficacy in a murine model of brain metastases of breast cancer (BMBC). Female NU/NU mice received a 1oo µl intracardiac injection of the human brain seeking Her2+ breast carcinoma cell line, JIMT-1BR3-Luc ($1.75x10^5$ cells). First, we characterized novel daily alterations in BBB permeability to a commonly used chemotherapeutic, ^{14}C-paclitaxel, within BMBC following injections given at four time points across the day. Peak and trough ^{14}C-paclitaxel concentrations within BMBC occurred during the mid-dark phase and at the beginning of the light phase, respectively. Next, we assessed the functional significance of increased ^{14}C-paclitaxel concentrations within BMBC by monitoring the onset of neurological symptoms and cell death within BMBC. Mice received a 1oo µl intracardiac injection of the cell lines, JIMT-1BR3-Luc ($1.75x10^5$ cells) or 4T1-BR5-Luc ($1.75x10^5$ cells) . Mice were randomly assigned to treatment groups, ZT0 or ZT17, and received once a week injections (2 weeks total) of paclitaxel (13 mg/kg) corresponding to either the peak or trough of BBB/BTB permeability (determined in Exp.1). Mice that received chemotherapy injections during the dark phase demonstrated increased cell death within BMBC and delayed onset of neurological symptoms relative to mice that received injections during the light phase. Together, these data provide strong evidence that chrono-chemotherapy is a novel and potentially effective treatment strategy for BMBC, and emphasizes the importance of time of day when administering chemotherapy to patients.

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Poster

P317. Pre-Clinical Therapeutic Strategies

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Program #/Poster #: P317.02

Topic: C.10. Brain Injury and Trauma

Support: PUNQ 1397/16
CONICET, PICT 2099-2014

Title: Is time the answer? Chonopharmacological approach of the novel drug 1A116 for glioblastoma treatement

Authors: *L. L. TREBUCQ[1], G. A. CARDAMA[2], P. LORENZANO MENNA[2], D. A. GOLOMBEK[1], L. MARPEGAN[3], J. CHIESA[1];
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Abstract: Glioblastomas (GBM) account for almost half of the malignant central nervous system tumors. Current therapy for GBM is generally very aggressive, combining chemotherapy, radiotherapy, and surgical removal and increased the life expectancy only from 11 to 14 months, but almost no significant advances were made in the last decades. Taking this into account, more effective compounds and new strategies need to be developed. The novel drug 1A-116 specifically blocks the interaction of the Rho GTPase Rac1 with some of its activators (GEFs proteins, such as Tiam1) inhibiting cell motility and proliferation of GBM. One strategy is to find a circadian time of maximal effectiveness and minimum toxicity (i.e., chronopharmacology). Thus, we studied the therapeutic response to 1A-116 at different circadian times, of LN229 glioblastoma cell line in vitro, and in xenogenic implants in nude mice. LN229 wild type and bmal1 knock down cell lines, obtained by Crispr/Cas9, were synchronized with serum shock (50% serum) and transfected with Bmal-luc plasmid for testing circadian rhythmicity. Cell lines treated at different circadian times with 1A-116 were assessed for proliferation, migration and apoptosis, and circadian oscillation of BMAL1 and TIAM1 mRNA and proteins was studied by qPCR and in cell western. In vivo studies were performed using xenogenic LN229 striatum implants in nude male mice (n=11), evaluating survival as result of daily treatment with 1A-116 at two circadian times (ZTs). Cell cultures displayed a functional circadian clock measured by Bmal1-luc activity and BMAL1 and PER1 protein expression. Circadian rhythm was also found for TIAM1 expression. We found that the effectivity of 1A-116

is rhythmic showing a minimum inhibition of proliferation when applied at 23 HPS after a serum shock, and a maximum when applied at 10 HPS. When applying the drug at 10 HPS, we found over 50% inhibition of migration and augmented apoptosis, with no significant results at 23 HPS. In LN229 bmal1 knock down cells, we found no difference in proliferation, when applying 1A-116 at different circadian times, and both BMAL and TIAM1 rhythms were abolished. In nude mice, we performed Kaplan-Meier survival curves, and found that the median survival of the mice treated at ZT3 was 68 days and at ZT12 was 73 days. These results suggest that a chronopharmacological approach based on the drug 1A-116 is a feasible strategy to improve GBM treatment outcomes. The loss of circadian variation of the effectiveness of 1A-116 and the rhythmicity of TIAM1 in bmal1 knock downs, suggests that 1A-116 is under circadian control, as well as Rac1 pathway.

Disclosures: L.L. Trebucq: None. **G.A. Cardama:** Other; Laboratory of Molecular Oncology, National University of Quilmes. **P. Lorenzano Menna:** Other; Laboratory of Molecular Oncology, National University of Quilmes. **D.A. Golombek:** None. **L. Marpegan:** None. **J. Chiesa:** None.

Poster

P317. Pre-Clinical Therapeutic Strategies

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Program #/Poster #: P317.03

Topic: C.10. Brain Injury and Trauma

Title: Invivo gene therapy in glioblastoma multiform: Using olfactory ensheathing cells containing thymidine kinase gene

Authors: *F. TIRGAR[1], Z. AZIZI[2], M. HADJIGHASSEM[1];
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Abstract: Gene therapy is a new approach to treat glioblastoma multiform (GBM), which has attracted many scientists around the world as an alternative to existing treatments. Suicide gene therapy is the most common gene therapy strategy against gliomas in preclinical studies and clinical trials. The main challenge here is how to transfer the desired genes. The fact that olfactory ensheathing cells (OECs) have high ability of migration even with long distances, makes these cells appropriate for gene therapy. In the current study, we used OECs as carrier for transfer of the lentivirus containing Herpes Simplex Virus-thymidine kinase (HSV-tk) gene to glioblastoma tumoral cells for the first time and examined the possible migration capacity and apoptosis induction ability of these cells in vivo. To achieve this goal, OECs were isolated from the rat olfactory bulbs, cultured and characterized by p75 antibody. OECs were then transfected with lentivirus containing thymidine kinase gene (OEC-tks). For In vivo studies, 49 male Wistar

rats (weight 200-220gr, about 8 weeks old) were divided into 7 groups. After inducing glioblastoma model with the injection of 106 of C6 cells in the frontal lobe of rats, 106 OEC-tks or PBS was injected ipsi-laterally or contra-laterally, followed by treatment with Ganciclovir (GCV; 50mg/kg) or saline intra-peritoneally daily for 5 consecutive days. Then Western blot analysis for Bcl-2 and bax proteins was performed to determine apoptosis (n=3/group). The tumor size was measured by Magnetic Resonance Imaging (MRI; n=4/group). The findings demonstrated that the injection of OEC-tks into the tumor area followed by GCV treatment, induced apoptosis. One-Way ANOVA analysis revealed that Bax expression as a proapoptotic protein was significantly upregulated ($p<0.001$) while Bcl-2 expression as an antiapoptotic protein was significantly downregulated ($p<0.01$) compared to the control. Injection of these cells into the contralateral hemisphere also showed similar effects on protein expression ($p<0.05$ for both proteins) which indicates high migration capability of these cells to the tumor site. On the other hand, One-Way ANOVA analysis showed significant reduction in tumor size in animals which received OEC-tks ipsi-laterally ($p<0.01$). There was a similar finding in tumor size reduction in animals which received this construct contra-laterally ($p<0.05$). Also there was no reduction in protein expression or tumor size in the case of injection of OEC-tks, GCV or PBS alone ($p>0.05$). Taken together, the results from this study supports this hypothesis that OEC-tks can provide a novel carrier for genetherapy and reduce in vivo glioma tumor progression.

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Poster

P317. Pre-Clinical Therapeutic Strategies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P317.04

Topic: C.10. Brain Injury and Trauma

Support: The Swedish Childhood Cancer Foundation (Barncancerfonden)
The Swedish Cancer Society (Cancerfonden)
The Swedish Medical Research Council (VR-MH)
Cancer Research KI

Title: Analysis of the transcriptome and DNA methylome of human neural stem and progenitor cells after irradiation and/or lithium treatment: Roles for BMP signaling and DNA demethylation

Authors: C. NEOFYTOU[1], V. RAILAITE[1], K. BLOMGREN[2], ***O. HERMANSON**[1]; [1]Dept. of Neuroscience, Karolinska Institutet, Stockholm, Sweden; [2]Karolinska Institutet, Stockholm, Sweden

Abstract: Brain tumors in kids show increasingly improved prognosis and survival, at least in part due to improvements in cranial radiotherapy. However, these survivors instead experience late side effects due to the irradiation, including cognitive deficits that prohibit a normal life quality. Even with retained intellectual capacity, the patients show tiredness and fatigue along with problems to focus which make education and normal working life very difficult. The population of adult childhood cancer survivors with late effects are now estimated to around 11,000 only in Sweden and soon millions world-wide. Efficient therapies for this population are thus urgent.
Lithium, a well-known mood stabilizer, has neuroprotective and antitumor effects. We have found lithium treatment to reverse radiation-induced damage in rodents, by among other things regulating the expression of glutamate decarboxylase 2 (GAD2) - associated with neuronal signaling and differentiation - via promoter demethylation (Zanni et al., Mol Psychiatry, 2021). It is however yet unknown whether such lithium-induced gene expression changes also occur in human neural progenitors.
We analyzed the transcriptome and DNA methylome of hiPSC-derived human neural stem and progenitor cells (hNSPCs) that had been exposed to irradiation (4Gy), lithium (3mM), or both, 48 hours after initiation of the experiment.
Our main findings were that lithium treatment of irradiated hNSPCs i) promoted neuronal differentiation by upregulating GAD2 expression similar to previous rodent data, ii) inhibited astroglial differentiation possibly by influencing BMP signaling that in turn was increased by irradiation, iii) increased expression of the DNA dioxygenase TET3 and decreased methylation of the TET3 activator GADD45A promoter in accordance with a decreased methylation of a subset of regulatory regions, iv) specifically decreased DNA methylation of the GREM1 gene, encoding the BMP inhibitor Gremlin-1.
These mechanistic insights provide the molecular basis to support lithium treatment of childhood cancer survivors suffering from cognitive deficits after irradiation.

Disclosures: C. Neofytou: None. **V. Railaite:** None. **K. Blomgren:** None. **O. Hermanson:** None.

Poster

P317. Pre-Clinical Therapeutic Strategies

Location: Virtual Only

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Program #/Poster #: P317.05

Topic: C.10. Brain Injury and Trauma

Support: Cerebral Palsy Alliance Research Foundation

Title: Cocktail Therapy with Neonatal Erythropoietin and Melatonin Prevents Executive Function and Attentional Deficits in Adult Rats with Cerebral Palsy

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Abstract: Cerebral Palsy (CP) is the most common motor disability in childhood. In the USA, CP is most commonly caused by very preterm birth, typically from chorioamnionitis. Children born preterm are prone to executive function deficits. They exhibit particularly poor performance as cognitive tasks become more challenging. Using our preclinical model of CP secondary to chorioamnionitis (CHORIO), we tested if cognitive and executive function abnormalities could be mitigated using a clinically relevant, extended course of erythropoietin (EPO) plus melatonin (MLT). CHORIO was induced in pregnant rat dams on embryonic day 18 by performing a laparotomy followed by uterine artery occlusion for 60min and intra-amniotic injection of lipopolysaccharide (4ug/sac). Shams underwent anesthesia and laparotomy only. On postnatal day (P)0, pups were randomly assigned to sex-balanced groups to receive a neonatal neuroreparative regimen of EPO plus MLT or saline vehicle. In adulthood, rats were trained on a touchscreen operant platform to assess executive function, including attention. Rats performed a visual discrimination task followed by a 5-Choice Serial Reaction Time Task (5-CSRTT), to assess performance on a progressively more challenging task. Passing criteria for each task was set a priori as 80% correct on one day for 5CSRTT and on two consecutive days for VD. Statistically significant differences were determined using Kruskal-Wallis with Dunn's corrections, with $p<0.5$ considered significant. Results confirm that vehicle-treated CHORIO rats grow up to have gait deficits similar to spastic-like CP, including toe-walking, shorter strides and ataxia. Touchscreen assessments reveal that adults in all groups (Sham (n=15), CHORIO veh (n=24), and CHORIO E+M (n=10)) can successfully pass VD. However, while all groups performed similarly during the 30s and 15s 5CSRTT time intervals, CHORIO veh rats had impaired performance as time intervals decreased and the task became more challenging. CHORIO veh rats needed more sessions to achieve passing criteria ($p=0.02$), and produced more errors than CHORIO E+M rats ($p=0.02$). Additionally, CHORIO veh rats made more omissions compared to shams ($p=0.03$), which was corrected by E+M treatment ($p=0.02$). CHORIO veh rats also had more premature responses compared to both Shams and CHORIO E+M rats ($p= 0.025$), indicative of poor inhibitory control. No difference was found between groups in reaction time or magazine latency. Our results show that a translatable dosing regimen of EPO+MLT can modulate deficits of attentional control, behavioral inhibition and executive function in adult rats with CP secondary to CHORIO.

Disclosures: S. Hamimi: None. **L.L. Jantzie:** None. **S. Robinson:** None.

Poster

P317. Pre-Clinical Therapeutic Strategies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P317.06

Topic: C.10. Brain Injury and Trauma

Title: Optimizing vagus nerve stimulation therapy to improve recovery of somatosensation

Authors: *A. RUIZ, K. MALLEY, F. AHMAD, C. MOTA BELTRAN, A. NABEEHA, M. KILGARD, S. HAYS;
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Abstract: More than 20 million people in the United States have some form of peripheral neuropathy. Vagus nerve stimulation (VNS) paired with rehabilitative training has been shown to enhance recovery after various neurological injuries, including nerve damage. This improved recovery is mediated by VNS-dependent facilitation of synaptic plasticity. Thus, paradigms of VNS therapy that promote greater plasticity may further improve recovery. Here, we systematically varied a number of parameters known to influence the magnitude of VNS-dependent plasticity, including stimulation intensity, interstimulation interval, and spaced and massed delivery, and explored the degree of recovery. These findings confirm that VNS paired with tactile rehabilitation enhances recovery of somatosensory function after peripheral nerve damage. Parameter exploration is important for clinical translation in order to create safe and effective therapies that reduce the burden patients carry.

Disclosures: A. Ruiz: None. **K. Malley:** None. **F. Ahmad:** None. **C. Mota Beltran:** None. **A. Nabeeha:** None. **M. Kilgard:** None. **S. Hays:** None.

Poster

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Program #/Poster #: P317.07

Topic: C.10. Brain Injury and Trauma

Support: VA Merit Award

Title: Alcohol and PARP inhibition on Ribosome-bound RNA pathways in cortical excitatory neurons

Authors: G. VALLERINI[1], H. R. KRISHNAN[2], H. E. GAVIN[3], D. P. GAVIN[4], ***R. P. SHARMA**[5];
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Abstract: We report on the effects of ethanol (EtOH) on the distribution of ribosomal-bound RNA transcripts in prefrontal cortical (PFC) pyramidal neurons. We hypothesized that the

pattern of RNA distribution aligned with either ribosomal attachment (ribosome-bound) or genomic expression (transcriptome, total-RNA) is altered by EtOH, and these distributions could be reversed via the inhibition of Poly (ADP-ribose) polymerase (PARP enzyme). We utilized the translating ribosome affinity purification (TRAP) technique to isolate cell-specific RNA. Transgenic mice with EGFP-tagged Rpl10a ribosomal protein expressed only in CaMKIIα-expressing pyramidal cells were administered EtOH or normal saline (CTL) i.p. twice a day, for four consecutive days. On the fourth day, a sub-group of mice that received EtOH in the previous three days received a combination of EtOH and the PARP inhibitor ABT-888 (EtOH+ABT). PFC tissue was processed to isolate CaMKIIα pyramidal cell-specific ribosomal-bound RNA (TRAP) and whole-tissue un-specified genomically expressed total-RNA (INPUT). Both TRAP and INPUT samples were submitted for RNA-seq. We observe EtOH effects on ribosomal-bound transcripts in pyramidal cells. We further demonstrate that treatment with a PARP inhibitor 'normalizes' the effect of EtOH on this ribosomal-bound pool. Further we report, that the PARP inhibitor ABT-888 reversed 82%, of the EtOH-induced changes in ribosomal engagement (more or less bound) (TRAP samples-translatome), and similarly 83% in the total-RNA transcripts (INPUT sample- transcriptome). We identify Insulin Receptor Signaling as highly enriched in the PARP regulated ribosome-bound pool (EtOH-affected) and validate five participating genes from this pathway. To our knowledge this is the first description of the effects of EtOH on the ribosomal cell-specific capture of transcripts from total-RNA and provide insights into PARP mediated regulation of this crucial step.

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Poster

P317. Pre-Clinical Therapeutic Strategies

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Program #/Poster #: P317.08

Topic: C.10. Brain Injury and Trauma

Support: NIH Grant R01NS094384
NIH Grant R01NS103803
DARPA BTO TNT Grant No. N66001-17-2-4011

Title: Functional recovery after peripheral nerve injury depends on vagus nerve stimulation parameters.

Authors: *K. MALLEY, A. RUIZ, T. DANAPHONGSE, S. HUSAIN, M. MIAN, A. SULE, N. VALMIKI, S. HAYS, M. KILGARD;
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Abstract: Vagus nerve stimulation (VNS) paired with rehabilitation represents an innovative approach to restore somatosensation and sensorimotor function after peripheral nerve injury (PNI). Recovery is dependent both on the timing of stimulation and the electrical stimulation parameters of VNS. Manipulating these parameters for optimal recovery is critical for clinical translation. In this study, we manipulated two of these parameters, the current amplitude and the interstimulus interval (ISI), and assessed their effect on somatosensory recovery after PNI-induced sensory loss. Animals received a PNI of their right forearm resulting in hyposensitivity of their forepaw. After an 8 week recovery, animals then underwent tactile rehabilitation, in which a brief mechanical stimulus applied to the affected paw was paired with VNS appropriate for their group . We measured sensorimotor recovery with an automated esthesiometer at multiple timepoints during treatment. A variety of other assessments were used to measure functional sensorimotor recovery and retention. In Experiment 1, rats were randomized to receive either 0 mA, 0.4 mA, 0.8 mA, or 1.2 mA VNS, with all other stimulation parameters held constant. In Experiment 2, a second cohort of animals received VNS with either a 2 second ISI or a 10 s ISI for 1 or 4 weeks, with all other parameters fixed. We found an inverted-U relationship between current intensity and recovery with a moderate intensity current being the most optimal for sustained recovery. This data corroborates other inverted-U observations, which suggests a central origin of the effect. Additionally, animals who received VNS once every 2 seconds recovered the majority of their sensorimotor function. When treatment was clustered into 1 week, animals did not recover their sensorimotor function compared to animals who received standard therapy. These results indicate that, with daily repetition, therapy can be done faster and still drive recovery if VNS is delivered at a moderate intensity, and clustering saturates after a certain number of stimulations. These data are readily translatable to a clinical setting and can be used in conjunction with other data from our lab to inform the appropriate clinical dosing and delivery of VNS.

Disclosures: K. Malley: None. **A. Ruiz:** None. **T. Danaphongse:** None. **S. Husain:** None. **M. Mian:** None. **A. Sule:** None. **N. Valmiki:** None. **S. Hays:** None. **M. Kilgard:** None.

Poster

P317. Pre-Clinical Therapeutic Strategies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P317.09

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Title: Emapunil attenuates microglial mitochondria membrane potential following LPS administration

Authors: *K. I. MCGLOTHEN, R. M. HINES, D. J. HINES;
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Abstract: The high metabolic demand of activated microglia is linked with changes in the mitochondrial membrane potential (ΔΨm) and indicate varying inflammatory states in the brain. Examining the dynamic function of ΔΨm in microglia could provide key insight to how and when brain microglia respond to periods of inflammation and aid in the innovation of pre-symptomatic screening techniques for inflammatory disorders. In the present study, a novel approach to examine spatio-temporal changes in microglial ΔΨm was utilized. ΔΨm was measured using cell permeable dye tetramethylrhodamine ethyl ester (TMRE). Acute cortical brain slices obtained from fractalkine mice were utilized to examine the spatial and temporal metabolic processes in microglia. Administration of Lipopolysaccharide (LPS) increased the ΔΨm at 3 timepoints differentially across the microglial soma, branches, and endfeet. The application of Emapunil attenuated the increase of the ΔΨm in microglia at each of these timepoints. Similarly, immunohistochemistry showed an increase in the 18 kDa translocator protein (TSPO), a protein that is upregulated in microglia during inflammatory activation, only at microglial branches following the administration of LPS. Application of TSPO ligand Emapunil at a specified temporal window attenuated the ΔΨm. The ability to examine alterations in microglial ΔΨm and TSPO during periods of neuroinflammation presents a novel tool with broad implications for many gliotic diseases.

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Poster

P317. Pre-Clinical Therapeutic Strategies

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Topic: C.10. Brain Injury and Trauma

Support: NIH Grant R33NS107785
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Children's Neuroscience Institute

Title: Development of Novel 20-HETE Formation Inhibitors for Neurotherapeutics

Authors: *B. P. NELSON[1,2], S. K. SEPPI[1], C. TANG[3], P. M. KOCHANEK[1,4], A. P. ROWLEY[1,2], L. A. MCDERMOTT[3], S. M. POLOYAC[5], M. D. MANOLE[1,2];
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Abstract: 20-hydroxyeicosatetraenoic acid (20-HETE) is a vasoconstrictive metabolite of arachidonic acid by cytochrome P450 (CYP) 4A11 and 4F2 in humans. Inhibition of 20-HETE

formation has been shown to produce neuroprotective effects in pre-clinical models of cardiac arrest and stroke; however, there are currently no 20-HETE inhibitors suitable for clinical translation.
Our primary objective was to develop clinically viable 20-HETE inhibitors that have strong potency and favorable physicochemical, metabolic stability, selectivity, solubility, CYP inhibition, and blood-brain barrier (BBB) permeability.
The study design consisted of designing, synthetizing, and evaluating 143 novel compounds in vitro. Compound 107 (UPMP107) was identified as a potential candidate, with suitable 20-HETE inhibition in liver microsomes. UPMP107 and 20-HETE levels were quantified in the brain and plasma of rats using solid phase extraction and UPLC-MS/MS analysis in a triple quadrupole mass spectrometer. Intravenous (IV) administration of UPMP107 in a pediatric rat model showed BBB penetration and exhibited a quick onset of brain 20-HETE inhibition sustained for 9 h after administration. Following initial pharmacokinetic and pharmacodynamic studies, our secondary objective was to utilize the selected compound in our pediatric rat model of asphyxial cardiac arrest (CA) to assess physiologic and neurologic outcomes. Anesthetized and intubated 17-day old rats had arterial and venous catheters placed. CA was induced for 9.5 min. Rats were resuscitated and were randomized to receive either UPMP107 (20mg/kg) or vehicle (13.3mL/kg 15% cyclodextrin in PBS) (n=6/group). Rats received a second dose of UPMP107 or vehicle at 8 h post-resuscitation. Mean arterial pressure, $PaCo_2$, PaO_2, pH, lactate, and HCO_3 were measured at baseline and at 10, 30, and 60min timepoints following the arrest. Physiological variables did not differ between the two groups. In conclusion, UPMP107 appears to be a suitable candidate for clinical translation. Further studies evaluating the neurological outcome, dosing, and administration of UPMP107 in the CA model are planned.

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Poster

P359. Peripheral Nerve Trauma: Mechanisms of Injury and Regeneration

Location: Virtual Only

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Program #/Poster #: P359.01

Topic: C.10. Brain Injury and Trauma

Support: FPU17/03657
SAF2017-84464-R

Title: Motor and sensory neuron subtypes differ in axon regeneration after nerve injury

Authors: *S. BOLÍVAR, E. UDINA;
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Abstract: Specificity in regeneration after peripheral nerve injuries is the major determinant of functional recovery. Unfortunately, regenerating motor and sensory axons rarely find their original pathways to reinnervate appropriate target organs. Despite the heterogeneity of these peripheral neuron populations, little is known about their differences in regeneration. Thus, we propose the comparative study of the regenerative capacity of the most relevant neuronal populations. We used double transgenic mice that express fluorescence in specific neurons: motoneurons (ChAT-Cre/Ai9), proprioceptors (Pv-Cre/Ai9), unmyelinated nociceptors (Trpv1-Cre/Ai9) and cutaneous mechanoreceptors (Npy2r-Cre/Ai9). We performed immunofluorescence analysis of neuronal markers (parvalbumin, neurofilament, CGRP, IB4, ChAT, calbindin) to confirm the validity of these transgenic mice as specific reporters of the desired neuronal populations. We obtained the regeneration rate of each neuron type by analysing the number of axons that regenerated up to the tibial nerve after a crush in the sciatic nerve. We found that unmyelinated nociceptors regenerated faster than any other peripheral neuron population 7 days after the injury, whereas proprioceptive neurons showed the worst regeneration among groups. Number of regenerating motor and unmyelinated nociceptive axons in the tibial nerve was similar to control values after 9 days, in contrast to cutaneous mechanoreceptors and proprioceptors, that regenerated incompletely. Evaluation of neurite growth in dorsal root ganglia organotypic cultures also showed different regenerative capabilities among $Trpv1^+$, $Npy2r^+$ and Pv^+ neurons. The preferential regeneration of these populations was assessed by applying retrotracers in the cutaneous and muscular branches of the femoral nerve after a transection. All the studied neuronal populations showed an early preference for the cutaneous branch that weakened at long term. Eight weeks after the lesion, motoneurons did not demonstrate any preference, whereas proprioceptors and cutaneous mechanoreceptors showed a preference for their correct branch (muscle and cutaneous branch, respectively). Our findings reveal novel intrinsic differences between peripheral neurons, which suggest unique regeneration mechanisms in each neuronal subtype. Future work will investigate in deep these mechanisms to modulate them and improve specific reinnervation.

Disclosures: S. Bolívar: None. **E. Udina:** None.

Poster

P359. Peripheral Nerve Trauma: Mechanisms of Injury and Regeneration

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P359.02

Topic: C.10. Brain Injury and Trauma

Title: Influence of tau on paclitaxel-induced neuropathy

Authors: E. OROZCO[1], B. BOBER[2], *S. SHAH[2];
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Abstract: Paclitaxel, a popular chemotherapeutic used with high efficacy to treat several metastatic cancers, causes a particularly insidious sensory peripheral neuropathy, resulting in acute pain, chronic tingling and burning sensations, and numbness. It is accepted that paclitaxel halts cancer cell division by binding and stabilizing microtubules. However, since neurons are post-mitotic, paclitaxel must impair neuronal function by a mechanism distinct from cell division. We hypothesized that by stabilizing neuronal microtubules, paclitaxel induces microtubule bundling, which then results in several downstream adverse structural and functional impacts. Further, we hypothesized that the microtubule associated protein tau, given its widely hypothesized role in microtubule stabilization and the proximity of its binding site in the microtubule lumen to that of paclitaxel, is a strong candidate to influence the progression of paclitaxel-induced neuropathy. To test these hypotheses, we performed a number of structural and functional assessments of wild-type or tau knockout (Mapt -/-) mice injected systemically with 10 mg/kg cumulative dose of paclitaxel or Cremaphor carrier over a 5 day period. Von Frey hair testing revealed increased mechanical sensitivity in paclitaxel-treated mice. Electron microscopy and centrifugation studies combined with Western blotting confirmed microtubule bundling in peripheral (sensory) nerves of these mice, and also revealed a hyper-connected neuronal cytoskeleton. The latter was confirmed by immunohistochemistry. Consistent with mechanical hypersensitivity, paclitaxel-treated cultured neurons from dorsal root ganglia (DRG) demonstrated persistent mechanically-gated calcium influx, in contrast to more transient activity in carrier-treated neurons. Tau knockout mice treated with paclitaxel, in contrast, displayed reduced mechanical hypersensitivity, reduced microtubule bundling, and a well-organized cytoskeleton. In addition, paclitaxel-treated cultured neurons from DRG of tau knockout mice were less responsive than their wild-type counterparts to mechanical stimuli. This may be due to an inherently lower baseline sensitivity to mechanical stimuli in control (carrier-treated) tau knockout neurons. Collectively, these findings reveal compelling roles for tau in cytoskeletal stability and mechanical gating of ion channels in the peripheral nervous system, and in modulating neuropathic progression in a mouse model of paclitaxel-induced neuropathy.

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Poster

P359. Peripheral Nerve Trauma: Mechanisms of Injury and Regeneration

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Program #/Poster #: P359.03

Topic: C.10. Brain Injury and Trauma

Support: VA Grant IK2RX002688
Neurological Degeneration Research Inc. Grant

Title: Carboxyl-terminal modulator protein promotes nerve injury-induced muscle atrophy

Authors: *C. L. WALKER[1,2], J. WANG[1], C. WILSON[1], L. TIERNEY[1];
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Abstract: The downregulation of Akt phosphorylation and activity in denervated muscle leads to increased proteins involved in muscle breakdown. Akt prevents the transcription of E3 ubiquitin ligases, such as muscle ring-finger protein 1 (Murf-1), involved in the structural degeneration of muscle cells. In addition, reduced Akt signaling leads to aberrant protein synthesis involved in muscle degeneration. Only recently has information regarding the intracellular regulation of Akt activity in muscle cells been further detailed. We previously discovered that a known negative regulator of Akt, carboxyl-terminal modulator protein (CTMP, also known as Them4), is highly elevated in late-stage atrophic muscle in a mouse model of amyotrophic lateral sclerosis (ALS). Furthermore, knocking down CTMP in an in vitro model of muscle atrophy reverses the downregulation of Akt phosphorylation. The present study investigated whether CTMP increases in atrophic muscle caused by complete transection sciatic nerve injury (SNI) and whether loss of CTMP reverses the atrophy process and acts through Akt signaling in a knock-out mouse model of SNI. We found that CTMP significantly increased in the gastrocnemius muscle 7 days post-unilateral nerve injury relative to the contralateral non-affected muscle ($p<0.05$). Using CTMP knock-out (CTMP-KO) and wild-type (WT) mice, sciatic nerve transection elicited significantly less atrophy in the CTMP-KO gastrocnemius compared to denervated WT muscle as determined by weight comparison and muscle fiber area relative to each animal's non-denervated muscle ($p<0.05$). The denervated muscle exhibited significantly increased Akt and glycogen synthase kinase 3β (GSK3β) phosphorylation relative to the non-affected muscle in CTMP-KO mice compared to WT mice ($p<0.05$). The phosphorylation of ribosomal protein S6 and 4E-BP1 proteins were also significantly relatively upregulated in denervated CTMP-KO muscle compared to WT mice ($p<0.001$ & $p<0.05$, respectively). WT mice also showed a significantly higher relative denervated muscle expression of myostatin and the atrophy-associated Murf-1 protein than CTMP-KO mice ($p<0.05$). Our results suggest that CTMP is important in the progression of muscle atrophy post-denervation. In addition, decreased Akt signaling, and subsequently reduced protein synthesis, and increased degradation processes are likely mechanisms of CTMP's action in promoting nerve injury-induced muscle atrophy.

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Poster

P359. Peripheral Nerve Trauma: Mechanisms of Injury and Regeneration

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Topic: C.10. Brain Injury and Trauma

Support: CIHR Canada Graduate Scholarship - Master's (CGS-MH)

Title: EGFR inhibition promotes nerve regeneration in a mouse hindlimb model of nerve injury and in neurite outgrowth assays

Authors: J. PELLER[1], A.-M. CROTTY[2], M. D. KAWAJA[3], *J. HENDRY[4];
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Abstract: The success of nerve regeneration following injury remains limited with no FDA-approved non-surgical methods to enhance the rate of recovery. Increasing the rate of axonal regeneration could limit the degree to which chronic denervation, chronic axotomy and denervation atrophy negatively impact functional recovery. Previous studies have demonstrated that EGFR inhibition may promote axonal regeneration by preventing chondroitin sulfate proteoglycan (CSPG) inhibition of regeneration. Furthermore, blockade of the tyrosine kinase receptor ErbB2 with the antibody inhibitor trastuzumab was found to accelerate multiple metrics of nerve regeneration. We hypothesized that these effects occur by disrupting an inhibitory heterodimer formation between epidermal growth factor receptor (EGFR) and ErbB2. In this study, we investigated the impact of the dual EGFR-ErbB2 small molecule inhibitor lapatinib and EGFR inhibitor gefitinib, FDA-approved cancer drugs, on nerve regeneration using a mouse hindlimb nerve injury model (only lapatinib) and *in vitro* neurite growth assays (both lapatinib and gefitinib). Twenty-four C57Bl/6 mice underwent sciatic nerve transection and repair. Animals were administered lapatinib or vehicle control for either 10 or 28 days, at which point regeneration was assessed by retrograde labelling and quantitative nerve histomorphometry. Mechanistic exploration took place with complimentary neurite outgrowth assays, whereby adult dorsal root ganglion (DRG) neurons were cultured in the presence of inhibitory chondroitin sulfate proteoglycan (CSPG), with or without lapatinib or gefitinib. Four weeks post-injury, lapatinib administration led to a significant increase in the number of regenerated myelinated axons distal to the repair site compared with vehicle controls (lapatinib: 1042 ± 108, vehicle: 729 ± 110; $p<0.05$). No significant differences were observed in the number of retrograde labelled sensory or motor neurons or other nerve histomorphometry parameters between vehicle and control. DRG neurons cultured in the presence of lapatinib and gefitinib had significantly increased mean neurite length compared to CSPG alone. This rescue effect was seen more strongly with gefitinib than lapatinib. Dual inhibition of the EGFR and ErbB2 receptors with the FDA-approved drug lapatinib increased the number of distally regenerated myelinated axons at 28 days following sciatic nerve transection and repair. Furthermore, in vitro neurite growth assays suggest that sole EGFR or dual EGFR-ErbB2 inhibition by gefitinib or lapatinib, respectively, may act be overcoming the inhibitory effects of CSPG on axonal growth.

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Poster

P359. Peripheral Nerve Trauma: Mechanisms of Injury and Regeneration

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Topic: C.10. Brain Injury and Trauma

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Grant of Saitama Chapter, Japanese Physical Therapy Association for Study Promotion 18-02

Title: Effects of exercise on muscle reinnervation and plasticity of spinal circuits in rat sciatic nerve crush injury models with different denervation periods

Authors: *Y. MINEGISHI[1,2], K. OZONE[1], Y. OKA[1], T. KOKUBUN[3], K. MURATA[3], N. KANEMURA[3];
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[2]Res. Fellowship for Young Scientists, Japan Society for the Promotion of Sci., Tokyo, Japan;
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Abstract: Peripheral nerves have the ability to regenerate after injury, but functional motor recovery is poor. The length of the denervation period after nerve injury is an important factor that defines motor function recovery. Failed muscle reinnervation and synapse withdrawal of primary afferent neurons from motoneurons inhibit motor function recovery. Physical exercise is an important strategy to regulate these changes and to improve motor function recovery after injury. However, the relationship between the denervation period and the effect of exercise is unclear. The purpose of this study was to examine the effect of the length of the denervation period before the initiation of exercise on the histological connectivity between reinnervation and spinal circuits, along with its relationship to motor function recovery. 10-week-old male Sprague Dawley rats (n = 32) were randomly divided into four groups: (1) sciatic nerve crush (SC group), (2) sciatic nerve crush and exercised (SC-EX group), (3) multiple sciatic nerve crush with prolonged denervation period (MSC group), (4) multiple sciatic nerve crush and exercised (MSC-EX group). Treadmill exercise was conducted at 10 m/min for 60 min, five times a week. The soleus muscle and L4-L5 spinal cord were collected 35 days after the last crush. The percentage of reinnervation was calculated by counting the neuromuscular junction (NMJ) with alpha-bungarotoxin that contacted neurofilament 200-positive fibers. Synaptic coverage of motoneurons in Lamina IX was determined from contacts between motoneurons and immunoreactive structures of synapse-associated proteins using anti-vesicular glutamate transporter 1 (VGLUT1) and anti-synaptophysin. The sciatic functional index (SFI) was used to assess motor function recovery. The percentage of SOL reinnervation tended to be lower with a longer denervation period. However, there was no significant difference between the exercise group and the non-exercise group. The synaptic coverage of VGLUT1 immunoreactivity was higher in the SC-EX group than in the SC group but did not show a significant difference

between the MSC-EX and MSC groups. In contrast, synaptic coverage of synaptophysin immunoreactivity did not show any significant difference between all groups. The SFI at 35 days after injury was higher in the SC-EX, SC, MSC-EX, and MSC groups, in that order. These results suggest that the length of the denervation period after axonotmesis affects muscle reinnervation and motoneuron synaptic changes differently, but exercise promotes motor function recovery regardless of the length of the denervation period.

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Poster

P359. Peripheral Nerve Trauma: Mechanisms of Injury and Regeneration

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Program #/Poster #: P359.06

Topic: C.10. Brain Injury and Trauma

Support: Funded by Science Foundation Ireland and Integra LifeSciences as part of the Advanced Materials and Bioengineering Research (AMBER) Centre (grant number TP27-1846B).

Title: Immuno-modulatory and pro-angiogenic extracellular matrix combinations improve outcomes in large defect peripheral nerve repair in a rat model

Authors: *Z. KOCI[1,2,3], A. J. HIBBITTS[1,2,3], S. L. KNEAFSEY[1,2,3], L. ZILIC[1,2,3], B. CAVANAGH[4], G. CHEN[5], C. T. BUCKLEY[1,2,3], S. J. ARCHIBALD[6], F. J. O'BRIEN[1,2,3]; [1]Anat. and Regenerative Med., Royal Col. of Surgeons In Ireland, Dublin, Ireland; [2]Trinity Ctr. for Bioengineering, Trinity Col. Dublin, Dublin, Ireland; [3]Advanced Meterials and Bioengineering Res. Ctr. (AMBER), RCSI and TCD, Dublin, Ireland; [4]Cell. and Mol. Imaging Core, [5]Dept. of Physiol. and Med. Physics, Ctr. for the Study of Neurolog. Disorders, Micros, Royal Col. of Surgeons in Ireland, Dublin, Ireland; [6]Integra Lifesciences, Plainsboro, NJ

Abstract: Biomaterial-based nerve guidance conduits (NGCs) have emerged as viable alternatives to autografts in peripheral nerve injuries; however, repair over long distances remains inconsistent. To address this, we have developed a filled NGC with an internal matrix consisting of aligned microporous channels containing combinations of macromolecules found in nerve extra-cellular matrix (ECM), namely fibronectin, laminin 1 and laminin 2. Using these combinations, we aim to prepare an off-the-shelf, acellular NGC with capacity to repair large nerve defects. NGCs were produced using an outer collagen conduit and an internal matrix of collagen-chondroitin sulphate augmented with ECM combinations. Porosity and pore alignment was assessed using scanning electron microscopy. The effects of the different ECM combinations were assessed for growth factor and inflammatory biomarkers using primary rat

dorsal root ganglia (DRG) cultures over 14 days. Following this, large 15 mm sciatic nerve defects were created in rats and the lead candidate NGCs were implanted and assessed after 8 weeks. Regeneration was assessed using foot withdrawal response, electrophysiology and immuno-histochemistry. Following fabrication, the internal matrix of filled conduits was found to be highly porous with unidirectional pores aligned along the longitudinal axis. When assessed over 14 days, an ECM combination of fibronectin, laminin 1 and laminin 2 at a ratio 1:4:1 significantly decreased DRGs' secretion of inflammatory markers such as interferon gamma-induced protein 10, monocyte chemoattractant protein 1 and macrophage inflammatory protein 1a and significantly increased DRGs' production of nerve growth factor, vascular endothelial growth factor (VEGF) and interleukin-6. Based on the *in vivo* analysis, regenerated tissue was found to be present throughout the NGC and functional recovery was comparable to autograft controls. Notably, ECM combinations of fibronectin, laminin 1 and laminin 2 at a ratio 1:4:1 significantly increased *in vivo* vascularisation inside NGCs when compared to NGCs without ECM, which may be associated with the capacity to increase VEGF production by DRGs observed *ex vivo*. In summary, the pro-regenerative effects of ECM in peripheral nerve repair were highly combination-dependent. This study has demonstrated that ECM combination of fibronectin, laminin 1 and laminin 2 at a ratio 1:4:1 has immune-modulatory and pro-angiogenic effect with capability to repair large peripheral nerve injuries to a level equivalent to autograft indicating its potential as a new clinical therapy for repairing large nerve defects.

Disclosures: Z. Koci: B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Integra Lifesciences. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Integra Lifesciences. **A.J. Hibbitts:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Integra Lifesciences. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Integra Lifesciences. **S.L. Kneafsey:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Integra Lifesciences. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Integra Lifesciences. **L. Zilic:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Integra Lifesciences. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Integra Lifesciences. **B. Cavanagh:** None. **G. Chen:** None. **C.T. Buckley:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Integra Lifesciences. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Integra Lifesciences. **S.J. Archibald:** A. Employment/Salary (full or part-time):; Integra Lifesciences. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current

grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Integra Lifesciences. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Integra Lifesciences. **F.J. O'Brien:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Integra Lifesciences. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Integra Lifesciences.

Poster

P359. Peripheral Nerve Trauma: Mechanisms of Injury and Regeneration

Location: Virtual Only

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Program #/Poster #: P359.07

Topic: C.10. Brain Injury and Trauma

Support: CIHR Grant 202003PJT-436747
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Title: Alpha B Crystallin plays a role in terminating the pro-inflammatory macrophage response in the injured aged peripheral nerve

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[1]Neurosci., [2]Clin. Neurosciences and Cell Biol. & Anat., Hotchkiss Brain Institute, Univ. of Calgary, Calgary, AB, Canada

Abstract: Peripheral nerve injury (PNI) affects millions of people worldwide, and those impacted are often left with debilitating motor and sensory impairments. More concerning is that regeneration of damaged peripheral axons is worse with advancing age. A decline in the functions of Schwann cells (SCs) as well as resident and infiltrating macrophages (MΦ) may contribute to the worsened regenerative capacity of the damaged aged peripheral nervous system (PNS). These cells are critical for effective PNS regeneration due to their release of neurotrophic factors and cell adhesion molecules by SCs, and together with MΦs, the phagocytosis of myelin and neuronal debris among other functions. We are interested in identifying the molecular factor(s) responsible for the impaired function of senescent SCs and MΦs. Towards this goal, we found that an age-related decline in the expression of a small heat shock protein called alphaB-crystallin (αBC) in SCs correlated with defective myelination and lipid synthesis in the injured PNS, as well as an increased presence of MΦs. Further, in younger global αBC knockout mice, there was a prolonged elevation of pro-inflammatory MΦs above that of wildtype (WT). To assess if the defect in myelination with age is related to alterations in the number of SCs and phenotype of MΦ after PNI, we quantified the number of SCs and pro- and anti-inflammatory MΦs after sciatic nerve crush injury in WT and αBC globally null mice across age. With respect

to MΦs, we found that the total number of MΦs was elevated in older mice at later time points post-injury in the αBC knockout mice, whereas older WT mice displayed a gradual decline in MΦ presence after damage, similar to that seen in younger animals. Furthermore, upon examination of the phenotype of these pathologically elevated MΦs in the αBC null animals, it appears that they are pro-inflammatory due to the significant increase in their numbers observed at the same time points as the general MΦ marker. In contrast, the number of anti-inflammatory MΦs was similar between the two genotypes after injury with an overall reduction in older animals relative to younger mice. These findings indicate that αBC may play a role in terminating the pro-inflammatory macrophage response following PNI, and that with increasing age the natural decline in αBC may result in an increased presence of pro-inflammatory macrophages that could contribute to adverse symptoms such as neuropathic pain. We are currently assessing the presence of myelinating and non-myelinating SCs.

Disclosures: K.M. Hagen: None. **S.S. Ousman:** None.

Poster

P359. Peripheral Nerve Trauma: Mechanisms of Injury and Regeneration

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Topic: C.10. Brain Injury and Trauma

Support: NIH Grant R01NS072171
NIH Grant 5R01NS098231-04

Title: Increased inhibitory interneuron activity in S1 after peripheral nerve injury visualized in transgenic GAD1-GCaMP rats

Authors: *V. KRISHNAN[1], **L. C. WADE**[2], R. R. ISRAELI[1], G. PELLED[1,3,2];
[1]Biomed. Engin., [2]Neurosci. program, [3]Radiology, Michigan State Univ., East Lansing, MI

Abstract: Using electrophysiology, we previously demonstrated that the activity of both excitatory neurons and inhibitory interneurons are impacted in the deprived S1. This could be a possible mechanism leading to phantom limb pain. However, the exact mechanism through which nerve injury affects this population remains unknown due to their lesser number and smaller sizes of inhibitory interneurons and the fact that it is difficult to identify and obtain electrophysiology recordings from them. In order to specifically identify and study this population, we generated transgenic Sprague Dawley rats encoding GAD1-GCaMP using CRISPR technology. GAD1 is a protein that is expressed in inhibitory interneurons and this transgenic rat provides an unparalleled opportunity to visualize neural activity through genetically encoded calcium indicators (GCaMP6S). We performed forepaw denervation (severing ulnar, median, and radial nerves) 4 weeks post birth in 5 rats. Additional 7 age-

matched rats were used as controls. Fluorescent imaging was performed 2 to 3 weeks after denervation on 300 µm-thick cortical slices. A bipolar tungsten electrode was positioned in layer 2/3, in layer 5, or in the corpus callosum in the deprived S1, and delivered 100 Hz stimulation for 5 s. We analyzed the fluorescence changes of 830 GAD1-expressing neurons that were located in an ROI of 700 X 400 µm adjacent to the stimulating electrode. Out of this population, 399 neurons were considered responsive as their change in fluorescence was greater than the standard error of mean (SEM) of that group. The results demonstrate that local stimulation of layer 2/3 and layer 5 leads to increases in the number of evoked responses of GAD1-expressing neurons in injured rats compared to controls: Stimulation of layer 2/3 led to 56.8% responses in injured rats in comparison to 40.0% responses in controls ($p = 0.001276$, chi-squared); Stimulation of layer 5 led to 91.3% responses in injured rats in comparison to 50.7% responses in controls ($p < 0.00001$, chi-squared). This suggests that inhibitory interneurons are susceptible to local, intrahemispheric stimulation. Stimulation of the corpus callosum led to 21.4% responses in layer 5 in injured rats compared to 41.1% responses measured in controls ($p = 0.007586$, chi-squared), suggesting that injury impacts interhemispheric communication, leading to an overall decrease in activity. Together, our results provide direct evidence that the inhibitory interneurons' activity is altered after injury, leading to increases in inhibition in the deprived S1.

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Poster

P359. Peripheral Nerve Trauma: Mechanisms of Injury and Regeneration

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Topic: C.10. Brain Injury and Trauma

Support: NIH U01NS115817

Title: Establishing a platform for neural culture of octopus neurons

Authors: *C. CYWIAK[1,2], T. VANBUREN[1,2], G. PELLED[1,2,3];
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Abstract: The octopus provides a unique model with unparalleled characteristics to study sensorimotor circuits associated with gripping behavior and decision making. The octopus's nervous system is highly developed with 350 million neurons in the eight axial cords and peripheral ganglia. The axial cord is similar to a vertebrate's spinal cord performing both sensing and motor planning and execution of movement. However, the neural ensembles encoding for different movements are unknown. We are working towards developing a platform that will enable visualizing neural activity using optical imaging methods. We have established a protocol to maintain a viable primary neural culture from neurons obtained from the arm of *Octopus*

bimaculoides. Octopus's arm neurons were dissected with collagenase P (4mg/ml), trypsin (1mg/ml) and accutase (4mg/ml) in 50% L15 - and 50% sea water, 15mM HEPES and 1mM glucose, each enzyme were incubated for 40 min. Afterwards, the cells were washed with 50% L15 - and 50% sea water and centrifuge three times to eliminated contamination. Finally, the cells were changed to a media solution containing 50% L15 - and 50% sea water, 15mM HEPES, 1Mm glucose, 1% Factor B-27, 1% PenStrip and 1% Amphotericin B and plated in a 24 well plate dish coated with matrigel. To test the viability of the neurons we used a multielectrode array system with 26,000 electrodes to record action potentials (AP) from the cultured cells (Maxwell biosystems). The cells were continuously perfused with a physiological solution. We were able record AP from these neurons up to 4 days after dissections. For neuronal validation we fixed the cells with 4% PFA for 40 min and changed to 20% sucrose media. The cells were sliced in 25µm sections, washed then incubated for 1hr with blocking solution (0.1% Triton-X 100 and 5% Goat Serum). The primary antibodies used were mouse polyclonal Antiβ III Tubulin antibody (1:250) and rabbit polyclonal poli (ADP-ribose) polymerase (PARP1) (1:250) overnight at 4◦C. Both antibodies are specific for neurons. After the cells were washed and incubated with second antibodies, FITC-conjugated goat anti-mouse IgG (1:200) and Rhodamine conjugated goat anti-rabbit IgG (1:200) for 2hrs and mounted in DAPI media solution then visualized on a Keyence Microscope. This work is important in establishing platforms to test neural function and neural assemblies in a control fashion. This preparation is also a critical step towards expressing genetically-encoded imaging probes (calcium and voltage indicators) that will be able to report on large-scale, and high resolution neural activity in the octopus.

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Poster

P359. Peripheral Nerve Trauma: Mechanisms of Injury and Regeneration

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Topic: C.10. Brain Injury and Trauma

Support: NIH Grant U01NS115817

Title: Spike recordings from an octopus disembodied arm

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Abstract: The incredible abilities of the octopus to perform complex movements and skillfully coordinate the behavior of eight arms simultaneously make it an excellent candidate for research focused on studying motion control. Furthermore, the octopus is thought to strongly utilize a

decentralized motion control mechanism, relying on much of the planning and execution to be performed in the arms themselves.
Central to understanding the mechanisms of such control is characterizing nerve firing patterns and relating them to observed motion. Here we explore approaches for recording such relevant patterns from stimulus evoked potentials in the axial nerve cord of an arm from the *Octopus bimaculoides*. Both a monopolar tungsten and carbon fiber multi-channel electrode array are used to record unit activity in the separated arm following numerous types of stimulation delivery, including chemical, electrical, and mechanical stimulation. We intend to further octopus electrophysiology work aimed at detecting patterns of motor activity (Gutfreund *et al.*, 2006; Fouke *et al.*, 2020) in an isolated octopus arm in response to stimulation.
An arm was removed and placed in a platform that was continuously perfused in a physiological solution. The arm was secured to reduce motion and the tungsten electrodes were positioned inside the axial cord. In response to chemical stimulation with NaOH, considerable activity was shown to be evoked in the axial cord, with as many as 27 spikes per second being recorded. Activity could be seen to persist for up to several minutes following stimulation. A brief tactile mechanical stimulation is also being investigated for it's ability to induce spiking activity. We are currently working on designing a multi-channel carbon fiber array which would be well suited for underwater octopus electrophysiology.
Characterization of multi-unit activity in the nerve cord of the octopus arm serves an important role in understanding the techniques used by the octopus to implement planning and motion control for it's arms to perform goal oriented behavior like hunting. Characterization of firing patterns in response to a battery of different stimuli using a variety of recording techniques may reveal novel control mechanisms and serve to advance the field of embedded soft robotic control.

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Poster

P360. Spinal Cord Injury: Therapeutic Strategies: Animal Models

Location: Virtual Only

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Title: Il10 overexpression in mesenchymal stromal cell transplants for spinal cord neuroprotection

Authors: *I. MALDONADO-LASUNCION[1,2], F. DE WINTER[4], J. VERHAAGEN[4], M. OUDEGA[5,2,3,6];
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Abstract: Traumatic spinal cord injuries (SCI) feature an initial mechanical injury followed by a chronic loss of nervous tissue surrounding the original injury epicenter. After injury, local immune cells become activated and waves of circulating immune cells infiltrate the injured tissue. While pro-inflammatory macrophages play an important role in clearing debris and dead tissue to prepare the spinal cord for repair, they eventually fail to switch their polarization to the anti-inflammatory and pro-resolving phenotypes that are necessary for tissue repair. The chronic presence of pro-inflammatory macrophages in SCI contributes to the expansive nature of the injury which significantly contributes to the lack of repair. Mesenchymal stromal cells (MSC) are multipotent stem cells that secrete bioactive molecules that may support nervous tissue repair. Acute intraspinal MSC transplants promote neuroprotection and immunomodulation after SCI, but the resulting therapeutic efficacy is suboptimal. In this study, we aimed at enhancing the immunomodulatory potential of MSC by inducing overexpression of the anti-inflammatory mediator interleukin-10 (IL10) using lentiviral vectors *in vitro*. We argued that this approach could increase the neuroprotective effects of an MSC transplant. We studied the effects of IL10 overexpression on MSC secretome, and their ability to induce anti-inflammatory macrophage polarization, *in vitro*. We also evaluated the neuroprotective and immunomodulatory potential of IL10-MSC transplants *in vivo*, using an adult rat model of contusive SCI.

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Poster

P360. Spinal Cord Injury: Therapeutic Strategies: Animal Models

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Program #/Poster #: P360.02

Topic: C.11. Spinal Cord Injury and Plasticity

Title: Three-dimensional painted microscaffold materials reduce inflammation following spinal cord contusion

Authors: *J. R. MOSS[1], R. D. FESSLER[1], Q. SHEN[1], Y. LOPEZ-RAMIREZ[1], D. TUCKER[1], A. E. JAKUS[2], R. N. SHAH[2], R. G. FESSLER[1], B. T. DAVID[1];
[1]Neurosurg., Rush Univ. Med. Ctr., Chicago, IL; [2]Dimension inx, Chicago, IL

Abstract: Spinal cord injuries (SCIs) are known to produce a myriad of effects over the acute, sub-acute, and chronic period following injury, including paralysis, altered sensation below the level of the injury, as well as aberrant autonomic responses. To date, the only FDA-approved treatment for acute SCI has been methylprednisolone, but that has fallen out of favor both domestically and internationally over the past decade. We have begun developing a cell-free, self-contained, acute-care, minimally invasive biomaterial treatment for both contusive and open SCIs that diminishes the onset and severity of secondary injury while simultaneously supporting and improving subsequent treatments (e.g., cell transplantation). Specifically, we investigated micro-scaffolds comprised of 3D-Painted Fluffy-PLG, an electrically conductive 3D-Graphene, and a blend of the two (Fluffy-Graphene). We have established the preliminary safety and lack of immunogenicity of micro-meshes made of 3D-printed Fluffy-PLG, 3D-Graphene, and Fluffy-Graphene when implanted in the spinal cords of both sham and injured rats. Additionally, all three materials demonstrate a reduction in the number of macrophages following contusive SCI when compared to injured rats without biomaterial implants. Further, we have shown that by acutely implanting the Fluffy-PLG into the injury site (via acute myelotomy), and then transplanting bone marrow mesenchymal stem cells (MSCs) directly into the biomaterial implant one day later, the immunomodulatory ability of MSCs is improved in comparison to those injured rats that received MSC transplants directly into the parenchyma at the same time point.

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Poster

P360. Spinal Cord Injury: Therapeutic Strategies: Animal Models

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Title: Dual functional hydrogel system for spinal cord regeneration with sustained release of arylsulfatase B alleviates fibrotic microenvironment and promotes axonal regeneration

Authors: *H. PARK[1,2], Y.-K. KIM[3,4], L. HONG[1,2], H. KIM[1,2], D. HWANG[1,5], X. JIN[1,2,6], M. KWON[1,5], S.-C. SONG[3,4], B. KIM[1,2,7];

[1]Ajou Univ. Sch. of Med., Suwon, Korea, Republic of; [2]Dept. of Brain Science, Dept. of Biomed. Sciences, Ajou Univ., Suwon, Korea, Republic of; [3]Ctr. for Biomaterials, Korea Inst. of Sci. and Technology, Seoul, 02792, Republic of Korea., Seoul, Korea, Republic of; [4]Div. of Bio-Medical Sci. and Technology, KIST School, Korea Univ. of Sci. and Technology, Seoul, 02792, Republic of Korea, Seoul, Korea, Republic of; [5]Dept. of Brain Science, Ajou Univ., Suwon, Korea, Republic of; [6]Dept. of Nephrology, Suqian First Hospital, Suqian 223800, Jiangsu Province, China., Jiangsu, China; [7]Dept. of Neurology, Ajou Univ. Sch. of Med., Suwon, Korea, Republic of

Abstract: Traumatic damage to the spinal cord does not spontaneously heal, often leading to permanent tissue defects with cystic cavities. Previously, WWwe previously showed have shown that injection of imidazole-poly(organophosphazene) hydrogel (I-5) bridges cystic spaces with newly assembled fibronectin-rich extracellular matrix (ECM) mediated by an imidazole moiety interacting with macrophages. The hydrogel-created ECM contains chondroitin sulfate proteoglycans (CSPGs), collagenous fibrils together with perivascular fibroblasts, and various fibrotic proteins, all of which could hinder axonal growth in the matrix. In an *in vitro* fibrotic scar model, cultured fibroblasts exhibited enhanced sensitivity to TGF-β1 when grown on CSPGs, which was prevented by recombinant arylsulfatase B (ARSB), a clinically approved enzyme to degrade CS carbohydrate chains. To alleviate the fibrotic microenvironment, the I-5 hydrogel was equipped with an additional function by making complex with ARSB via a hydrophobic interaction. Complexation of ARSB with the hydrogel allowed sustained release of the enzyme. Delivery of the I-5/ARSB complex significantly diminished the fibrotic ECM components in the lesioned spinal cord. The complex promoted serotonergic axonal growth into the hydrogel-induced matrix and enhanced serotonergic innervation of the lumbar motor neurons. Robust rRegeneration of the propriospinal axons labeled by AAV8-tdTomato was observed deepdeep into inside the matrix in animals with the I-5/ARSB complex, and some of the propriospinal axons grew to the lumbar spinal cord. and to the lumbar spinal cord was robustly increased. The enhanced axonal growth and neuronal innervation were accompanied by improved locomotor recovery in rats with the I-5/ARSB complex. Therefore, our dual functional system upgraded the functionality of the hydrogel for spinal cord regeneration by creating ECM to bridge tissue defects and concurrently facilitating axonal connections through the newly assembled ECM.
Keywords : Fibrotic microenvironment; injectable hydrogel; spinal cord injury; axon regeneration; arylsulfatase B; chondroitin sulfate proteoglycan

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Poster

P360. Spinal Cord Injury: Therapeutic Strategies: Animal Models

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Topic: C.11. Spinal Cord Injury and Plasticity

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Title: Assessing consequences of intra-spinal delivery of rAAV-retro to delete PTEN on recovery of upper extremity function following cervical spinal cord injury in mice

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Abstract: Previous studies have documented that conditional genetic deletion of phosphatase and tensin homolog (PTEN) enables regeneration of axons in the spinal cord after spinal cord injury (SCI). Here, we test efficacy of an approach using AAV vectors that are transported retrogradely from an injection site in the spinal cord to the majority of neurons that give rise to spinal tracts. We report results of 3 separate studies that use $PTEN^{f/f}$;$Rosa^{tdTomato}$ mice in which Cre induces expression of tdT and simultaneously deletes PTEN. In two studies, controls were $Rosa^{tdTomato}$ mice that received rAAV-retro/Cre; in the other, controls were $PTEN^{f/f}$;$Rosa^{tdTomato}$ mice that received rAAV-retro/GFP. 8-week old female and male mice received different genome copies (GC) of rAAV-retro at the same time as a dorsal hemisection injury at C5 (1E10 GC: $Rosa^{tdTomato}$ n=13, $PTEN^{f/f}$;$Rosa^{tdTomato}$ n=12; 6E9 GC: rAAV-retro/Cre n=9; rAAV-retro/GFP n=7; 3E9 GC: $Rosa^{tdTomato}$ n=15, $PTEN^{f/f}$;$Rosa^{tdTomato}$ n=25). The surgeon doing the injections was blind with respect to strain and vector. Forelimb motor function was assessed over time using a grip strength meter by individuals who were blind to treatment group. Following C5 lesions, average grip strength decreased to <15% of pre-operative control immediately after injury and then gradually recovered. In studies involving higher (1E10 GC) and mid (3E9 GC) doses of rAAV-retro/Cre, $PTEN^{f/f}$;$Rosa^{tdTomato}$ mice recovered more than respective controls. Across studies, the extent of recovery was dose dependent. Mice that received 1E10, 6E9, and 3E9 GC, recovered to a peak of 88.8%, 38%, and 12.4% of pre-operative control values respectively. Although $PTEN^{f/f}$;$Rosa^{tdTomato}$ mice that received rAAV-retro/Cre exhibited a greater initial recovery, a complication was the development of severe scratching behaviors suggesting intense sensations of itch and hindlimb motor dysfunction beginning at about 50-60 days post-injury. These late developing abnormalities were not observed in $Rosa^{tdTomato}$ control mice that received rAAV-retro/Cre or in $PTEN^{f/f}$;$Rosa^{tdTomato}$ mice that received rAAV-retro/GFP, indicating that the abnormalities are not due to the AAV itself or the Cre cargo it expresses. We conclude that, although intra-spinal injections of rAAV-retro/Cre in $PTEN^{f/f}$;$Rosa^{tdTomato}$ mice can lead to initial improvements in forelimb motor recovery after SCI, there are late-developing functional abnormalities with the experimental conditions used here.

Disclosures: M. Metcalfe: None. **O. Steward:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds);

OS is a co-founder and has economic interests in the company Axonis Inc, which holds a license on patents relating to PTEN deletion and axon regeneration..

Poster

P360. Spinal Cord Injury: Therapeutic Strategies: Animal Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P360.05

Topic: C.11. Spinal Cord Injury and Plasticity

Support: CIHR

Title: Development of a novel herpes simplex virus-thymidine kinase system for selection ablation of neural progenitor cells to facilitate mechanistic gain and loss of function studies

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Abstract: Background: Neural progenitor cell (NPC) transplants are a promising therapy for treating spinal cord injury (SCI), yet their long-term role after engraftment and their relative contribution to ongoing functional recovery remains a key knowledge gap. Selective human cell ablation techniques, currently being developed to improve the safety of progenitor cell transplant therapies in patients, may also be used as tools to probe the regenerative effects attributable to individual grafted cell populations. The Herpes Simplex Virus Thymidine Kinase (HSV-TK) and ganciclovir (GCV) system has been extensively studied in the context of SCI and broader CNS disease. However, the efficacy of brivudine (BVDU), another HSV-TK prodrug with potentially reduced bystander cytotoxic effects and in vivo toxicity, has yet to be investigated for NPC ablation. Purpose: This study aims to optimize the ablational efficiency of the HSV-TK system in the spinal cord through investigating the use of BVDU separately and in combination with GCV. Hypothesis: Combinatorial treatment of HSV-TK expressing NPCs with GCV and BVDU leads to increased cell ablation over treatment with only one prodrug. Results: In the in vitro cell ablation assay, we detected an approximately 80% reduction in the number of surviving HSV-TK+ NPCs when compared to controls for both GCV and BVDU treatments at the highest concentrations used (1 µg/mL) after 96 hours. Through a TUNEL assay, we found that there were differences in bystander effect and killing efficiency between GCV and BVDU at 1 µg/mL. GCV was found to ablate cells approximately 4% more than BVDU at 73.6% and 69.8% respectively, and the bystander killing effect of GCV was 27.6% compared to 12.1% with BVDU. We observed the highest killing efficiency of 89.3% when using a combination of 0.5 µg/mL of GCV and 0.5 µg/mL of BVDU, while the bystander effect was observed to be 25.4%,

2.2% below that of GCV alone. Following transplantation of HSV-TK+ hiPSC-NPCs into the cervical spine of the rat model, we were able to demonstrate that the largest elimination of the cells occurred when the rats were injected IV with a combination of GCV and BVDU over treatment with either prodrug alone. Conclusions: In this study, we demonstrated successful generation and ablation of HSV-TK-expressing human iPSC-derived NPCs. Our findings show enhanced ablation efficiency and reduced bystander effects when targeting all rapidly dividing cells with combinatorial GCV and BVDU treatment. However, for use in loss of function studies for a single proliferating cell subpopulation, BVDU alone is optimal due to reduced nonselective cell ablation.

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Poster

P360. Spinal Cord Injury: Therapeutic Strategies: Animal Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P360.06

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH Grant R01 NS101105 to WJA
University of Kentucky Professional Student Mentored Research Fellowship to ALS

Title: Intermittent hypoxia, sustained hypoxia, and normoxia induce differential patterns of HIF-1a and VEGF expression in the central nervous system of uninjured adult rats

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Abstract: Spinal cord injuries (SCIs) most commonly occur at the cervical level and often render injured people unable to breathe. Mechanical ventilation can preserve life despite respiratory motor paresis, but detrimentally affects quality of life and can lead to long-term complications including lung damage and respiratory muscular atrophy. Promisingly, moderate intermittent hypoxia (mIH) treatment can be used to induce increased respiratory output known as long term facilitation (LTF) after SCI, but recovery is transient. Seemingly subtle modifications of hypoxia treatment paradigm activate distinct cellular signaling pathways and thus respiratory motor response, even as originally demonstrated in uninjured laboratory animals. A better understanding of these signaling pathways will likely contribute to development of hypoxic dosing patterns more adaptive for achievement of full respiratory motor recovery after SCI. One key pathway involves hypoxia inducible factor 1α (HIF-1α) and vascular endothelial

growth factor (VEGF) activity. HIF-1α is a transcription factor induced by cellular hypoxia and increases in neurons after mIH treatment while VEGF is downstream and can induce LTF-like plasticity by exogenous application to the spinal cord both alone and combined with mIH treatment. However, it is unknown how endogenous HIF-1α and VEGF levels compare between animals treated with mIH and moderate sustained hypoxia (mSH). Our prediction is that a moderate level of HIF-1α and VEGF expression is adaptive for LTF and that both the low and high protein levels likely induced by normoxia and mSH treatments, respectively, will be outside the ideal range induced by mIH. Thus, with hypoxia treatment type as our independent variable, we hypothesize that mSH will induce the most HIF-1α and VEGF expression in the spinal cord and brainstem, followed by mIH and normoxia, in descending order. We treated uninjured adult Sprague-Dawley female rats with 5 consecutive days of either normoxia, mIH, or mSH (n=3 per treatment group) and harvested fresh whole central nervous system tissue (CNS) immediately following final treatment, measuring HIF-1α and VEGF via MSD and ELISA assays, respectively. Interestingly, results demonstrated that mSH induced the least expression of HIF-1α across CNS regions while paradoxically inducing the greatest VEGF expression of the three treatments. To understand this, future experiments should explore whether VEGF exerts feedback inhibition on HIF-1α expression following mSH by use of pharmacological inhibition or genetic knockout of VEGF prior to treatment with mSH and measurement of HIF-1α.

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Poster

P360. Spinal Cord Injury: Therapeutic Strategies: Animal Models

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Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH NS096514
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Title: Daily acute intermittent hypoxia therapy improves spontaneous urination pattern and reflex micturition event behaviors in rats with mid-thoracic moderate contusion spinal cord injury

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Abstract: Spinal cord injury (SCI) has a profound negative impact on bladder function, resulting in problems with urine storage and emptying of the bladder that lead to detrusor overactivity, inefficient voiding, and urine retention. Current approaches to treat bladder dysfunction following SCI are inadequate, and novel therapeutic strategies aimed at restoring bladder

function are needed. To this end, preclinical rat studies in our laboratory have been examining the impact of exposure to neuroplasticity-inducing acute intermittent hypoxia (AIH) as a strategy for improving bladder function following SCI. We have found that exposure to a single bout of AIH results in immediate short-term beneficial improvements in bladder function characterized by a reduction or elimination of non-voiding bladder contractions (NVCs), increased void volume and inter-contraction intervals (i.e., improved voiding efficiency), and decreased minimum bladder pressure (i.e., reduced urine retention). Here, we investigate whether daily AIH (dAIH) therapy for 7 consecutive days beginning at 1 week after SCI produces sustained beneficial improvements in bladder function in our contusion SCI model; daily normoxic exposures (dNx) served as a control. Examination of awake urination pattern using serial weekly overnight metabolic chamber measurements revealed that most rats exhibited functional voids that were infrequent but with large volumes at 1 week after SCI, and subsequent dAIH but not dNx treatment led to recovery of void frequency pattern (i.e., number of voids and inter-void interval) to pre-SCI levels by 4 weeks post-SCI, but with slightly increased void volumes. Further, in a subset of rats that exhibited no effective voiding events (i.e., small overnight total urine volume) at 1 week after SCI, dAIH but not dNx treatment produced partial recovery of micturition pattern behavior in a small subset of these rats by 4 weeks post-SCI. Examination of reflex micturition event (rME) behaviors elicited by continuous infusion of saline (0.04-0.09 ml/min) into the bladder under urethane anesthesia revealed that at 4 weeks after SCI (which corresponds to 2 weeks after dAIH), dAIH-treated rats exhibited predominantly a mild degree of bladder dysfunction characterized by a low incidence of NVCs and enhanced voiding efficiency, and multiple premature rMEs could be elicited by brief exposure to hypoxia. These findings demonstrate that dAIH treatment can lead to beneficial improvements in spontaneous and reflex bladder function behaviors that persist for at least two weeks following completion of dAIH therapy, and support the use of dAIH therapy to treat SCI-induced bladder dysfunction.

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Poster

P360. Spinal Cord Injury: Therapeutic Strategies: Animal Models

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Program #/Poster #: P360.08

Topic: C.11. Spinal Cord Injury and Plasticity

Support: DoD CDMRP Grant W81XWH-18-1-0753

Title: Contrast-enhanced ultrasound imaging to assess spinal cord injury severity and the efficacy of dural decompression in rodents

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Abstract: Traumatic spinal cord injury (tSCI) is characterized by a near complete loss of blood flow at the injury epicenter and substantial hypoperfusion in the penumbral zone, resulting in sustained ischemia and cell death. This ischemic deficit arises from progressive hemorrhage, edema, and associated increased intraspinal pressure (ISP). We hypothesize that the extent and magnitude of hemodynamic disruption following tSCI is indicative of the injury severity, and that acute dural decompression will alleviate elevated ISP and ameliorate tissue perfusion in the spinal cord parenchyma, with the intent of preserving the maximal amount of healthy tissue. In order to quantitatively evaluate spinal hemodynamics, we developed an ultrafast contrast-enhanced ultrasound imaging modality (CEUS) capable of interrogating microcirculatory blood flow within the entire spinal cord with high spatiotemporal resolution (100 um, 20 kHz). Epidural imaging was conducted following a three-level laminectomy (T7-T9) in female Long Evans rats. CEUS data were acquired at baseline, following an experimentally-induced contusion SCI (Infinite Horizon device; 100, 150, or 200 kDyn impact force), and after dural decompression (45 min post-injury) or no intervention. Behavioral testing was conducted weekly until 8 weeks post-injury (wpi). Preliminary results indicated that the area of perfusion deficit increased in response to increasing administered impact force (e.g., 0.42 mm^2, 1.9 mm^2, and 3.4 mm^2 perfusion deficit for mild to severe injuries, respectively). Area of perfusion deficit also correlated strongly with 2- and 8-wpi behavior in the “no intervention” groups as measured using the Basso, Beattie, Bresnahan locomotor rating scale ($R^2 = 0.76$; $p < 0.05$; N=6 animals per severity). With respect to decompression, resistance to blood flow in the penumbral zone rostral to the epicenter (quantified from CEUS using bolus inflow kinetics) increased significantly following tSCI (175% change from baseline); this was immediately ameliorated following durotomy (110% change from baseline). Similar results were observed with blood flow velocity (-28% change from baseline after SCI; -20% change from baseline after durotomy), suggesting this may be a promising neuroprotective intervention. Additional studies are currently underway to evaluate the effects of dural decompression on chronic functional outcomes and tissue perfusion.

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Poster

P360. Spinal Cord Injury: Therapeutic Strategies: Animal Models

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Program #/Poster #: P360.09

Topic: C.11. Spinal Cord Injury and Plasticity

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NINDS T32 5T32NS100663-04
2020 Susan and Isaac Wakil Foundation John Monash Scholarship

Title: Ultrasound Examination of Spinal Cord Perfusion in an Ovine Model

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Abstract: Ultrasound Examination of Spinal Cord Perfusion in an Ovine Model

Introduction

Patients with spinal cord injury require adequate spinal cord perfusion post-injury to avoid secondary damage to the cord from inflammation and ischemia. Currently, real time bedside imaging of spinal cord perfusion is non-existent. Post-operative imaging in SCI patients consists of MRIs to assess extent of decompression and CT scans to assess hardware integrity. Acute SCI patients are managed by augmenting blood pressure using pharmacological means and measuring mean arterial pressures. Developing an ultrasound based platform for real-time, portable perfusion measurements would help manage this patient population. We aim to develop such a platform using an ovine model.

Methods

To investigate spinal tissue perfusion, we used a GE Vivid™ iq Ultra Edition portable ultrasound system with a L8-18i intraoperative probe to image the spinal cords of two sheep. High resolution (sub-mm), broadband (16 MHz center frequency) 2D image cross-sections of the spinal cord were visualized in real time. We performed measurements of blood perfusion using the Doppler function. The sheep had an epidural electrode placed on the spinal cord and we made blood flow measurements during spinal cord stimulation.

Results

We were able to successfully characterize the intact and contused spinal cord using US intra-operatively. The white and grey matter were distinctly visualized. Hemorrhage had a distinct appearance on US from normal neuronal tissue. Doppler functionality allowed the visualization of the blood flow to the cord. When the animal was at rest, the blood flow pattern was symmetric, with regions of blood flow in both directions. When EES was applied, spikes in blood flow could be observed at distinct times when the stimulation was applied. Interestingly, the spatial locations of increased blood flow were related to the regions of stimulation.

Conclusion

Ultrasound imaging of the spinal cord has the potential to revolutionize post-injury care for patients with spinal cord injury. In preliminary work, we show that US is able to distinctly visualize the white and grey matter of the spinal cord. Moreover, spinal cord perfusion can be measured using US. Variations in spinal cord perfusion correlate with areas of increased activity.

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Poster

P360. Spinal Cord Injury: Therapeutic Strategies: Animal Models

Location: Virtual Only

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Topic: C.11. Spinal Cord Injury and Plasticity

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NINDS T32 Postdoctoral Training Program in Recovery and Restoration of CNS Health and Function (5T32NS100663-04)

Title: Mediolateral spinal local field potentials enable real-time decoding of gait cycle in an ovine model

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Abstract: Financial Support: This work was sponsored in part by the Defense Advanced Research Projects Agency (DARPA) BTO under the auspices of Dr. Alfred Emondi through the (Space and Naval Warfare Systems Center, Pacific OR DARPA Contracts Management Office) grant/contract nos. D15AP00112. SRP was supported by the 2020 Susan and Isaac Wakil Foundation John Monash Scholarship. JSC was supported by the NINDS T32 Postdoctoral Training Program in Recovery and Restoration of CNS Health and Function (5T32NS100663-04) under guidance of DAB.

Abstract: Spinal Cord Stimulation (SCS) delivered at titrated phases of the gait cycle has previously been shown to restore locomotor kinematics in the hindlimb of Rhesus monkeys and humans following a spinal cord injury. These techniques show great promise for restoring mobility and independence in people living with a spinal cord injury. Historically, real-time gait cycle detection algorithms have relied on electromyographic (EMG), kinematic, or corticographic recording techniques to identify the current gait phase. These techniques require additional integration of several sensors, resulting in a more complex system. In our work, we demonstrate lateralization of spinal Local Field Potentials (LFPs) recorded from two 24-contact epidural SCS arrays implanted along the lumbosacral spinal cord of two ovine models during

relevant hindlimb movements, including flexion, extension, adduction, abduction, rotation, and treadmill walking. Spinal LFPs were simultaneously recorded from all 48 electrodes, spanning the mediolateral width of the spinal cord, and extending along the L4-L6 vertebral segments. Following the identification of lateralized, time-locked responses to manipulated movements, spinal LFPs recorded during treadmill walking were decoded to predict gait cycle phase during locomotion. A decoder was constructed for each hind leg, and both decoders were trained on the same training dataset. The training data consisted of autoregressive (AR) coefficients computed over 300ms sliding epochs from approximately two minutes of treadmill walking LFP signals. Dimensionality was reduced by applying principal component analysis (PCA). The decoders predicted the gait cycle of each hindlimb independently. On a held-out validation data set, the swing-stance state fore each leg was predicted to an accuracy greater than 70%. Our results demonstrate promise for future real-time closed-loop SCS control schemes solely reliant on epidural spinal arrays for both gait prediction and restorative stimulation.

Disclosures: S.R. Parker: E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Patent Pending for Related Work. **J.S. Calvert:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Patent Pending for Related Work. **R. Darie:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Patent Pending for Related Work. **E. Shaaya:** None. **L.N. Govindarajan:** None. **M. Jung:** None. **T. Serre:** None. **B. McLaughlin:** A. Employment/Salary (full or part-time):; Employee of Micro-Leads Medical, who supplied equipment used in this study. **D.A. Borton:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Patent Pending for Related Work.

Poster

P360. Spinal Cord Injury: Therapeutic Strategies: Animal Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P360.11

Topic: C.11. Spinal Cord Injury and Plasticity

Support: The Paul Kalmanovitz Central Nervous System Repair Research Program

Title: Bilateral electrical stimulation of sciatic nerves in rat thoracic spinal cord contusion model as treatment for functional recovery

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Abstract: Spinal cord injuries (SCI) result in partial or complete loss of efferent and/or afferent connections between the brain and peripheral nerves, often yielding severe functional deficits for patients. While more than 300,000 people are currently living with a traumatic SCI in the U.S., there are currently no treatments that significantly aid in functional recovery. Presently, the only approved treatments for functional improvements post-SCI rely on rehabilitative strategies. Included in these strategies are functional electrical stimulation (FES) devices which utilize electrical stimulators to control specific muscles movements (i.e. stand, walk, grasp). There is evidence from our lab, using a stroke model, to suggest that electrical stimulation of peripheral nerves following neurological dysfunction promotes functional recovery via neuroplastic repair mechanisms. Thus, we hypothesized that a similar approach could be beneficial to functional recovery in a SCI model. Here, we used male, Long Evans adult rats to investigate peripheral nerve electrical stimulation as a treatment for functional recovery and/or neural plasticity after moderate, incomplete, thoracic spinal cord contusion. Daily electrical stimulation (30 min.) was applied bilaterally to the sciatic nerves via nerve cuff electrodes for 8 weeks and functional recovery assessed weekly by BBB, horizontal ladder walk, and von Frey filament testing. The SCI contusion model resulted in diminished hindlimb functional capacity as assessed by BBB and horizontal ladder walk. Additionally, SCI resulted in hindlimb hypersensitivity 9 days post-SCI. Preliminary results suggest that the electrical stimulation parameters used here (the same used for median nerve stimulation post-stroke) were not effective in improving functional recovery beyond spontaneous levels in our SCI model. We are still analyzing the lesion site and biotinylated dextran amine tracing from the hindlimb motor cortex to determine if peripheral nerve electrical stimulation effected neuroplasticity. Our results indicate that functional improvements via peripheral nerve electrical stimulation may rely on differing parameters based on the type and location of neurological deficit. Further work is continuing to test longer daily stimulation times, which may be needed post-SCI, in contrast to the improvement seen in the impaired forelimb as we have previously reported post-stroke.

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Poster

P360. Spinal Cord Injury: Therapeutic Strategies: Animal Models

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Topic: C.11. Spinal Cord Injury and Plasticity

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Title: Multisite targeted, activity-dependent spinal stimulation (MTADSS) promotes sex-dependent restoration of motor function after chronic cervical spinal cord injury

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Abstract: Cervical spinal cord injuries (SCI) damage descending motor pathways that control movements of the hands and arms. We are developing a novel therapy for chronic SCIs that uses targeted activity-dependent electrical spinal stimulation (TADSS) to improve motor recovery. TADSS is designed to strengthen spared descending pathways, based on principles of spike timing-dependent plasticity. We previously showed that a single site of intraspinal TADSS synchronized to volitional contractions of an impaired forelimb muscle improved performance of female rats in a reach-and-grasp task. For this study, we sought to improve TADSS therapy and investigate mechanisms of recovery. Four weeks after a moderate-severe cervical SCI, rats were implanted with an array of penetrating platinum-iridium microwires in the ventral horn of spinal segments C5-C7 (or epidural array of electrodes) and wires in 3 muscles of the impaired forelimb for electromyographic (EMG) recording. Starting 2 weeks after implantation, animals received therapy for 5 or 14 weeks. During the therapy period, TADSS animals received 3-channels of closed-loop spinal stimulation contingent on muscle activity of the impaired forelimb muscles. For 5 hours per day, including during reaching tasks, EMG from each muscle triggered sub-movement-threshold stimulation through a spinal electrode that could elicit contractions of that muscle with higher stimulus currents. Non-stimulated control animals received only behavioral training. Multi-site TADSS (MTADSS) promoted robust functional improvements in fine motor control and strength, but only in females. These functional benefits persisted with reduced physical training and no additional stimulation for 16 weeks after then end of the 14 week therapy period. Interestingly, 5 weeks of MTADSS therapy also restored arm function but the benefits did not persist after the end of the therapy, suggesting a minimum amount of therapy is needed to produce lasting behavioral effects. Recovery after 5 weeks was similar for TADSS delivered via intraspinal or epidural electrodes. In the 5 week study, males continued to show little functional recovery. We also periodically stimulated the contralesional motor cortex and recorded evoked potentials in the spinal cord. In animals that received MTADSS therapy, there was potentiation of the evoked corticospinal response. These results demonstrate optimizations for long term functional benefits of TADSS and identify plasticity in the corticospinal pathway as a potential mechanism driving recovery. Moreover, these data identify a profound need to understand why females show greater recovery than males.

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Poster

P360. Spinal Cord Injury: Therapeutic Strategies: Animal Models

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NIH Fellowship 5T32NS100663-04
2020 Susan and Isaac Wakil Foundation John Monash Scholarship

Title: Spatiotemporal distribution of electrically evoked spinal compound action potentials during spinal cord stimulation

Authors: *J. S. CALVERT[1], R. DARIE[1], S. R. PARKER[1], E. SHAAYA[2], S. SYED[2], B. L. MCLAUGHLIN[3], J. S. FRIDLEY[2], D. A. BORTON[1,4,5];
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Abstract: Recent studies using Spinal Cord Stimulation (SCS) have demonstrated restoration of motor function in individuals previously diagnosed with chronic spinal cord injury (SCI). In parallel, recent advances in SCS have indicated the use of spinal evoked compound action potentials (ECAPs) in closed-loop devices to titrate delivery of stimulation in the context of chronic pain and in further understanding the mechanisms of SCS function. However, the previous characterizations of the ECAPs recorded during SCS were performed with one-dimensional electrode leads (i.e., percutaneously placed leads). Here, we describe the spinal ECAPs elicited using clinically and physiologically relevant SCS parameters and stimulation locations across both the rostral-caudal and medial-lateral directions of the sheep spinal cord. In two sheep, two 24-contact epidural SCS arrays were implanted along the lumbosacral spinal cord. Spinal ECAPs were recorded during SCS from the non-stimulating contacts of the epidural array, which were synced to bilateral electromyography (EMG) recordings from six back and lower extremity muscles. We observed a distinct triphasic P1, N1, P2 peak morphology and propagation in the ECAPs during midline and lateral stimulation. Spinal ECAP recruitment spatially and temporally correlated with lower extremity EMG recruitment. Interestingly, distinct regions of lateral stimulation produced both triphasic spinal ECAPs and motor evoked responses measured via EMG, while others exhibited neither response type. Furthermore, when high frequency SCS (e.g., 10kHz) was used to "block" low frequency SCS at a caudal location, spinal

ECAPs could still be observed rostral to the high frequency SCS. These results suggest that spinal ECAPs can be used to investigate the effect of SCS on spinal structures, and may be informative in strategies to optimize the clinical benefit of SCS in the context of both managing chronic pain and restoration of function after SCI.

Disclosures: **J.S. Calvert:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Patent Pending on Related Work. **R. Darie:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Patent Pending on Related Work. **S.R. Parker:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Patent Pending on Related Work. **E. Shaaya:** None. **S. Syed:** None. **B.L. McLaughlin:** A. Employment/Salary (full or part-time):; Employee of Micro-Leads Medical Inc., a medical device technology company whose technology was used in this study. **J.S. Fridley:** None. **D.A. Borton:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Patent pending on related work.

Poster

P409. Plasticity and Reorganization: Cutaneous Neurons to Cortex

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P409.01

Topic: D.04. Somatosensation – Touch

Support: NINDS F31NS103439 to RCC
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Title: In vivo time lapse imaging reveals dynamics of sensory neuron structural remodeling in adult mouse skin

Authors: *R. C. CLARY, E. A. LUMPKIN;
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Abstract: Sensory afferents that innervate the skin and encode thermal, noxious, itch, and tactile stimuli must maintain neuronal architecture despite the continuous postnatal renewal of target tissues. For example, myelinated Aβ afferents that innervate epidermal Merkel cells and respond to gentle touch stimuli must maintain connections with Merkel cells even though the cells that make up the epidermis, including Merkel cells, undergo constant renewal. To address this gap in knowledge, we developed a technique utilizing time-lapse *in vivo* two photon imaging of Merkel cells and afferent terminals.
We imaged adult transgenic mice expressing Atoh1-GFP to label Merkel cells and TrkC-tdTomato to label Aβ afferents every three days for approximately one month. Over this time

period, we noted that both Merkel cells and afferent terminals were highly plastic, with afferent terminals undergoing frequent growth and regression, as well as both Merkel cells and terminal branches being added or removed. Plasticity of Merkel cells and terminal neurites was positively correlated, but almost every time-lapse session had a higher percentage of neurites displaying plasticity than Merkel cells. Surprisingly, there were many datapoints where terminal neurites displayed plasticity even though Merkel cells were stable. Analysis of the temporal trajectories of plasticity in neuronal terminals and Merkel cells showed that plasticity occurs in phases with afferents going through periods of active remodeling and periods of quiescence. Ongoing analyses will test whether there are any biological factors that relate to temporal profiles of plasticity.
Using the TrkC-tdTomato reporter allowed us to visualize the entire extent of afferent terminals, where previous studies using Neurofilament Heavy antibodies only label the intermediate filament cytoskeleton. This reporter revealed two morphologically distinct types of axonal terminals that make apparent contacts with Merkel cells. The canonical Merkel-cell contact, which we term a kylix, cups the majority of the Merkel cell from underneath. A subset of endings lack this cup and instead appear to contact Merkel cells with a modest swelling; we term these boutons. We show that kylikes form long-lasting connections with Merkel cells while boutons are more transient. Ongoing analyses will test whether there is a defined trajectory, similar to development, in which branches grow out into boutons before becoming full kylikes.

Disclosures: R.C. Clary: None. **E.A. Lumpkin:** None.

Poster

P409. Plasticity and Reorganization: Cutaneous Neurons to Cortex

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P409.02

Topic: D.04. Somatosensation – Touch

Support: Medical Student Summer Research Program

Title: Cutaneous nerves depend on their target for proper innervation pattern in a time-dependent fashion

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Abstract: The current study was designed to address the question of whether a normal target is required to provide the necessary cues for a proper innervation pattern. The spontaneous mutation of the skin in nude mice that lack hair and exhibit defective formation of hair follicles

provides an excellent experimental model to test for defective targeting for cutaneous nerves. The innervation of skin in normal and nude mice was compared using three areas of the skin: dorsal skin, paw skin, and the skin of the snout. To label all populations of nerves, pan-neuronal markers such as monoclonal anti-acetylated tubulin, polyclonal anti-PGP 9.5 and anti-neurofilament were used to label nerve fibers. In addition, rabbit polyclonal anti-keratin14 was used to label keratinocytes of the hair follicle and surface epidermis in double labeling experiments. The patterns of the cutaneous nerves were disrupted in the dorsal skin in the nude mice with complete loss of the normal plexiform arrangement shown in normal mice. Innervation patterns of the paw skin and snout were normal in nude mice. We concluded that normal hair, and hair follicles are required for normal cutaneous nerve plexus formation in the skin. To address whether a normal target can correct the innervation errors of nude skin, normal skin was grafted on the dorsal interscapular region of nude mice. Analysis of innervation patterns of grafted skin after three weeks suggested that grafting normal skin in adult nude mice cannot recapitulate the patterns achieved during normal development.

Disclosures: K. Rejent: None. **W.R. Gherman:** None. **P.W. Frank:** None. **D.R. Giovannucci:** None. **A. Maklad:** None.

Poster

P409. Plasticity and Reorganization: Cutaneous Neurons to Cortex

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P409.03

Topic: D.04. Somatosensation – Touch

Title: Plasticity of feed-forward pathways during whisker dependent sensory association training

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Abstract: Sensory responses are constructed from the interaction of both feed-forward sensory input as well as feedback information from the thalamus and other cortical areas. Previous work has shown that feedback pathways, including input from higher-order thalamic areas, are potentiated during sensory learning (Audette et al., 2018; Pardi et al., 2020), but it remains unknown how learning influences feedforward pathways in primary sensory cortex. Here we evaluated changes in synaptic strength from layer 4 (L4), the main input layer of the cortex, onto L2/3 pyramidal neurons during sensory association training in a whisker-dependent learning task (Bernhard et al., 2020). Because prior studies have shown that L4 to L2/3 synapses can be potentiated both in vivo and in vitro by whisker stimulation (Gambino and Holtmaat, 2012; Williams and Holtmaat, 2019), we predicted that sensory learning in a whisker-dependent association task would be associated with a significant increase in L4 input to L2/3 pyramidal neurons. Using the L4-restricted Scnn1a-Cre transgenic mouse for cell-type specific ChR2

expression, we evaluated changes in the strength L4 inputs across the learning trajectory. Surprisingly, we found that sensory association training did not potentiate L4 inputs onto L2/3 pyramidal neurons, despite robust synaptic plasticity at other cortical synapses. Based on these data, we hypothesize that sensory learning does not augment feedforward sensory inputs. Previously defined increases in synaptic strength from higher-order thalamic inputs (Audette et al., 2019) suggests that feedback pathways, but not feed-forward pathways, may be selectively enhanced during learning.

Disclosures: J.A. Christian: None. **E. Park:** None. **A.L. Barth:** None.

Poster

P409. Plasticity and Reorganization: Cutaneous Neurons to Cortex

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Program #/Poster #: P409.04

Topic: D.04. Somatosensation – Touch

Support: JST ERATO JPMJER1801
JSPS Grants-in-Aid for Scientific Research 20K15926
JSPS Grants-in-Aid for Scientific Research 18H05525

Title: Assessment of texture representation in the rat primary somatosensory cortex using deep learning

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Abstract: In this study, we took a deep-learning-based approach to decode complex tactile information from neuronal activity of a rat. Texture of a surface is high-dimensionally encoded in the somatosensory cortex (S1) and cannot be decoded from activity of a single neuron. Therefore, we focused on the local field potentials (LFPs), which collectively reflect the synaptic activity of thousands of neurons. To record neural activities from a wide region of the S1, we designed a novel 32-channel electrode array. The electrode array was implanted on the right primary somatosensory cortex, specifically including forelimb (S1FL) and hindlimb (S1HL) regions and was lowered to the depth of cortical layers 2/3. Then the animal was placed in an open field chamber (90 x 90 cm) with two different floor textures in either “dark” or in “light” conditions. LFPs were continuously recorded while the rat explored the field. All the electrophysiological data were collected under free-moving conditions at a sampling rate of 2 kHz. For classification, the channels that successfully recorded LFPs were selected for each rat. A convolutional neural network (CNN) model was used to classify the LFPs, and the score was evaluated using *k*-fold cross-validation ($k = 5$). Data were separated into 2-second bins and were predicted for each bin. From the LFPs in the “light” condition, our CNN model yielded average

classification accuracy up to 0.72. Further analysis will reveal how experiencing “dark” condition affects the representations of the floor textures.

Disclosures: K. Yamashiro: None. **Y. Ikegaya:** None. **N. Matsumoto:** None.

Poster

P409. Plasticity and Reorganization: Cutaneous Neurons to Cortex

Location: Virtual Only

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Program #/Poster #: P409.05

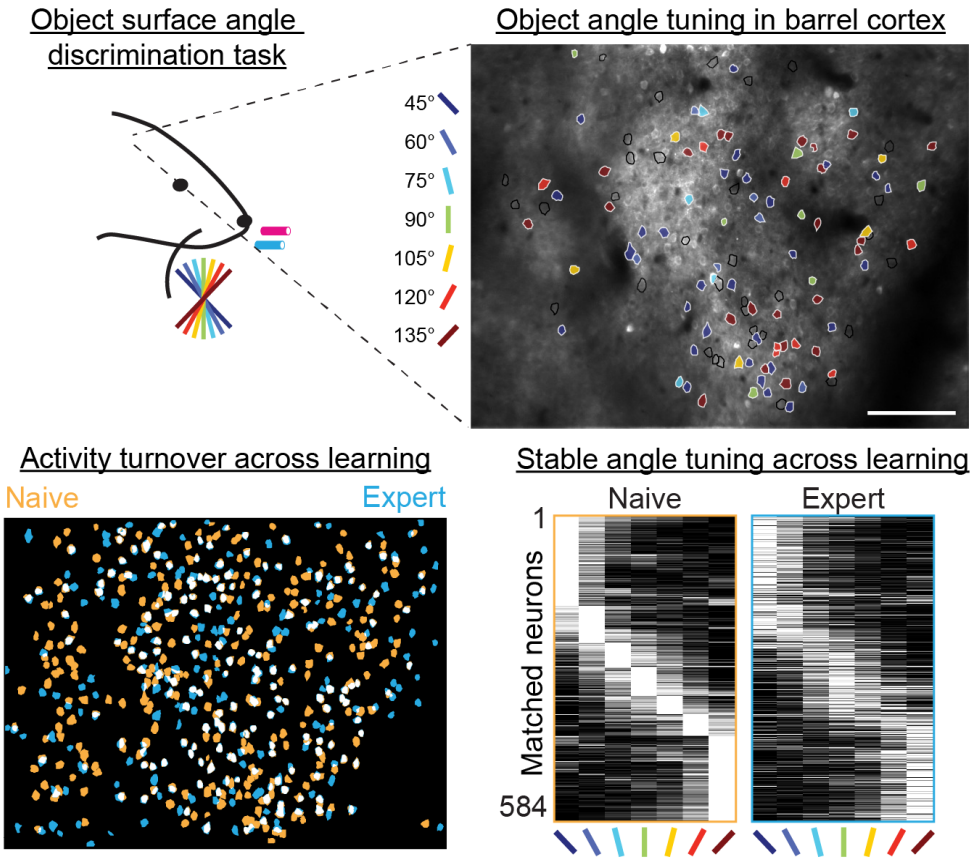
Topic: D.04. Somatosensation – Touch

Support: NIH/NINDS New Innovator Award 1DP2NS105554

Title: Behavioral and neural bases of tactile shape discrimination learning in head-fixed mice

Authors: *J. KIM[1], A. ERSKINE[1], J. CHEUNG[1], S. A. HIRES[2];
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Abstract: Tactile object recognition is an active behavior that requires both the perception of three-dimensional object shape and the association of its meaning through learning. The primary element that defines three-dimensional shape is object surface angle. Thus, we investigated how object surface angles are represented in the brain, how this representation is constructed from sensory inputs, and how it reorganizes across learning. Head-fixed mice learned to discriminate object surface angles by active exploration with one whisker. Two-photon calcium imaging of excitatory neurons in layers 2-4 of barrel cortex revealed maps of object-angle tuning before and after learning. Three-dimensional whisker tracking and mathematical modelling demonstrated that the sensory input components that best discriminate angle (vertical bending and slide distance) also have the greatest influence on object-angle tuning. This match between behavioral component and neural component suggests efficient encoding scheme of object shape in primary somatosensory cortex. To investigate how learning influences the representation of object surface angles, we compared response patterns of the same set of neurons before and after learning. About 40% of active neurons in naïve sessions became silent in expert sessions, and vice versa. Despite this high activity turnover rate, both the population distribution of object-angle preferences and individual preferences remained stable after learning. Object angle discrimination learning did not involve changes in motor strategy or sensory inputs. However, population activity pattern became better at discriminating object surface angles after learning. These results show how discrimination training enhances stimulus selectivity in the primary somatosensory cortex while maintaining perceptual stability.



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Poster

P409. Plasticity and Reorganization: Cutaneous Neurons to Cortex

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P409.06

Topic: D.04. Somatosensation – Touch

Support: European Research Council (ERC) Consolidator grant (ARTTOUCH No. 772242)

Title: Effect of multidigit scanning on tactile discrimination

Authors: M. DIONE, H. DAUMAS, *R. ACKERLEY;
Lab. de Neurosciences Cognitives UMR7291, CNRS - Aix-Marseille Univ., Marseille, France

Abstract: Intensive synchronous multidigit tactile training, for example when reading Braille with several fingers or playing a music instrument, typically leads to experience-dependent

changes both at behavioral and cortical levels. At a behavioral level, tactile sensitivity and static spatial acuity are improved for the fingers stimulated conjointly during training. However, a tendency to confuse which finger is touched by a light tactile stimulus (mislocalization) has also been observed. At a cortical level, changes have been observed in the primary somatosensory cortex. The cortical representations of the trained fingers enlarge, superimpose, merge and reorganize in a way that does not respect the somatotopy. Reorganization at a cortical level has been linked to the mislocalization phenomenon. It is unclear how these changes affect tactile discrimination capacities, especially when considering dynamic tasks such as Braille reading. How do we reconcile multidigit Braille reading with mislocalizations? Is there an advantage in using multiple fingers to process tactile information or are the changes observed side effects disadvantageous for the performance? In the present study, to begin addressing those questions, we evaluated the tactile discrimination abilities of non-expert participants (N = 21) on a dynamic active task of tactile discrimination (gratings composed of parallel lines with different inter-band spacing) using one or several fingers (index, middle, ring). Tactile sensitivity and mislocalizations were evaluated before, after and in between the blocks of the discrimination task. Synchronous multidigit tactile experience (SMTE) was characterized using a questionnaire adapted from Guest et al. (2014). Results revealed that discrimination abilities were better with the index or the middle finger, compared to the ring finger in the unidigit blocks. The multidigit performance was as good as with the best finger. Mislocalizations tended to increase after the multidigit block, with the middle finger named more than the other fingers. This suggests that the mechanisms described earlier may have been initiated while performing the task with several fingers. Tactile sensitivity was not affected by the task. Finally, the participants with more SMTE had lower multidigit discrimination abilities. Altogether, our results support an equivalence rather than a disadvantage in using multidigit scanning strategies to process tactile information. However, SMTE may be disadvantageous to multidigit scanning discrimination. Further studies, including Braille readers or musicians are needed to confirm the latter result.

Disclosures: M. Dione: None. **H. Daumas:** None. **R. Ackerley:** None.

Poster

P409. Plasticity and Reorganization: Cutaneous Neurons to Cortex

Location: Virtual Only

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Program #/Poster #: P409.07

Topic: D.04. Somatosensation – Touch

Support: BMBF INOPRO project, 16SV7656
DFG EXC 1086

Title: Corticomuscular coherence during upright standing in unilateral transfemoral amputees

Authors: *C. PASLUOSTA[1], B. MEYER[1], T. KRAUSKOPF[1], K. FUCHS[1], T. BALL[2], M. MUELLER[3], L. KLEIN[2], G. W. HERGET[2], T. STIEGLITZ[1];
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Abstract: Restoring sensory feedback by electrically stimulating nerve pathways improves balance and locomotion in lower limb amputees. Yet, bidirectional prostheses for the lower limb fail to completely restore function and sensory feedback. We argue that a major drawback in the advancement toward near-natural limb function restoral is our insufficient understanding of the structural and neurophysiological alterations that occur post-amputation. These neuromuscular adaptations occur to cope with a disrupted feedback system, loss of mechanical degrees of freedom and psychophysical alterations. Thus, quantifying these adaptations by objective measures may improve the design of stimulation paradigms to restore sensory feedback in amputees. In this work, we investigated corticomuscular coupling (CMC) during quiet standing to study adaptations in the driving neural mechanisms behind balance control in lower limb amputees. Ten patients with a unilateral transfemoral amputation and ten age-matched able-bodied control subjects were recruited. 32-channel electroencephalographic (EEG) recordings and electromyographic (EMG) data from the trunk and from the intact/dominant lower limb muscles were collected during quiet standing. Participants performed five 30-second trials with eyes open, eyes closed and while performing dual task. Data were band-pass filtered from 1 Hz to 100 Hz, notch filtered around 50 Hz and downsampled to 250 Hz. Large artefacts within the EEG data were removed by subtracting the artefact components from the original signal using wavelet transform. Further artefacts in the EEG data were removed through independent component analysis, after re-referencing the EEG data to a common average reference. To asses CMC, a multivariate autoregressive (MVAR) model was fitted, from which the normalized partial directed coherence (nPDC) was computed using the in Matlab implemented toolbox SIFT. We focused on CMC in the alpha, beta and gamma frequency ranges. Further, nPDC provides a measurement of the strength of the corticomuscular coupling as well as the direction of the coupling. This made it possible to investigate sensory feedback loops that occur during the control of posture. We observed distinct differences in coherence between the group of amputees and controls for the efferent and the afferent directions. Distinct differences in coherence were also observed between the afferent and the efferent direction within the group of amputees. Results are discussed in the context of balance control to explain sensorimotor mechanisms possible disrupted by the amputation.

Disclosures: C. Pasluosta: None. **B. Meyer:** None. **T. Krauskopf:** None. **K. Fuchs:** None. **T. Ball:** None. **M. Mueller:** None. **L. Klein:** None. **G.W. Herget:** None. **T. Stieglitz:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Cortec, neuroloop.

Poster

P409. Plasticity and Reorganization: Cutaneous Neurons to Cortex

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P409.08

Topic: D.04. Somatosensation – Touch

Support: ERC Starting Grant (715022 EmbodiedTech)
Wellcome Trust Senior Research Fellowship (215575/Z/19/Z)

Title: Are phantom referred sensations a perceptual consequence of S1 remapping?

Authors: *E. AMORUSO[1], D. B. TERHUNE[2], M. KROMM[1], D.-A. MURET[1], T. R. MAKIN[1]; [1]Univ. Col. London, London, United Kingdom; [2]Goldsmiths, Univ. of London, London, United Kingdom

Abstract: Seminal studies (Ramachandran et al., 1992, *Science;* Ramachandran et al., 1993, *PNAS)* have shown that touches on the face can evoke topographically organized sensations on the phantom hand in some amputees (referred sensations). This striking phenomenon has been classically operationalized as the perceptual correlate of the cortical remapping of the face onto the neighboring missing-hand territory in the primary somatosensory cortex (S1). Here, we investigate whether reports of referred sensations can be, at least partially, influenced by previously uncontrolled experimental factors, such as demand characteristics and suggestibility (Lush et al., 2020, *Nature Communications*). Unilateral upper-limb amputees (N=17), congenital one-handers (N=19), and able-bodied controls (N=20) were repeatedly stimulated with PC-controlled vibrations on several body-parts. They were asked to report on each trial the occurrence of any dual sensation on the hand(s) (including the phantom in amputees). Before the experiment, participants were told that they would receive 'special' vibrations (found to be able to evoke dual sensations) intermixed with 'classical' vibrations. Results show that manipulating expectations about the occurrence of the phenomenon was sufficient to induce a significant amount of referred sensations in all groups. Crucially, referred sensations reports were not more frequent in amputees than in the other groups and did not occur more often following stimulation of the face or body-parts ipsilateral to the missing hand. Together, these findings generally weaken the hypothesis that referred sensations are a behavioural consequence of post-amputation S1 remapping.

Disclosures: E. Amoruso: None. **D.B. Terhune:** None. **M. Kromm:** None. **D. Muret:** None. **T.R. Makin:** None.

Poster

P409. Plasticity and Reorganization: Cutaneous Neurons to Cortex

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Program #/Poster #: P409.09

Topic: D.04. Somatosensation – Touch

Support: Prodex (BELSPO)
Fonds de la Recherche Scientifique – FNRS

Title: Fast grip force adjustment to friction relies on localized fingerpad strains

Authors: *B. DELHAYE[1], F. SCHILTZ[2], F. CREVECOEUR[3], J.-L. THONNARD[4], P. LEFEVRE[5];
[1]Univ. catholique de Louvain, Woluwe-saint-Lambert, Belgium; [2]Univ. catholique de Louvain, Brussels, Belgium; [3]Univ. Catholique de Louvain, Louvain-la-Neuve, Belgium; [4]Univ. Catholique De Louvain, Brussels, Belgium; [5]ICTEAM and Inst. of Neuroscience, Univ. catholique de Louvain, Louvain-la-Neuve, Belgium

Abstract: When lifting objects of different mass or surface, humans adjust the grip force applied to the object such that the amount of force is minimal. In particular, it has been shown that, after changing the friction of the object-skin interface, humans can quickly adjust their grip force such that a slippery surface is squeezed more firmly than a sticky surface. To perform this adjustment, the central nervous system relies on the feedback from the mechanoreceptive afferents innervating the fingertip skin at or near the contact surface with the object. These tactile afferents encode information related to skin deformation and therefore the nature of the feedback signaling a change in friction must somehow originate from a difference in the way the skin deforms when contacting and interacting with objects of different friction.
To better understand the nature of this tactile feedback, we asked human participants to perform a grip-lifting task with a manipulandum equipped with an optical imaging system. This system enabled recording images of the fingertip-object contact and monitoring the time-varying skin deformations through transparent plates of glass. The plates had different levels of friction and were interchanged unexpectedly between blocks of 5 trials.
We observed that, following a change in friction, participants adjusted the level of grip force accordingly within 370ms after contact with the surface. By comparing the deformation patterns when switching from a high to a low friction condition, we found a significant increase in skin deformation inside the contact area, resulting from significantly larger partial slips. This increase became significant more than 100ms before the adjustment of grip force in response to changes in friction, suggesting that local and partial deformation patterns before lift-off are used by the central nervous system to adjust the grip force as a function of the frictional condition.

Disclosures: B. Delhaye: None. **F. Schiltz:** None. **F. Crevecoeur:** None. **J. Thonnard:** None. **P. Lefevre:** None.

Poster

P409. Plasticity and Reorganization: Cutaneous Neurons to Cortex

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Program #/Poster #: P409.10

Topic: D.04. Somatosensation – Touch

Support: European Research Council (ERC) Consolidator grant (ARTTOUCH No. 772242)

Title: How drops of water are encoded and perceived by humans

Authors: *L. SAMAIN-AUPIC, M. DIONE, J.-M. AIMONETTI, R. H. WATKINS, R. ACKERLEY;
Lab. de Neurosciences Cognitives - UMR 7291, Aix-Marseille Univ. - CNRS, Marseille, France

Abstract: We have receptors in our skin that allow us to sense and interact with liquids in our environment, for example, changing our grip force on a wet object so that it does not slip or for determining if it is raining. As humans do not have hygroreceptors, wetness perception relies on the integration of tactile information conveyed by low-threshold mechanoreceptors and/or temperature information conveyed by thermoreceptors. Here, we investigated how participants perceived drops of water at room temperature, by modulating the height and the mass of a drop. This acuity was tested on 20 healthy young adults using 5 drops of different masses (10, 20, 25, 40, and 50mg) or by applying the same mass of drop, but at 4 different heights (6, 12, 18, and 24 cm). The drops were applied to the palm of the hand using 1 ml syringes with different end attachments and the skin was dried between drops. For the drop mass task, participants had to compare 48 pairs of drops with one reference on each pair and had to say which one was more intense. Furthermore, in a different task, they were asked to estimate the intensity of 60 single drops of water using an unbounded scale. In the drop height task, they were asked to do the same, but estimate the intensity on 48 drops released at 4 different heights. In addition, we tested tactile sensitivity (calibrated monofilaments) and cool/warm thermal perception (QST, France) of the same skin, to compare tactile and thermal sensitivity to the perception of drops of water. We showed, with psychometric measures that the minimum difference for perceiving a difference in drop mass is 9.2mg. The second task showed that the participant needed a drop mass between 5-10 mg to perceive a difference. The height task showed that the drop intensity was perceived to increase with height. We also investigated how and which mechanoreceptors are activated by a drop of water with microneurography. We inserted a microelectrode in the median nerve of healthy participants in order to locate and stimulate mechanoreceptors. For each single afferent identified, we released 10 drops of water at room temperature 5 cm above the receptive field of the unit. Our preliminary results showed that only fast-adapting type II (Pacinian) mechanoreceptive afferents are activated following drops of water. In conclusion, there exists very little in the literature concerning our perception of wetness and we presently show that humans are able to discriminate very well between different intensities of sensation provided by changing the mass and height of drops of water. Moreover, we the perception of these drops of water seems to be encoded mainly by Pacinian mechanoreceptors.

Disclosures: L. Samain-Aupic: None. **M. Dione:** None. **J. Aimonetti:** None. **R.H. Watkins:** None. **R. Ackerley:** None.

Poster

P424. Chemical Senses: Behavior and Perception

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Program #/Poster #: P424.01

Topic: D.05. Olfaction and Taste

Support: Wu Tsai Neuroscience Institute 'Big Ideas'
HEAL supplement to R35NS105092-S1

Title: Exploring the C. elegans response to plant-derived compounds salvinorin A and analogs

Authors: *T. A. LOGAN-GARBISCH[1], S. GUHA[4], E. FRYER[6], L. ROGEL[4], E. REZAEI[4], I. MOLLHOFF[5], A. NEKIMKEN[4], A. XU[6], S. FECHNER[4], A. DAS[4], J. CASAR[4], H. FARAH[4], L. A. O'CONNELL[2], S. DRUCKMANN[2], T. R. CLANDININ[3], S. Y. RHEE[6], M. B. GOODMAN[1]; [1]Mol. and Cell. Physiol., [3]Neurobio., [2]Stanford Univ., Stanford, CA; [4]Mol. and Cell. Physiol., [5]Biol., Stanford, Stanford, CA; [6]Carnegie Inst. for Sci., Stanford, CA

Abstract: Plants and animals have co-evolved over millions of years, which has provided tools to the immobile plants in the form of chemical compounds that affect animal behavior. Additionally, humans have traditionally cultivated plants for both food and medicinal purposes, allowing for changes in relative abundance of the plants themselves as well as alterations in the amounts of specific chemicals (e.g. capsaicin content in peppers). *Salvia divinorum*, or diviner's sage, is a cultivated, clonal plant with psychoactive properties (hallucination, pain relief) in humans, known to act primarily via ***K***;-opiod receptors binding the terpenoid salvinorin A. However, *Salvia divinorum* didn't evolve to relieve pain in humans, and we know much less about its mechanism of action in non-mammalian animals. We are exploring the chemotactic responses of *C. elegans* nematodes to salvinorin A and several analogs, as well as beginning to categorize the neurons that might detect these compounds. We focused on the use of chemotaxis behavioral assays to determine attraction, repulsion, or indifference to plant-derived compounds, assessing wild-type (N2) animals alone, or N2 and mutant animals together. We conducted these behavioral assays with a minimum of three independent trial replicates, targeting a sample size of 150-300 worms per trial. We use solvent as the compound control (DMSO or water), and for larger screens, we also used known chemical attractants and repellents as reference compounds. Our team developed automated image analysis code to determine the positions of the worms along a chemical gradient and used that information to compute a standard chemotaxis index; genotype and compound remained masked until we completed data analysis. We used day 1 adult hermaphrodites reared in standard conditions for this study. So far, we have discovered that salvinorin A acts as a chemoattractant to wild-type (N2) animals, and preliminary data suggest

that all 3 analogs tested do as well. Interestingly, the salvinorin A response is mediated through two sets of chemosensory neurons: tax-4 expressing neurons and osm-9 expressing neurons, which are mutually exclusive categories. This suggests that in *C. elegans*, there is at least one, (and possibly more than one) receptor responding to salvinorin A which is certainly not a ***K***;-opiod receptor. We are currently exploring the response to the analogs in more depth, and we plan to genetically dissect which specific chemosensory neurons are involved in the detection of salvinorin A for our immediate next steps. Our data provide some intriguing insights into an invertebrate response to salvinorin A and its analogs.

Disclosures: T.A. Logan-Garbisch: None. **S. Guha:** None. **E. Fryer:** None. **L. Rogel:** None. **E. Rezaei:** None. **I. Mollhoff:** None. **A. Nekimken:** None. **A. Xu:** None. **S. Fechner:** None. **A. Das:** None. **J. Casar:** None. **H. Farah:** None. **L.A. O'Connell:** None. **S. Druckmann:** None. **T.R. Clandinin:** None. **S.Y. Rhee:** None. **M.B. Goodman:** None.

Poster

P424. Chemical Senses: Behavior and Perception

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P424.02

Topic: D.05. Olfaction and Taste

Support: NIH Grant GM129576
NIH Grant DA047645

Title: Mechanism for the munchies: endocannabinoid modulation of food preferences in C. elegans

Authors: *A. LEVICHEV, S. FAUMONT, R. Z. BERNER, S. R. LOCKERY;
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Abstract: The mammalian endocannabinoid system, which is comprised of cannabinoid receptors CB1 and CB2 and their endogenous ligands, is thought to integrate internal energy state and sensory food cues to modulate feeding. For example, cannabinoids acting on CB1 can increase preference for more palatable, calorically dense food: a response called *hedonic amplification*, colloquially known as “the munchies.” In mammals, cannabinoids can increase sensitivity to odors and sweet tastes, which may underlie hedonic amplification. We are developing *C. elegans*, an omnivorous bacterivore, as a model in which to investigate the neurophysiology of hedonic amplification. We have found that exposure to N-arachidonoyl-ethanolamine (AEA), an endogenous cannabinoid common to both mammals and *C. elegans*, increases the worms’ preference for more palatable bacteria over less palatable bacteria, mimicking hedonic amplification in mammals. Furthermore, AEA acts bidirectionally, increasing consumption of more palatable bacteria while decreasing consumption of less

palatable bacteria. We also found that deletion of the CB1 ortholog, *npr-19*, eliminates hedonic amplification, which can be rescued by expression of the human CB1, establishing a humanized worm for cannabinoid signaling studies. Deletion of the olfactory neuron AWC, which directs chemotaxis to food, abolishes hedonic amplification. Consistent with this finding, calcium imaging revealed that AEA bidirectional modulates AWC activity, increasing its responses to more palatable food and decreasing its response to less palatable food. However, GFP expression analysis revealed that *npr-19* is expressed ~21 neuron classes but, surprisingly, not in AWC. Although AEA's effect could be mediated by NPR-19-expressing neurons presynaptic to AWC, nearly complete elimination of fast synaptic transmission, via the mutation *unc-13(e51)*, has no effect on modulation. Instead, AEA's effect on AWC is mediated via *unc-31*-dependent dense-core vesicle release. We are now attempting to disambiguate the other neurons involved in AEA's effect on AWC and the worms' feeding preferences.

Disclosures: A. Levichev: None. **S. Faumont:** None. **R.Z. Berner:** None. **S.R. Lockery:** None.

Poster

P424. Chemical Senses: Behavior and Perception

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P424.03

Topic: D.05. Olfaction and Taste

Support: NSF/CIHR/DFG/FRQ/UKRI-MRC Next Generation Networks for Neuroscience Program

Title: Novelty detection in early olfactory processing of the honey bee, Apis mellifera

Authors: *H. LEI[1], S. HANEY[2], C. M. JERNIGAN[3], X. GUO[4], C. N. COOK[5], M. V. BAZHENOV[6], B. H. SMITH[1];
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Abstract: Animals are constantly bombarded with stimuli, which presents a fundamental problem of distinguishing between pervasive uninformative stimuli and novel, possibly meaningful stimuli. Novelty and familiarity are commonly thought as percepts produced at higher-order synaptic levels such as the mushroom bodies of insects. We took a multidisciplinary approach to test the hypothesis that novelty and familiarity are already present in early signal processing stages. We first verified that bees respond with different behavior to novel and familiar odors. Using a video tracking software (SwarmSight), we evaluated novelty detection behaviorally in honey bees as they position their antennae differentially in an air stream carrying familiar or novel odors. Upon stimulation of the antennae of naïve bees, the bees moved their

antennae to a position lateral to the head and perpendicular to the airflow. The bees resumed movement of the antennae when odor delivery was terminated. After familiarization to one odor, stimulation with a novel odor led bees to maintain the lateral position well beyond odor delivery, which is the basis for novelty detection in our study. Bees failed to show this pattern to the familiar odor. We then used a multiunit recording method (TDT RZ2) to characterize neuronal responses to familiar and novel odors in the first synaptic relay in the brain - the antennal lobes. We found that the neurons that exhibited stronger initial responses to the odor that was to be familiarized are the same units that later distinguish familiar and novel odors. These units, including both projection neurons and local interneurons, showed a decreased response to the familiar odor but an increased response to the novel odor. In summary, our results suggest that the antennal lobe may assign a category of familiarity or novelty to an odor stimulus in addition to its chemical identity code. Therefore, the mechanisms for novelty detection may be present in early sensory processing, either as a result of local synaptic interaction or via feedback from higher brain centers.

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Poster

P424. Chemical Senses: Behavior and Perception

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Program #/Poster #: P424.04

Topic: D.05. Olfaction and Taste

Support: JSPS 19J12156

Title: Encoding and control of odor value by Drosophila dopamine neurons

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Abstract: Animals behave based on the value of sensory cues, which is constantly re-evaluated through experiences. Such adaptive behavior depends on dopaminergic neurons (DANs), whose activity has been most extensively studied in the context of associative learning. Specifically, DANs respond to an unconditioned stimulus (US) and later to a conditioned stimulus (CS) through learning. However, because some commonly used CS including odors has innate value, DANs may respond to CS and drive plasticity outside the context of associative learning. Here, we addressed whether DANs represent innate value of odors and induce neural and behavioral plasticity in a purely olfactory environment using Drosophila whose brain structure allows us to comprehensively characterize DANs. We focused on DANs innervating the mushroom body (MB) where olfactory information is modulated by reward or punishment. The MB comprises 15

compartments each of which receives input from a distinct type of DANs and provides a place for synaptic plasticity underlying learning. We performed in vivo two-photon Ca2+ imaging in a fly expressing GCaMP6f in the entire DANs innervating the MB. We recorded responses of DANs to 27 odors with a wide range of innate odor values, which were quantified based on the behavior of flies navigating in a virtual flight arena (Badel et al., 2016). We found that DANs in specific compartments differentially respond to odors with positive and negative values. Regression analysis showed that information about odor values can be decoded from the activity of a population of DANs. Because we found that odors alone can activate both olfactory and DAN pathways, moment-to-moment adaptive changes in the downstream MB neurons and behavior may take place. To test this hypothesis, we asked whether repetitive application of odors was sufficient to induce neural plasticity and indeed observed that the neural activity of MB output neurons showed plastic changes in an odor value-dependent manner. Finally, we are investigating if flies navigating in an olfactory environment exhibit DAN-dependent, online adaptive changes in behavior. In sum, our results highlight a novel role of DANs in driving odor value-based, non-associative control of neural circuits and behavior.

Disclosures: A. Kato: None. **H. Kazama:** None.

Poster

P424. Chemical Senses: Behavior and Perception

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P424.05

Topic: D.05. Olfaction and Taste

Support: NIDCD 900026
NSF 920022
NINDS 900107

Title: Sensorimotor transformations underlying odor-guided locomotion in Drosophila

Authors: *L. TAO[1], S. OZARKAR[2], V. BHANDAWAT[1];
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Abstract: The ability to flexibly modulate essential behaviors, such as locomotive food search, using sensory information is crucial to the survival of animals. The *Drosophila* olfactory system is an excellent model organism to study the role of sensory processing on behaviors due to its simplicity, the availability of genetic tools, and because odors have a marked effect on its locomotion. While much is known about the structural connectivity of the early olfactory circuit in flies, how the signals from ORNs modulate locomotion and the mechanisms by which these ORNs interact to perform this modulation are still being actively studied. In particular, while olfaction alone in the absence of directional cues such as wind has been shown to be sufficient in

eliciting broad changes in locomotion based on binary activation of ORNs, how moment by moment changes in ORN activity modulates locomotion is not well understood. Here, we investigate how optogenetic activation of precise subsets of ORNs that are activated by apple cider vinegar (an attractive odor) causes changes in locomotion in freely walking flies. Using electrophysiology, computational, and agent-based modeling, we developed a framework to capture the sensorimotor transformation from ORN activity to locomotion. We first show that both the firing rate (f) and change in firing rate (df) are necessary to describe the dynamics of the ORN response. We then show that the locomotor behavior of the fly is well described by four locomotor states (sharp turns, curved walks, stops, and boundary) each parameterized by kinematics such as speed and curvature. For any given ORN, there is a probabilistic mapping between the locomotor state kinematics and ORN activity in the f-df space. Using this framework, we show simple rules of combination between the f-df space of different ORNs. In addition to kinematics, we show that large temporal changes in ORN activity give flies a sense of their orientation relative to the stimulus and cause flies to stop walking and start turning. Based on these results, we develop an agent-based model of *Drosophila* locomotion that was able to capture much of the spatial-temporal distribution of experimental flies in our arena. These results provide insight into both sensorimotor transformations underlying olfactory locomotion and how this transformation is integrated across olfactory inputs.

Disclosures: L. Tao: None. **S. Ozarkar:** None. **V. Bhandawat:** None.

Poster

P424. Chemical Senses: Behavior and Perception

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Program #/Poster #: P424.06

Topic: D.05. Olfaction and Taste

Support: NIH Grant R34 NS116731-01
University of Oregon Peter O'Day Fellowship
University of Oregon Summer Undergraduate Research Fellowship
Undergraduate Research Opportunities Program Mini Grant

Title: Active olfactomotor responses in head-fixed mice

Authors: *I. CULLEN[1], A. RANDHAWA[2], D. YEH[3], J. ACOSTA-KING[6], A. WELCH[3], A. SINGH BALA[4], M. SMEAR[5];
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Abstract: Olfactomotor responses are respiratory, orofacial, and locomotive movements for sampling and reacting to odors (Rabell et al. 2017, Kurnikova, Deschênes, and Kleinfeld 2019, Findley et al. 2020, Johnson et al 2003, Wesson et al 2008, Jones and Urban 2018). Altered sensory sampling behaviors, such as eye movement, temperature insensitivity, and pain insensitivity, have been identified in individuals with Autism Spectrum Disorder (ASD). In addition, Rozenkrantz et al. (2015) showed that olfactomotor behavior is affected in children with ASD. These children do not modulate sniffing behavior to aversive odors despite correctly identifying odors as unpleasant, suggesting an altered unconscious motor response. To investigate the neural mechanisms underlying olfactomotor sampling, we investigated respiratory and orofacial responses to odor using wildtype mice. Wildtype mice are exposed to 2-phenylethanol (attractive odor), 2-methylbutyric acid (aversive odor), alpha- pinene (neutral odor), or clear air in the course of a behavioral session. We record respiration with an intranasal thermistor, and track orofacial movements using DeepLabCut. Our preliminary results in wildtype mice (n=2) suggest that mice alter their sniffing and nose movement in response to odor stimuli. This work will shed light on active olfaction and help us understand more about naturalistic olfactomotor behaviors.

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Poster

P424. Chemical Senses: Behavior and Perception

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Program #/Poster #: P424.07

Topic: D.05. Olfaction and Taste

Support: NIH-NIDCD R01 5R01DC014487-05

Title: Understanding the neuronal substrates of sensorimotor transformations via a novel closed-loop olfactory task for mice

Authors: *M. DUSSAUZE, P. GUPTA, U. LIVNEH, D. ALBEANU;
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Abstract: Throughout life, via repeated experience of the closed-loop relationship between sensory inputs and movements, the brain builds sensorimotor models that predict the sensory consequences of its own actions (von Helmholtz, 1867; Craik, 1947; Barlow 1961). As such, perception can be thought of as the continuous comparison of expected (sensorimotor predictions) and actual sensory inputs. This process generates prediction errors that can be minimized by learning increasingly accurate models of the world. While odorant sampling (sniffing) is tightly coupled to motor actions (head and body movements), to date, it remains

unknown how olfactory representations are regulated by motor signals and whether such control supports sensorimotor predictions. To investigate the neural substrates of sensorimotor predictions both at behavioral and circuit-level, we established a novel closed-loop olfactory task (*Smellocator*) where head-fixed mice are trained to steer the left-right location of an odorant source by controlling a light lever with their forepaws. In this task, mice learn to link motor action to well-defined sensory expectations (odor location). Mice master the task quickly (~2 weeks, >90% accuracy), perform numerous trials per session (500-700) and show precise movements that are locked to the instantaneous sensory (odor) feedback. In expert animals, we violate these learnt sensorimotor expectations by transiently decoupling the stimulus from the current action (open-loop replay, halting of odor source, changing the gain), thus creating sensorimotor prediction errors (mismatches) **(**Schneider et al, 2014; Keller and Mrsic-Flogel, 2018). Additionally, across longer timescales (blocks of trials), we modulate the sensorimotor mapping (direction), so as to trigger updates in the animal's internal model and engage sensorimotor adaptation. Strikingly, mice readily counter these sensorimotor errors and re-center the odor via precise corrective movements which provide a behavioral read-out of the animal's internal model. To understand the sensorimotor transformations which enable this behavior, we have started monitoring neural activity in the olfactory cortex (Anterior Olfactory Nucleus and Piriform Cortex) using chronically implanted tetrode micro-drives.

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Poster

P434. Auditory Processing: Vocalizations and Natural Sounds I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P434.01

Topic: D.06. Auditory & Vestibular Systems

Support: NIH-NIDCD Grant
NSF Graduate Research Fellowship

Title: Modeling the adaptation of the human auditory cortex to background noise changes with deep neural networks

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Abstract: The human auditory pathway displays a robust capacity to adapt to sudden changes in the statistics of an auditory scene, an ability that helps in many real-world scenarios, such as

tracking a conversation while walking through a door from a street to a loud restaurant. Previous work has demonstrated that the auditory cortex rapidly suppresses new acoustic features arising from unexpected background noise changes. While this neural ability has been identified and characterized, its mechanism is not well understood due to the difficulty of interpreting such a nonlinear behavior. In order to understand how the brain achieves this adaptation, an important first step is to identify the simplest model that can still mimic the brain's behavior. Traditional linear models such as the spectro-temporal receptive field (STRF) are highly interpretable, due to their linearity, but fail to capture the nonlinear dynamics of neural adaptation. To overcome this limitation, we employ recent advances in a certain class of deep neural network models (DNN). These models have greatly improved capacity to learn nonlinear transformations and can be easily interpreted as dynamic STRFs (DSTRF), since a linear equivalent function to the DNN can be found for each stimulus instance. This provides a means of interpreting the nonlinear operations of the network as stimulus-dependent linear functions. In this study, we recorded intracranial EEG (iEEG) from neurosurgical patients who were asked to attend to speech overlaid on background noise that kept changing between several categories. We first demonstrate that a DNN trained to predict neural responses can produce the same rapid adaptation phenomenon as the auditory cortex significantly better than a linear STRF model. Additionally, this model can replicate a more complex phenomenon arising from adaptation, where phoneme discrimination momentarily drops and background noise encoding increases following a noise change, after which noise information is suppressed and phoneme discrimination is restored. Using the equivalent piecewise linear function of the DNN model, we analyze the computations performed by these models and show that some neural regions react to a sudden change in background noise by altering their filters to deal with the new sound statistics. By looking at how these filters change over time particularly when adapting to a new background noise, we provide evidence that can explain how the brain adapts to changing noise conditions. Hence, we show that modeling the auditory cortex with deep neural networks can help us understand the neural computations that underlie noise-robust speech perception in real-world environments.

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Poster

P434. Auditory Processing: Vocalizations and Natural Sounds I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P434.02

Topic: D.06. Auditory & Vestibular Systems

Support: MHRD/IIT KGP/Institute Fellowship
IITKGP/SRIC/SGIGC-2015/DMN

IA/I/11/2500270

Title: Coding of two-tone harmonic complexes by excitatory and inhibitory neurons in the mouse auditory cortex

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Abstract: Two-tone harmonic complexes (TTHC) are a common syllable type in mouse vocalization, especially in the context of courtship. Differences in the representations of each of the component tones (F_0 and F_1, fundamental and the first harmonic) and the TTHC in AC neurons would help us understand how a unified percept may form from two simultaneously presented component stimuli. Further, underlying mechanisms in such binding can be better understood from differential representation of TTHC and tones in excitatory and inhibitory neurons (EXNs and INNs) neurons as well as from the EX-EX and IN-IN network modulation by such stimuli. In the current study we first used widefield fluorescence imaging, through a cranial window, in mice expressing GCaMP6f either in Thy-1 positive EXNs or in specific INNs to first localize the primary AC (A1) using pure tone stimuli. Next with widefield imaging of responses to TTHC and responses to corresponding component pure tones, regions within A1 were found to show higher selectivity to TTHCs compared to the summed response of component tones. To further investigate such harmonic selectivity in single neurons and properties of their micro-networks, chronic 2-photon Ca^{2+} imaging was performed in the same mice. We found that EXNs and INNs showed a variety of response properties with respect to the 3 stimuli, F_0, F_1 and the TTHC depending on the relative location of the F_0 and F_1 from the BF of neurons. In particular, we find higher normalized responses to TTHC in somatostatin (SST) positive INNs compared to EXNs near BF and not away from BF when neurons respond to all 3 stimuli. However, both EXNs and SOM INNs showed higher normalized responses to TTHCs away from BF when one of F_0 and F_1 did not elicit a response indicating nonlinear interactions. Similarly a large subpopulation of neurons also showed inhibitory effects of TTHC with no response to TTHC but moderate to strong responses to either or both F_0 and F_1. Further, EXNs showed higher noise correlations and SST INNs showed no change in noise correlations during TTHC presentation compared to that during tone presentation, indicating recurrent excitatory connections being involved in coding of TTHC. Thus we find a differential representation of TTHCs in EXNs and SST INNs and their local networks compared to that of component pure tones.

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Poster

P434. Auditory Processing: Vocalizations and Natural Sounds I

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Program #/Poster #: P434.03

Topic: D.06. Auditory & Vestibular Systems

Support: NIDCD grant to FET NIH R01DC018321

Title: Ensemble Neural Code of a Complete Vocal Repertoire

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Abstract: Social species are able to quickly identify vocalizations and respond accordingly. This vocal communication requires the ability to categorize sounds into meaningful units. Due to their well described auditory system and large behaviorally relevant vocal repertoire, Zebra Finches are a great model organism to study categorization of species specific vocalizations. Using a wireless lightweight electrophysiological recording device, we recorded from primary and secondary auditory regions of awake and unrestrained zebra finches as they listened to playbacks of conspecific communication calls. We used decoding analyses to understand how well single neurons and neural ensembles represent behaviorally defined call types in auditory regions. We find that the activity of single units in secondary auditory regions do not contain enough information to decode semantic call categories. However, increasing the size of ensembles of neurons quickly increases decoder performance. This result supports a selective yet somewhat distributed code hypothesis, akin to results in primate studies (Chang L & Tsao DY 2017; M. Rigotti et al. 2013) in which individual cells are selective for features that do not correspond to particular calls (or categories), but together, neural ensembles very efficiently code for behaviorally relevant categories. Finally, using a novel hierarchical clustering based approach, we compare the neural population responses to the acoustic and semantic structure of the stimuli. We find that neural ensembles represent the semantic category of calls better than the acoustic features, implicating these brain regions in the sound to meaning transformation.

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Poster

P434. Auditory Processing: Vocalizations and Natural Sounds I

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Program #/Poster #: P434.04

Topic: D.06. Auditory & Vestibular Systems

Support: R01-DC-017690

Title: Unsupervised learning of stimulus invariance of natural auditory stimuli via slow feature analysis

Authors: *R. DITULLIO[1], C. K. PARTHIBAN[1], E. PIASINI[1], V. BALASUBRAMANIAN[1], Y. E. COHEN[2];
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Abstract: A fundamental goal of sensory systems is to extract sensory information from the environment and convert it into perceptual representations. Because the brain does not have enough resources to simply transduce and represent all of the information present in the environment, it must find a means to extract only the sensory information that is most relevant to the animal's survival; that is, sensory systems must perform feature selection. One way a sensory system could perform feature selection is to try to encode particular statistical regularities in the sensory environment. An important statistical regularity of natural auditory stimuli is that they tend to have low temporal modulation; i.e. the powers of the frequencies that comprises natural stimuli tend to change slowly over time. Despite their ubiquitous presence in natural auditory stimuli, it is unknown whether such slow temporal regularities are sufficient to enable learning and perception of auditory object classes.To test this idea, we adapted an unsupervised temporal learning algorithm, Slow Feature Analysis (SFA), to extract the auditory features that change most slowly over time (i.e., those having the lowest temporal variance). We then used this algorithm to computationally evaluate the hypothesis that extracting these slowly varying features will capture the variance inherent within a stimulus class and the variability that differentiates between stimulus classes. We tested this hypothesis by first learning a set of slow features from rhesus macaque vocalizations and then projecting these vocalizations into the learned feature space. We then instantiated a linear classifier to determine whether the projected stimuli are linearly separable. We found that (1) pairs of vocalizations in the SFA-generated feature space were linearly separable; (2) this feature space is robust to clutter (noise) in the training data set; (3) this feature space captures enough variability for the classification of novel exemplars; (4) classes of vocalizations tend to naturally cluster together in this feature space. Together, our results suggest that if the brain can extract the slow temporal features from auditory stimuli, it may be sufficient for and underlie important components of perception.

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Poster

P434. Auditory Processing: Vocalizations and Natural Sounds I

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Program #/Poster #: P434.05

Topic: D.06. Auditory & Vestibular Systems

Support: NIDCD Grant DC00937

Title: Representation of mouse vocalizations across subdivisions of the inferior colliculus

Authors: *M. ATTARHAIE TEHRANI, S. J. SHANBHAG, R. PATEL, J. J. WENSTRUP; Dept. of Anat. and Neurobio., Northeast Ohio Med. Univ., Rootstown, OH

Abstract: Subdivisions of the inferior colliculus (IC) participate in several circuits that analyze social vocalizations. We explored the representation in IC subdivisions of both ultrasonic vocalizations (USVs) and several categories of non-USV, mostly broadband calls. We examined: 1) how responsiveness to different vocal categories relates to frequency tuning, 2) vocal selectivity beyond that related to frequency tuning, and 3) the distribution of vocal responses and frequency tuning across IC subdivisions. In head-fixed, urethane-anesthetized adult CBA/CaJ mice, we used multichannel probes to extracellularly record single- and multi-unit responses to acoustic stimuli. Frequency tuning and sound level responses were assessed using tones and broad band noise. We then obtained responses to pre-recorded syllables containing a broad range of spectral components. Most IC sites were tuned below 30 kHz, with differences across subdivisions. In the external IC, all sites were tuned below 30 kHz, while dorsal and central IC also contained tuning >30 kHz. Generally, IC sites responded to vocal signals with energy corresponding to their frequency response areas. Sites tuned below 20 kHz responded to the non-USV calls only: the LFH call (fundamental 4 kHz), MFV call (fundamental ~9-15 kHz), and Noisy call (broad band >15 kHz). Sites tuned to 30-40 kHz also responded to stepped USVs that contain frequencies below 50 kHz, but not to chevron or tonal USVs limited to frequencies above 55 kHz. Only high frequency sites (>55 kHz) responded to chevron or tonal USVs. However, several of these sites also responded to the non-USVs, due either to signal energy near the peak of high frequency tuning curves or associated with the tails of these tuning curves. Thus, responses to non-USVs were observed throughout IC, but USV responses were found mostly in higher frequency regions of the central IC. Further, the more complex spectrotemporal structures of stepped USVs and noisy calls were represented in temporal response patterns of some neurons within IC populations, including both excitatory and suppressive responses. These results indicate that information about a wide range of mouse vocal signals is present both in the lemniscal projection to auditory thalamus and in non-lemniscal circuits through the medial auditory thalamus to structures such as the amygdala. Future research will explore these representations in the higher order structures of the auditory thalamus and the cortex, as well as the amygdala. These findings in normal-hearing mice are key to evaluating changes in neural mechanisms underlying acoustic communication that occur in mouse models of impaired auditory perception.

Disclosures: M. Attarhaie Tehrani: None. **S.J. Shanbhag:** None. **R. Patel:** None. **J.J. Wenstrup:** None.

Poster

P434. Auditory Processing: Vocalizations and Natural Sounds I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P434.06

Topic: D.06. Auditory & Vestibular Systems

Title: Simultaneous perturbation of fundamental and formant frequencies in auditory feedback of speech

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Abstract: In speech production, vocal cord vibration and vocal tract shape are controlled simultaneously. These are expressed by speech parameters called fundamental frequency (f0) and formant frequency, respectively. In order to verify the importance of auditory feedback in each parameter, an altered auditory feedback (AAF) experiment has been conducted, and it was found that a compensatory response to the perturbation occurred. However, in these experiments, the fundamental and formant frequencies were perturbed independently. Although auditory errors at the fundamental and formant frequencies can occur simultaneously in daily speech production, there have been few AAF experiments with these perturbations simultaneously. In this study, we investigate the compensatory response when both fundamental and formant frequencies are perturbed simultaneously during vowel production. Native Japanese speakers were participated in the experiments. They were instructed to repeat 140 Japanese syllable /he/ speech. Both fundamental and formant frequencies of speech sound are gradually shifted up to the maximum perturbations. The maximum amount of perturbation of fundamental and formant frequencies was decided based on the threshold of discrimination in the speech perception. The experiment was carried out under the conditions of raising both f0 and the first formant frequency (F1) or lowering f0 and raising F1. In this experiment, we used a phase equalization-based autoregressive exogenous (PEAR) model, which is a formant estimation algorithm that is robust to f0, and a pitch synchronous overlap and add (PSOLA), which is a f0 shift algorithm, to enable highly accurate perturbation. The experimental results showed that the compensatory response to f0 occurred irrespective of the coincidence mismatch in the perturbed direction of F1, but that to F1 is affected by the perturbation direction of f0. This suggests that the f0 error may have a greater impact on speech production than the formant frequency and that F1 control is affected by the f0 error. Therefore, the auditory feedback control mechanisms of f0 and formant frequencies may be different.

Disclosures: S. Hiroya: None. **T. Mochida:** None.

Poster

P434. Auditory Processing: Vocalizations and Natural Sounds I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P434.07

Topic: D.06. Auditory & Vestibular Systems

Title: Effect of novel acoustic environment on lateralized activity patterns and new neuron influx

Authors: *B. FUREST CATALDO[1], J. OVETSKY[2], P. DADIKA[2], M.-N. BROOKS[2], B. BLAIR-WHIATKER[2], D. S. VICARIO[3];
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Abstract: Lateralization is widely studied to elucidate the functions that each hemisphere serves in the context of language perception and production. However, hemispheric asymmetries are not unique to humans or to language and may subserve many functions. Like humans, songbirds are vocal learners that also exhibit lateral differences during sensory and motor processing of communication signals. In addition, the introduction of new neurons in adulthood is lateralized in the higher auditory area NCM, and the degree of lateralization is correlated with the ability to form good copies of the birds' tutor song, which in turn depends on the ability to form a proper auditory representation/memory of the tutor song. Our laboratory has used electrophysiological recordings in adult Zebra finch NCM to show that activity in response to song playback is lateralized and that the asymmetry shifts in birds passively exposed to novel heterospecific auditory environments for extended periods (days to weeks). Furthermore, shifts in lateralization are associated with enhanced ability to discriminate between heterospecific sounds, suggesting that passive exposure can evoke dynamic changes in neural representations that subserve behavior. The current study has three Aims: 1) Determine the timecourse of dynamic shifts in lateralized activity in birds exposed to a heterospecific environment; 2) Explore whether exposure to a heterospecific environment changes the lateralized introduction of new neurons in NCM; and 3) Assess the correlation between shifts in lateralized activity and the influx of new neurons into NCM in the two hemispheres. To address Aim 1, Zebra finches were implanted with a custom epidural array that enables lateralized activity, measured as event related potentials (ERPs), to be assayed longitudinally in birds exposed to a novel heterospecific environment for extended periods. To address Aims 2 and 3, a separate cohort of Zebra finches was injected with BrdU, to label new cells, and exposed to the heterospecific environment. Then, lateralized NCM activity and new neuron influx were quantified after discrete exposure periods (9,14, or 30 days). The results confirm that lateralized activity transiently shifts in birds exposed to the heterospecific environment as a function of exposure period. Preliminary observations indicate that exposure to the heterospecific environment for different periods may transiently affect the lateralized bias for new neurons in NCM, compared to birds exposed to a control environment. Furthermore, the data suggests an inverse relationship between lateralized new neuron counts and lateralized electrophysiological activity.

Disclosures: B. Furest Cataldo: None. **J. Ovetsky:** None. **P. Dadika:** None. **M. Brooks:** None. **B. Blair-Whiatker:** None. **D.S. Vicario:** None.

Poster

P434. Auditory Processing: Vocalizations and Natural Sounds I

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Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P434.08

Topic: D.06. Auditory & Vestibular Systems

Title: Encoding of changes in frequency for conspecific vocalizations in auditory core and belt areas of the common marmoset

Authors: *J. LEE, X. WANG;
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Abstract: In early stages of auditory processing, neurons faithfully encode acoustic features of sounds. As we move up the auditory pathway, the auditory system is thought to represent behaviorally relevant stimuli (e.g., species-specific vocalizations) in a manner that is invariant to differences or changes in their basic acoustic parameters when such differences fall within the range of natural stimuli. This invariance is believed to be the underlying mechanism for perceptual phenomena such as categorical perception or the perceptual magnet effect that have been documented in human speech perception (Kuhl 1991). Although there have been studies investigating the emergence of such invariance regarding vocalizations in the auditory system, there is much to discover in terms of where and how it is represented by both neural populations and individual neurons in various areas of auditory cortex. In this study we investigated how core and belt areas of marmoset auditory cortex encode changes in center frequency of both pure tones and synthesized marmoset phee calls and compared the neural responses with corresponding behavioral measures. Marmoset is a highly vocal non-human primate species with a hearing range similar to that of humans. We observed that in the core area, responses to a given pair of stimuli (pure tones or phees) reflect their differences in center frequency. In the belt area, however, this was the case for the responses to pure tones, but not to phees. In particular, offset responses to frequency-shifted phees were invariant to changes in center frequency when the frequency shift was within the range of the center frequency of phee population samples recorded in our colony (Agamaite et al., 2015).

Disclosures: J. Lee: None. **X. Wang:** None.

Poster

P491. Eye Movements and Visuomotor Processing

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P491.01

Topic: D.08. Visual Sensory-motor Processing

Support: ARL/DSO W911NF-10-2-0022 (Subaward from DCS Corp: APX02-N013)

Title: Saccading does not reset neural processing of visual targets in natural viewing task

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Abstract: Relatively little is known about visual processing during free-viewing visual search in realistic dynamic environments. Free-viewing is characterized by frequent saccades. During saccades, visual processing is thought to be suppressed, yet we know that the pre-saccadic visual content can modulate post-saccadic processing. To better understand these processes in a realistic setting, we study here saccades and neural responses elicited by the appearance of visual targets in a realistic virtual environment. While subjects were being driven through a 3D virtual town they were asked to discriminate between targets that appear on the road. We found that the presence of a target enhances early occipital as well as late frontocentral saccade-related responses. The earlier potential, shortly after 125ms post-saccade onset, was enhanced for targets that appeared in peripheral vision as compared to central vision, suggesting that fast peripheral processing initiated before saccade onset. The later potential, at 195ms post-saccade onset, was strongly modulated by the visibility of the target. Together these results suggest that, during natural viewing, neural processing of the pre-saccadic visual stimulus continues throughout the saccade, apparently unencumbered by saccadic suppression.

Disclosures: A.D. Stankov: None. **J. Touryan:** None. **S. Gordon:** None. **A. Ries:** None. **J. Ki:** None. **L.C. Parra:** None.

Poster

P491. Eye Movements and Visuomotor Processing

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Program #/Poster #: P491.02

Topic: D.08. Visual Sensory-motor Processing

Support: NIH EY014924

Title: Foveal viewing influences subsequent peripheral stimulus processing by visual cortical neurons

Authors: *R. XIA[1], D. JONIKAITIS[1,2], T. MOORE[1,2];
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Abstract: In humans and other primates, visual acuity decreases dramatically from the point of fixation (fovea) to the visual periphery. As a result, targets of interest must be fixated sequentially to accumulate detailed visual information contained within a scene, and this is accomplished largely by way of saccadic eye movements. Psychophysical evidence suggests that at least some visual information is integrated across sequential fixations, but the neural mechanisms underlying trans-saccadic integration remain largely unknown. In this study, we asked whether detailed visual information acquired at fixation exerts an influence on the subsequent processing of that information in the periphery by neurons in visual cortex. To address this question, we recorded neuronal activity in area V4 of rhesus monkeys using 32-channel linear probes (Plexon, Inc.) while the monkeys performed a triple-saccade task. In the task, each trial began with the monkey fixating a central fixation spot, after which a gray-scale drifting grating (3° in diameter) was presented at a peripheral location coinciding with the locations of the receptive fields (RFs) of recorded V4 neurons (5 - 10º eccentricity). The monkey maintained fixation for 500 ms (pre-foveation period) until the fixation spot disappeared, which cued the monkey to make a saccade to the grating. After fixating the grating for 500 ms, the monkey re-fixated the initial fixation spot as it re-appeared. Following another 500 ms (post-foveation period), the fixation spot disappeared again, and the monkey was rewarded for making a saccade back to the grating to complete the trial. Across trials, we varied both the direction of motion and the spatial frequency of the grating target, and compared the neuronal responses to the gratings during pre- and post- foveation periods. As expected, the magnitude of visual responses to the target gratings was diminished from the pre-foveation to the post-foveation periods, consistent with the repetition suppression effects previously described. Nonetheless, neurons remained selective to the orientation and direction of motion of the grating targets in the post-foveation period. Moreover, thus far, we find that stimulus selectivity (e.g. direction of motion) increases during the post-foveation period compared to the pre-foveation period for gratings presented at higher spatial frequencies whereas selectivity decreases for lower spatial frequency gratings. These results suggest that high acuity visual information obtained through foveal viewing influences subsequent peripheral processing and is integrated across saccades.

Disclosures: R. Xia: None. **D. Jonikaitis:** None. **T. Moore:** None.

Poster

P491. Eye Movements and Visuomotor Processing

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Program #/Poster #: P491.03

Topic: D.08. Visual Sensory-motor Processing

Support: Marie Skłodowska-Curie Individual Fellowship No. 846392

Title: The trans-saccadic preview effect: Neural adaptation or evidence for active vision?

Authors: C. HUBER-HUBER[1], *F. DE LANGE[1], D. MELCHER[2];
[1]Donders Inst. for Brain, Cognition and Behaviour, Radboud Univ., Nijmegen, Netherlands;
[2]New York Univ. Abu Dhabi, Abu Dhabi, United Arab Emirates

Abstract: Most neurocognitive research on human visual perception up until now has been carried out while the eyes of the participants were still, usually focused at a fixation cross at the center of the screen. In real life, however, we move our eyes about three times per second, each time leading to drastic and abrupt changes in visual stimulation of which we are not aware. These gaze dynamics entail that every time we perceive something foveally with high resolution we have previously seen the same with much worse resolution further in the visual periphery. There is always a visual preview. We demonstrated recently, using face stimuli and combined EEG and eye-tracking, that this pre-saccadic preview affects post-saccadic processing, which is in line with earlier work in the domain of reading research. Here, we ask whether this effect is contingent on active vision or whether it results simply from adaptation of neurons with large receptive fields which would be unrelated to eye movements. Our results show a larger preview effect in the post-saccadic N170 component for an active vision condition compared to a passive stimulation condition which suggests that real-life visual perception is fundamentally different from passive visual perception usually studied in the laboratory. In addition, we discuss further control conditions designed to rule out other types of adaptation.

Disclosures: C. Huber-Huber: None. **F. de Lange:** None. **D. Melcher:** None.

Poster

P491. Eye Movements and Visuomotor Processing

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Topic: D.08. Visual Sensory-motor Processing

Support: Departmental pilot funding
NIH Grant 5R01NS079518-09

Title: Dissecting the visuomotor circuit for saccadic suppression

Authors: *J. HUNT[1], G. FELSEN[2], A. POLEG-POLSKY[3];
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Abstract: Many species, including mice, use saccades to actively explore their visual environment. Saccades result in a transient modification of visual perception such that perceptual thresholds are elevated relative to extra-saccadic performance, a phenomenon referred to as saccadic suppression. Saccadic suppression is associated with a decrease in the visual

responsiveness of neurons across the visual system which is believed to be the neural basis of the perceptual phenomenon. While several sources of neural suppression have been identified, including a visual-only mechanism originating in the retina, a long-standing complementary hypothesis proposes that a premotor corollary discharge (CD) broadcast by the motor (deep) layers of the superior colliculus (SC) to other nodes in the visual system triggers suppression of neuronal responses across the visual system hierarchy. Our overall goal is to test this hypothesis by dissecting the visuomotor circuitry in the SC in head-fixed mice making saccades. In particular, we seek to test the idea that premotor neurons in the deep SC relay a CD to visual neurons in the superficial SC via GABAergic interneurons. Here, in a set of preliminary experiments we used the optokinetic reflex (OKR) to elicit conjugate saccades in head-fixed mice and found that unilateral inactivation of the SC with muscimol arrested saccades directed toward contralateral visual space. This result suggests that saccades elicited through the OKR are SC dependent. We next recorded single-unit activity across the dorsal-ventral axis of the SC in response to visual stimuli presented around the time of saccades using high-density Neuropixels probes. We identified visual responses in the visual (superficial) layers of the SC, which we predict will be suppressed by coincident saccades. This predicted result would show saccadic suppression of visual responses in the mouse SC. Future experiments can then use cell-type specific tools to dissect the collicular and retinal circuitry that underlies saccadic suppression.

Disclosures: J. Hunt: None. **G. Felsen:** None. **A. Poleg-Polsky:** None.

Poster

P491. Eye Movements and Visuomotor Processing

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Program #/Poster #: P491.05

Topic: D.08. Visual Sensory-motor Processing

Support: NIH Grant EY025172

Title: Encoding of salience and informed choice colludes and conflicts across FEF neuronal types in the guidance of urgent saccadic decisions

Authors: *E. E. OOR, E. SALINAS, T. R. STANFORD;
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Abstract: Scanning eye movements are guided by both the physical salience and behavioral relevance of items in a visual scene. Thus, each saccadic choice is the result of dynamic interplay between exogenous and endogenous mechanisms and their respective influences on evolving motor plans. Saccadic choice tasks typically preclude such interplay by serializing perceptual evaluation and motor choice stages in that order. To expose specific psychophysical and neural manifestations of this interaction, we designed an urgent color-matching task in which motor

planning precedes the perceptual evaluation. The design tracks both the subject's choice and the underlying neuronal activity as they unfold over time. By also manipulating the relative salience of the target/distracter pair, we can distinguish exogenous from endogenous contributions to target selection. Previous findings from our lab—that visual neurons in the Frontal Eye Field (FEF) primarily signal the salience of items in their response field (Scerra et al., 2019), and that exogenous signals capture attention automatically within ~100 ms of stimulus onset (Salinas et al., 2019)—make specific predictions for how a salience-driven signal would bias choices based on endogenous information. Here, we tracked the timing of oculomotor activity via single-neuron recordings from the FEF in two monkeys. Consistent with our model, we found that during an ongoing saccadic choice, exogenous mechanisms boosted performance when the salient item was the target and hindered it when it was the distracter—rapid, involuntary effects that were superseded by endogenous mechanisms ~40 ms later. Activity recorded from FEF confirmed that visual neurons select salient items regardless of target/distracter identity and thus can aid or impede selection of the endogenously defined target. Assuming FEF motor activity reflects a choice biased by the activity of visual neurons, we predicted motor activity to be accelerated or attenuated depending on whether the motor plan was congruent or opposed to the salient item. Remarkably, the activity of pure motor cells resisted such saliency selection to unambiguously signal the target regardless of its physical attributes. In contrast, the activity of visuomotor cells was most revealing of the predicted exogenous-endogenous interaction by signaling stimulus salience and then stimulus identity (target vs distracter) in rapid temporal sequence. The results reveal correlates of the interactions between exogenous and endogenous signals in FEF that are distinct across specific neuronal types and are finely orchestrated over a few tens of milliseconds to yield each saccadic choice.

Disclosures: E.E. Oor: None. **E. Salinas:** None. **T.R. Stanford:** None.

Poster

P491. Eye Movements and Visuomotor Processing

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Topic: D.08. Visual Sensory-motor Processing

Support: NSF Grant BCS-1261433
NIH Grant T32EY018080
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Title: Active vision and the effects of saccades on perception, V1 spiking, noise correlations, and choice probability

Authors: *S. AKERS-CAMPBELL[1], J. E. NIEMEYER[2], A. GREGOIRE[1], M. A. PARADISO[1];
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Abstract: Perception in multiple sensory modalities is an active process that involves exploratory behaviors such as sniffing and whisking in rodents. In humans and other primates, vision results from active sensory sampling guided by saccadic eye movements. Neural recordings with EEG, LFPs, spikes, and BOLD signals show that saccades modulate activity in multiple brain areas. Psychophysical experiments in humans have revealed effects of saccades on perception and it is hypothesized that a motor signal associated with eye movements speeds and enhances object recognition and contributes to our sense of visual stability across saccades. We investigated the integration of saccades into visual processing and, more specifically, the contribution of an eye-movement related signal associated with active sensory sampling. Recordings were made from networks of macaque V1 neurons as animals performed a contrast detection task. Perceptual and neural measures were compared when an animal made real saccades that brought a stimulus into V1 receptive fields and when simulated saccades were made (identical retinal stimulation but no eye movement or corollary discharge signal). The average activity of the neurons was strongly influenced by saccades in a stimulus-specific manner. When low spatial frequency stimuli were presented, both perceptual contrast sensitivity and V1 activity were lower on saccade trials than simulated saccade trials. Conversely, with higher spatial frequency stimuli, contrast sensitivity and V1 activity were greater on saccade trials. These effects may underlie the suppression of vision during saccades and the enhancement of perception at the start of fixations. Saccades were also found to decrease pairwise noise correlations and choice probability across all spatial frequencies. Taken together, the findings suggest that an extraretinal signal related to the saccadic eye movement alters both V1 spiking and network correlations in a manner consistent with changes in visual sensitivity. More broadly, effects of motor activity (saccades) on visual perception and noise correlations appear to parallel effects observed in other sensory modalities. In rodents, active whisking and sniffing are also associated with improved sensory discrimination and decreased noise correlations and choice probability. Thus it appears that corollary-discharge signals in vision, olfaction, and somatosensation reach sensory cortex, decrease noise correlations, and modulate functional connectivity, leading to overall improved information flow and sensory discrimination.

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Poster

P491. Eye Movements and Visuomotor Processing

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Topic: D.08. Visual Sensory-motor Processing

Support: NEI R01EY017699
NIMH 1P50MH109429

Title: Visual and motor responses in mediodorsal pulvinar in comparison to LIP and FEF

Authors: *R. CHEN[1], *K. DOUGHERTY[1], B. WU[1], A. B. MARTIN[2,1], S. KASTNER[1];
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Abstract: The mediodorsal pulvinar (mdPul) is a thalamic nucleus exclusive to primates, and anatomically connected with dozens of cortical areas (Homman-Ludiye, 2019). mdPul processes both visual and motor signals and is involved in attention through interactions with the frontoparietal network (Fiebelkorn, 2019). To understand the specific role of mdPul relative to its interconnected cortex, we simultaneously recorded neuronal signals in mdPul, frontal eye field (FEF) and the lateral intraparietal (LIP) cortex with multi-contact linear arrays, while monkeys performed a change detection task. In each trial, a cue was presented briefly to indicate the location of an upcoming target. After a variable delay, a circular array of six barrel and bowtie shapes appeared, one of these items changed its contrast after a second delay, monkeys were required to make a saccade to the target to get a juice reward. We identified hundreds of well-isolated single units in each area. We classified neurons into visual, visuomotor and motor groups based on each neuron's responsiveness to visual stimuli and eye movement. We further sub-divided neurons into narrow- and broad-spiking classes according to a bimodal distribution of spike waveform widths to consider properties of putative inhibitory and putative excitatory neurons, respectively. For visual and visuomotor neurons, we found that the visual response latencies of broad-spiking units followed a hierarchical order, with responses initiating in LIP, then mdPul and finally FEF. The narrow-spiking units, in contrast, showed highly overlapping distributions of visual latencies across all three areas. Similar ratios of visuomotor units, exhibiting pre-saccadic activity, were observed across all regions for both broad- and narrow-spiking units. In contrast, a much smaller proportion of pre-saccadic motor units were present in LIP than in mdPul and FEF. In addition to considering the proportion of functional neurons across areas, and their visual latencies, we also examined variability in spiking following the visual cue and prior to saccades by computing Fano factor. We observed that the variability of neuronal responses in LIP and FEF decreased after visual stimuli or saccade onset for both broad- and narrow-spiking units. However, variability reduction in mdPul was only observed for narrow-spiking units. These results show that mdPul shares common functional properties with interconnected cortex, with the exception of spiking variability. This functional difference may result from differences in the anatomical organization of cortex and pulvinar.

Disclosures: R. Chen: None. **K. Dougherty:** None. **B. Wu:** None. **A.B. Martin:** None. **S. Kastner:** None.

Poster

P491. Eye Movements and Visuomotor Processing

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Topic: D.08. Visual Sensory-motor Processing

Support: NIH Grant F30-EY027684
NIH Grant R01-EY037272

Title: Pontine integration of visual motion and visual-motor gain signals in the primate smooth pursuit eye movement system

Authors: *T. R. DARLINGTON, S. G. LISBERGER;
Dept. of Neurobio., Duke Univ., Durham, NC

Abstract: Voluntary movement systems combine many types of signals related to motivation, expectation, and sensory information to generate proper motor commands. Smooth pursuit eye movement is an example of such a sensory-motor behavior and allows primates to smoothly track moving objects of interest with their eyes. Two primary signals are responsible for driving smooth pursuit: visual motion and visual-motor gain. Visual-motor gain controls access of visual motion signals to the oculomotor machinery and may allow extrasensory features like expectation and reward/motivation to influence pursuit. In the cerebral cortex, the middle temporal visual area (MT) provides the visual motion input to the pursuit system; the smooth eye movement region of the frontal eye fields (FEF_{SEM}) controls visual-motor gain.
Our present goal is to move downstream in the pursuit circuit to understand how visual motion and visual-motor gain are combined to drive smooth pursuit eye movement. We recorded the extracellular activity of 63 single neurons in the dorsolateral pontine nucleus (DLPN) and 114 neurons in the nucleus reticularis tegmenti pontis (NRTP) of two monkeys. Both nuclei receive convergent cortical input, though DLPN and NRTP receive most of their input from area MT and FEF_{SEM}, respectively. Monkeys performed smooth pursuit eye movements in a speed context task that consisted of alternating blocks of trials where targets moved predominantly at either fast (80% 20 deg/s, 20% 10 deg/s) or slow (80% 2 deg/s, 20% 10 deg/s) speeds. Preparatory modulation of activity like that in FEF_{SEM} was present in both NRTP and DLPN and encoded expectation of target speed: the modulation of preparatory activity was larger during the fast- compared to the slow-context. We probed the state of visual-motor gain during motor preparation in the fast- and slow-context with brief, 50-ms pulses of local coherent motion at 5 deg/s delivered either early or late after fixation onset. In NRTP, neuronal response amplitudes to the pulses of motion were larger during the fast- versus slow-context and later versus earlier during fixation, consistent with eye speed responses to the pulses. In DLPN, responses were affected by fixation time but not by speed context. In both areas there was a continuum from small to large effects. Our results imply that either: (i) the combination of visual motion and visual-motor gain occurs gradually within NRTP and DLPN with the outputs provided by the neurons with the largest effects; or (ii) NRTP and DLPN represent an early stage of integration of visual motion and visual-motor gain signals with further integration at downstream nodes.

Disclosures: T.R. Darlington: None. **S.G. Lisberger:** None.

Poster

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Topic: D.08. Visual Sensory-motor Processing

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JSPS KAKENHI 18KK0286

Title: Predictive control of smooth pursuit eye movements based on stimulus and behavioral histories

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Abstract: The human oculomotor system requires approximately 100 ms from the onset of target motion to the onset of eye movement, and this delay can lead to a loss of visual information because visual acuity declines as the target moves from the fovea to the retinal periphery. To overcome the inherent delay, the oculomotor system has the ability to perform predictive feedforward control of eye movements. Several studies have demonstrated that preceding changes in eye velocity are generated based on weighted averaging of past stimulus timing even when the target motion is unpredictable due to velocity or direction changes. Here, we attempted to clarify whether not only stimulus history but also behavior history affects predictive pursuit to randomized target velocity. Healthy 12 observers with normal or corrected to normal vision were instructed to track a small target moving horizontally over a distance of 24 deg as accurately as possible. The target moved in an irregular triangular waveform, in which the rightward velocity was fixed at 16 deg/s while the leftward velocity was either fixed (predictable) at one of seven velocities (4, 8, 12, 16, 20, 24, 28 deg/s) or randomized (unpredictable). Eye movements were evaluated when the target reversed direction from right to left. When the future target velocity was predictable, predictive pursuit corresponding to the target velocity was observed. Predictive pursuit was also found even when the future target velocity was unpredictable, but of course the responses did not correspond to the target velocity. To examine the influences of stimulus and behavioral histories on predictive pursuit to the randomized target velocity, a total of 15 linear mixed-effects models were set up with the predictive pursuit response in the current trial (n) as the dependent variable, and the target velocity (stimulus) and the pursuit response (behavior) back to the last 5 trials (n-5) as the

independent variables. These models included those in which each history alone was incorporated into the model and those in which both were incorporated. The model adopted based on the AIC, which incorporated the stimulus and behavioral histories up to the last 5 trials, showed that both histories of the previous two (n-2) or three trials (n-3) had influences on the predictive pursuit responses in the current trial. Our results suggest that the predictive pursuit system allows to track randomized target motion using the information from several previous trials, and the information of target velocity (stimulus) and motor output (behavior) in the past time sequences have partially different influences on predictive pursuit.

Disclosures: T. Miyamoto: None. **Y. Hirata:** None. **A. Katoh:** None. **K. Miura:** None. **S. Ono:** None.

Poster

P491. Eye Movements and Visuomotor Processing

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P491.10

Topic: D.08. Visual Sensory-motor Processing

Support: KAKEN Grant 18KK0286
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Title: Acquisition of predictive optokinetic eye movements in mice and humans

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Abstract: Predictive optokinetic response (pOKR) was found to be acquired by goldfish after repeated temporally periodic visual stimulation (Marsh & Baker, 1997). A recent study demonstrated that the cerebellum is required for the acquisition of pOKR (Miki et al., 2018). However, despite having almost identical cerebellar neural circuitry to goldfish, zebrafish and medaka hardly acquired pOKR, while carp did (Miki et al., 2020). It was also found that all animals that acquired pOKR presented robust optokinetic after nystagmus (OKAN) which is considered as a manifestation of the velocity storage mechanism (VSM). Therefore, the VSM was hypothesized to be another determinant for the acquisition of pOKR (Miki et al., 2020). Here we conducted experiments to test the hypothesis in mice and humans. In the mice experiment, optokinetic visual stimulus (OKS) rotating at 20 deg/s horizontally was given to 3

wild type mice (C57BL/6) for 1 min and suddenly turned off to measure OKAN. The same mice were trained for 90 min with periodic velocity square-wave OKS that rotated bilaterally for 4 s (±5 deg/s) and paused for 2 s in between. Before and after the training, test OKS with twice the training stimulus period was presented for 10 cycles. As the results, the duration of OKAN was found to be very short, and pOKR was hardly acquired in all the mice tested, supporting strongly the hypothesis that pOKR is dependent on the presence of the VSM. In the human experiment, 12 participants wore a head mount display (HTC vive) in which horizontally rotating random dots were projected as OKS. OKAN was measured in each participant by giving the OKS at a constant velocity (40 deg/s) for 18 s, followed by complete darkness. We then presented 5 cycles of test OKS rotating bilaterally (±40 deg/s) for 8 s, followed by 80 cycles of OKS whose period was a half of the test OKS. Immediately after the training, another test OKS was given for 1 cycle. It was found that the duration of OKAN in humans was highly correlated with the strength of the predictive eye movement. However, learning and forgetting the predictive behavior in humans occurred far much rapidly than goldfish and carp, suggesting other mechanisms taken place than reflexive OKR, such as smooth pursuit. To confirm, we trained 5 participants with the same OKS except that foveal regions were occluded binocularly by real-time eye tracking and masking technique. We demonstrate that no predictive component was acquired after the training, confirming that smooth pursuit may play a main role in the predictive eye movement. Taken together, the VSM is required in addition to the cerebellum to acquire pOKR, and it may also determine the ability to acquire predictive smooth pursuit.

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Poster

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Topic: D.08. Visual Sensory-motor Processing

Support: Gösta Fraenckel Foundation for Medical Research
Sigvard and Marianne Bernadotte Research Foundation for Children's Eye Care

Title: Visual attention and general alertness exhibit different modulating influences on the optokinetic reflex

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Abstract: Reflexive neural responses, such as the optokinetic reflex (OKR), represent a valuable clinical and psychometric tool to depict a range of neurological conditions. While previous

studies have illustrated that optokinetic responses are influenced by different levels of attention and alertness, there is currently no study illustrating how the OKN is affected throughout the visual attention spectrum while controlling for general alertness. This study measures torsional OKN (tOKN) to characterize how general alertness and visual attention affect the OKR in healthy individuals. Twelve healthy subjects (8m, 4f; mean age 33, SD = 9.36) with intact gaze-stability, visual acuity, and binocularity were recruited. Subjects were exposed to a rotating visual scene while wearing a head-mounted eye tracker equipped with an accelerometer to ensure head fixation. They were instructed to place the chin over a chin rest and maintain the fixation on a dot placed at the centre of the screen. All subjects were given three tasks requiring low, neutral, and high levels of visual attention. To control for the general level of alertness, the low visual attention engagement was achieved by submitting a distracting task, the Paced Auditory Serial Addition Test (PASAT), which had subjects shift their attentional focus to auditory cues. A repeated-measures ANOVA revealed a significant effect of visual attention on the OKN frequency responses ($F(2, 22) = 12.353$, $p < 0.001$). The number of OKN responses increased with higher attention levels and differed significantly across all conditions. General alertness significantly modulated the OKN gain, i.e., torsional velocity, ($F(2, 22) = 7.144$, $p = 0.006$). Specifically, both low and high attentional tasks, both requiring substantial cognitive alertness, increased OKN gain compared to neutral trials. Higher visual attention level resulting in higher OKN frequency hints at a more efficient integration of visual motion. Such a relationship may be expected from an evolutionary perspective, coupling an increased readiness with faster updating sensorimotor reflexes. Regarding the OKN gain, during the high attention task, it is likely caused by an increase in visual motion processing, whereas during the low attention task it may reflect the heightened alertness caused by auditory cues according to the same evolutionary principles. In conclusion, high attentional and alertness levels independently influenced different characteristics of the OKN, supporting cortical modulation on subcortical sensorimotor processes fundamental for visual perception and visually guided behaviours.

Disclosures: D. Frattini: None. **T. Pansell:** None. **T. Wibble:** None.

Poster

P491. Eye Movements and Visuomotor Processing

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Support: Canadian Foundation of Innovation and Ontario Research Fund (CFI/ORF project no. 37597)
NSERC (RGPIN-2019-06479)
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Connaught New Researcher Awards

CIHR Project Grant

Title: Circuit mechanisms underlying the cortical modulation of optokinetic reflex

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Abstract: In mammals, the visual cortex has an unsurpassed capability of discriminating visual features, such as orientation, depth, and motion. This cortical area sends massive innervation to the brainstem, namely corticofugal projections. Through this pathway, the visual cortex can theoretically impact the behavior mediated by brainstem circuits. But the functional role of those projections was not fully understood. Recently evidence showed that the corticofugal projection from the visual cortex indeed modulated an innate behavior, optokinetic reflex (OKR) which is essential to stabilize retinal images. This cortical modulation can expand the behavioral repertoire of mammals and therefore help animals survive in dynamically changing environments. We decided to use this system to investigate the circuit mechanisms of cortical modulation on innate behaviors. We used viral-based circuit tracing and two-photon functional imaging to explore the structure and function of the circuit involved in this cortical modulation. We found that cortical neurons innervating the OKR circuit primarily came from specific areas. Moreover, those neurons shared similar functional properties. This functionally specific connection will provide insight into the mechanism of how the cortex modulates innate behaviors.

Disclosures: J. Liu: None. **Y. He:** None. **B. Liu:** None.

Poster

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Topic: D.08. Visual Sensory-motor Processing

Support: NIH/NEI Grant R01 EY027857
Research to Prevent Blindness (RPB) / Lions' Clubs International Foundation (LCIF) Low Vision Research Award

Title: Transformer-based computational model of human eye movements for reading

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Abstract: Reading is a highly complex process that involves the integration of visual sensory, oculomotor, linguistic, and cognitive information. Our brain acquires relevant information during

reading by making a series of rapid ballistic eye movements called saccades, interleaved with short fixations. Thus, understanding human eye movements in reading can help us elucidate how our brain searches for and integrates relevant low-level sensory and high-level cognitive information. However, the pattern of eye movements in reading has been primarily explained by low level sensory or oculomotor factors. Thus, little is known about how language context (e.g., the relationship between words & semantic knowledge of words) influences human eye movements for reading.

To address the issue, here we develop an ideal-observer computational model that uses visual sensory, oculomotor, lexical, memory, and language contextual information to simulate human reading behaviour. This ideal-observer model can optimally utilize the statistical properties of all the information available in the text to make saccades as it reads through the text.

To incorporate language contextual information into the model, we adopt a transformer-based language model GPT2 (Radford et al., 2019) that computes the likelihood of upcoming words based on preceding words and contextual information present in the text. We consider the memory component for reading by restricting the amount of context the language model can use. The task of our model is to read through the text in a minimum number of saccades by minimizing entropy introduced in the Mr Chips model (Legge, Klitz & Tjan, 1997). We calculate this entropy using the text information available through a retina, word alternatives using lexicon information, prediction of next word using the previously identified words as context, and the knowledge of oculomotor noise distribution.

We test our model on text excerpts from books (e.g., Alice in Wonderland). The model's saccade strategies were evaluated in terms of saccade lengths, reading speed and regression saccades compared to the Mr Chips model. The simulation results revealed a benefit of using language contextual information as observed in human readers: lesser regressive saccades, faster reading speed, and longer saccade lengths. More details on the emerging properties of the model were analyzed and compared between the two models.

Our findings suggest that our transformer-based computational model may serve as a valuable tool for studying human eye movements for natural reading in normal and clinical populations.

Disclosures: A. Dipani: None. **F. Shamsi:** None. **M. Kwon:** None.

Poster

P491. Eye Movements and Visuomotor Processing

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Program #/Poster #: P491.14

Topic: D.08. Visual Sensory-motor Processing

Support: NIH Grant EY027373

Title: Responses of frontal cortical neurons to changes in stimulus reliability

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Abstract: The responses of frontal cortical neurons depend on the reliability of sensory signals that drive motor behavior. Models based on responses during perceptual decisions suggest the strength of sensory inputs to frontal cortical neurons is set according to stimulus reliability. Models based on responses during smooth pursuit eye movements suggest that reliability modulates the strength of shunting inhibition. While different in implementation, the predictions of both classes of models are quite similar in terms of neural responses at the onset of sensory and/or motor events. However, because shunting inhibition depends on the response of the neuron at any given moment, the predictions of the models diverge as steady-state pursuit behavior and the response of individual frontal neurons evolve dynamically over time.
To evaluate the mechanisms behind the sensitivity of frontal cortical neurons to stimulus reliability, we recorded from neurons in the smooth pursuit region of the frontal eye fields (FEFsem) of a monkey during the initiation and steady-state pursuit of targets that were patches of moving dots. Motion reliability was controlled by adjusting the percentage of dots coherently moving in the pursuit direction. Motion coherence had a strong effect on FEFsem responses throughout the pursuit response. At stimulus onset and during the initiation of pursuit, FEFsem neurons often responded more strongly to higher coherence than lower coherence, replicating previous results that changed reliability using stimulus contrast. The responses, however, were quite diverse if we increased or decreased the coherence of the patch of dots during steady-state pursuit. The strength and sign of the response to the change in motion reliability often was unrelated to the strength and sign of the effects during pursuit onset. We take the disparate effects at pursuit initiation versus steady-state as evidence that the pursuit-related activity of neurons in FEFsem has a non-linear effect on their responses to sensory inputs. We plan to use this result to constrain models of individual neurons that will reveal how sensory reliability and motor responses interact in the frontal cortex.

Disclosures: S.W. Egger: None. **S.G. Lisberger:** None.

Poster

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Program #/Poster #: P491.15

Topic: D.08. Visual Sensory-motor Processing

Support: Vista

Title: Mechanisms for integrating allocentric and egocentric visual information for goal-directed movements: A neural network approach

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Abstract: Allocentric (landmark-centered) and egocentric (eye-centered) visual information are optimally integrated for goal-directed movements (Byrne et al., 2010; Klinghammer et al., 2017). Additionally, neural recordings found landmark-specific coding in supplementary and frontal eye fields when monkeys performed saccades toward a remembered visual target (Schütz et al., 2020; Bharmauria et al., 2020). Nevertheless, the intrinsic coordinate representation and underlying processes for this combination remain a puzzle, mainly due to inadequacy of current theoretical models to explain data at different levels (i.e., behavior, single neuron, and distributed networks). Here, we propose a neural network with physiologically inspired retinal / extraretinal inputs, outputs based on frontal eye field (FEF) properties, and two major 'hidden' components: First, a Convolutional Neural Network (CNN) is used to extract the allocentric and target information. Our CNN performs repeated (2 layers) convolution, rectification, and normalization to first extract low-level features from input images. With the features extracted, the challenge is to combine them to generate an abstract representation (allocentric cues and target position). We address this challenge by training a feature pooling layer at the end of our CNN network. Second, a Multi-Layer Perceptron network (MLP) is used to combine allocentric (extracted feature maps from the CNN) and egocentric information (initial gaze position fed to the network as an additional input). Our MLP consists of two fully connected hidden layers that incrementally transform the allocentric and egocentric representations into an integrated motor response. Finally, a decoder transforms motor responses into final gaze positions. The network input included retinal projection of images and somatosensory coding of initial gaze location while the output was designed to replicate frontal eye field's motor responses. The network was trained on both synthetic and actual monkey gaze behavior. MLP output units accurately simulated FEF motor responses (open-ended response fields that shifted partially with the landmark) and their decoded output achieved good correspondence (MLP: R2 = 0.80) with actual gaze behavior (Bharmauria et al. 2020). These results suggest that our framework provides a suitable tool to study the underlying mechanisms of allocentric and egocentric integration.

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Poster

P492. Perception Studies

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Program #/Poster #: P492.01

Topic: H.02. Perception and Imagery

Support: European Research Council (Advanced iConnect Project Grant ADV 320708) the Netherlands Organisation for Scientific Research (Language in Interaction Project Gravitation Grant 024.001.006)

Title: Open multimodal iEEG-fMRI dataset from naturalistic stimulation with a short audiovisual film

Authors: *J. BEREZUTSKAYA[1,2], M. J. VANSTEENSEL[1], E. J. AARNOUTSE[1], Z. V. FREUDENBURG[1], G. PIANTONI[1], M. P. BRANCO[1], N. F. RAMSEY[1];
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Abstract: Human intracranial (iEEG) data are a valuable but rare source of information about the brain. The few medical centers in the world that acquire iEEG for research purposes suffer from low patient rates (5-10 subjects a year), and due to various reasons cannot easily share the data. This limits iEEG potential for neuroscientific research and hampers scientific reproducibility. Here, we present the first openly shared large multimodal iEEG-fMRI dataset that is unique in several ways. First, it contains a large set of iEEG data (51 subjects who all did the same task). Second, it provides additional fMRI data from the same task (30 subjects). Third, the data were collected using a rich naturalistic stimulus - a short audiovisual film, for which detailed video and audio annotations are provided. The dataset can be used to study neural signal during speech, perception, social discourse and more; develop new analysis techniques and study neurovascular coupling. The dataset is available online (https://openneuro.org/datasets/ds003688). It includes 51 iEEG and 30 fMRI subjects, of which 18 subjects have both iEEG and fMRI data. The majority of the data was recorded from subdural electrode grids. Data have been de-identified (MRI defaced, personal metadata removed) and are provided in the iBIDS format. Handedness and language dominance hemisphere are provided per subject. As the main task patients watched a short film (6.5 minutes) edited together from scenes of Pippi Longstocking (1970) with interleaved blocks of speech and music. For iEEG, additional resting state data (3 minutes) are provided. Rest data was either acquired during a resting state task (26 subjects) or cut from natural rest in continuous 24/7 recordings (24 subjects). FMRI data contains only the audiovisual film task. Detailed annotations are provided with time stamps for spoken sentences, words and phonemes and labeling of the visual content (characters, objects and actions per frame).Subsets of IEEG data have been previously analyzed and published with respect to visual and auditory processing. For the full dataset we report strong responses to the film that are similar across iEEG and fMRI.The present dataset offers an unprecedentedly large amount of iEEG data accompanied by fMRI recordings during a naturalistic cognitive task. Extensive use of these data can help us understand neural processing that supports different aspects of high-level cognition including speech processing, which is central to research on brain-computer interfaces for communication. We believe this shared dataset will help promote collaborative and open science in the field of iEEG as well as the neuroscientific community as a whole.

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Poster

P492. Perception Studies

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Topic: H.02. Perception and Imagery

Support: ANR #18-CE37-0010-01 CONFI-LEARN
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Agence de l'Innovation de Défense - Ministère des Armées
Ecole Doctorale FIRE - Programme Bettencourt

Title: Human brain networks for probabilistic learning explored with ultra-high field fMRI

Authors: *T. BOUNMY[1,2], E. EGER[1], F. MEYNIEL[1];
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Abstract: Faced with a changing and uncertain world, the human brain is able to estimate the probability of future events based on past observations. This ability is key to interacting with our environment from perception to decision making. According to the Bayesian brain hypothesis [1], the brain relies on uncertainty (or inversely, confidence) to correctly update its beliefs. This Bayesian approach characterizes the computational level but the representational and implementational levels of probabilistic learning in humans remain to be elucidated. Here, we probed the neural activity of 26 human adults on a probability-learning task with ultra-high field (7T) MRI. Subjects were shown binary sequences of visual stimuli that follow a Bernoulli process whose probability parameters changed abruptly and unpredictably. Subjects were asked to learn this changing probability and to occasionally report it along with the associated confidence. We analyzed the latent variables of learning with a Bayesian Ideal Observer model to distinguish surprise (log improbability of each observation) and confidence (log posterior precision). Subjects' probability estimates and confidence levels tightly followed the Bayesian model. Confidence levels also showed different normative features of Bayesian inference. fMRI revealed distinct brain networks correlating with confidence (positively in ventromedial prefrontal and frontopolar cortex, anterior temporal lobe, hippocampus and negatively in intraparietal sulcus) and surprise (positively in prefrontal and visual cortices). These results extend and corroborate a previous study that used 3T MRI and a similar task (with an order-1 Markov process instead of Bernoulli process and both auditory and visual sequences) [2]. The brain correlates of confidence (which characterizes epistemic uncertainty) were not confounded by aleatoric variability (which characterizes how predictable the next stimulus is, and which was included in the regression model). By identifying brain networks correlating with the confidence that accompanies learning, our findings contribute to unraveling the neural mechanisms of

probabilistic learning. References: [1] Knill, D. C., & Pouget, A. (2004). *TRENDS in Neurosciences* 27(12), 712-719. [2] Meyniel, F., & Dehaene, S. (2017). *PNAS* 114(19), E3859-E3868.

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Poster

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Topic: H.02. Perception and Imagery

Support: JSPS Kakenhi 20H00235

Title: Partial face cognition in rapid serial visual presentation paradigm

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Abstract: Objective: Under the COVID-19 pandemic situation, wearing masks became a must-do action in our daily life. Regardless that some parts of the face are being covered, humans can still cognize people's faces precisely. On this account, we assumed that people could also cognize faces by only seeing some parts of the face but seeing the whole face would help them to cognize easier. This study aims to examine and compare the response of the brain related to the whole and part face cognition using rapid serial visual presentation (RSVP). Method: The part face conditions examined were faces with eyes covered, nose covered, mouth covered, eyes and nose covered, eyes and mouth covered, and nose and mouth covered to identify the part of the face that provides the most critical information for face cognition. In the experiment, we recorded an electroencephalogram (EEG) on 18 healthy volunteers, 9 male and 9 female (mean age 25.72 ± 2.66, min 21 and max 31). The experiment consisted of two cognition tasks, images displayed at 1000 ms with a button press when the target is shown, and images displayed at 200 ms without any physical responses. Each task contained six unknown faces presented at 20 times each, where one face was set as a target, and all subjects were trained to remember that face priorly. The obtained data were re-reference with an average of earlobe electrodes, filtered using a bandpass filter at 1 to 30 Hz. The artifacts such as eye blinks were removed by independent component analysis (ICA), and an event-related potential (ERP) was calculated by grand averaging among all subjects. Results: We found that part face stimulus provided a more significant event-related response, P300, than whole face cognition in both the tasks, which contrasted with our hypothesis. The face with the nose covered yielded the largest P300 among all face conditions in both a slower RSVP or button press task and a faster RSVP or no button press task. We also compared the ERP obtained in a whole face presentation and face with nose

and mouth covered condition, resembling wearing a mask. The P300 amplitudes in these conditions were produced at a similar level. This result supports the fact that wearing masks or faces with nose and mouth covered does not lower face cognition ability.

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Poster

P492. Perception Studies

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Topic: H.02. Perception and Imagery

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Title: Neural responses to naturalistic stimuli are heritable and related to the brain's intrinsic functional organization

Authors: *D. GRUSKIN[1], G. H. PATEL[1,2];
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Abstract: How do our genes shape our perceptions of the world around us? Recent work using retinotopic and task-free paradigms has established that various aspects of sensory-relevant brain function are under genetic control. However, the extent to which genetic factors underly individual differences in brain responses to dynamic, naturalistic stimuli is still unclear. In the present study, we address this gap by applying multidimensional heritability analyses to movie watching 7T fMRI data from 176 Human Connectome Project participants. First, using a pairwise intersubject correlation (ISC) analysis, we show that BOLD responses to naturalistic stimuli are heritable across both unimodal and, to a lesser degree, heteromodal cortex (average parcel-level $h^2 = .07$; maximum parcel-level $h^2 = .18$, $p_{perm} < .0001$). Network- and region-level analyses reveal that heritability effect sizes and spatial topologies are consistent within functional networks and across different stimuli and days of data collection (average spatial correlation $\rho = .92$, $p_{spin} < .001$). Next, we use functional alignment to demonstrate that this heritability in part reflects shared latent representations of sensory information that are independent from genetic similarity in functional topology. Finally, we describe relationships between individual differences in movie-evoked brain activity and variability in the brain's intrinsic functional architecture, illustrating how intra-regional activations are related to heritable patterns of inter-regional communication. Taken together, these findings establish a genetic basis for individual

differences in the neural processing of audiovisual stimuli and advance the study of genetic influences on sensory processing into the naturalistic domain.

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Poster

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Topic: H.02. Perception and Imagery

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Title: Towards a Mechanistic Account of Perceptual Curiosity

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Abstract: Realistic environments are rife with uncertainty. Often the uncertainty is sensory, requiring us to clarify ambiguous inputs. Is the shadowy figure I see on my hike a bear or a rock? Recent results suggest that perceptual curiosity, the intrinsic desire to reduce sensory uncertainty, is crucial for sensory processing and memory but its neural basis is poorly understood. Using a novel behavioral task, we asked whether and how perceptual curiosity is related to neural representations of confidence and sensory uncertainty and how it modulates episodic memory. During fMRI, participants viewed a distorted image ("texform") of an animal or inanimate object and rated their confidence in recognizing the image and their curiosity to see the original (undistorted) image. At the end of the session, participants took a surprise memory test for the ambiguous texforms. We show that, like curiosity about trivia questions, perceptual curiosity has a negative quadratic (inverted-U) relation with confidence. Moreover, perceptual curiosity was predicted by two separate neural representations of sensory uncertainty. One was a multivariate representation of uncertainty about stimulus category in occipitotemporal cortex (OTC), measured based on the relative correlations between a multivoxel pattern evoked by a texform and separately acquired animate and inanimate template activity patterns. In addition, a portion of the perigenual anterior cingulate cortex (pgACC) provided a univariate response that correlated with subjective reports of confidence and predicted curiosity independently of the OTC. The results reveal the neural signatures of uncertainty that predict subjective judgments of

perceptual curiosity. Future analyses will uncover how the brain's memory systems interact with OTC and pgACC to support memory for ambiguous events.
+ These authors are co-PIs.

Disclosures: M. Cohanpour: None. **M. Aly+:** None. **J.P. Gottlieb+:** None.

Poster

P492. Perception Studies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P492.06

Topic: H.02. Perception and Imagery

Title: The role of entropy, mutual information and synchronization in perception and action

Authors: *D. GUPTA;
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Abstract: A large and growing volume of experimental data have shown that during action and perception there are increases in the spiking activities in the cortical areas, which are synchronized by neural oscillations of different bandwidths. The spiking activities are random (high entropy) if there is no knowledge about actions or stimulus characteristics. However, the knowledge about the action or the stimulus characteristics will resolve the surprise in the spiking patterns. The knowledge about the action or the stimulus is an outcome of a successful interaction with the physical surroundings. A successful outcome for the organism will increase the knowledge about the external stimulus or object, such as the juiciness of a fruit due to changing shape, but fixed volume - processed during interaction as a low-dimensional feature. Furthermore, note that spiking patterns, given no knowledge about the source, such as stimulus or action-initiation, are stochastic due to sparse interconnections between the cortical neurons. The decrease in entropy has an energy equivalent when there is a gain in the knowledge about stimulus (perception) and action (voluntary movement). This gives a physical basis to perception and voluntary movements that are comparable between individuals. We will investigate, using theoretical considerations, the changes in mutual information and entropy due to sensory processing and synchronization to understand the information theoretic basis of action and perception. There are increased correlations between pairs of neurons activated by different stimulus characteristics and/or low-dimensional stimulus features, which will decrease the joint entropy between pairs of neurons in the cortex, increasing the energy equivalent of the gain in the knowledge about action and stimulus (perception). The processing of sensory stimulus is responsible for the increase in entropy measured in the spiking activities. Moreover, synchronization will selectively increase the number of pairs of neurons showing correlated activity specific for the task. Consequently, there will be fewer number of possible spiking states

in the brain as a result of synchronization since synchronized neurons will represent a single macrostate, resulting in a reduction of entropy.

Disclosures: D. Gupta: None.

Poster

P492. Perception Studies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P492.07

Topic: H.02. Perception and Imagery

Support: BioTechMed-Graz Young Researcher Group Grant

Title: Human parietal cortex responses to static illusory figures and their modulation by attention

Authors: *A. ARSENOVIC[1,2], A. ISCHEBECK[1,2], N. ZARETSKAYA[1,2];
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Abstract: Investigating visual processing of illusory figures is an important approach for identifying the mechanisms of perceptual organisation and their neuronal correlates. Until now, neuronal representations of static illusions, such as the Kanizsa figure, have been demonstrated in the early visual cortex and in the ventral stream of the visual processing hierarchy. On the other hand, the dorsal stream, including the superior parietal areas, has been shown to process illusory figures defined by motion. It remains unclear whether parietal cortex only reacts to motion-defined illusory shapes or to illusory shapes in general. In the current study we tested if parietal cortex also responds to static Kanizsa figures. In addition, since parietal cortex is known to be involved in attention, we also tested whether and how this shape-related activity is modulated by attention. Seventeen healthy human participants took part in one fMRI session. The participants performed a central detection task, which varied in difficulty on two levels (easy - detection of a specific colour; hard - detection of a specific symbol). Irrelevant to the task, four flickering black inducers were presented simultaneously on the display around the fixation task. The inducer configurations were either forming an illusory diamond in the centre, a triangle in the left or the right hemifield, or were inverted such that no illusory figure was formed. All conditions were presented for 12 s per trial in a block design. The task difficulty varied between the blocks. We performed a surface-based GLM analysis comparing responses to each illusory figure with the non-illusory condition for the parietal regions of interest (IPS0, IPS1, IPS2, IPS3, IPS4, IPS5, SPL1). Most of these areas showed stronger responses to the diamond compared to inverted inducers. Interestingly, IPS0-3 also showed a significant response modulation by the triangle location, preferring the triangles in the contralateral hemisphere. While the overall response to the triangles (compared to inverted inducers) was attenuated by attention, the

contralateral triangle preference was not. Our results demonstrate that, along with the motion-defined illusory shapes, parietal cortex also responds to static illusory Kanizsa figures. These responses were attenuated, but not eliminated by diverting attention to a central fixation task. Importantly, our results also show that diverting attention away from the illusory figure attenuates the overall shape response, but not the spatially specific response to the exact location of illusory triangles. Overall, they imply that the parietal cortex plays a general role in representing illusory content.

Disclosures: A. Arsenovic: None. **A. Ischebeck:** None. **N. Zaretskaya:** None.

Poster

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Program #/Poster #: P492.08

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Title: Causal roles of prefrontal cortex during spontaneous perceptual switching depend on brain state dynamics

Authors: *T. WATANABE;
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Abstract: Dynamics are among the essential properties of human cognition, and the prefrontal cortex (PFC) is thought to control such cognitive fluctuation. However, in bistable visual perception using a structure-from-motion stimulus, no direct evidence for such presumable prefrontal causal roles has been reported, resulting in debates on PFC functions in spontaneous perceptual inference. To tackle this controversy, by combining an energy landscape analysis with a non-invasive transcranial magnetic brain stimulation (TMS), we tracked brain state dynamics in an online manner and intervened a focal neural activity when the whole-brain activity pattern was staying in a specific brain state. This experiment revealed that the prefrontal causality during bistable visual perception depends on brain state dynamics. For three PFC regions—right frontal eye fields (FEF) and anterior/posterior dorsolateral PFCs (a/pDLPFCs)—, the causal behavioural responses were detectable only when we modulated the PFC activity in state-/state-history-

dependent manners. These behavioural effects were explainable by transient changes in the brain state dynamics that underpinned the fluctuating perception, and such neural responses were attributable to structural transformations of hypothetical energy landscapes. Moreover, these findings revealed how the three PFC areas affected the brain state dynamics: aDLPFC enhances the integration of the two PFC-active brain states, whereas pDLPFC supports the diversity between them; FEF stabilises one of the PFC-active states. Furthermore, we found that the state-/state-history-dependent neural stimulation had accumulative effects, indicating its clinical potential. This work resolves the controversy over the PFC roles in spontaneous perceptual switching and highlights the importance of considering brain state dynamics in investigation of brain-behaviour causality.

Disclosures: T. Watanabe: None.

Poster

P492. Perception Studies

Location: Virtual Only

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Program #/Poster #: P492.09

Topic: H.02. Perception and Imagery

Support: NICHD K25HD096116
NU-URAP 1380URAS199082120817
NU-SURG 1271SUMMER2119608

Title: Effects of volitional activity on detecting a tactile stimulus in individuals post-hemiparetic stroke: a pilot study

Authors: *N. CAI[1], E. G. MEDINA[2], S. A. GONZALEZ[2], A. DUONG[2], N. GURARI[3]; [1]Dept. of Physical Therapy and Human Movement Sciences; NUIN Program, [2]Dept. of Physical Therapy and Human Movement Sci., [3]Dept. of Physical Therapy and Human Movement Sciences; Dept. of Mechanical Engin., Northwestern Univ., Chicago, IL

Abstract: In populations without neurological impairments, the detection threshold of tactile stimuli can be altered during volitional muscle activation. Literature suggests that an increased threshold during movement would arise due to gating of tactile sensory signals, likely, in part, from presynaptic afferent inhibition by interneurons. In individuals with hemiparetic stroke, the baseline activity of interneurons is heightened, which could amplify the gating of tactile sensory signals. However, it remains unclear whether volitional muscle activation post stroke would further modulate interneuron activity, altering the gating of tactile sensory signals and, subsequently, ability to detect the stimulus. Therefore, here we determined the impact of volitional activation on the ability to detect a tactile stimulus in individuals with hemiparetic stroke.Four males with hemiparetic stroke (age: 63±2 years, UE-FMA: 43±10, years since

stroke: 8±2) participated in this study. Participants were situated in a Biodex chair with their forearm affixed to a mechatronic device, which was instrumented with a torque sensor. Participants verbally indicated when they detected a force stimulus, which was administered via von frey filaments (0.07g, 0.4g, 2.0g, 4.0g, 10g, and 300g). This task was performed when participants remained at rest and when they isometrically flexed at the elbow to 25% and 50% of their maximum voluntary torque (MVT). The detection threshold of this tactile stimulus was obtained at each arm using an up-down staircase method.Participants had a detection threshold with a mean±standard deviation (μ±σ) of 1.58±0.90g at the non-paretic arm and 1.56±0.90g at the paretic arm when at rest. When participants flexed their elbow to 25% MVT, their μ±σ detection threshold increased to 2.56±2.44g at the paretic arm and 2.00±0.00g at the non-paretic arm. Finally, when flexing their elbow to 50% MVT, their μ±σ detection threshold was 3.56±2.79g at the paretic arm and 2.05±0.74g at the non-paretic arm.
In both arms, the detection threshold increased during volitional activation in our participants with hemiparetic stroke, in accordance with previous studies in individuals who are neurologically intact. Additionally, when our participants increased their flexion torque, their detection threshold correspondingly increased. Finally, the increase in the detection threshold as a function of generated torque was greater at the paretic arm than at the non-paretic arm. Combined, these results provide early evidence that sensory gating differs between the paretic and non-paretic arms, possibly because of altered interneuron activity post stroke.

Disclosures: N. Cai: None. **E.G. Medina:** None. **S.A. Gonzalez:** None. **A. Duong:** None. **N. Gurari:** None.

Poster

P492. Perception Studies

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Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P492.10

Topic: H.02. Perception and Imagery

Title: Characteristics of speech transmission of distantly-presented bone-conducted sounds in the audible-frequency range

Authors: *H. ISHIKAWA, S. OTSUKA, S. NAKAGAWA;
Chiba Univ., Chiba, Japan

Abstract: Although the upper frequency limit of human hearing is believed to be approximately 20 kHz, several studies have reported that high-frequency sound up to at least 100 kHz can be perceived clearly by bone-conduction (BC). This audible ultrasound through BC is referred to as bone-conducted ultrasound (BCU). BCU can be perceived even by the profoundly sensorineural hearing-impaired and has been applied to a novel hearing aid. On the other hand, in BC, a vibrator needs to be pressed onto the osseous parts of the temporal bone, such as the mastoid

process. Therefore, BC is often accompanied by pain and esthetic problems. In order to solve these problems, “distant presentation” has been proposed. In the distant presentation, vibrators are presented to the neck, upper limb or trunk. Our previous studies focused on the perception and propagation characteristics of distantly-presented BCU and an application to a novel audio-interface. On the other hand, a limited number of studies have been conducted on distantly-presented BC in the audible-frequency range. We have measured hearing thresholds and frequency- and temporal resolutions of distantly-presented BC sound in the audible-frequency range and showed that although distance attenuation is occurred largely as compared to BCU, no significant degradation of frequency and temporal information occurs in the propagation process of BC sound in the body. However, the clarity and sound quality of the transmitted speech sound should be investigated in more detail. In this study, to examine the characteristics of speech transmission of distantly-presented BC sound in the audible-frequency range, word intelligibility and monosyllable articulation tests were conducted in Japanese. As results, the intelligibility and articulation decreased as the stimulus placement got further from the head. This result indicates that amplitude information of the words and monosyllables was lost in the propagation process in the body. In addition, to examine the distortion of speech generated during the propagation inside the body, we composed confusion matrices based on the result of the monosyllable articulation test. In the phonemes of /tj/, /sj/, /zj/, significant differences were observed between percent corrects when the speech sounds presented to the mastoid process and trunk. The result indicates that degradation of high-frequency components of the speech sound occurred in the propagation process and it became more difficult to perceive the phonemes whose energy is concentrated in high-frequency range.

Disclosures: H. Ishikawa: None. **S. Otsuka:** None. **S. Nakagawa:** None.

Poster

P492. Perception Studies

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Program #/Poster #: P492.11

Topic: D.07. Vision

Support: SC & JM: NIH EY030998
GS: NSF GRFP 2020305476
JY: K99 EY032179

Title: Laminar Organization of Pre-Saccadic Attention in Marmoset Area MT

Authors: *S. H. COOP[1], G. SARCH[2], J. L. YATES[3], J. F. MITCHELL[1];
[1]Brain and Cognitive Sci., Univ. of Rochester, Rochester, NY; [2]Neurosci., Carnegie Mellon Univ., Pittsburgh, PA; [3]Univ. of Maryland Col. Park, College Park, MD

Abstract: Attention leads eye movements producing perceptual enhancements at the saccade target immediately before saccades (Deubel and Schneider, 1996; Kowler et al, 1995; Rolfs and Carrasco, 2012). This pre-saccadic attention has been related to enhanced neural responses before saccades made into a neuron's receptive field in macaque visual area V4 (Moore and Chang, 2009), but it is unknown how general the phenomena is and how it is organized across cortical layers. Here, we examined pre-saccadic attention in the middle temporal area (MT) of the marmoset monkey and took advantage of their smooth cortical surface to measure neural effects as a function of laminar position. In one animal, we used high density silicon arrays (NeuroNexus, 64 channel) to record from the full cortical depth. We find that current source density (CSD) methods, traditionally used in primary visual cortex, provide estimates of the input layer in area MT based on an early latency sink. Next we examined pre-saccadic attention in a saccade foraging paradigm. In each trial, the marmoset made a saccade from a central fixation point to one of three equally eccentric stimuli (full coherence dot fields with motion sampled independently between apertures, each from 16 directions). We positioned stimuli such that one foraged location overlapped the receptive fields of neurons under study and examined how tuning functions for motion direction changed when a saccade was planned towards or away from the receptive field. We fit the neural responses from -50 to 30ms around saccade onset with an adjusted Von Mises function that estimated both additive and gain changes in the tuning. We found that saccades towards the receptive field were associated with increases in gain that were prominent only in superficial layers while additive increases were shared across all layers. We also examined the extracellular waveform shapes and found a bi-modal distribution of peak-to-trough durations (i.e., narrow and broad spiking categories). In particular, the gain increases among superficial layer neurons were specific to the broad spiking category. These results indicate a diversity of function across layer and cell type that will be important towards constraining circuit level models of pre-saccadic attention.

Disclosures: S.H. Coop: None. **G. Sarch:** None. **J.L. Yates:** None. **J.F. Mitchell:** None.

Poster

P559. Posture and Gait: Kinematics, Muscle Activity, Exercise and Fatigue, and Biomechanics I

Location: Virtual Only

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Program #/Poster #: P559.01

Topic: E.06. Posture and Gait

Support: NIH Grant -R01AG050672

Title: Neuromuscular mechanisms of motor adaptation to repeated gait-slip perturbations in older adults

Authors: S. WANG, ***T. BHATT**;
Univ. of Illinois at Chicago, Chicago, IL

Abstract: Background: Individuals can rapidly develop adaptive skills for fall prevention after their exposure to the repeated-slip paradigm, and the fall-resisting skills acquired from this training session can be retained for at least 6 months. However, the changes in neuromuscular control contributing to such motor adaptation after the perturbation training remains unclear.Aim:This study investigated changes in neuromuscular control across different stages of slip-adaptation by examining changes in muscle synergies (temporal and spatial components).Method:Electromyography signals during repeated slip trials in gait were collected for 30 healthy older adults. Muscle synergies in novel slip trial, early (6 to 8) and late trials (22 to 24) were extracted respectively using non-negative matrix factorization algorithm, and then the similarity between the recruited muscle synergies in three different phases (no adaptation, early-adaptation, and late-adaptation) was analyzed.Results:Our results showed that participants made significant improvements in their balance outcomes from novel slips to the adapted slips (balance loss rate dropped from 100% to 0%), Correspondingly, there was a significant increment in the muscle synergy numbers from no-adaptation slips to the adapted slips. The participants retained majority of muscle synergies (5 out of 7) used in novel slips post adaptation (both early- and late-adaptation stages), while the activation of these common synergies became lower in the adapted slips. A few specific patterns (n=8) of muscle synergies presented in the early-adaptation stage to compensate the motor errors due to external perturbation, in the late-adaptation stage, only 2 out of these 8 new synergies were retained. Conclusion:We found that with repeated slip adaptation individuals could acquire new muscle synergies through refinement of the pre-existing ones in the late adaptation phase could produce motor strategies that could effectively assist in recovery from the slip perturbation. During the early adaptation phase the CNS generates redundancy synergies that are eliminated as the adaptation process progresses with repeated exposure to the slips. This study enhances our understanding of the neuromuscular basis for adaptation to slips, which could assist with designing perturbation-based fall prevention training.

Disclosures: S. Wang: None. **T. Bhatt:** None.

Poster

P559. Posture and Gait: Kinematics, Muscle Activity, Exercise and Fatigue, and Biomechanics I

Location: Virtual Only

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Program #/Poster #: P559.02

Topic: E.06. Posture and Gait

Support: Clinical and Translational Science Award (CTSA) program, through the NIH National Center for Advancing Translational Sciences (NCATS), grant UL1TR002373. The content is solely the responsibility of the authors.

Title: Vision and proprioceptive effects on force intersection point in quiet human standing

Authors: *K. GRUBEN[1], J. BARTLOFF[1], C. GROVE[2];
[1]Univ. Wisconsin, Madison, WI; [2]Johns Hopkins Univ., Baltimore, MD

Abstract: This study evaluated effects of reduced and disruptive visual stimuli and altered proprioception on sagittal-plane coordination during quiet human standing. Ten human female volunteers (aged 23 to 62 years) 5 with motion sickness and 5 age-matched controls, stood quietly for 30s in various conditions, repeated on four separate days within a 6-week period. The volunteer participants provided informed consent to a protocol approved by the local IRB. Coordination was assessed with the frequency-dependent intersection point analysis which characterizes the emergent property of the ground-on-feet force (F) to exhibit an intersection point (IP) within narrow frequency bands. The height of the IP (zIP) reflects the ratios of muscle torque at the primary joints and summarizes the complex interaction between the multi-segmental inverted pendulum dynamics and neuromuscular coordination strategies. Three representative frequencies (1, 2.2, 3.4 Hz) were chosen to characterize the zIP dependence on frequency. Previous work showed that sloped surfaces and volitional co-activation alter the zIP in a systematic frequency-dependent manner. Visual and proprioceptive inputs typically contribute to feedback during quiet standing, though the contributions vary across individuals. This study manipulated visual input via reduced complexity and disruptive character. The reduced complexity stimulus displayed a single world-fixed vertical bar of white light directly in front of the individual. The disruptive stimulus was an annulus of dots that filled most of the visual field and spun counterclockwise (30 deg/s) about the visual axis. The stimuli were delivered in a visual void with a virtual reality headset. This study also altered proprioceptive feedback with a foam pad under the feet. F was measured with custom 6-axis force plates. Student's t-tests (alpha = 0.05) with Holm-Bonferroni adjustment assessed the comparisons. Results show that on a firm surface viewing the light bar in the VR headset as compared to viewing the laboratory raised the zIP at the higher two frequencies. Standing on foam, compared to firm, increased zIP at the higher two frequencies when viewing the laboratory. However, with VR, standing on foam decreased zIP at 1 Hz, regardless of visual stimulus. These results provide evidence that the zIP provides a means of discriminating among the effects of visual and proprioceptive influences on the frequency components of coordination that ensure upright posture during quiet standing. Future work is needed to elucidate the mechanisms by which visual and proprioceptive feedback are integrated into this metric of coordination.

Disclosures: K. Gruben: None. **J. Bartloff:** None. **C. Grove:** None.

Poster

P559. Posture and Gait: Kinematics, Muscle Activity, Exercise and Fatigue, and Biomechanics I

Location: Virtual Only

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Program #/Poster #: P559.03

Topic: E.06. Posture and Gait

Support: NIH RO1 NS113746
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MnDrive Postdoctoral Fellowship to JWC

Title: Progression of gait disturbances in people with Parkinson's disease and REM sleep without atonia

Authors: D. J. WEAVER[1], S. L. AMUNDSEN HUFFMASTER[1], J. CHUNG[2], C. LU[1], M. E. LINN-EVANS[1], S. E. COOPER[1], A. VIDENOVIC[4], M. J. HOWELL[1], *C. D. MACKINNON[3]; [1]Neurol., [2]Dept. of Neurol., [3]Univ. of Minnesota, Minneapolis, MN; [4]Neurol., Massachusetts Gen. Hospital, Harvard Med. Sch., Boston, MA

Abstract: Approximately 30 to 40% of people with Parkinson's disease (PD) have REM sleep behavior disorder (RBD), a parasomnia characterized by increased muscle activity during REM sleep (termed REM sleep without atonia, RSWA) and dream enactment. The co-expression of PD with RBD is associated with increased motor and cognitive impairment, including postural instability and gait disturbances, compared to those without RBD. Little is known about the association between the presence and expression of RSWA (measured by the level of muscle activity during REM sleep) and the rate of progression of gait impairment in people with PD. This study examined the progression of gait impairment in people with mild-to-moderate PD, with and without RSWA, and healthy controls over the course of three years. We hypothesized that: (i) people with PD with RSWA (PD+RSWA) would show a significantly higher rate of progression than the group without RSWA (PD-RSWA) and controls and (ii) baseline RSWA scores would predict the rate of progression of gait impairment. Forty-one people with PD (22 PD+RSWA; 19 PD-RSWA) and 21 age- and sex-matched controls participated. Testing was conducted off Parkinson's medications. Spatiotemporal parameters of steady-state gait were obtained using a GAITRite Electronic Walkway (CIR Systems, Inc.) (minimum of 31 steps). A linear mixed model tested for differences in progression (change from baseline to follow-up) between groups and sides (least affected vs. more affected). Linear regression examined the relationship between baseline RSWA scores and gait progression in the PD groups. Forty-eight individuals were tested at follow-up (16 in each group). Of those lost to follow-up, 43% were in the PD+RSWA group (PD-RSWA: 21%, controls: 36%). The mean time between baseline and follow-up tests was 35.3 ± 6.8 months. Stride length decreased over time in both PD groups, but the change was only significantly different from controls in the PD+RSWA group ($p < 0.030$). The average percentage change in stride length was -7.9, -3.9, and -1.2% in the PD+RSWA, PD-RSWA and control groups, respectively. There were no main effects of group or side, nor

interactions, for cadence and gait speed. RSWA scores at baseline did not predict the changes in gait over three years. These findings provide evidence that the presence of RSWA in early PD is a harbinger of rapid progression in gait impairment.

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Poster

P559. Posture and Gait: Kinematics, Muscle Activity, Exercise and Fatigue, and Biomechanics I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P559.04

Topic: E.06. Posture and Gait

Title: Mechanical coordination of wrist and MP joint motion during typing

Authors: *T. ITO, M. YOSHIDA, H. FUNAKOSHI, T. KOKUBUN;
Grad. Sch. of Saitama Prefectural Univ., Koshigaya, Japan

Abstract: [1. Motivation]Repetitive typing movements are associated with the development of tendonitis of the hand (Gerr 2002). Coordinated movements have been evaluated in single keystrokes (Dennerlein 2006), but mechanical coordination has not been evaluated in freestyle typing. In this study, we use the Induced acceleration analysis (IAA) method, which consists of inverse and forward dynamics calculations to assess the effects of the wrist and metacarpophalangeal (MP) joint muscle torques on the generation of wrist acceleration. This study aims to clarify the mechanical interaction between hand segments.
[2.Methods]The subject was one healthy adult male. He provided written informed consent, following a detailed explanation of the study's purpose and risks according to the Declaration of Helsinki. He was 21 years old, right-handed, and a 10-finger typist. We used a 3D motion capture system (Vicon) and a QWERTY keyboard (Microsoft). The task was 5,000 Japanese characters. We recorded the sagittal plane of the right side of the wrist and the MP joint of the index finger angle, velocity, and acceleration during the task. The analysis interval was the time of five consecutive keystrokes in the right-hand region. The analysis items were (1) joint angles, (2) correlation coefficients, and (3) mean values (standard deviations; SD) and coefficients of variation of the contribution of joint muscle torques by IAA. This study was approved by the ethics review committee.
[3.Results]The mean (SD), maximum, minimum value, and coefficient of variation of the wrist joint extension angle were 41.8° (2.54), 45.0°, 37.3°, 0.06. The MP joint angle was 32.7° (4.11), 37.3°, 25.1°, 0.13. The wrist joint extension angle and the MP joint flexion angle were 42.5° and 34.4°. The correlation coefficient was 0.98. The mean (SD) and coefficient of variation of the

contribution were 83.2% (0.01) and 0.01 by the wrist muscle joint torques, and 16.5% (0.01) and 0.04 by the MP joint muscle torques.
[4.Conclusion/implications]The inertia force (centrifugal force and Coriolis force) generated by joint motion distal to the wrist joint had little effect on wrist motion at any time of the keystroke. The ratio of the contribution of the wrist and MP muscle torques to the velocity of wrist flexion/extension was kept constant, and the wrist and MP joint angles changed synchronously. This kinematic strategy is thought to be related to the efficiency of hand movements and to ensure the steadiness of successive keystrokes. The disruption of this motor strategy may be one of the motor mechanisms involved in developing tendonitis of the hand.

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Poster

P559. Posture and Gait: Kinematics, Muscle Activity, Exercise and Fatigue, and Biomechanics I

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Program #/Poster #: P559.05

Topic: E.06. Posture and Gait

Support: NIH F31AG069412
American Society of Biomechanics Graduate Student Grant-in-Aid

Title: Neural control of ankle impedance is more efficient during movement than posture

Authors: *K. JAKUBOWSKI[1,2,4], D. LUDVIG[1,4], S. S. M. LEE[2,5], E. J. PERREAULT[1,3,4]; [1]Biomed. Engin., Northwestern Univ., Evanston, IL; [2]Physical Therapy and Human Movement Sci., [3]Physical Med. and Rehabil., Northwestern Univ., Chicago, IL; [4]Shirley Ryan AbilityLab, Chicago, IL; [5]Dept. of Biomed. Physiol. and Kinesiology, Simon Fraser Univ., Burnaby, BC, Canada

Abstract: Regulating ankle impedance—the torque evoked by an imposed displacement—is critical for ambulation and postural control. The impedance of the triceps surae muscles and Achilles tendon are the dominant contributors to ankle impedance in the sagittal plane during plantarflexion, but the nervous system can only regulate muscle impedance. Since these structures are connected in series, the most compliant structure will largely determine ankle impedance. However, their relative impedance during the posture and movement tasks commonly performed by the ankle is unknown. The objective of this study was to quantify these relative impedances during posture and movement to determine the limits on how the nervous system can leverage the tunable mechanical properties of muscle to regulate ankle impedance. We quantify triceps surae, Achilles tendon, and ankle impedance using a technique that combines joint-level measurements with ultrasound imaging. Ten healthy young adults had their

right foot attached to a motor that controlled ankle position and measured torque. Since muscle and tendon impedance change with torque, subjects were instructed to produce a constant plantarflexion torque aided by visual feedback. Subjects completed two tasks: a movement task where the motor moved the subject's ankle through a 20-degree sinusoidal motion at 0.5 Hz and a posture task where subjects performed isometric contractions at four ankle angles within the same range as the movement task. Ultrasound imaging of the medial gastrocnemius muscle-tendon junction was used to quantify muscle and tendon motion. The motor imposed small stochastic perturbations in all tasks to estimate impedance using time-varying system identification. We present the stiffness component of these impedances.
Across all angles, we observed a 39%, 55%, and 17% decrease in ankle, muscle, and tendon stiffness during movement compared to the posture task (all $p < 0.001$). During the posture task, the muscle was 2.6 times stiffer than the tendon, but the muscle was only 1.4 times stiffer during movement. Therefore, the Achilles tendon, a passive structure, is the dominant contributor to ankle stiffness during posture, but the triceps surae and Achilles tendon contribute more equally during movement. Additionally, a unit change in muscle activity would cause only a 28% change in ankle stiffness during the posture task but a 42% change during movement. Given that the nervous system can only actively regulate muscle stiffness, the nervous system will have better control of ankle stiffness during movement compared to posture even though the net stiffness for a given level of muscle activation is lower during movement.

Disclosures: K. Jakubowski: None. **D. Ludvig:** None. **S.S.M. Lee:** None. **E.J. Perreault:** None.

Poster

P559. Posture and Gait: Kinematics, Muscle Activity, Exercise and Fatigue, and Biomechanics I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P559.06

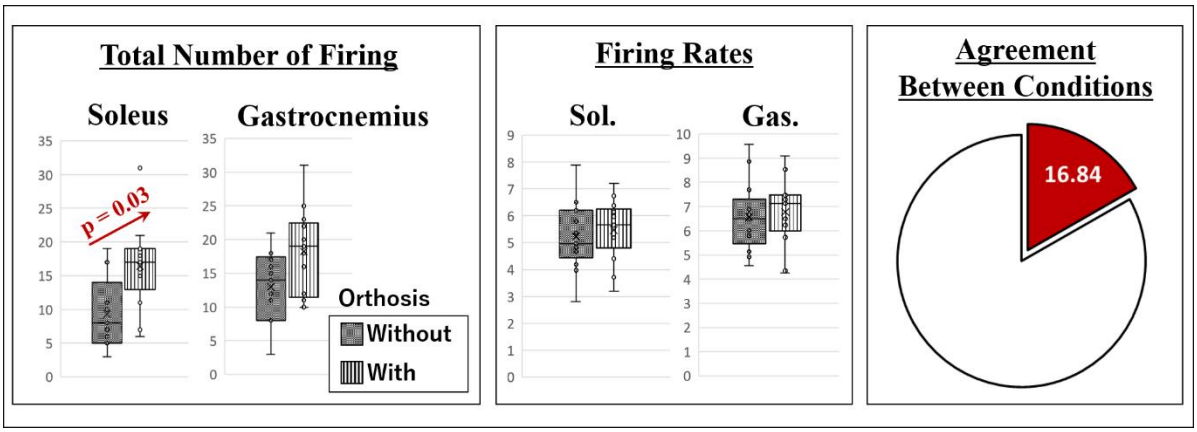
Topic: E.06. Posture and Gait

Support: JSPS KAKENHI Grant 20K23260
JSPS KAKENHI Grant 21K17461

Title: Behavior of motor units of the triceps surae in orthotic-assisted heel-raise

Authors: *H. HANAWA[1], K. HIRATA[2], T. MIYAZAWA[1], T. FUJINO[1], K. KUBOTA[3], M. YOKOYAMA[3], Y. MATSUMOTO[3], N. KANEMURA[4];
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Abstract: The heel-rise is frequently performed to train patients who are unable to plantar flex their ankle joints. Patients are often assisted by therapists because their whole-body muscle strength is impaired. However, it is unclear how the muscle activity of the ankle plantar flexors (i.e., triceps surae) changes with the assistance. Furthermore, since patients often have neurological problems, it is important to observe not only the amplitude of the compound action potential but also the behavior of the motor units. In this study, we examined the number and frequency of motor unit firings in the triceps surae during the heel-raise in 14 healthy adults (all were females, 18-20 ages) with and without a knee contracture brace. In addition, the Peason's correlation coefficient was used to verify whether the same motor units were detected across conditions; $r > 0.8$ was considered identical. The high-density surface electromyogram used to detect the motor units was Trigno Galileo (DELSYS, Inc.). Correspondence t-test was used for comparison between conditions. Significance level was set at $p = 0.05$. As a result, the number of motor unit firings of the triceps surae was higher under the condition with orthosis ($p = 0.03$). On the other hand, there was no significant difference in the frequency of firing of motor units between the conditions. In addition, the number of the same motor units detected between the conditions was only 16.84%. We cannot conclude the efficacy of using the orthosis during training, and further verification is necessary because of the possible limitation of detecting motor units from four electrodes of the measurement device.



Disclosures: H. Hanawa: None. **K. Hirata:** None. **T. Miyazawa:** None. **T. Fujino:** None. **K. Kubota:** None. **M. Yokoyama:** None. **Y. Matsumoto:** None. **N. Kanemura:** None.

Poster

P559. Posture and Gait: Kinematics, Muscle Activity, Exercise and Fatigue, and Biomechanics I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P559.07

Topic: E.06. Posture and Gait

Support: NSF Grant 2015317

Title: Neural control of rhythmic limb motion is shaped by size and speed

Authors: G. P. SUTTON[1], ***N. S. SZCZECINSKI**[2], R. D. QUINN[3], H. J. CHIEL[4];
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Abstract: Over many size and time scales, behaviors such as locomotion or feeding require cyclic mechanical movements. Size and time in turn determine a behavior's dominant mechanical properties: mass, stiffness or viscous damping. The constraints for limbed behaviors can thus be quantified by two variables: limb size and limb cycle time, defining a 'mechanics space' that shows the relative magnitude of each mechanical property for animals ranging from fruit fly to elephant. The mechanics space is continuous, but exhibits three primary regions: 1) an inertia-dominated region; 2) a gravity or elastic-force-dominated region; and 3) a viscous-force-dominated region. In the inertia-dominated region, associated with large limbs moving rapidly, muscle work is translated primarily into kinetic energy. Thus, stable motion requires compensatory control and active damping. In the elastic-force-dominated region, associated with small limbs moving slowly, muscle energy is translated primarily into gravitational or elastic potential energy. These motions are relatively insensitive to external perturbations and may require less active control than inertia-dominated motions. Lastly, in the viscous region, associated with small limbs moving quickly, joint viscosity acts to damp actuation, enabling the joint to dissipate most disturbances passively. A consequence of this analysis is that the relevant zone for a given behavior can be determined from not only the mechanics of the behavior (directly measuring forces within the limb), but also from analysis of the phase relationship between muscle force and limb position; with a 180 degrees relationship between force and position being a hallmark of the inertial dominated region, a 0 degree (in phase) relationship being a hallmark of the elastic-dominated region, and an intermediate phase relationship being a hallmark of the viscous dominated region. The relevant biomechanical region thus depends almost entirely on the size and cycle time of limb movement. In turn, these relationships between force and motion, and responses to perturbation have major implications for neural control.

Disclosures: G.P. Sutton: None. **N.S. Szczecinski:** None. **R.D. Quinn:** None. **H.J. Chiel:** None.

Poster

P559. Posture and Gait: Kinematics, Muscle Activity, Exercise and Fatigue, and Biomechanics I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P559.08

Topic: E.06. Posture and Gait

Support: Chaire ANR Quantici

Title: Determinants of falls and recoveries following platform translations

Authors: *P.-P. VIDAL[1,2,3], D. WANG[4,2], M. LE GOIC[1], J. DUYSENS[5];
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Abstract: The ability to react to sudden unpredictable multidirectional support-surface translations was investigated in young volunteers while standing upright quietly in a standard position with their eyes open. The perturbation velocities (35 and 90 cm/s) were sufficiently challenging to trigger non-recoverable falls. The surface electromyography activity (EMG) of 14 muscles was recorded and analyzed both for latency and amplitude. Without corrective steps, all slow forward or backward translations ended in a fall. With one or more corrective steps the recoveries increased. With less steps, there were better chances of recovery. Furthermore, recovery steps tended to be faster. The longer, higher or the larger the first step, the better chances of recovery. These corrective movements were achieved by flexing all joints. The compensatory flexion occurred earlier in successful recoveries than falls. However, the peak flexion amplitudes were not significantly different between falls and recoveries at the ankle, knee and hip level. In addition, fall trials kinematics displayed higher variability than recoveries. In fast forward trials with successful recovery, the functional arm movements occurred earlier than in falls. Moreover, the arms abduction was of greater peak amplitude and reached earlier than during a failed recovery. In contrast, in falls, peak elbow flexion was significantly greater, with a similar time to peak angle. As for EMG, the latencies of most responses to fast-forward did not differ much between the 2 types of trials. Nevertheless, in the swing leg the latency of TA was significantly shorter in the fall trials as compared to the recovery trials as in erector spinae and in deltoids. Hence, fast responses do not predict success in recovery. Overall, for other muscles, the EMG responses tended to have higher amplitude in recovery, in particular for SOL and RF in the swing leg and for TA and RF in the stance leg. These higher activities were associated with larger corrective steps. Backward translations were easier to correct for and, in fact, the fast-backward translations accounted for only 11% of all falls, as compared to 31% of all falls for fast-forward translations. Furthermore, when the translation was backward, the duration of the first step was significantly shorter than in the other conditions. Leg muscles activation muscles had latencies which were similar to those seen in forward translation. Following mediolateral perturbation, side-stepping was the most popular strategy while the crossover step was seen to be favoured in fall trials. Leg muscles activation in medio-lateral perturbations showed that recovery trials were generally faster.

Disclosures: P. Vidal: None. **D. Wang:** None. **M. Le Goic:** None. **J. Duysens:** None.

Poster

P559. Posture and Gait: Kinematics, Muscle Activity, Exercise and Fatigue, and Biomechanics I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P559.09

Topic: E.06. Posture and Gait

Support: NSF CAREER 1847891

Title: Generalization of motor memories is governed by the contextual similarity between training and testing environments

Authors: Y. AUCIE, ***G. TORRES-OVIEDO**;
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Abstract: Generalization of movements from experienced to novel situations is a critical aspect of motor adaptation. It has been shown that augmenting the extent of motor adaptation and increasing the similarity between training and testing environments would enhance the generalization of movements from the trained to untrained contexts. However, little is known about how augmenting the extent of adaptation while decreasing the contextual similarity between the training and the testing environment would affect the transfer of movements between environments (i.e., generalization). Here we investigated the effect of augmenting the extent of adaptation on the generalization of learned movements while decreasing the similarities between the training and the testing environments. It has been demonstrated that incline walking would augment the extent of adaptation while decreasing the similarity compared to flat overground walking. To this end, a group of young, unimpaired individuals was adapted on the treadmill either incline or flat. We compared the generalization of movements from these two adaptation settings by quantifying the adaptation effects (i.e., after-effects) overground. We found that the generalization of aftereffect was larger when participants were adapted flat compared to those who adapted incline. Our results suggest that increasing the similarity between the training and the testing contexts is a more important factor than the extent of adaptation in the generalization of recalibrated movements in split-belt walking paradigms. This suggests that training that maximizes the similarity between the training and the testing settings would lead to better outcomes in new situations.

Disclosures: Y. Aucie: None. **G. Torres-Oviedo:** None.

Poster

P559. Posture and Gait: Kinematics, Muscle Activity, Exercise and Fatigue, and Biomechanics I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P559.10

Topic: E.06. Posture and Gait

Title: Distinct perturbation-evoked cortical potentials associated with balance recovery during unexpected slipping while walking: a feasibility study

Authors: *R. M. PUROHIT[1], S. WANG[1], S. SANGANI[3], J. FUNG[4], T. S. BHATT[2];
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[3]Feil/Oberfeld/CRIR Res. Ctr., Jewish Rehabil. Hosp., Laval, QC, Canada; [4]McGill Univ., Montreal, QC, Canada

Abstract: Background: Cortical contributions to reactive balance control have recently been suggested through electroencephalography (EEG) based analysis of beta oscillatory activations and large-amplitude N1 evoked potentials in response to unexpected surface translations in standing. However, cortical dynamics involved in real-life like unexpected environmental perturbations during walking (slips and trips) have never been studied. We have previously identified different muscle synergies between falling or recovering from overground slips while walking. The present study examines cortical evoked potentials in response to large-scale overground slip perturbations in walking to contrast between balance recoveries and falls. **Methods:** Fifteen healthy young adults (8 female, 7 male; ages 18-35) were exposed to a single, novel, unannounced large-size (slip distance=65cm) right-sided overground gait-slip. Analysis of cortical responses from 32 wireless EEG channels acquired at 500Hz filtered with a band pass filter (2-60Hz) were focused bilaterally over supplementary motor (FC3, FC4) and motor areas (C3-C4, Cz) as well as posterior parietal areas (P3, P4, Pz). EEG responses were analyzed corresponding to perturbation onset and recovery responses (0-150 ms window for the negative N1, positive P1 responses; and 150-300 ms for polysynaptic (N2) responses). **Results:** All subjects experienced a backward loss of balance (BLOB) with a slip exposure. Following an unpredicted perturbation, distinct large-amplitude evoked potentials (N1 responses) were identified from baseline to the trough of first negative wave (N1) . N1 cortical responses were consistently followed by large-amplitude positive peak (P1) . N1 responses were largest (200.23 ± 50μV) for subjects who experienced a fall (7 out of 15) as compared to smaller amplitude N1 responses in those who successfully recovered by exhibiting an aborted (28.75 ± 24.17 μV) or a backward (73.75 ± 44 μV)step. The N2 ERP component could clearly be identified and robustly corresponded to the time of recovery stepping in all individuals. This polysynaptic cortical response known to reflect executive cognitive control was found to be larger in amplitude for individuals who executed a backward stepping response as compared to those who aborted their step to recover from balance loss. **Conclusion:** Distinct N1-P1 and N2 cortical responses can be identified following an unpredicted over ground slip exposure in healthy young subjects. Balance-perturbation evoked negative peak cortical responses are modulated and seem to scale according to balance recovery or fall.

Disclosures: R.M. Purohit: Other; University of Illinois at Chicago, Graduate student,PhD program in Rehab Sciences, Physical Therapy. **S. Wang:** Other; University of Illinois at

Chicago, Department of Physical Therapy. **S. Sangani:** Other; Centre for Interdisciplinary Research in Rehabilitation (CRIR) of Greater Montreal, 6363 Hudson Road #061, Lindsay Pavilion of the IURDPM, Montreal (QC) Canada H3S 1M9, Jewish Rehabilitation Hospital (site of CRIR and CISSS Laval), 3205 Place Alton Goldbloom, Laval (QC) Canada H7V 1R2. **J. Fung:** Other; School of Physical and Occupational Therapy, McGill University, 3653 Prom. Sir William Osler, Montreal (QC) Canada H3G 1Y5, Centre for Interdisciplinary Research in Rehabilitation (CRIR) of Greater Montreal, 6363 Hudson Road #061, Lindsay Pavilion of the IURDPM, Montreal (QC) Canada H3S 1M9, Jewish Rehabilitation Hospital (site of CRIR and CISSS Laval), 3205 Place Alton Goldbloom, Laval (QC) Canada H7V 1R2. **T.S. Bhatt:** Other; University of Illinois at Chicago, Associate Professor, Physical Therapy Program Coordinator, MS in Rehab Sciences, Physical Therapy.

Poster

P559. Posture and Gait: Kinematics, Muscle Activity, Exercise and Fatigue, and Biomechanics I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P559.11

Topic: E.06. Posture and Gait

Support: ZOLL Medical Corp

Title: Neuromuscular influences on the divergence of joint angles during repetitive motion

Authors: *B. SENDERLING, N. STERGIOU, A. LIKENS;
Ctr. for Res. in Human Movement Variability, Univ. of Nebraska at Omaha, Omaha, NE

Abstract: Introduction: Human movement contains an inherent level of variability resulting from a complex nervous system. Because of this, repetitive motions never repeat the same path and have some level of mathematical divergence. While neuromuscular activity and divergence during varied movements have been frequently investigated, the influences of one on the other have not been articulated. Therefore, we hypothesized that the divergence in human movement is related to coordination of muscles around a joint. We analyzed data from a previous study to investigate the correlation between muscle coordination and joint angle divergence during cardiopulmonary resuscitation. A reliable correlation would provide insight on a potential source of human movement variability. Furthermore, it could allow joint angle divergence to be quantified with alternative technologies. **Methods:** Sixteen individuals (11 lay-rescuers and 5 professional) performed two minutes of chest compressions on a manikin. The first and last 3 s were not used in the data analysis. Elbow, hip and knee angles were measured bilaterally with an Xsens (Enshede, The Netherlands) inertial motion capture system. Muscle activity was recorded bilaterally through 8 Delsys Trigno (Natick, MA, USA) sensors placed on the agonist-antagonist muscle pairs. State-space reconstruction and Lyapunov Exponents were used to quantify joint

angle divergence. Muscle activity was band-pass filtered between 10 and 500 Hz and enveloped with a 4 Hz low-pass filter. The variation in muscular coordination between agonist-antagonist muscle pairs was quantified as the circular standard deviation of the continuous relative phase between pairs. Bayesian linear regression with Jeffreys priors was used to find the relationship between joint angle divergence and relative phase circular standard deviation.
Results/Discussion: Regression results reported somewhat low R squared values, 0.126 ± 0.085. Relative phase standard deviation was positively related to joint angle divergence for all joint angles except the left elbow. The posterior probabilities averaged 0.797 ± 0.271. These exploratory results provide weak evidence that deviations in coordination between agonist-antagonist muscle pairs tend to be positively correlated with higher joint angle divergence values. However, the results also suggest a larger sample size will be needed in order to draw strong conclusions. If replicable, though, the results provide promising evidence that joint angle divergence could be quantified through muscle activity.

Disclosures: B. Senderling: None. **N. Stergiou:** None. **A. Likens:** None.

Poster

P559. Posture and Gait: Kinematics, Muscle Activity, Exercise and Fatigue, and Biomechanics I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P559.12

Topic: E.06. Posture and Gait

Support: JSPS KAKENHI Grant Number JP 21J13335

Title: Modulation of muscle synergies during locomotor adaptation in humans

Authors: *A. OSHIMA[1,3], Y. NAKAMURA[2], T. WAKAHARA[2,4], K. KAMIBAYASHI[2]; [1]Grad. Sch. of Hlth. and Sports Sci., [2]Fac. of Hlth. and Sports Sci., Doshisha Univ., Kyoto, Japan; [3]Res. Fellow of Japan Society for the Promotion of Sci., Tokyo, Japan; [4]Human Performance Lab., Waseda Univ., Saitama, Japan

Abstract: Modulation of the walking pattern (Locomotor adaptation) has been investigated using a split-belt treadmill that has two belts. Previous studies measuring kinematics data have indicated that humans can adapt to a new walking condition by modifying their walking spatiotemporal parameters and that the central nervous system (CNS) such as the cerebellum plays a critical role in locomotor adaptation. However, it is still unclear how the CNS controls multiple muscle activities in the lower extremities during locomotor adaptation. Regarding control of numerous muscle activities in human locomotion, the muscle synergy concept has been proposed. Therefore, we aimed to investigate neural control of numerous muscle activities underlying locomotor adaptation based on the muscle synergies concept. Healthy young males

were asked to walk on the split-belt treadmill which was operated either tied-belt (two belts moved at the same speed) or split-belt (two belts moved at the different speed) condition. The walking speeds were set to 3.0 (slow) and 5.4 (fast) km/h. The baseline period was tied-belt condition at fast and slow for each 2 min. Subsequently, the adaptation period was split-belt condition with the left belt at slow and right belt at fast for 9 min. We defined the leg on the slow or fast belt during adaptation period as “slow leg” or “fast leg”, respectively. The post-adaptation period was again tied-belt condition at slow for 5 min. Electromyograms (EMGs) were recorded from 26 lower limb muscles bilaterally using surface EMG electrodes. We then extracted the muscle weightings and activation coefficients of the muscle synergies from the EMG data in each period using a non-negative matrix factorization. Approximately four muscle synergies were sufficient to explain the original EMG data in each period. In the muscle synergy which was mainly activated in the initial swing phase in the slow leg, the center of activity (CoA) shifted to the later phase within a gait cycle during adaptation period. The full width of half maximum (FWHM) of the same muscle synergy decreased during adaptation period. The CoA of the muscle synergy which was mainly recruited in the late stance phase in the slow leg shifted to the earlier phase within a gait cycle during adaptation period. On the other hand, the CoA and the FWHM of the muscle synergy which was mainly activated in the late swing phase in the slow leg were unchanged in the adaptation period. These results suggest that the CNS flexibly regulates the activity timing and the duration of neural input in some specific muscle synergies when adapting to the new walking condition.

Disclosures: A. Oshima: None. **Y. Nakamura:** None. **T. Wakahara:** None. **K. Kamibayashi:** None.

Poster

P559. Posture and Gait: Kinematics, Muscle Activity, Exercise and Fatigue, and Biomechanics I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P559.13

Topic: E.06. Posture and Gait

Title: Concurrent gait adaptation to visual and split-belt perturbations

Authors: S. HOWSDEN, N. BARTELS, ***S.-J. KIM**;
California Baptist Univ., Riverside, CA

Abstract: Gait asymmetry is a common symptom in populations with neurological disorders and significantly reduces gait efficiency. To improve gait symmetry, we propose a novel gait rehabilitation paradigm that combines distinct perturbation strategies, specifically, visual feedback distortion (VFD) with split-belt treadmill (SBT) walking. In SBT walking, spatiotemporal gait adaptation can be readily made, but it quickly fades. Gait adaptation to VFD

tends to persist longer, potentially due to more involvement of implicit learning processes. Thus, we aimed to determine whether combined strategies lead to more effective gait symmetric adaptation with a longer retention. Our pilot experiment had 20 subjects participate in two trials (SBT only & VFD+SBT conditions). Each trial consisted of 3-min base, 7-min adaptation, and 15-min post-adaptation periods. During the adaptation period, only the right leg speed was increased from 2 to 3 mph by 0.2 every minute and stayed at 3 mph until the post-adaptation period started (both belt speeds became the same). Under the VFD+SBT condition, the visual bar representing only the right step length was distorted (shortened) by 8% every minute down to a total of 60% distortion. The subjects were unaware of the distortion and looked at the visual bars while adapting to SBT walking. The VFD and SBT perturbations were then removed during the post-adaptation period. Figure below shows the changes in step length symmetry and aftereffects between the two conditions. We found 9 subjects who showed adaptation (Fig. A, group A) and 8 subjects who did not show adaptation (Fig. B, group B) and three outliers (not included). Group A clearly showed an increase in the aftereffects of the new gait pattern. Although more testing will be needed, the results of this pilot study demonstrated a potential advantage of combining VFD into SBT walking to facilitate gait adaptation and improve the retention effect.

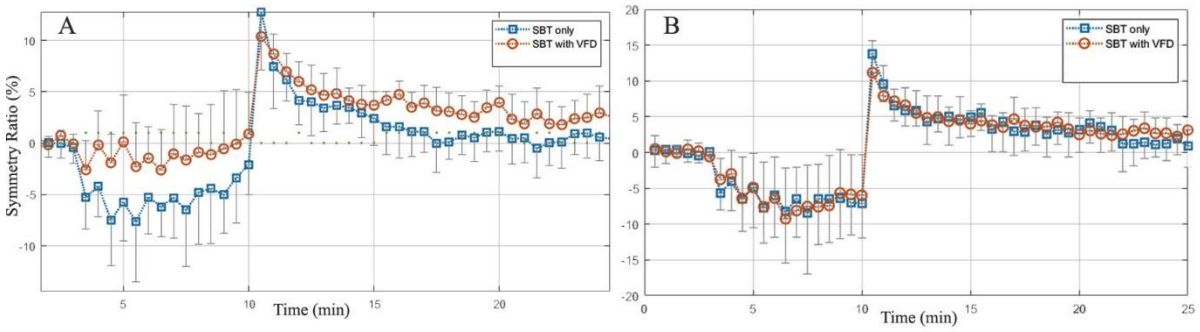


Figure. Changes in step length symmetry during adaptation and post-adaptation periods for SBT only & VFD+SBT conditions. The symmetry ratio (%) was measured by 2*(R-L)/(R+L).

Disclosures: S. Howsden: None. **N. Bartels:** None. **S. Kim:** None.

Poster

P559. Posture and Gait: Kinematics, Muscle Activity, Exercise and Fatigue, and Biomechanics I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P559.14

Topic: E.06. Posture and Gait

Support: NIH Grant R01NS115487

Title: Trunk support improves walking in children with cerebral palsy

Authors: *S. YAN[1,2], S. PARK[1,2], R. REED[1], W. DEE[1], A.-M. ROJAS[1,2], W. Z. RYMER[1,2], M. WU[1,2,3];
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Abstract: Children with cerebral palsy (CP) often show impaired walking ability and excessive trunk movements. Excessive trunk movements are considered not only compensation for lower limb deficits but also poor trunk control. It is unclear whether providing lateral trunk support during walking would improve gait in children with CP. The purpose of this study was to determine the effects of lateral segmental trunk support on walking in children with CP. We hypothesized that providing lateral trunk support during treadmill walking would improve gait performances in children with CP. Twelve children with CP (mean = 11yr (5yr-16yr), 4 boys) participated. Two belts were attached to the children's trunk. One belt was below the armpits for supporting the upper trunk (T7-T10), the other was above the pelvis for supporting the lower trunk (L1-L4). A D-shape ring was sewed on both sides of the belts, allowing a rope to hook and provide pulling forces to the left and right sides. On each side, the D-shape ring and the rope were connected by an extension spring (stiffness = 1.31N/mm) so that various pulling forces were applied by extending the spring. After walking on a treadmill for 1 minute at a comfortable speed (baseline), children then walked at the same speed under three randomized conditions: the springs were attached to the belt(s) positioned at the upper trunk (UTS), the lower trunk (LTS), and a combination of the upper and lower levels of the trunk (CTS). The extended length of the springs was decided based on children's tolerance before the three conditions and remained the same on both sides and all conditions. Each condition lasted for 1 minute and a 1-min break was given between conditions. We recorded kinematic signals using Xsens wearable sensors, propulsive forces with a Bertec treadmill, and muscle activities using Trigno wireless EMG system. Variables included step length, step height, sagittal hip range of motion (ROM), and the integrated muscle activity during the single-limb stance phase. For the weak side, children had greater step length and step height in CTS than those in baseline, and greater step length in UTS and CTS than that in LTS ($p < 0.05$). Changes of soleus activity of the single-limb stance phase across conditions mirrored changes of step length across conditions ($p < 0.05$). Greater sagittal hip ROM on the strong side occurred in CTS compared to baseline ($p < 0.05$). We concluded that lateral trunk support during treadmill walking can improve gait performances in children with CP. Results suggest that targeted trunk control may be applied in future training to improve gait performances in children with CP.

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Poster

P559. Posture and Gait: Kinematics, Muscle Activity, Exercise and Fatigue, and Biomechanics I

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Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P559.15

Topic: E.06. Posture and Gait

Support: Ontario Neuro-trauma Foundation 2016-RHI-PREV-1019
Discovery Grant from the Natrual Sciences and Engineering Research Council of Canada DGRGPIN-2017-06790
Ontario Graduate Scholarship

Title: Reactive Stepping Balance Following Balance Training for Individuals with Incomplete Spinal Cord Injury

Authors: *J. W. LEE[1,4], J. YANG[1], S. MAUCERI[1], K. CHAN[4], J. UNGER[2,4], K. E. MUSSELMAN[2,3,4], K. MASANI[1,4];
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Abstract: Introduction Individuals with incomplete spinal cord injury (iSCI) often experience impairment of lower-limb sensorimotor functions, resulting in compromised reactive balance and increased fall risk. We previously reported that during a reactive stepping task, those with iSCI more frequently required multiple steps to recover from a forward fall, compared to able-bodied (AB) individuals. We also reported that inadequate foot placement for those with iSCI may contribute to the need for multiple steps to recover balance. This inadequate foot placement may improve after balance training, resulting in improved reactive balance and reduced fall risk. Here we investigated the effect of balance training on foot placement during a reactive stepping task, elicited by a simulated forward fall. We hypothesized that following balance training, foot placement of those with iSCI would become similar to that of AB individuals. Method Twenty-one individuals with iSCI participated in eight weeks of balance training. Data from ten participants were analyzed. Participants performed up to 10 trials of the Lean-and-Release (L&R) test. Participants adopted a forward lean while 8-12% of their body weight was supported by a tether, which was unexpectedly released. The L&R test was performed before and after balance training as well as 3-months after completing balance training. Kinematic and kinetic data were recorded using a motion capture system and force platform. We compared the average margin-of-stability (MOS) and intra-subject standard deviation (SD) of MOS at foot contact (FC) and 0.05s after FC. In addition, correlation of MOS at FC with scores on the Mini Balance Evaluation Systems Test (mini-BESTest) and Community Balance and Mobility (CB&M) scale were calculated. Results and Discussion The average MOS were not significantly different across three time points (FC: p=0.175; FC+0.05s: p=0.200). There was a significant effect of balance training on SD of MOS at FC (p=0.042) while not at FC+0.05s (p=0.087). In addition, average MOS was significantly correlated with both mini-BESTest scores (pre: p=0.019; post: p=0.008; follow-up: p=0.014) and with CB&M scores at post and 3-months after balance training (pre: p=0.072; post: p=0.030; follow-up: p=0.028). MOS was used to analyze the

dynamics of how foot placement changed after balance training for those with iSCI. Our findings suggest that balance training could improve foot placement during a reactive stepping task. This study contributes to a better understanding of dynamic postural control for individuals with iSCI, which may lead to the development of effective therapies and assessments for this population.

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Poster

P566. Motor Systems: Neuromodulation of Neuronal and Synaptic Properties

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P566.01

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NINDS NS066587
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Title: Multiple modes of generating ingestive motor programs in the feeding network of Aplysia

Authors: *C. G. EVANS[1], M. A. BARRY[1], M. H. PERKINS[1], J. JING[2], K. R. WEISS[1], E. C. CROPPER[1];
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Abstract: In many CPGs activity is reconfigured to generate different outputs. We study this process in the feeding network of Aplysia. In this system an ingestive configuration can be achieved if cycles of activity are repeatedly triggered by a command-like neuron, CBI-2. In this situation, repetition priming is observed. Cycles of activity gradually switch from an intermediate to an ingestive configuration, and a persistent state is created. Priming is largely mediated by slow modulatory effects of neuromodulators released from CBI-2.
We recently showed that there are preparations in which priming is absent (Evans 2021). Ingestive activity can be generated in these preparations by co-activation of a higher order neuron CBI-3, with CBI-2. With CBI-3 co-activation, reconfiguration occurs immediately, and rapid switches between ingestive and intermediate activity are possible. We have hypothesized that there are multiple modes of generating ingestive activity in the feeding network that serve different purposes.
To probe this idea, we sought to determine whether the absence of priming is due to deficits in neuromodulation. When priming occurs, it results from progressive increases in the firing frequency of the B8s. In part, changes in B8 activity result from neuromodulator induced excitability increases that occur during the first few cycles of activity (Dacks, 2012). However,

changes in B8 firing frequency occur subsequently, as a result of disinhibition, i.e., there are progressive decreases in the firing frequency of the inhibitory B4/5 neurons presynaptic to B8. Disinhibition is independent of neuromodulation. It results from activity dependent decreases in B4/5 intrinsic excitability.
We characterized B4/5 activity in preparations that would not prime, and found that firing frequency did not decrease despite the presence of the activity dependent mechanism that decreases intrinsic excitability. This suggests that when there is no priming, B4/5 receive increased excitatory synaptic input. Consistent with this idea we found that the phasic (presumably synaptic) depolarization in B4/5 increased after failed attempts to induce priming, but did not increase when priming was successful. Our data suggest that the absence of priming is not simply due to the lack of a neuromodulator induced increase in B8 excitability. Instead, it is due to a mechanism that actively maintains the default state inhibition of B8, i.e., maintains B4/5 activity. These data support the idea that the absence of priming does not reflect a pathology, but suggest in some conditions, ingestive responses may require CBI-3 activity.

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Poster

P566. Motor Systems: Neuromodulation of Neuronal and Synaptic Properties

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Program #/Poster #: P566.02

Topic: E.07. Rhythmic Motor Pattern Generation

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Title: Modulator capacity corresponds with dietary diversity in three species of decapod crustaceans

Authors: *D. J. POWELL, A. I. MILLER, J. S. KAZMI, G. BUKOWSKI-THALL, P. S. DICKINSON;
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Abstract: The central nervous systems of decapod crustaceans contains numerous amines, amino acids, and peptides that modulate the central pattern generating (CPG) circuits found within its stomatogastric nervous system (STNS) and cardiac ganglion (CG); over 30 modulators have been identified in one species of crab. CPGs in the STNS are responsible for internally chewing and filtering food into the midgut, while the CG drives the heartbeat. Neuromodulation allows these CPGs to flexibly produce a variety of output patterns; however, the variety of

modulators exceeds the variety of induced output, leading us to ask why so many modulators are present. Although modulatory capacity, a measure of the extent to which a neuronal circuit can be modulated, may be solely an inherited trait, we hypothesize a functional correlation, with decapods that need more behavioral flexibility utilizing a broader range of modulators than those with more stereotyped behaviors. We thus predicted that decapods with broad diets would have a larger modulatory capacity in the STNS, but not the CG, than those with limited diets. To test this, we compared responses of the STNS and CG CPGs in three species of majoid crabs (*Pugettia producta*, *Libinia emarginata*, and *Chionoecetes opilio*) to the same neuromodulators. *L. emarginata* and *C. opilio* both have broad diets, while *P. producta* primarily eats kelp. With one exception, each species' CG exhibited similar responses to all modulators. The STNS of crabs with broader diets responded similarly to all modulators, whereas the *P. producta* STNS showed limited responses to the same set of neuromodulators. CG responses did not follow this trend, as all crabs showed varied CG response to modulation. Modulators: *Cancer borealis* Tachykinin Related Peptide (CabTRP), Crustacean Cardioactive Peptide (CCAP), Red Pigment Concentrating Hormone (RPCH), Myosuppressin (Myo), Oxotremorine (Oxo), Dopamine (DA), Proctolin (Proc).

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Poster

P566. Motor Systems: Neuromodulation of Neuronal and Synaptic Properties

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P566.03

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NSF IOS 1755098

Title: Habitat temperatures determine the functional temperature range of central pattern generators in the crustacean stomatogastric nervous system

Authors: *L. VINK-LAINAS, W. STEIN;
Illinois State Univ., Normal, IL

Abstract: Rapid temperature fluctuations are a major challenge to neuronal function. Among other effects, temperature changes cause an imbalance of ionic conductances that are key to neuronal and synaptic activity, leading to failure of neuronal function. This is a particular threat for ectothermic species that experience rapid environmental temperature fluctuations due to natural and climate change-induced extreme weather conditions. Thus, possessing physiological mechanisms to mitigate the effects of temperature changes is essential for survival. However, the limits within which neurons can respond adequately to temperature changes and the evolutionary

pressures to temperature robustness are not well understood.
We are investigating temperature responses in the pyloric and gastric mill central pattern generators of the crustacean stomatogastric nervous system. These pattern generators control aspects of feeding, serve a vital function in survival, and have been characterized in several species. The main question driving our research is whether species' habitat temperatures affect neuronal temperature responses and robustness against failure. Using a comparative approach, we record the pyloric and gastric mill rhythms across temperatures in several related crab species: *Cancer borealis*, *Cancer magister*, and *Carcinus maenas*. We identify the range of temperatures at which the rhythms remain functional in each species and compare to their distinct habitat temperatures.
The species that experiences a greater range of temperatures in its environment had a pyloric rhythm that was functional at a greater range of temperatures. Specifically, *C. maenas*, which experiences habitat temperatures ranging 0-35°C, had a rhythm that was functional over an average range of 6-33.4 ± 1.3°C (n=10). *C. borealis*, which experiences habitat temperatures ranging 3-25°C, had a functional rhythm over an average range of 6-26.9 ± 1.4°C (n=9). The pyloric rhythm of *C. magister*, which lives in similar habitat temperatures as *C. borealis*, remained functional between 6 and 24.9 ± 3.7°C (n=9). Thus, our results show that the maximum temperatures at which the pyloric rhythm remained functional corresponded to environmental temperature ranges experienced, suggesting that habitat temperature determines the range of temperatures at which the nervous system can function. We are currently testing whether this holds true for the gastric mill rhythm as well.

Disclosures: L. Vink-Lainas: None. **W. Stein:** None.

Poster

P566. Motor Systems: Neuromodulation of Neuronal and Synaptic Properties

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Program #/Poster #: P566.04

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NIH MH-060605
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Title: Excitatory neuromodulation reduces inter-animal variability of neuronal activity

Authors: *O. ITANI, A. C. SCHNEIDER, D. BUCHER, H. G. ROTSTEIN, F. NADIM; Federated Dept Biol. Sci., New Jersey Inst. of Technol., Newark, NJ

Abstract: Neural circuit output can be remarkably consistent across animals despite significant variability in the circuit building blocks. Recent experiments suggest that neuromodulators may

play a significant role in promoting such consistent circuit output. However, the mechanisms through which neuromodulators may do so remain unknown. We focused on consistency of individual neuronal activity, and used mathematical modeling to demonstrate how neuromodulation of cellular excitability may promote consistent cellular activity from a population of cells with variable underlying conductances.
We used a population of leaky integrate-and-fire (LIF) models, with their leak conductance chosen from a uniform distribution, and considered the frequency-current (fI) relationship as a measure of cellular activity. The coefficient of variation (CV_{fI}) of firing frequency measured across a range of input currents was used as the measure of variability across the population, so that a reduction in CV_{fI} was interpreted as a more consistent inter-population variability. Excitatory neuromodulation was modeled by adding an excitatory ionic current of a fixed conductance.
We could readily demonstrate that adding a fixed level of excitatory neuromodulation always reduces CV_{fI} across the population of LIF neurons. However, it is known that neuromodulatory excitation itself can be variable. To address how the LIF population responds to variable levels of excitatory neuromodulation, we examined how CV_{fI} changed with the mean and standard deviation (SD_m) of the modulatory conductance. We found that excitatory neuromodulation reduced CV_{fI} so long as SD_m remained below a threshold value.
To compare our findings with the effect of an modulatory neuropeptide acting on the crab stomatogastric lateral pyloric neuron, we extended our LIF model to include a refractory conductance within the same framework, and investigated the influence of the refractory parameter variability on CV_{fI}. This approach proved to be amenable to interpreting biological data and variability and allowed for the determination of ranges for which SD_m reduced CV_f, which could then be compared to the experimentally measured variability of the modulatory conductance.

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Poster

P566. Motor Systems: Neuromodulation of Neuronal and Synaptic Properties

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P566.05

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NSF IOS: 1755283

Title: Neuropeptide-elicited neuronal switching involves modulation of multiple intrinsic properties

Authors: *R. R. SNYDER, D. M. BLITZ;
Biol., Miami Univ., Oxford, OH

Abstract: Neuronal switching contributes to internetwork coordination and flexibility of oscillatory networks to drive context-appropriate rhythmic behaviors (Dickinson, 1995, *Curr Opin Neurobiol*). In the established switching mechanism, synaptic strength modulation recruits a neuron into another network. Alternatively, in the crab *Cancer borealis* the LPG neuron switches from only fast rhythm activity (1 Hz; food filtering) to dual fast/slow (0.1 Hz; food chewing) bursting when membrane properties are altered by a modulatory neuron or its bath-applied neuropeptide Gly^1-SIFamide (SIF) (Fahoum & Blitz, 2021, *BioRxiv*). Thus far, membrane property(ies) and ion channel(s) that enable intrinsic slow bursting, which must periodically escape from fast rhythm electrical synaptic input, are unknown. We isolated LPG from both networks and first examined intrinsic properties with depolarizing current injections. We found bath-applied SIF (5×10^{-6} M) enhanced the firing frequency/current relationship (0.5 - 2.5 nA, n=7; $p<0.01$; two-way ANOVA) and decreased Spike Frequency Adaptation (SFA; Control rate of change: 0.014 ± 0.003; SIF: 0.006 ± 0.01 sec^{-1}, n=9; p=0.004, signed-rank). This increased intrinsic excitability and decreased SFA may prolong burst duration. Regarding mechanisms of burst initiation, we found that SIF enhanced post inhibitory rebound, evident as a larger depolarization above baseline post-hyperpolarization (Control integrated area: 4.1 ± 1.0; SIF: 20.5 ± 3.0 mV*ms, n=9; $p<0.001$, paired t-test). SIF increased a corresponding sag potential (Control: 1.93 ± 0.42; SIF: 5.12 ± 0.71 mV, n=10; $p<0.001$, paired t-test) indicating a hyperpolarization-activated (I_h) current. SIF + CsCl (5µM; I_h blocker) had a variable effect on LPG burst duration (n=2/5 longer, 3/5 shorter than SIF). However, SIF + CsCl elicited a longer LPG interburst interval in 5/5 experiments thus far (p=0.063, power=0.33, paired t-test), suggesting I_h contributes to burst initiation. We also used riluzole (30 µM) to block the persistent sodium current (I_{NaP}) which can support burst maintenance (Paton et al, 2006, *Nat Neurosci*). SIF + riluzole elicited a shorter burst duration than SIF only (SIF: 6.0 ± 0.4; SIF+ril: 1.5 ± 0.3 s, n=8; $p<0.001$, paired t-test) but did not alter interburst interval (n=8; p=0.063, paired t-test), implicating I_{NaP} in burst maintenance but not initiation. Thus, in a switching neuron recruited by intrinsic property modulation, we identified multiple intrinsic properties that are modified. Future goals are to determine if these modulatory actions are sufficient for intrinsic bursting capable of escaping faster, electrically-coupled rhythmic input.

Disclosures: R.R. Snyder: None. **D.M. Blitz:** None.

Poster

P566. Motor Systems: Neuromodulation of Neuronal and Synaptic Properties

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Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P566.06

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NSF IOS: 1755283

Title: Multiple neuron types mediate internetwork coordination via distinct synaptic mechanisms

Authors: *S.-R. H. FAHOUM, D. M. BLITZ;
Biol., Miami Univ., Oxford, OH

Abstract: Internetwork coordination between distinct rhythmic networks is important for overall function in many systems (Steriade et al., J Neurosci 1993; Larson et al., J Neurophys 1994; Tryba et al., J Neurophys 2008; Rangel et al., Elife 2016; Blitz et al., J Neurophys 2019), but cellular mechanisms are poorly understood. Some identified mechanisms include rhythmic pre-synaptic inhibition of a modulatory input and a pivotal internetwork synapse (Bartos et al., J Neurosci 1997; Bartos and Nusbaum, J Neurosci 1999). Here, we examine intra-/internetwork interactions in the crab *Cancer borealis* stomatogastric nervous system, which contains two small, well-defined networks (pyloric [fast, ~1 s cycle period (CP)]; gastric mill [slow, ~10 s CP]) and identified modulatory inputs. In an established biological model, bath-applied neuropeptide Gly^1-SIFamide (SIF: 5 µM) mimics modulatory neuron activity by recruiting the fast-only LPG neuron into dual (fast/slow) activity that is coordinated with both networks (Fahoum and Blitz, BioRxiv 2021). We use this model to examine whether dual activity of the switching neuron LPG is required for rhythm generation (RG), intra- and/or internetwork coordination. First, we removed the two copies of LPG using photoinactivation (SIF-kill). Both rhythms occurred in the SIF-kill condition (n = 4/4), suggesting that LPG is not necessary for RG in either network. In SIF-intact, LPG slow activity is coordinated with slow network neurons in a three-phase pattern: LG/IC, LPG, DG (n = 4/4). These timing relationships are not altered in SIF-kill (SIF-intact vs SIF-kill: $p > 0.05$, n = 4, paired t-test). Thus, LPG is not necessary to coordinate slow network neurons. However, LPG may play a role in fast/slow coordination, as LPG is electrically coupled to the fast network pacemaker neurons (Shruti et al., J Neurophys 2014), and the slow network regulates fast network CP in SIF-intact (Blitz et al., J Neurophys 2019). We used fast network CP variability (coefficient of variation, CV) to measure slow network regulation of the fast network. SIF-intact increased fast network CV compared to saline ($p = 0.007$, n = 4). SIF-kill decreased CV ($p = 0.01$), but not to saline level ($p = 0.01$), suggesting LPG contributes partially to internetwork coordination. In SIF-intact, hyperpolarizing slow network neurons LG ($p = 0.004$, n = 8) or IC ($p = 0.013$, n = 10), but not DG ($p = 0.212$, n = 8), decreased CV, indicating that LG and IC also contribute to fast network regulation. These data suggest that multiple neurons, active in distinct phases of a slow rhythm and using electrical vs chemical inhibitory synapses, collectively contribute to regulating a related faster rhythmic network.

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Poster

P566. Motor Systems: Neuromodulation of Neuronal and Synaptic Properties

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Topic: E.07. Rhythmic Motor Pattern Generation

Support: NIH grant R35NS 097343

Title: Activity and energy: The effect of K-ATP channel activity on network output of the pyloric circuit in the C.borealis stomatogastric ganglion

Authors: *S. KEDIA, E. MARDER;
Brandeis Univ., Waltham, MA

Abstract: Prolonged changes in neuronal activity are triggers for homeostatic processes involved in maintaining activity levels around arbitrary 'set points'. An essential component of the homeostatic machinery is a means by which neurons can sense their own activity levels. ATP, the cell's energy currency, is intimately involved in maintaining a neuron's internal ionic environment and membrane hyperpolarization, especially in times of heightened activity. Fluctuations in levels of available ATP might serve as a signal for changing neuronal activity to balance its demand and supply. ATP-sensitive K channels have been implicated in regulating neuronal activity in response to changing glucose levels and after events such as hypoxia to balance activity levels with available energy resources. The channel is activated by increasing levels of ADP, the product of ATP hydrolysis indicating decreasing ATP levels, upon which it participates in hyperpolarizing the neuron to reduce its firing. We explore the role of K-ATP channels in regulating activity of an oscillatory system, the pyloric circuit in the *Cancer borealis* stomatogastric ganglion (STG). The network produces a rhythmic output continuously *in vivo* and continues to do so *in vitro* and has been studied for its remarkable homeostatic and adaptive properties to changing neuromodulatory and environmental conditions, however the molecular underpinnings of these adaptations remain uncovered. We wanted to examine pathways of neuronal activity modulation as a consequence of energy use in this system. We studied the effects of blocking and activating K-ATP channels pharmacologically using known sulfonylureas, tolbutamide and glibenclamide to block channel activity, and diazoxide to activate the channel. We found that pharmacological activation of K-ATP channels can result in a complete cessation of rhythmic activity in the intact circuit seen as a loss of both firing and slow wave oscillations of membrane potential of pacemaker neurons. Channels blockers, on the other hand, caused minor increases in oscillation frequency in the intact network. Removal of neuromodulatory inputs to STG neurons can result in a dramatic reduction of network activity, even a loss of network oscillations. Under these conditions we found that blocking K-ATP channels could restore oscillatory activity in the network, and in some cases, activate gastric mill neurons which are largely stimulated in the animal in the presence of food and in vitro by certain neuromodulators. Hence, we report the presence of a potent pathway in STG neurons for the modulation of their activity on the basis of their intracellular ATP levels.

Disclosures: S. Kedia: None. **E. Marder:** None.

Poster

P566. Motor Systems: Neuromodulation of Neuronal and Synaptic Properties

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Topic: E.07. Rhythmic Motor Pattern Generation

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Arnold and Mabel Beckman Foundation
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Title: Structural variation among neuropeptide isoforms affects function in the lobster cardiac system

Authors: D. POWELL[1], A. J. MUSCATO[1], W. BULHAN[1], E. MACKENZIE[1], A. PUPO[1], M. ROLPH[1], A. E. CHRISTIE[2], ***P. S. DICKINSON**[1];
[1]Bowdoin Coll, Brunswick, ME; [2]Univ. of Hawaii at Manoa, Honolulu, HI

Abstract: Neuronal responses to peptide signaling are determined by the specific binding of a peptide to its receptor(s). For example, isoforms of the same peptide family can drive distinct responses in the same circuit by having different affinities for the same receptor, by having isoforms bind to different receptors, or by a combination of these scenarios. Small changes in peptide composition can alter the binding kinetics and overall physiological response. The heart of the American lobster, *Homarus americanus*, is neurogenic, and is controlled by the nine neurons that make up the cardiac ganglion. In the lobster, native isoforms of C-type allatostatins (AST-Cs) usually decrease heartbeat frequency and alter contraction force. However, one of the three AST-C isoforms, AST-C II, drives a cardiac response that is distinct from that elicited by the other two. This isoform differs from AST-C I and III not only in specific amino acids, but also because AST-C II is amidated, whereas AST-C I and III are not amidated. To investigate these differential responses, we altered various features of each peptide sequence. Although the presence of an amide group at the end of a peptide sequence (amidation) is often essential for determining physiological function, we demonstrate that C-terminal amidation does not dictate the AST-C response in the lobster cardiac system. However, a single amino acid substitution within the consensus sequence did account for at least a portion of the differences in specific response characteristics (e.g. contraction frequency or force).

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Poster

P566. Motor Systems: Neuromodulation of Neuronal and Synaptic Properties

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Topic: E.07. Rhythmic Motor Pattern Generation

Support: NIH award P20GM0103423 from NIGMS
NSF grant IOS-1121973
NSF grant IOS-1856433
Henry L. and Grace Doherty Charitable Foundational Coastal Studies Research Fellowship

Title: Is stretch feedback in the heart of Homarus americanus modulated by neuropeptides?

Authors: *M. THIES, E. S. DICKINSON, X. QU, D. POWELL, P. S. DICKINSON;
Bowdoin Col., Brunswick, ME

Abstract: The heart of the American lobster, Homarus americanus, is controlled by the cardiac ganglion (CG), which is responsible for generating patterned bursts of action potentials, each of which results in a contraction of the heart. The cardiac ganglion includes four premotor and five motor neurons, as well as stretch-sensitive dendrites, which extend from the CG neurons (Cooke, 2002). All of the nine neurons are electrically and chemically coupled so that the bursting of the neurons is coordinated. Two intrinsic feedback pathways modulate the bursting pattern of the CG. A negative feedback pathway using nitric oxide decreases contraction frequency, while feedback from the stretch-sensitive dendrites generally increases contraction frequency. In addition, extrinsic modulation via neuromodulators plays a key role in achieving flexibility. However, the extent to which the heart's feedback system is modulated is still unknown. Here, we asked whether neuromodulators alter the positive stretch feedback system itself. Two FMRF-amide-like peptides, SGRNFLRFamide (SGRN) and GYSDRNYLRFamide (GYS), have previously been shown to exert concentration and force dependent modulatory effects on the CG and the periphery. The previous data suggested that these peptides may alter feedback as well, with SGRN favoring positive feedback and GYS favoring negative feedback. Here we used a preparation that included the CG and the muscle surrounding the premotor neurons to record the responses to stretch of those muscles. In response to stretch, cycle frequency most often increased while burst duration decreased. However, the responses varied considerably across preparations, with some stretches inducing an excitatory response, and others inducing an inhibitory response. Application of GYS appeared to decrease the changes in the burst duration and interburst interval elicited by stretch, resulting in a significant decrease in variation in the presence of the peptide compared to the control. GYS thus may reduce positive feedback, favoring negative feedback, as was previously predicted. SGRN did not enhance feedback, as we had predicted, but instead also generally decreased the variation of change in the burst duration and interburst interval elicited by stretch. These data suggest that any change in the balance of

positive and negative feedback elicited by the peptide is more likely mediated by the negative feedback pathway.

Disclosures: M. Thies: None. **E.S. Dickinson:** None. **X. Qu:** None. **D. Powell:** None. **P.S. Dickinson:** None.

Poster

P624. Respiratory Control I

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Program #/Poster #: P624.01

Topic: E.08. Respiratory Regulation

Support: AHA (PI Arble 20IPA35320195)

Title: Setmelanotide improves the hypercapnic ventilatory response in diet-induced obese mice

Authors: *S. N. FRAMNES-DEBOER, A. R. SPEARS, R. NOUR, D. M. ARBLE; Marquette Univ. Dept. of Biol. Sci., Milwaukee, WI

Abstract: Setmelanotide, a melanocortin-4 receptor agonist, was recently FDA approved for weight loss in individuals with pro-opiomelanocortin, proprotein subtilisin/kexin type 1, or leptin receptor deficiency. While ongoing efforts continue to investigate the role of setmelanotide for weight loss, the effect of setmelanotide on obesity-related disease states remain largely unknown. In particular, obstructive sleep apnea occurs in nearly 70% of the obese population and is the most common form of sleep disordered breathing. Here we test the hypothesis that setmelanotide will improve the hypercapnic ventilatory response of obese mice. Setmelanotide or vehicle was provided via an intraperitoneal injection (0.2 mg/kg or 1.0 mg/kg) for 9 days in diet-induced obese C57Bl/6J mice. The hypercapnic ventilatory response was measured by whole-body plethysmography before and after treatment. We found that while both doses of setmelanotide decreased body weight, the higher dose resulted in significantly greater weight loss. Additionally, the higher dose of setmelanotide (1.0 mg/kg) significantly improved the hypercapnic ventilatory response. These data suggest that melanocortin receptor agonists may have additional physiological effects beyond weight loss alone.

Disclosures: S.N. Framnes-DeBoer: None. **A.R. Spears:** None. **R. Nour:** None. **D.M. Arble:** None.

Poster

P624. Respiratory Control I

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Program #/Poster #: P624.02

Topic: E.08. Respiratory Regulation

Support: NIH R01 NS101105 to W.J.A.

Title: Mice expressing human ApoE4 genotype exhibit greater baseline respiratory rate and less robust hypoxic ventilatory response than ApoE3 mice as measured by whole body plethysmography

Authors: *C. E. TAYLOR[1], J. S. NEWTON[2], C. M. CALULOT[1], A. L. SILVERSTEIN[3], L. A. JOHNSON[1], L. E. STRATTAN[4], W. J. ALILAIN[5];
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Abstract: Breathing is vital for life and is both uniquely complex and vulnerable to disruption. Disorders of breathing such as those that occur in sleep apnea and even high cervical spinal cord injury (SCI) can impair quality of life and potentiate the ill effects of comorbidities like Alzheimer's disease (AD). Indeed, studies have shown greater cognitive and cardiovascular impairments in subjects with sleep apnea and AD. Apolipoprotein E (ApoE) is a ubiquitous protein found throughout the body and CNS which aids in the regulation and transport of cholesterol and lipids. Moreover, the *E4* allele confers a higher risk of early onset AD than either the *E2* or *E3* alleles. Previous research in our lab has shown the interaction between ApoE genotype and sex differentially affects breathing motor plasticity following SCI in mice. However, it is unknown how baseline respiratory pattern and capability during hypoxic challenge may differ between uninjured individuals expressing *APOE4* and *E3* alleles. Thus, with *APOE* genotype as our independent variable, we measured the respiratory parameters of male *E3* and *E4* targeted replacement mice using whole body plethysmography (WBP), hypothesizing that *E3* animals would display a more adaptive, heightened response to hypoxic exposure than those expressing *E4*. Preliminary data comparing *E3* animals (n=9) and *E4* animals (n=8) demonstrated differences across several key measurements. At baseline, *E4* animals were shown to breathe in an energy inefficient manner as exemplified high measures in tidal volume and frequency. During sustained hypoxia, frequency, tidal volume, and minute volume initially increased across all animals, but *APOE3* animals mounted a much more robust and sustained response with *APOE4* values falling well below baseline measures prior to termination of hypoxia. These preliminary results suggest a critical role of the *APOE* genotype in regulation of baseline breathing and in response to hypoxic conditions and that possessing the *APOE4* allele may confer vulnerabilities in respiratory function and during environmental challenges leading to cognitive decline. Future directions include examining anatomically critical CNS respiratory regions and neural cell function.

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Poster

P624. Respiratory Control I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P624.03

Topic: E.08. Respiratory Regulation

Support: NIH F31-HL156470
NIH R01-HL137094

Title: KCNQ2 channels preferentially regulate activity of chemosensitive neurons in the retrotrapazoid nucleus

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Abstract: The KCNQ1-5 (Kv7.1-7.5)family of potassium channels regulates cellular activity at multiple levels ofthe respiratory circuit including neurons in the retrotrapezoid nucleus (RTN)that control breathing in response to CO_2/H^+ (i.e.,function as respiratory chemoreceptors). In particular,clinical evidence suggests both loss- (LOF) and gain-of-function (GOF)mutations in KCNQ2 disrupt respiratory function. Therefore, the goalof this study is to understand how conditional ablation (*Phox2b*$^{cre/+}$*::Kcnq2*$^{f/f}$; *Kcnq2* cKO) or expression of a human KCNQ2 GOF variant (R201C) inchemosenstive RTN neurons (*Phox2b*$^{cre/+}$*::Kcnq2*$^{R201C/+}$)effects baseline breathing and the ventilator response to CO_2. Forthese experiments RTN chemoreceptors were targeted based on Phox2b expressionand breathing was measured in 30 day old mice (mixed sex) by whole bodyplethysmography. We found that *Kcnq2*cKO hyperventilated under room air conditions (control: 189 ± 6 bpm vs *Kcnq2* cKO: 236 ± 6.37 bpm) and minuteventilation (MV) increased by 0.74 ml/min/g but with no change in theventilator response to CO_2. Conversely, *Kcnq2*$^{R201C/+}$ mice showednormal respiratory activity under room air conditions but failed to increasebreathing in response to CO2 (MV at 7% CO2 control: 10.7 ± 0.5 ml/min/g vs. *Kcnq2*$^{R201C/+}$: 7.8 ± 0.9ml/min/g). Consistent with these functional results,fluorescent *in-situ* hybridization showed that Phox2b+ RTNneurons express KCNQ2 but not KCNQ3 transcript. These resultssuggest KCNQ2 channels preferentially regulate activity of RTNchemoreceptors.

Disclosures: J. Soto: None. **D.K. Mulkey:** None. **A. Tzingounis:** None.

Poster

P624. Respiratory Control I

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Program #/Poster #: P624.04

Topic: E.08. Respiratory Regulation

Support: PA DOH Grant SAP#410079760

Title: Cerebral cortical control of the posterior cricoarytenoid muscle in nonhuman primates

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Abstract: Introduction: The posterior cricoarytenoid (PCA) muscle is the sole abductor of the vocal folds. It plays a critical role in valving the airway during gross and fine control of breathing and phonation. The neural substrates underlying cortical control of the PCA have not yet been defined in nonhuman primates. We used the rabies virus as a retrograde transsynaptic tracer to define regions of the cerebral cortex that are involved in the descending control of the PCA muscle.
Methods: We injected rabies virus (N2c strain, 700-1000 µL, 1 x10^10 pfu) into the left PCA muscle in juvenile (n=2) and adult (n=2) female cynomolgous monkeys. We set the survival time (106-121 hours) to allow virus to infect putatively third- and fourth-order neurons in the cerebral cortex. Rabies virus was transported transneuronally in the retrograde direction in a time-dependent manner, revealing chains of synaptically-connected neurons. Then, we processed serial sections through the brain and spinal cord to reveal the location of neurons infected with rabies virus. We plotted labeled neurons in every fourth cortical section under bright-field microscopy into Neurolucida, then translated the sections into two-dimensional reconstructions to create maps of the neural substrates underlying motor control of the PCA.
Results: We found prominent densities of neurons labeled by retrograde transport of rabies virus from the PCA in multiple cerebral cortical regions. At the earliest stage of virus transport, labeling was largely restricted to the lateral aspects of the primary motor cortex (M1). At later stages of transport, labeling expanded to include premotor areas on the lateral surface, dorsal and ventral premotor cortex, and on the medial wall, the supplementary motor area (PreSMA) and the cingulate motor areas. At the same time, we also observed neurons labeled in the anterior cingulate cortex, areas of the medial and ventrolateral prefrontal cortex, amygdala, agranular and granular insula, and auditory association regions.
Conclusions: Motor control of the PCA is coordinated with that of muscles of respiration, articulation, and phonation. The apparent co-localization of the neuronal substrate for PCA motor control with that of other intrinsic laryngeal muscles and the thoracic diaphragm substantiate a functional coordination. This detailed first description of the neural substrates underlying motor control of the PCA has theoretical and clinical implications for respiratory and vocal motor control.

Disclosures: **L.B. Helou:** A. Employment/Salary (full or part-time):; University of Pittsburgh. **J.M. Michlich:** None. **A.C. Bostan:** A. Employment/Salary (full or part-time):; University of Pittsburgh.

Poster

P624. Respiratory Control I

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Program #/Poster #: P624.05

Topic: E.08. Respiratory Regulation

Support: ARC Grant DP180101890

Title: Inflammatory responses in central respiratory regions of offspring following maternal cigarette smoke exposure in mice.

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Abstract: Accruing evidence suggests that prenatal cigarette smoke or nicotine exposure generates reactive oxygen species or promotes apoptosis in the brainstem areas that participate in respiratory homeostasis. Maternal smoking contributes to CNS developmental delays and blunted central chemoreflex in cases of sudden infant death syndrome (SIDS). However, the relationship between pre- or postnatal cigarette smoke exposure (SE) and the pathogenesis of diminished ventilatory efforts is unclear. There is no direct experimental evidence to substantiate whether maternal SE exerts deleterious effects, specifically on inflammatory processes in respiratory regions in adult offspring following maternal SE in mice. Therefore, we aimed to explore the inflammatory responses in the brain respiratory regions (nucleus tractus solitarius (NTS), retrotrapezoid nucleus (RTN) and ventral respiratory column (VRC)) or non-respiratory regions (hypothalamus, cerebellum) in SE dams and their adult offspring. In our study, adult female BALB/c mice (6 weeks old) were exposed to 12 cigarettes/day (Winfield, VIC, Australia) in the InExposure smoke exposure system for 6 weeks before mating until the offspring were weaned. After weaning, offspring (both sexes) were exposed to 12 cigarettes/day from postnatal day (p) 63 to p91 for 1 month and then culled on p92. Non-SE dams and offspring were exposed to room air as controls. Then, fresh brain tissues (hypothalamus, cerebellum, NTS, RTN, and VRC) were micro-dissected and processed for cytokine assays. The level of interleukin-1beta (IL-1β) in the hypothalamus, cerebellum, NTS, and RTN was unchanged in SE dams compared to the control dams. IL-1β levels in the VRC of the SE dams decreased compared with non-SE dams ($P < 0.05$, Mann-Whitney U test, n =12 , 2 mice samples pooled for the cytokine assay). In adult SE offspring, IL-1β levels were increased ($P < 0.05$, Mann-Whitney U test, n = 16-18, 2 mice samples pooled for the cytokine assay) in the VRC compared with the non-SE offspring.

The levels of IL-1β in the hypothalamus, cerebellum, NTS and RTN remained unchanged in the SE compared to non-SE offspring. We propose that immune activation in the VRC from prenatal or postnatal SE offspring could potentially predispose them to breathing dysfunction in adulthood. Maternal SE may modify brain immune responses in offspring at an early age, driving brain impairment in adulthood.

Disclosures: L. Bulbul: None. **H. Chen:** None. **N.M. Jones:** None. **N.N. Kumar:** None.

Poster

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Title: A Closed Loop Feature Detection Platform for Automated Neonate Cardio-Respiratory Measurements

Authors: *C. WARD[1], E. AISSI[3], D. PATEL[2], B. RUIZ[2], R. RAY[2];
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Abstract: Sudden Infant Death Syndrome (SIDS, a leading cause of death for neonate infants 1 month to 1-year-old) and some forms of neonate death (ND, the leading cause of death for newborns 28 days old or less) are thought to result, in part, from unseen brain abnormalities affecting cardio-respiratory function, but for which no clear genetic or environmental mechanisms are known. Mouse genetic and exposure models present an opportunity to uncover potential molecular and environmental mechanisms in SIDS and ND. However, measuring cardio-respiratory function to model SIDS and ND in neonate mice is expensive, difficult, and inefficient. Previously, we have developed for in-lab facile production an inexpensive face-mask pneumotachograph system for neonate respiratory measurements. A remaining challenge is the time and effort needed to carry out neonate respiratory measurements. For example, the neonate autoresuscitation assay consisting of repeated anoxic exposures followed by recovery requires the full attention of a single observer for the duration of the assay (2-3hrs) for a single subject, thus limiting the number of pups in a litter that can be assayed. To address these inefficiencies, we sought to develop a closed-loop feature detection platform for automated neonate cardio-

respiratory measurements that can be deployed in large numbers for high throughput parallel studies by a single person. The platform design consists of three sub-systems. First, we leveraged our previously published pneumotachograph face-mask system for precise respiratory measurements (PMID: 28213294). Second, we developed a micro-controller automated bell-housing gas switching system for near square wave induction of respiratory challenges. Third, we developed a micro-computer-based data-acquisition system with real-time feature detection and response outputs for controlling gas exposure times. Through the rotational and linear actuation driven by a pair of stepper motors, the gas challenges are administered via a bell housing that is rotated into position. A python program detects specific waveform features in real-time (lag = 100ms), such as apnea and bradycardia. Upon apnea detection, the system is directed to switch to a rescue gas. The resumption of normal breathing and heart rate can also be detected and incorporated into the criteria for the automated initiation of the next anoxic exposure trial. Data is stored for later offline automated analysis. The described apparatus offers an inexpensive approach for automated high throughput neonate cardio-respiratory assessment for facile screening to yield important clues in developmental pathophysiology.

Disclosures: **C. Ward:** A. Employment/Salary (full or part-time):; Baylor College of Medicine. **E. Aissi:** None. **D. Patel:** None. **B. Ruiz:** None. **R. Ray:** A. Employment/Salary (full or part-time):; Baylor College of Medicine.

Poster

P624. Respiratory Control I

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Topic: E.08. Respiratory Regulation

Support: Research was funded by the Canadian Institutes of Health Research and AF was supported by an Ontario Graduate Scholarship

Title: Neural circuits regulating opioid-induced motor hyperactivity and respiratory depression

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Abstract: Opioid-induced respiratory depression (OIRD) is characterized by a severe decline in breathing rate that can be lethal during opioid overdose. In addition to their inhibitory effects, opioid drugs also have excitatory effects illustrated by locomotor hyperactivity in rodents. While multiple neural circuits may be involved, the distinct cellular subpopulations mediating these opioid effects remain unclear. We aimed to determine the role of mu-opioid receptors (MORs) and somatostatin (SST)-expressing cells, which are critical for normal breathing and behaviours,

in regulating opioid effects in mice. We hypothesized that deletion of MORs and MORs in SST-expressing cells will reduce OIRD and opioid-induced hyperactivity. Transgenic knockout mice lacking MORs in SST-expressing cells (SST-MOR$^{-/-}$) were developed by crossing SST-Cre$^{+/+}$ mice (Sstt$^{m2.1(cre)Zjh/J}$) with Oprm1$^{fl/fl}$ mice (Oprm1$^{tm1.1Cgrf/KffJ}$). SST-Cre$^{+/+}$ and MOR$^{-/-}$ mice (Oprm1^{tm1Kff}) were used as controls. Mice (male, 3-4 months old) were placed in a whole-body plethysmography chamber to record respiratory rate and intraperitoneal injections of fentanyl (0.3 mg/kg) or saline were administered to quantify respiratory changes. Video recordings of mouse movements were captured simultaneously to assess behaviours. We found that intraperitoneal injection of fentanyl induced a significant respiratory rate depression in SST-Cre$^{+/+}$ ($p<0.001$, n=9), MOR$^{-/-}$ ($p<0.05$, n=7) and SST-MOR$^{-/-}$ mice ($p<0.001$, n=8) but was more pronounced in SST-Cre$^{+/+}$ and SST-MOR$^{-/-}$ mice. Fentanyl injection also induced a hyperactive state (increased locomotor behaviour) in SST-Cre$^{+/+}$ and SST-MOR$^{-/-}$ mice but not in MOR$^{-/-}$ mice. To further determine whether SST-expressing cells may be mediating subtle changes in breathing and behaviours, we are currently assessing respiratory changes during different behavioural states (inactive, active and hyperactive). In conclusion, our data showed that MORs mediate the effects of fentanyl on breathing and behaviours. Although SST is highly expressed in brainstem neurons that generate breathing, such as in the preBötzinger Complex, SST-expressing cells do not mediate the effects of fentanyl on breathing and behaviours.

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Poster

P624. Respiratory Control I

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Program #/Poster #: P624.08

Topic: E.08. Respiratory Regulation

Support: R01 HL144801
R01 HL126523
R01 HL151389

Title: Coordinated neural dynamics of breathing across the medulla and pons

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Abstract: The neural control of breathing involves the coordinated activity of diverse populations of medullary neurons distributed along the rostrocaudal aspect of the ventrolateral medulla: the so called Ventral Respiratory Group (VRG). The VRG participates in circuits with extra-medullary circuits, such as the pons, to generate robust—yet labile—rhythms.
Here, we use high-density electrophysiological recordings (Neuropixels) to record spiking

activity simultaneously from hundreds of neurons across both the VRG and pons in anesthetized, freely breathing mice. We couple this with an optogenetic tagging approach to identify ChAT, DBX1, Vgat, and Vglut2 positive neurons, and reconstruct the three dimensional location of each recorded neuron. We find diverse patterns of respiratory-related neural activity that continuously tile the entire breathing phase. Further, we observe intermingling of inspiratory and expiratory neurons across the VRG. By analyzing activity across simultaneously recorded neurons, we uncover dynamically coordinated ensembles of neurons that define "cell assemblies". These cell assemblies give rise to discretely different patterns of respiratory activity, including specific, distributed populations recruited during hypoxia induced gasping. In addition, we record from these distributed neural populations during administration of opiates (morphine) and observe that the respiratory network as a whole slows, but does not drastically reconfigure, in response to opiates. Together, these data provide an integrated view of the dynamic coordination of neural activity driving breathing in the intact animal.

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Poster

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Program #/Poster #: P624.09

Topic: E.08. Respiratory Regulation

Support: NIH R01 AG057052
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Title: Inhibition of TrkB Kinase Activity Impairs Autophagy Flux in Cervical Motor Neurons

Authors: *M. PAREJA-CAJIAO[1], N. J. HELLYER[2], E. C. DUCAJI[1], H. M. GRANSEE[1], G. C. SIECK[1,3], C. B. MANTILLA[1,3];
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Abstract: Autophagy is a catabolic process that eliminates damaged cytoplasmic structures. Aging-related autophagy impairment is suggested by the accumulation of damaged proteins and organelles in motor neurons and associated neuromuscular dysfunction. Signaling through the tropomyosin-related kinase receptor subtype B (TrkB) is important in maintaining neuromuscular function in the phrenic motor system including in old age. Our overarching hypothesis is that impaired autophagy flux contributes to aging-related neuromuscular dysfunction in phrenic motor neurons and that inhibiting TrkB kinase activity mimics the aging effects on autophagy in young mice. We previously reported increased expression of LC3 and p62 by 24 months of age in cervical motor neurons, indicating impaired autophagy with

autophagosome accumulation. In the present study, we leveraged an aging colony of *TrkB*^{F616A} mice, which possess a mutation that renders TrkB kinase activity susceptible to rapid and robust inhibition by 1NMPP1. Male and female *TrkB*^{F616A} mice at 6- 18- and 24- months of age received a 1-week treatment with vehicle or 1NMPP1. Relative expression of the autophagy markers LC3 and p62 in cervical motor neurons in the region of the phrenic motor nucleus was determined by immunofluorescence intensity. The effect of TrkB kinase inhibition was dependent on age. In 6-month old mice, there was an effect of 1NMPP1 treatment on both LC3 and p62 expression in motor neurons ($p < 0.01$ in both cases), and a sex difference in LC3 ($p<0.01$) but not p62 expression. There was no treatment*sex interaction. At 18- or 24- months of age, there were no effects on LC3 or p62 expression of 1NMPP1 treatment, sex, and no treatment*sex interaction. In 6-month old mice, treatment with 1NMPP1 increased the relative expression of both LC3 and p62 in cervical motor neurons by ~25% compared to control, consistent with the change in expression evident in 24-month old mice compared to 6-month old mice. Our results indicate that inhibition of TrkB kinase activity impairs autophagy flux in the motor neurons of young mice and this effect is similar to the effect of age on autophagy in motor neurons. Accordingly, the aging-related loss of TrkB signaling may impair autophagy flux and contribute to neuromuscular dysfunction in old age.

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Poster

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Program #/Poster #: P624.10

Topic: E.08. Respiratory Regulation

Support: FAPESP
CNPq
CAPES
Serrapilheira Institute

Title: Tumor Necrosis Factor alpha receptor 1 is necessary for respiratory functional and neuroanatomical changes in mice submitted to 6-OHDA model of Parkinson’s Disease

Authors: L. CABRAL[1], L. OLIVEIRA[1], T. S. MOREIRA[1], ***A. TAKAKURA**[2];
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Abstract: Introduction: Respiratory deficits are responsible for considerable deaths in Parkinson´s Disease patients. Previous studies have shown brainstem degeneration which is

probably linked to respiratory function impairment in a rat model of PD induced by 6-hydroxydopamine (6-OHDA) injection in caudate-putamen (CPu). Many mechanisms are responsible for neuronal death, including neuroinflammation which is present in many neurodegenerative diseases. **Objective:** This study aims to investigate the role of neuroinflammation mediated by glial cells in respiratory functional and neuroanatomical deficits in the animal model of PD induced by 6-OHDA. **Methods:** C57BL/6 or tumor necrosis factor receptor 1 knockout mice ($TNFR1^{-/-}$) were anesthetized with isoflurane 2.5% and received bilateral injection of 6-OHDA (10 µg/µl) or vehicle in CPu through stereotaxic surgery. 20 days after surgery, mice were submitted to whole body plethysmography under normal and hypercapnic conditions (7% CO_2). Animals were perfused and the brains were removed to anatomical analysis. **Results:** 6-OHDA destroyed TH^+ neurons in Substantia nigra (SNc) in all groups analyzed (6-OHDA WT: 77%; 6-OHDA $TNFR1^{-/-}$: 79%). Degeneration was observed in preBötzinger complex (preBötC) and in retrotrapezoid nucleus (RTN) in 6-OHDA WT group, but not in 6-OHDA $TNFR1^{-/-}$ group (preBötC: 6-OHDA WT: 43%, 6-OHDA $TNFR1^{-/-}$: 22%; RTN: 6-OHDA WT: 52%; 6-OHDA $TNFR1^{-/-}$: 15%). Astrocyte's density in SNc is enhanced in 6-OHDA WT group but not in 6-OHDA $TNFR1^{-/-}$ group (6-OHDA WT: 122%) and is reduced in preBötC in all groups analyzed ($p<0.05$), while in RTN, no change was observed. Microglia analysis indicates neuroinflammation in SNc and RTN in all groups. Respiratory assessment reveals reduction in basal respiratory rate in 6-OHDA WT group but not in 6-OHDA $TNFR1^{-/-}$ group (6-OHDA WT: 25%; 6-OHDA $TNFR1^{-/-}$: 17%). No change was observed between groups in respiratory parameters under hypercapnia. **Conclusions:** In this model of PD, respiratory deficits are associated with neurodegeneration in respiratory brainstem areas. Inhibiting TNF-α inflammation pathway, reduction in respiratory rate is prevented indicating the involvement of this pathway in the death of brainstem nucleus in this model.**Support:** FAPESP, CNPq, CAPES and Serrapilheira Institute.

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Poster

P624. Respiratory Control I

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Program #/Poster #: P624.11

Topic: E.08. Respiratory Regulation

Title: Purinergic signaling mediates neuroglial interactions to generate sighs

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Brain Res., Seattle Children's Res. Inst., Seattle, WA; [3]Dept. of Pediatrics, [4]Dept. of Neurolog. Surgery, The Univ. of Washington Sch. of Med., Seattle, WA

Abstract: Sighs are large amplitude breaths that occur periodically during normal breathing and are necessary for preventing atelectasis. Normal 'eupneic' inspirations and sighs originate from the same medullary network, the preBötzinger Complex (preBötC). Despite the importance of sighing in maintaining normal breathing, it is not understood how one circuit can produce two independently modulated and distinctly timed rhythmic behaviors. Sighs are modulated by many mechanisms both intrinsic and extrinsic to the brainstem circuits. However, a fundamental understanding of rhythm generation within the preBötC circuit is missing. We hypothesize that sighing originates within the preBötC and is regulated in part by astrocytic Ca2+ dynamics. Computational simulations predict that sighs occur with the coincidence of fast and slow Ca2+ oscillations. Experimental studies come to the same conclusion: pharmacologically increasing Ca2+ mimics in silico sigh generation. In vitro optogenetic stimulation of glia in Aldh1l1cre/Ai32 mice elicits sighs, which along with spontaneous sighs are inhibited with P2Y1 antagonists. Likewise, in vivo optogenetics stimulates sighs, but incomplete block with P2Y1 antagonists supports the idea of tightly regulated network activity and overriding influence from outside modulation. Furthermore, P2Y1 antagonist applied during short anoxic exposures in vitro eliminates the onset of sighs and reduces the augmenting eupneic phase. Our experiments currently focus on 2photon imaging of network calcium dynamics to more precisely understand the role of purinergic signaling in sigh generation, such as how timing of astrocytic Ca2+ fluctuations contribute to the temporal correlation of sighs and eupnea during different network states. Preliminary data suggests that astrocytes exhibit calcium fluctuations that loosely correspond to eupnea and sigh onset. These studies will focus on determining the source of fast and slow Ca2+ oscillations and the mechanisms by which increased glial Ca2+ through purinergic signaling contributes to both normal breathing and sighing. Taken together, our experiments suggest that the timing characteristics underlying sigh generation are a coordinated action between glial calcium oscillations and the recruitment of preBötC neurons along the inspiratory column through purinergic signaling.

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Poster

P624. Respiratory Control I

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Program #/Poster #: P624.12

Topic: E.08. Respiratory Regulation

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Title: Chronic maternal exposure to titanium dioxide nanoparticles alters breathing in newborn offspring

Authors: *E. COLNOT[1], L. CARDOIT[1], M.-J. CABIROL[1], L. ROUDIER[2], M.-H. DELVILLE[2], A. FAYOUX[1], P. DE DEURWAERDÈRE[1], M. THOBY-BRISSON[1], L. JUVIN[1], D. MORIN[1];
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Abstract: Over the last two decades, nanotechnologies and the use of nanoparticles represent one of the greatest technological advances in many fields of human activity. Titanium dioxide nanoparticles (TiO2 NPs) are one of the nanomaterials most frequently found in everyday consumer products. But, due to their particular physicochemical properties, TiO2 NPs are prone to cross biological barriers and potentially lead to adverse health effects. The presence of TiO2 NPs found in human placentae and in the infant meconium has indicated unequivocally the capacity for a materno-fetal transfer of this nanomaterial. Although chronic exposure to TiO2 NPs during pregnancy is known to induce offspring cognitive deficits associated with neurotoxicity, the impact of a gestational exposure on a vital motor function such as respiration, whose functional emergence occurs during fetal development, remains unknown. Here, using in vivo whole-body plethysmographic recordings from neonatal mice, we show that a chronic exposure to TiO2 NPs during pregnancy alters the respiratory activity of offspring, characterized by an abnormally elevated rate of breathing. Correspondingly, ex vivo experiments performed on isolated brainstem-spinal cord preparations of newborn mice and medullary slice preparations containing the central components of the breathing command, showed that the spontaneously generated respiratory-related rhythm is significantly and abnormally accelerated in animals prenatally exposed to TiO2 NPs. Moreover, whole-cell patch clamp recordings revealed that maternal exposure to TiO2 NPs during gestation affect the bioelectrical properties of pre-Bötzinger Complex inspiratory neurons by elevating their resting membrane potential and abnormally increasing their cellular excitability. Finally, such a chronic prenatal exposure was found to impair the capacity of respiratory neural circuitry to effectively adjust breathing rates in response to excitatory environmental stimuli. Our findings thus demonstrate that a maternal exposure to TiO2 NPs during pregnancy dramatically affects the normal development and operation of the respiratory centers in progeny.

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Poster

P640. Sleep and Stress

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P640.01

Topic: F.08. Biological Rhythms and Sleep

Support: European Research Council – FRONTHAL - 742595

Title: Optogenetic inhibition of calretinin positive neurons in the midline thalamus influences stress induced sleep disorder

Authors: *A. JÁSZ[1,4], L. BIRO[1], Z. BUDAY[1], O. SZALÁRDY[5,4], B. KIRALY[2,6], R. BODIZS[7,4], B. HANGYA[3], L. ACSADY[8];
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Abstract: Anna Jász, Laszlo Biró, Zsolt Buday, Robert Bódizs, Orsolya Szalárdy, Bálint Király, Balázs Hangya Laszlo Acsády **Post-stress activity of calretinin positive cells in the paraventricular thalamic nucleus is required for long term, stress induced disturbance of sleep behavior** Sleep disorders caused by stress affect millions of people around the world, but its neurobiological bases are still unclear. The calretinin-positive neurons of the paraventricular thalamus (PVT/CR+) are in a unique position to participate in stress induced sleep disturbances since their activity is significantly affected both by stress and by sleep-wake transitions. In this study we aimed to determine the activity of PVT/CR+ cells before and after the exposure to a natural stressor (fox odor, 2MT, 10 min) and to test causal relationship between post-stress PVT/CR+ activity and post-stress sleep behaviour using optogenetic inhibition (1 hour) of PVT/CR+ cells after the stress situation. Since sleep disturbances not only involve sleep but also the pre-sleep behaviour we separately analysed neuronal activity (216 units so far) and behaviour in the nest before sleep. Recordings involved 3 hours sessions for five days before and after stress using movable tetrodes and optogenetic tagging of PVT/CR+ cells. Sleep depth was measured by spectral slopes calculated on the basis of the power law scaling of the Fourier spectra.PVT/CR+ cells displayed strongly state dependent activity during pre-stress days. Wake activity in the nest was lower than outside the nest and firing further decreased at the onset of sleep. At the day of the exposure to 2MT both firing rate and synchrony among PVT/CR+ cells increased. Firing rate remained elevated for six days after the stress, with strongest change in the nest. Bursting activity of PVT/CR+ cells decreased during NREM sleep after the stress. These changes were accompanied by altered locomotor activity, nesting behaviour and a decrease in the spectral slope during NREM sleep, showing disturbed sleep. Optogenetic inhibition of PVT/CR+ neurons for one hour after 2MT presentation prevented altered locomotion before sleep and the normal nesting behavior were reinstated on the poststress days. These data together strongly indicate that altered post-stress activity of PVT/CR+ cells is crucial to establish the neuronal network responsible for the emergence of stress induced sleep behaviour.

Disclosures: A. Jász: None. **L. Biro:** None. **Z. Buday:** None. **O. Szalárdy:** None. **B. Kiraly:** None. **R. Bodizs:** None. **B. Hangya:** None. **L. Acsady:** None.

Poster

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Program #/Poster #: P640.02

Topic: F.08. Biological Rhythms and Sleep

Support: NIGMS NIH Award Number U54GM133807
NSF Award Number 1633184

Title: Does the "Night-float rotation" alter sleep/wake cycles and circadian rhythms of UPR physician residents: Modulation by chronotypes

Authors: *J. R. MARRERO[1], Y. DE JESUS[3], A. SEGARRA[4], L. ORTA ANÉS[5], C. M. NAZARIO[5], F. RAMIREZ[2], J. L. AGOSTO[1];
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Abstract: Introduction: Sleep disruption during medical residency is recognized as a significant risk factor for medical trainees' exhaustion. Burnout increases the risk of medical errors and mental health deterioration, potentially leading up to physician suicide. Coaching interactions with physicians in-training revealed that some residents are struggling with sleep difficulties during the night-float rotation. We aim to explore the relationship between residents' chronotype and their misalignment with work shift as a possible reason for this observation.
Methods: This is a longitudinal cohort study of repeated measures in twenty medical residents before, during, and after the night-float rotation. Participants will complete the Composite Scale of Morningness to determine their chronotype; and the Pittsburg Sleep Quality Index to measure perceived sleep quality during each rotation. To explore confounding variables a standard questionnaire and the Demand Control Support Questionnaire will be completed. Physiologic parameters will be monitored using an actigraph and a dermal patch (Biosensor) to measure sleep quality and determine internal circadian synchronization. Salivary cortisol and melatonin levels will be measured to assess stress and explore circadian fluctuations as related to sleep disruption.
Analysis: The measure of outcome variation according to hazard exposures and other characteristics will be performed by collecting information on each participant. To increase power, measurements will be repeated on the same participants at various points in time. We anticipate a small sample size, therefore, the importance of study results would not be based solely on statistical significance, but also on clinical significance.
Conclusions: Exploring the effects of a night-float rotation in residents' sleeping pattern and its relationship to their chronotypes may reveal missed opportunities to prevent burnout syndrome in medical residencies.

Disclosures: J.R. Marrero: None. **Y. de Jesus:** None. **A. Segarra:** None. **L. Orta Anés:** None. **C.M. Nazario:** None. **F. Ramirez:** None. **J.L. Agosto:** None.

Poster

P640. Sleep and Stress

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Program #/Poster #: P640.03

Topic: F.08. Biological Rhythms and Sleep

Title: Desynchronization of wake-sleep rhythm and eating patterns during the social isolation by Covid-19 in a Mexican Population

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Abstract: Overweight and obesity has become one of the most serious global health issues due to the high rates of prevalence that presents in different populations. The social isolation during COVID-19 pandemic had an impact of on sleep, dietary patterns and physical health. Sleep is an essential process for the brain and the maintaining of metabolism. Inadequate sleep and obesity are important public health problems. Studies provide consistent evidence that recurrent sleep curtailment and shifting time of sleep may increase the risk of obesity and diabetes. In this sense the circadian clock, controls sleep, onset of sleep, also regulates energy homeostasis and the disruption is associated with obesity.During the social isolation many people shift their sleep and had more irregular eating patterns. The aim of the present study was to investigate relationship between circadian rhythm of sleep and eating patterns during social isolation caused by the contingency Covid-19 in a Mexican population. For this purpose, a total of 2385 subjects (18-80 years) participated. The study was conducted in according to standards established by the declaration of Helsinki as well as their applied the informed consent. All subjects completed a questionnaire in a google forms, and scale parameters Pittsburght for quality and quantity of total sleep, also completed a questionnaire for identify eating patterns, light exposure and use of electronic devices. The results suggest that the population presented desynchronization of wake-sleep rhythms during the social isolation due the COVID-19 contingency and presented desynchronization of feeding-fasting rhythms, also the people had a variation in the patterns of exposure to light. Our data suggest that the social isolation impact in the energetic metabolism and circadian control of sleep, this condition might promote development and increase obesity rates and other chronic diseases in this population, promoting comorbidity with infectious diseases such as COVID-19.

Disclosures: L. Perez-Puente: None. **A. Montezano-Lopez:** None. **F. Sandoval-Gómez:** None. **A. Pavon-Rosado:** None. **C. Gutierrez-Garcia:** None. **M. Melgarejo-Gutierrez:** None.

Poster

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Program #/Poster #: P640.04

Topic: F.08. Biological Rhythms and Sleep

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KAKENHI 20K22297

Title: The first-night effect impairs neurochemical processing of visual plasticity during NREM sleep, revealed by simultaneous magnetic resonance spectroscopy and polysomnography

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Abstract: How sleep disturbances affect learning and memory has remained elusive. We have previously found that performance on a visual perceptual learning (VPL) task improved after nonrapid eye movement (NREM) sleep (offline gains), and that NREM sleep plays a role in enhancing visual plasticity. Here, we investigated whether non chronic sleep disturbances impair visual plasticity by utilizing the well documented phenomenon, the first night effect (FNE) that refers to a temporary sleep disturbance that occurs only during the first session of sleep experiments in healthy young human adults. First, we tested whether the FNE affects a neurochemical plasticity index during NREM sleep. Previous studies demonstrated that increases in the neurochemical plasticity index, the ratio of excitatory (glutamate) to inhibitory (GABA) neurotransmitter concentrations (E/I ratio) in early visual areas (EVA) measured by magnetic resonance spectroscopy (MRS), were closely related to enhanced visual plasticity in VPL. Moreover, the E/I ratio increased during NREM sleep compared to wakefulness, in correlation with offline gains of VPL over sleep. We tested whether the FNE prevents the E/I ratio from increasing during NREM sleep, by measuring concentrations of glutamate and GABA in EVA during sleep using simultaneous MRS and polysomnography on day 1 (with FNE) and 2 (without

FNE). Subjects were asked to sleep inside the MRI bore for 90 min, twice with an interval of a week. There was no significant increase in the E/I ratio during NREM sleep compared to wakefulness on day 1. The E/I ratio during NREM sleep relative to wakefulness was significantly smaller on day 1 than day 2 ($p=0.004$), suggesting that the FNE impairs visual plasticity during NREM sleep. Next, we behaviorally tested whether the FNE impairs offline gains in VPL over sleep, by comparing offline gains over sleep with and without the FNE. Before subjects were asked to sleep for 90 min, they were trained on the texture discrimination task (TDT), a standard VPL task, at different visual hemifields on days 1 and 2 with an interval of a week. Because learning of TDT is specific to the trained retinal location, the state of learning on each day is safely assumed to be independent. Test sessions on TDT were performed before and after the sleep session. While significant offline gains over sleep on day 2 without the FNE ($p<0.001$) replicated previous finding, no significant offline gains found on day 1 with the FNE supports that the FNE impairs visual plasticity during sleep. Altogether, these results clearly suggest that non chronic sleep disturbances alter neurochemical processing and impair visual plasticity during sleep in healthy young adults.

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Poster

P640. Sleep and Stress

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Program #/Poster #: P640.05

Topic: F.08. Biological Rhythms and Sleep

Support: KAKENHI 17H06095

Title: Involvement of microglia in sleep/wake regulation under acute social defeat stress in mice

Authors: *K. MIYANISHI[1], H. FUNATO[2], M. YANAGISAWA[1];
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[2]Toho Univ. Sch. of Med., Tokyo, Japan

Abstract: Microglia, the brain-resident macrophages, produce inflammatory cytokines and remove dead cells and foreign materials in diseased brains. Recently, we reported circadian changes in the morphology and function of microglia. In the prefrontal cortex, microglia displayed hyper-ramified morphology and increased phagocytosis of synapses during sleep. These results suggested that the circadian changes in microglial activity may contribute to sleep/wake regulation. However, how microglia are involved in sleep/wake behavior remains to be elucidated. Here, we examined sleep/wakefulness in mice administered with selective colony-stimulating factor-1 receptor inhibitor, PLX5622 (PLX), which depletes microglia. We further

examined whether social defeat stress alters the effect of microglial elimination on sleep/wake behavior. EEG/EMG recordings were performed after mice were fed with the PLX-formulated chow for 14 days. As a result, PLX-treatment did not change wake, NREM sleep, and REM sleep amounts. Thus, microglial elimination by PLX-treatment did not appreciably change sleep/wake behavior in adult healthy mice. Next, we examined the effects of microglial ablation on sleep/wake cycle in socially defeated mice. Social defeat stress is well-known to induce microglial activation, and previous studies reported that acute social defeat stress increased NREM sleep amounts. Following the previous studies, in our social defeat stress model, C57BL/6N mice were briefly encountered with highly aggressive ICR mice 4 times in 1 hour. We exposed mice with or without PLX-treatment to 1-hour social defeat stress during ZT11-12 and performed EEG/EMG recordings. After social defeat stress, PLX-treated mice showed further increased NREM sleep amounts compared with mice without PLX-treatment. These results suggested the possibility that microglia may be involved in sleep/wake regulation in response to social stress in mice.

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Poster

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Topic: F.08. Biological Rhythms and Sleep

Support: European Research Council – FRONTHAL - 742595

Title: The role of calretinin positive neurons of the paraventricular thalamus in sleep disturbance induced by innate fear

Authors: *L. BIRO[1], A. JASZ[1], Z. BUDAY[1], G. KOMLOSI[1], S. FERENCZI[2], K. KOVACS[1], L. ACSADY[1];
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Abstract: Exposure to severe stress could lead to the emergence of stress-related psychiatric disorders such as insomnia and PTSD. The paraventricular thalamic nucleus (PVT) is a midline thalamic region involved in sleep, fear, anxiety, and stress response regulation. Calretinin-positive cells of the PVT (PVT/CR+) display elevated c-Fos expression upon aversive stimulus exposure and send highly collateralized projections to stress regulatory structures. In this study, we tested whether the post-stress activity of PVT/CR+ cells is required for the manifestation of disturbed sleep behavior induced by exposure to a natural stressor (fox odor, 2MT). We studied how post-stress optogenetic photoinhibition of PVT/CR+ neurons affects nesting and sleep

behaviors, stress hormone levels, and neuronal activity in stress-responsive downstream brain regions. PVT/CR+ neurons were inhibited using the inhibitory step-function opsin SwiChR. After five pre-stress days SwiChR and control, EYFP mice were subjected to a predator odor (2MT, 10 min) followed by 1 hr laser exposure in their homecage. During stress exposure, both SwiChR and control groups exhibited similar levels of defensive behaviors (~80% time), particularly freezing and escape jumps. Compared to the pre-stress behavior after stress exposure, the EYFP group showed increased locomotion, freezing-like behavior, and disturbed sleep which was associated with marked c-Fos expression in PVT/CR+ neurons and elevated corticosterone levels. Photoinhibition of PVT/CR+ cells after the stress exposure prevented all these changes. We also tested whether post-stress photoinhibition of PVT/CR+ cells is able to reverse elevated c-fos expression in their major postsynaptic targets such as nucleus accumbens (NAc), central amygdala (CeA), prelimbic cortex (PrL), and bed nucleus of the stria terminalis (BNST). Collectively, our findings indicate that post-stress photoinhibition of PVTCR neurons resulted in decreased neuronal activity, which was sufficient to decrease stress-induced discomfort and sleep disturbance and alleviate hormonal stress reactivity.

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Poster

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Topic: F.08. Biological Rhythms and Sleep

Support: NIH Grant MH60670

Title: Post-traumatic stress REM sleep features and heart rate variability contributing to fear extinction memory

Authors: *Y. CABRERA[1], J. A. HOLLOWAY[1], S. N. AYALA ROSARIO[1], J. URE[2], K. KIM[2], G. R. POE[1];
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Abstract: Research into rapid eye movement (REM) sleep has revealed its role in synaptic plasticity mechanisms supporting the updating of memory schema and attenuating emotionality linked to experiences. Absence of the stress neurotransmitter, norepinephrine (NE), during REM sleep is paramount to the maintenance of the REM sleep state and the depotentiation mechanisms believed to underlie adaptive emotional memory processing. REM sleep is normally characterized by large theta (5-10 Hz) frequency oscillations in the associative memory and emotion processing limbic system as well as muscle atonia and increased heart rate variability

(HRV). Evidence from our lab and others supports the hypothesis that REM sleep disturbance impairs emotional memory processing via aberrant NE signaling, causing abnormal memory retention and altered autonomic system regulation following stress which in turn lead to retention deficits in fear extinction and increase the risk of developing post-traumatic stress disorder (PTSD). Interestingly, a top symptom of PTSD is sleep disturbance like insomnia, characterized by fragmentation, non-restorative sleep, and decreased HRV. These disturbances likely reflect heightened vigilance and arousal during sleep. REM sleep micro-features and autonomic physiology after stressor exposure have not been completely characterized nor examined holistically. Thus, this study looked at sleep architecture, REM sleep theta features, and HRV to examine differences between baseline and post-traumatic stress conditions in rats exhibiting adaptive versus maladaptive responses in subsequent fear extinction learning. We hypothesized that rats showing the greatest REM sleep and HRV disruption relative to baseline would show greater deficits in fear extinction retention. Rats were implanted with electrodes over parietal and frontal brain regions, in nuchal muscles to measure atonia, and over intercostal muscles to measure heart rate. After recovery, we analyzed 24-hour recordings under baseline conditions as well as after stress enhanced fear learning (SEFL), a rodent model of PTSD. We found that increased REM sleep fragmentation, increased phasic muscle twitches, and decreased HRV relative to baseline were correlated with impaired fear extinction memory retention. Also, increased numbers of transitions to wake and decreased REM theta frequency and power were correlated with poorer extinction learning outcomes. These results, all associated with abnormal presence of NE, corroborate the importance of REM sleep stability in adaptive mnemonic and autonomic processes following stress, and may contribute to targeted sleep treatments for PTSD.

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Poster

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Program #/Poster #: P640.08

Topic: F.08. Biological Rhythms and Sleep

Title: At-home sleep quality measurements correlate with next-day seizure counts in individuals with Epilepsy

Authors: *K. PATEL[1,2], C. N. KATZ[1,2], M. GARINGO[2], S. ZUM[2], X. FAN[2], J. STERN[3,2], P. SABETIAN[2];
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Abstract: Epilepsy is a common, complex neurological condition that does not respond to treatment with seizure freedom for about 30% of patients. Beyond seizures, epilepsy also is associated with an array of neurological and behavioral comorbidities that collectively reduce quality of life (QoL). Among the co-morbidities, sleep disorders is one that is a well-known predictor of reduced QoL, and sleep is particularly relevant for individuals with epilepsy because of the association between sleep deprivation and seizure likelihood. These known interactions between sleep and epilepsy have been mostly established through studies that are either diary based or performed in controlled monitoring environments, such as in epilepsy monitoring units. In this study, we used daily physiologic monitoring to measure the interaction between sleep quality and seizure frequency in individuals with epilepsy in their home environments. The monitoring utilized wearable devices to record motion from 22 individuals with epilepsy over the course of 338 nights with more than 4 hours of continuous data per night. We developed an algorithm that utilizes the principles of actigraphy to identify periods of restlessness during sleep and used this algorithm to calculate total sleep duration and sleep efficiency for each night. These metrics were compared to expected values of a healthy individual of a similar age to calculate a comprehensive sleep quality score for each night. Linear regression was performed between nightly sleep quality scores and seizure counts the following day collected with an interactive data platform. We found that sleep quality had a significant negative relationship with next-day seizure counts across all patients and all sleep sessions (Pearson's R = -0.1653, p-value = 0.0023). These results suggest that a simple actigraphy-based approach can effectively capture sleep quality in this patient population via off-the-shelf wearables, and that sleep quality may be an important predictor of risk for subsequent seizures on the following day. The results also support the use of wearable physiological monitoring devices to assist in epilepsy care through sleep analysis

Disclosures: K. Patel: A. Employment/Salary (full or part-time):; Novela Neurotechnologies. **C.N. Katz:** A. Employment/Salary (full or part-time):; Novela Neurotechnologies. **M. Garingo:** A. Employment/Salary (full or part-time):; Novela Neurotechnologies. **S. Zum:** A. Employment/Salary (full or part-time):; Novela Neurotechnologies. **X. Fan:** A. Employment/Salary (full or part-time):; Novela Neurotechnologies. **J. Stern:** F. Consulting Fees (e.g., advisory boards); Novela Neurotechnologies. **P. Sabetian:** A. Employment/Salary (full or part-time):; Novela Neurotechnologies.

Poster

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Topic: F.08. Biological Rhythms and Sleep

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Title: Early propranolol administration following fear extinction training reduces sleep spindle-slow oscillation coupling and enhances fear extinction recall

Authors: *R. GUTHRIE[1], J. HOLLOWAY[1], Y. CABRERA[1], J. URE[1], S. N. A. ROSARIO[1], K. KIM[1], G. R. POE[2];
[1]Univ. of California, Los Angeles, CA; [2]Dept. of Integrative Biol. and Physiol., UCLA, Los Angeles, CA

Abstract: Sleep is a critical period for the cortical transfer and memory consolidation of recent experiences involving hippocampal dependent learning. One mechanism thought to underlie this process is the coupling of hippocampal replay during ripples to cortical slow waves and spindles during non-REM sleep following learning. Exposure to psychological trauma can cause sustained norepinephrine release across sleep, which can disturb sleep continuity. Norepinephrine prevents synaptic depotentiation processes that are necessary for reversal learning including fear extinction learning and facilitates potentiation. *We hypothesized that elevated norepinephrine release during post-trauma sleep would prevent fear extinction memory consolidation and alter spindle and slow oscillation dynamics.* The present study probes the effect of sleep features following psychological trauma treatment using the beta-adrenergic receptor blocker propranolol during the critical post-learning sleep memory consolidation window. Animals implanted with electrodes in the parietal and frontal cortex were exposed to stress enhanced fear learning (SEFL) then, 8 days later, to 5 pairs of tone-shock fear conditioning. 24 h later they were fear extinction (FE) trained in a separate context. One group was given 40 mg/kg propranolol immediately after training and the other 4 h later, after the critical consolidation window. 5 days later, animals were tested in the extinction context for contextual and cued fear extinction recall (FER) as measured by freezing. Preliminary results show that animals given propranolol immediately after FE had lower freezing the next day compared to those administered propranolol after the consolidation window. Early propranolol animals showed less contextual and cued freezing on extinction recall day, a trend correlated with a reduction in sleep spindle density, slow wave density, and spindle-slow oscillation coupling in the parietal cortex. Amount of time spent in slow wave sleep did not predict freezing. Spindle-slow oscillation coupling in high-freezing animals may reflect continued consolidation of the fear memory traces in the hippocampus despite extinction training. These results highlight the importance of taking into account specific sleep features that may govern consolidation of emotional memory instead of total time spent in each state. Future work is needed to explore the mechanisms through which propranolol reduces spindle-slow oscillation coupling and the content of the memory replay in treated vs untreated animals that could support resilience from overgeneralization and sustained fear.

Disclosures: R. Guthrie: None. **J. Holloway:** None. **Y. Cabrera:** None. **J. Ure:** None. **S.N.A. Rosario:** None. **K. Kim:** None. **G.R. Poe:** None.

Poster

P655. Amygdala and Related Systems for Defensive Behavior

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Topic: H.07. Long-Term Memory

Support: National Research Foundation of Korea (NRF) grant 2018R1A2B3004486

Title: Dissociated role of thalamic and cortical input to the lateral amygdala for consolidation of long-term fear memory.

Authors: *Y. LEE, J.-P. OH, J.-H. HAN;
KAIST, Yuseong-Gu, Daejeon, Korea, Republic of

Abstract: Coordinated communications of inter-regional brain networks during post-learning period are thought to be critical for the consolidation of memories. However, the role of specific neural circuit pathways in this process has been scarcely explored. It has been well established that auditory input to the lateral nucleus of amygdala (LA) is essential for the acquisition and retrieval of auditory fear memory. Accumulating evidence suggests that fear conditioning induces associative plasticity not only in the amygdala but also in regions outside the amygdala, including the various stage of the auditory pathways. Further, post-learning engagement of the auditory cortex is necessary for the formation of remote fear memories and the coherent cortico-amygdala theta during remote memory retrieval. Despite these findings, it is unknown whether auditory inputs to the LA is necessary during a post-learning period for consolidation of fear memory. To investigate this question, we optogenetically silenced the inputs from both auditory cortex (AuV/TeA) and thalamus (MGm/PIN) in the LA for 15 min immediately following auditory fear conditioning (FC) and examined its effect on fear memory formation in mice. Optogenetic inhibition of both inputs disrupted long-term fear memory formation tested 24 h after FC. This effect was specific such that the same inhibition did not affect short-term memory and context-dependent memory. Moreover, long-term memory was intact if the inputs were inhibited at much later time points after FC (3 h or 1 d after FC), indicating that optical inhibition for 15 min itself does not produce any non-specific deleterious effect on fear memory retrieval. Selective inhibition of thalamic input was sufficient to impair fear memory formation. In contrast, selective inhibition of cortical input disrupted remote fear memory without affecting recent memory. These results reveal a dissociated role of thalamic and cortical input to the LA during early post-learning periods for consolidation of long-term fear memory.

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Poster

P655. Amygdala and Related Systems for Defensive Behavior

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Topic: H.07. Long-Term Memory

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Samsung Science and Technology Foundation SSTFBA1801- 10
NRF Korea (ME) Grant 2019R1A6A3A01097213

Title: Potentiated neurons preferentially participate in a fear memory engram during memory formation

Authors: *Y. JEONG, H.-Y. CHO, M. KIM, J.-P. OH, M. KANG, M. YOO, H.-S. LEE, J.-H. HAN;
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Abstract: Memory engram, defined as physical changes elicited by learning, is consisted of sparse neuronal networks across the brain. Such changes mainly occur on synapses in the form of long-term potentiation (LTP). LTP is a persistent strengthening of synaptic transmission between neurons and is considered a neural substrate of memory. However, due to the technical limitations of current engram labeling methods which required sufficient new protein synthesis, thus take at least hours and days, the role of synaptic plasticity during memory engram formation remained untestable. In this study, we developed an optogenetic method to prime sparse auditory-to-LA synapses which were not originally potentiated by auditory fear conditioning. We first confirmed these primed synapses were potentiated by the following learning. The activity of the primed synapses was necessary and sufficient for auditory fear memory recall. Next, visualization of neuronal activity marker, c-Fos, after retrieval revealed that the potentiated auditory neurons by priming more preferentially participated in a fear memory engram than non-primed neighboring neurons. Such preferential memory encoding was reversed by optical long-term depression (LTD) shortly after learning, while memory formation was normal. However, LTD 30 min after learning prevents fear memory recall. Finally, optical LTP shortly after learning also sufficiently induced preferential memory encoding into the potentiated neurons. Notably, the number of c-Fos-positive cells was not changed among all stimulation conditions. Taken together, these results indicate that LTP during memory formation directs which neurons will preferentially encode memory.

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Poster

P655. Amygdala and Related Systems for Defensive Behavior

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Topic: G.04. Emotion

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The Brain and Behavior Research Foundation
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Title: A genetically-defined population in the lateral and ventrolateral periaqueductal grey selectively promotes active avoidance from threat

Authors: *M. LA-VU, E. SETHI, S. PEREIRA, B. TOBIAS, F. REIS, W. WANG, S. LEONARD, P. SCHUETTE, L. LIN, A. ADHIKARI;
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Abstract: When encountering threat in the external world, an animal's survival depends on the engagement of appropriate defensive reactions to minimize harm or potential injury. There are major clinical implications for identifying the neural circuitry and activation patterns that produce such defensive reactions, as abnormal overactivation of these circuits is thought to underlie pathological human anxiety and fear responses. Studies using electrical stimulation, immediate early gene analysis, lesions, and excitatory amino acid injections strongly implicate the periaqueductal grey (PAG) in defense. Varying defensive functions mediated by the PAG have long been associated with anatomical column-shaped subregions along its rostrocaudal axis, generally with the dorsolateral column (dlPAG) controlling active avoidance and the ventrolateral column (vlPAG) controlling passive avoidance. Despite decades of studying the functions of these anatomical PAG subdivisions, the roles of various genetically-identified PAG populations have not been explored. Here, we show that optogenetic activation of a population of cholecystokinin-expressing (cck+) neurons in the mouse lateral and ventrolateral PAG (l/vlPAG) promoted active avoidance of open spaces and live predators, while inhibition of these cells decreased avoidance of threats, without altering any other defensive behaviors such as freezing. Finally, using in vivo fiber photometry we show cck+ l/vlPAG population activity ramped down with proximity to a predator and encoded aspects of relative safety within an environment. In contrast, activating pan-neuronal (synapsin-expressing) l/vlPAG cells promoted freezing and synapsin-expressing cells were activated in proximity to threat. Together these data show that in the l/vlPAG, a region classically known to control passive avoidance, there is a cck+ neuronal population that is sufficient and necessary for selectively controlling active avoidance from threat. These data underscore the paramount importance of investigating genetically-identified PAG cells, and show that, in addition to this region's anatomical columnar architecture, the molecular identity of PAG cells may confer an additional axis of functional organization.

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Poster

P655. Amygdala and Related Systems for Defensive Behavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P655.04

Topic: G.04. Emotion

Support: ARC Discovery Grant DP170103952
Australian Government Research Training Fellowship

Title: Higher-order fear learning in the rat basolateral amygdala complex is independent of NMDA receptors, and instead requires the activation of calcium-permeable AMPA receptors on GABAergic interneurons

Authors: *M. WILLIAMS-SPOONER[1], A. J. DELANEY[2], F. WESTBROOK[3], N. M. HOLMES[3];
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Abstract: In Pavlovian fear conditioning, learning about cues that signal aversive events, such as a footshock, requires the basolateral amygdala complex (BLA). In naive rats, acquiring conditioned freezing to a discrete cue paired with shock involves glutamatergic receptor activity in the BLA, including NMDA receptors (NMDAR). During fear conditioning, sensory inputs carrying information about pairings of conditioned and unconditioned stimuli (CS-US pairings) converge on principal cells in the BLA. The resulting depolarization releases the magnesium block from the NMDAR channel, enabling it to be activated by glutamate. This permits influx of calcium and triggers intracellular cascades which direct the cellular plasticity that is believed to underlie fear memories. Thus, NMDAR activation on BLA principal cells is thought to act as a coincidence detector for salient events (e.g., CS-US pairings) and a gateway for the plasticity that encodes fear learning in memory. However, recent studies on context fear conditioning in the dorsal hippocampus indicate that NMDAR activation is unnecessary when the delivery of the US is fully predicted by what the rat has already learned in prior training. Contrary to the standard view, these results suggest that NMDAR are not simply activated by the coincidence of sensory events, but by the coincidence of unexpected sensory events. To assess whether this is also true of cued fear conditioning in the BLA, we used a two-stage fear conditioning protocol, recently developed by our lab, in which rats receive S1-shock pairings in stage one, and serial-order pairings of S2-S1-shock in stage two. Here, using bilateral cannulations of the rat BLA and micro-infusions of D-AP5 to block NMDAR activation, we show that acquiring conditioned fear

to S2 in this protocol does not require NMDAR. We further show that this NMDAR-independent fear learning to S2 instead requires the activation of calcium-permeable AMPAR receptors (CP-AMPAR) in the BLA, using the CP-AMPAR antagonist, NASPM. Interestingly, CP-AMPAR were also necessary for NMDAR-dependent fear learning to S2 and S1. Finally, we use electrophysiological experiments to demonstrate that NASPM selectively inhibits interneurons, and not principal cells, in the rat BLA. These findings indicate that activation of NMDAR on principal cells and CP-AMPAR on interneurons are both necessary for learning when fear conditioning involves unexpected events, while activation of CP-AMPAR on interneurons is sufficient when fear learning to a novel cue involves events that are expected based on prior learning.

Disclosures: M. Williams-Spooner: None. **A.J. Delaney:** None. **F. Westbrook:** None. **N.M. Holmes:** None.

Poster

P655. Amygdala and Related Systems for Defensive Behavior

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Program #/Poster #: P655.05

Topic: F.04. Stress and the Brain

Support: NIH Grant R01MH100536

Title: Sex differences in small conductance calcium-activated potassium channel (SK) expression in rat basolateral amygdala neurons following repeated restraint stress

Authors: *J. VANTREASE[1], J. KO[2], J. H. URBAN[3], J. ROSENKRANZ[1];
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Abstract: Mood disorders disproportionally affect women more than men. The basolateral amygdala (BLA) is critical for the expression of fear and anxiety-like behaviors and is hyperactive in mood disorders. Stress is a leading factor in the development of mood disorders and BLA activity following stress also varies between the sexes. Specifically, stress leads to an increase in BLA spontaneous activity in males, but a reduction in activity in females. Prior work has shown greater BLA activity in naïve females compared to males, but the mechanisms by which this occurs remain unknown. BLA activity is regulated, in part, by small conductance calcium-activated potassium (SK) channels, which mediate afterhyperpolarizations (AHPs) and reduce neuronal firing. Activation of the SK2 channel inhibits BLA neuronal firing in males, but the role of SK2 in females is relatively unexplored. We hypothesized that SK2 expression would

be lower in females resulting in higher spontaneous firing rate of BLA neurons compared to males, and that the expression of SK2 will be altered by stress in a sex-specific manner. In order to test this, we utilized qPCR and Western blot analysis to determine the relative expression of SK2 in the BLA of adult naïve male and female rats. Preliminary results reveal BLA SK2 mRNA levels are similar between the sexes, but BLA SK2 protein levels are lower in females compared to males. We further hypothesize that lower SK2 expression in females will correspond with a smaller SK2 channel activity contributing to increased spontaneous BLA activity. To test this we performed in vivo electrophysiology of BLA neural activity following local injections of a SK2 channel inhibitor (apamin; 1uM) or saline. Additionally, SK2 expression is reduced in response to stress in males, but it is unknown whether this trend occurs in females. Therefore, we also sought to determine whether sex differences exist in SK2 expression and function in rats following repeated restraint stress (20 min/day for 7 out of 9 days), compared to control-handled rats. Our initial findings suggest that repeated restraint stress increases spontaneous BLA activity in males and this activity is less sensitive to apamin injection compared to control-handled rats. In contrast, repeated restraint stress appears to reduce spontaneous BLA activity in females, which is then more sensitive to apamin injection. A more thorough understanding of sex differences in SK2 channel expression and activity following stress may contribute to a better understanding of the sex disparities observed in mood disorders.

Disclosures: J. Vantrease: None. **J. Ko:** None. **J.H. Urban:** None. **J. Rosenkranz:** None.

Poster

P655. Amygdala and Related Systems for Defensive Behavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P655.06

Topic: G.04. Emotion

Support: Stichting Incontinence Foundation

Title: Amygdala Control of the Emotional Motor System

Authors: *G. HOLSTEGE[1], H. H. SUBRAMANIAN[2];
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Abstract: The amygdala regulates fear and anxiety. Expression of fear and anxiety is associated with changes in the emotional motor system control of breathing, cardiovascular and pelvic organ function. The amygdala via its strong projections to the periaqueductal gray (PAG) could function as one of the emotional motor pathways for the modulation of fear and anxiety. In that case, stimulation of the amygdala should have a direct causal influence on breathing as well as on cardiovascular and pelvic organ functions. This was investigated by stereotaxic glutamate

stimulation of different portions of the amygdala in both cats and rats. Glutamate only stimulates the cell bodies and not the fibers of the passage. Using this technique it was possible to find out whether within the amygdala a topographical organization of emotional functions exist. **Results from the cat studies** **Respiration:** In the cat, stimulation of the lateral amygdala generated hyperpnea and tachypnea, while stimulation of the central amygdala induced dyspnea, bradypnea, inspiratory apneusis and double diaphragm breathing patterns. Stimulation in the amygdala never generated apneas. **Cardiovascular effects:** Stimulation in both the lateral and central amygdala induced hyper- as well as hypotension. **Micturition:** In the cat stimulation of central amygdala generated micturition, but stimulation in the lateral amygdala did not. **Results from the rat studies** Focal stimulation of the central amygdala in the rat induced both tachypnea/hypertension as well as apnea/hypotension. Topical stimulation in the lateral portion of the central amygdala produced breath-hold. Focal glutamate stimulation in the lateral amygdala did not result in any cardiorespiratory effect. In the rat micturition was never generated in either the lateral or the central amygdala. **Conclusion** The lateral and the central amygdalar areas produce distinct modulations of the emotional motor system depending on its association with stress, fear and anxiety. Differences between the cat and rat exist in terms of modulation of the emotional motor system. Acknowledgement The animal studies were undertaken by GH and HHS wholly at The University of Queensland between 2013-2017 under UQ institutional ethics approval. HHS and GH designed the project, performed the experiments, analysed primary data and made figure illustrations. GH curated and approved the final data/figure representation in this presentation/poster. None of work described here were undertaken at either of the current work designations of Hari Subramanian or Gert Holstege.

Disclosures: G. Holstege: None. **H.H. Subramanian:** None.

Poster

P655. Amygdala and Related Systems for Defensive Behavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P655.07

Topic: E.07. Rhythmic Motor Pattern Generation

Support: Stichting Incontinence Foundation

Title: The midbrain periaqueductal gray (PAG): one stop-shop for emotional motor modulation?

Authors: *H. H. SUBRAMANIAN[1], R. J. BALNAVE[2], G. HOLSTEGE[3];
[1]Boston Scientific, Valencia, CA; [2]The Univ. of Sydney, Sydney, Australia; [3]Emeritus Professor, Rijksuniversiteit Groningen, Haren, Netherlands

Abstract: The midbrain PAG is a critical relay center of the emotional motor system and controls neural circuits that regulate breathing, blood pressure, bladder function, vocalization,

coughing, sneezing, vomiting and maintenance of abdominal and intrathoracic pressure. Dysfunction of PAG circuits lead to dysreflexia such as dyspnea, ataxic breathing, hyper & hypotension and micturition disruption (urinary incontinence). Circuit specific neuromodulation of the PAG could reverse specific types or combined dysfunctions paving way for development of neurologic clinical therapy. However, PAG circuit dynamics are unkown. For this reason, we stereotaxically mapped the PAG in the rat in vivo to investigate its neural architecture, circuit physiology and anatomical topography. The PAG was found to be predominantly quiescent in the resting state and could be activated by iontophoresis of excitatory amino acid glutamate agonists. Cells thus activated ceased function when glutamate ejection was terminated or by co-iontophoresis of muscimol (GABA agonist). Very few spontaneously active cells were found in the PAG, mainly in its dorsal region and these cells, typically fired in a slow and irregular pattern. Activation of either behavioral and/or emotional motor interventions caused immediate activation of PAG neurons mainly in the lateral and ventrolateral PAG, showing two distinct types of activity patterns; 1) single spike firing and 2) burst firing, The cells fired both tonically and phasically when correlated with specific motor output such as the diaphragm EMG. Predominantly the non-bursting PAG neurons had a near normal distribution around 200 to 250 msec, while burst-firing cells typically showing a bimodal distribution. The functional implications of PAG neuronal activity are discussed in terms of descending emotional motor control and effective translation for application of neuromodulation clinical therapy for specific emotional motor diseases. **Acknowledgement**This work was wholly undertaken in the laboratories of HHS-RJB at The University of Sydney and The University of Queensland with respective institutional ethics approvals. HHS, RJB and GH conceived and designed the projects, performed the experiments, analyzed primary data and made figure illustrations. GH curated and approved the final data/figure representation in this presentation/poster. None of the work described here were undertaken at either of the current work designations of Hari Subramanian or Gert Holstege.

Disclosures: H.H. Subramanian: None. **R.J. Balnave:** None. **G. Holstege:** None.

Poster

P686. Motivation: Social Communication and Behavior Across Species

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P686.01

Topic: G.03. Motivation

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Title: Hippocampal volume and intrinsic functional connectivity are associated with social comparison orientation of opinion, but not ability, in older adults

Authors: *H. SUGIMOTO, T. SEKIGUCHI, M. OTAKE-MATSUURA;
RIKEN, Tokyo, Japan

Abstract: This study aimed to examine whether cognitive function and brain states in older adults are associated with social comparison orientation (SCO), which refers to the tendency to compare oneself with others. To achieve this purpose, we collected multi-modal MRI data, including T1-weighted images, resting-state fMRI, and DTI, from ninety-seven community-dwelling older adults, as well as self-reported questionnaire data of the SCO scale that contains two subscales of opinion (e.g., "I often like to talk with others about mutual opinions and experiences") and ability (e.g., "I often compare myself with others with respect to what I have accomplished in life") and behavioral data of the Japanese version of the Montreal Cognitive Assessment (MoCA). All of the MRI analyses were conducted using the general linear model with a covariate of the SCO score (opinion or ability). Age, sex, and years of education were also included in the model as nuisance covariates (total intracranial volume was additionally entered into the model for VBM and TBSS analyses). Seven participants were excluded from all analyses, due to having probable arachnoid cyst, metal artifact, or meningioma. Thus, a total of ninety participants (55 females; age, mean ± SD = 73.5 ± 4.5 years; years of education, mean ± SD = 14.5 ± 2.2 years) were included in the analyses. Behavioral results showed that the MoCA score had a significantly positive correlation with the score of opinion ($r = 0.22$, $p < .05$), but not with that of ability ($r = -0.06$, $p = .56$). DARTEL-based VBM analysis demonstrated that the left hippocampal volume positively correlated with the score of opinion (ROI-based correction by FWE, $p < .05$), but not with that of ability. No significant correlation was observed in the right hippocampal volume. In the analysis for resting-state fMRI data, the functional connectivity between the left hippocampus as a seed region and the left angular gyrus, right ventromedial prefrontal cortex, and left posterior middle temporal gyrus showed a significantly positive correlation with the score of opinion (FWE, $p < .05$), but not with that of ability. Such a functional connectivity pattern was not found when we employed the right hippocampal seed. Finally, TBSS analysis for DTI data revealed that neither the score of opinion nor that of ability had significant correlation with the DTI metrics, including fractional anisotropy, mean diffusivity, axial diffusion, and radial diffusion. These findings suggest that older adults' general cognitive function and hippocampal structure and functional connectivity are modulated by the degree to which they compare their opinion with others, but not ability, in their daily lives.

Disclosures: H. Sugimoto: None. **T. Sekiguchi:** None. **M. Otake-Matsuura:** None.

Poster

P686. Motivation: Social Communication and Behavior Across Species

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Title: Bonding scenes trigger motivation for social touch

Authors: O. GRICHTCHOUK, J. M. OLIVEIRA, C. FRANKLIN, M. F. CORRREA, V. ROCHA-REGO, *E. VOLCHAN;
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Abstract: Proximity and interpersonal social contact are prominent components of social connection in human beings. Social interaction scenes were shown to facilitate the recruitment of pre-set cortical motor repertoires related to caress-like movements. Here, we investigated implicit predispositions for giving social touch associated with exposure to visual bonding stimuli. We explored the visuo-motor affective interplay at the final effector by monitoring the electromyographic activity of fingers flexor muscle in the absence of any overt movements. Photographs of dyads in social interaction attitudes and matched photographs of the same dyads not interacting were presented while electromyographic activity of fingers flexor muscle from the right hand was recorded. Exposure to dyads socially interacting significantly and sustainably increased participant electromyographic activity compared to exposure to dyads non-interacting, in the absence of any overt movement. When comparing each pair of matched pictures, the one depicting the interacting dyad consistently evoked higher electromyographic activity in comparison with the one depicting the same dyad non interacting. In the giver perspective, the flexion of fingers is a very important component of social touch. Here, using bonding visual priming and while participants were not making any overt movement, we were able to probe a motor output tightly implicated in the predisposition for social touch.

Disclosures: O. Grichtchouk: None. **J.M. Oliveira:** None. **C. Franklin:** None. **M.F. Corrrea:** None. **V. Rocha-Rego:** None. **E. Volchan:** None.

Poster

P686. Motivation: Social Communication and Behavior Across Species

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Topic: G.03. Motivation

Support: Spectrum Health- MSU Alliance
MSU-CRAIND Seed Funding

Title: Combined behavioral approaches for probing social motivation in young children

Authors: *B. THOMPSON[1], M. CHAVEZ[1], E. WOLFRUM[2], A. CARBAYO[1];
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Abstract: Background: Determining the neural mechanisms that underlie the heterogeneity of social-affective behaviors is challenging in developing children. The factors that influence the heterogeneity of these social behaviors in both typically developing (TD) children and children with Autism Spectrum Disorder (ASD), for whom disruptions in social behaviors are among the core deficits, are currently unknown. Objectives: We aimed to determine whether alterations in social behavior in children with ASD are driven by an aversion to, or lack of reward from, social interactions using a combination of remote eye-tracking techniques and detailed behavioral coding of social play. Methods: We adapted our previously established associative learning paradigm of conditioned place preference (CPP) in young children for use with a social unconditioned stimulus (US). In this paradigm, during an initial preference test (IPT), TD children (30-60 months) and children with ASD (36-66 months) freely explored two neutral rooms in our child-friendly castle. Then, across training trials, children alternated play between the two rooms while one of the rooms, the conditioned stimulus (CS), was paired with the US, a novel and interactive social experimenter. During the final preference test (FPT), children again freely explored both rooms, in absence of the US. Social CPP (sCPP) scores were calculated as the difference in time spent in the social-paired room during the IPT and FPT. Additionally, these children watched a 6-minute video comprised of social and non-social naturalistic scenes as well as cartoon animations. Eye movements were recorded using a Tobii Pro Spectrum eye-tracker and software. Advanced regressions were run to determine relationships between eye-tracking metrics and performance in the sCPP task. Results: We found significant heterogeneity in sCPP scores in both TD children and in children with ASD, with no differences in sCPP scores between groups. The two groups did demonstrate differences in gaze patterns across the video stimuli, though social scenes produced more frequent fixations in both groups. Preliminary statistical analyses revealed that eye-tracking metrics correlated with sCPP in these same children. Conclusions: This work provides a more comprehensive understanding of the mechanisms driving social motivation. The heterogeneity of sCPP scores provides greater insight into social motivation across young children and may help transform assessment and intervention approaches in young children.

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Poster

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Topic: G.03. Motivation

Support: NIMH Grant R01MH61285
NIMH Grant T32MH018931-30
NICHD Grant U54 HD090256

Title: Perceived social isolation, resting parasympathetic activity, and motivated behaviors

Authors: *K. E. SMITH, S. D. POLLAK;
Univ. of Wisconsin - Madison, Madison, WI

Abstract: A critical aspect of adaptive behavior is the ability to use cues in the environment to approach and avoid potential rewards and threats. Perceived social isolation has been theorized to change how individuals approach and avoid salient information. Specifically, acute social isolation is thought to act as a signal one lacks sufficient social relationships and increase approach motivations. However, findings in this area are conflicting. Parasympathetic cardiac regulation is associated with variability in approach and avoidance behaviors, with individuals with higher resting parasympathetic activity demonstrate more flexible adaptation to salient cues. Thus, the effects of perceived social isolation on approach and avoidance may be moderated by parasympathetic functioning. Here, we examined how parasympathetic functioning and perceptions of social isolation influence approach and avoidance behaviors. We collected measures of resting parasympathetic activity and perceptions of social isolation from 74 adults (18 - 46 years). Participants underwent an instrumental conditioning paradigm assessing how they approach and avoid rewards and threats. Higher levels of perceived social isolation were associated with increased approach behaviors. This was most pronounced in individuals with higher resting parasympathetic activity. In contrast, individuals with lower resting parasympathetic activity demonstrated increased avoidance behaviors. In sum, these findings suggest that social isolation does shift motivational behaviors. Additionally, individuals with higher resting parasympathetic activity may demonstrate more adaptive changes in motivation linked to perceived social isolation.

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Poster

P686. Motivation: Social Communication and Behavior Across Species

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Topic: G.03. Motivation

Support: Language in Interaction, financed by the Netherlands Organisation for Scientific Research (NWO)

Title: Tracing the cognitive sources of communicative challenges in Autism Spectrum Disorder

Authors: K. CAI[1], S. KOCH[1], M. MANGNUS[1], I. TONI[1], J. BASNAKOVA[1], *A. STOLK[2]; [1]Donders Inst., Nijmegen, Netherlands; [2]Dartmouth Col., Hanover, NH

Abstract: Autism Spectrum Disorder (ASD) is diagnosed on the basis of communicative challenges experienced in everyday interactions, yet the cognitive sources of those challenges remain largely unknown. A popular suggestion holds that autistic individuals struggle to predict other people's behaviors, either due to bottlenecks in bottom-up sensory processing or attenuated sensitivity to top-down priors when interpreting communicative behaviors (Pellicano & Burr, 2012). However, studies of predictive processing have produced mixed results in the perceptual domain, and few studies to date have investigated ASD predictive capabilities during live social interaction.

This eye-tracking study aims to assess bottom-up and top-down contributions to neurotypical and autistic interpretations of genuinely interactive communicative behaviors. Predictive abilities are assessed while participants solve a series of coordination problems in the two-player "tacit communication game" (Wadge et al., 2019). In each round of this game, players have to jointly reproduce a target configuration of their two given shapes on a digital game board. However, the target configuration is shown to one of the players only, the Sender. Given that the other player, the Receiver, cannot see the target configuration, the Sender needs to move her shape in a way that conveys the location and orientation of the Receiver's shape. As when interpreting everyday ambiguous words and gestures, the Receiver needs to rapidly identify relevant features from the Sender's movements to infer her target position. A benefit of the novel communicative medium is that it offers the possibility to manipulate access to top-down priors during communicative interpretation by introducing coordination problems that are more easily solved in light of previous interactions. Furthermore, the computer-controlled interactions allow for a temporally precise overlay of eye gaze and communicative behavior to determine any bottlenecks in bottom-up sensory processing.

Preliminary results (25 neurotypical and 19 ASD pairs) suggest that the communicative challenges experienced by autistic individuals cannot be attributed solely to limitations in bottom-up processing, as seen in matched durations of task-relevant fixations between neurotypical and ASD participants. Second, autistic and neurotypical individuals' eye gaze behaviors exhibited a similar susceptibility to top-down priors established during previous interactions. These findings open the way to investigate the cognitive mechanisms that integrate bottom-up and top-down processes to produce accurate inferences about other people's behavior in ASD.

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Poster

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Program #/Poster #: P686.06

Topic: G.03. Motivation

Support: UMD VFIN
NINDS 1 R01 NS104008-01

Title: Influence of HVC Dopamine Receptor Subtypes on Song Production in Canaries

Authors: *C. M. HAAKENSON[1], A. E. MARQUARDT[2], J. W. VANRYZIN[3], M. M. MCCARTHY[2], G. F. BALL[4];
[1]Univ. of Maryland, Col. Park, College Park, MD; [2]Univ. of Maryland Sch. of Med., Baltimore, MD; [3]Univ. of Maryland, Baltimore, Baltimore, MD; [4]Univ. of Maryland, College Park, MD

Abstract: Communication is essential for social species that rely on coordinated behavior for survival and reproduction, however relatively little is known about the neural mechanisms underlying the social and motivational aspects of this behavior. We examined a species that uses vocal communication to relay social information, the canary (*Serinus canaria*), and a potential modulator of the motivation to sing - the neurotransmitter dopamine. There is evidence that dopamine in some song nuclei can influence the amount of singing directed at potential mates. In addition, activation of dopamine type 1 receptors (D1R) can have different electrophysiological effects than activation of dopamine type 2 receptors (D2R). Yet it is currently unknown how differences in the effects of D1R versus D2R action manifest in singing behavior. Therefore, in order to investigate the effects of dopamine receptor activation on song, we implanted guide cannula targeting the song control nucleus HVC. We injected drugs to both activate and inhibit dopamine receptors in this region. Each bird received injections of a D1R agonist, a D1R antagonist, a D2R agonist, a D2R antagonist, and saline with a 48-hour washout period between treatments. From songs recorded following injections, we quantified the latency to initiate singing and the quality of songs produced in the hour following drug injection. In addition, we applied a novel computational approach to identify changes in song structure following treatment – employing new dimensionality reduction techniques, density-based spatial clustering, and network diagrams. This experiment indicates that D1 receptor activation in HVC increases the latency to sing. Furthermore, song following D1 receptor agonist treatment had a reduction in three particular syllable types, while these syllable types were increased in song following D2 receptor agonist and antagonist treatment. In addition, we performed RNAscope in situ hybridization in order to characterize the cells responsible for these effects. Brains were collected 30 minutes following song and sections containing HVC were labeled with a probe for the immediate early gene Egr1, probes for dopamine receptor subtypes (D1R, D2R or D3R), a probe for GAD2 to identify inhibitory cells, and a probe for VGLUT2 to identify excitatory cells. We found that the majority of cells that expressed dopamine receptors were excitatory and a large proportion were active during singing. These results indicate that activation of dopamine

receptor types have profoundly different effects on song production, potentially modulating the type of vocalization produced to be appropriate for the social context.

Disclosures: C.M. Haakenson: None. **A.E. Marquardt:** None. **J.W. Vanryzin:** None. **M.M. McCarthy:** None. **G.F. Ball:** None.

Poster

P686. Motivation: Social Communication and Behavior Across Species

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Topic: G.03. Motivation

Support: NIMH R21-111998

Title: Molecular underpinnings of paternal behavior in prairie voles

Authors: *N. WADDELL[1], J. M. CHITAMAN[1], Y. LIU[2], Z. WANG[2], J. FENG[1];
[1]Biol. Sci., [2]Psychology, Florida State Univ., Tallahassee, FL

Abstract: Throughout the animal kingdom most species are polygamous, yet some form social bonds with sexual partners, resembling a monogamous social relationship. Prairie voles form such strong social bonds during mating, with both father and mother showing rearing behaviors to their offspring and aggression to stranger individuals. In addition, before pup-exposure, virgin males also present split behavioral phenotypes, where some of these unbonded males demonstrate parental behavior to novel pups (i.e. “virgin paternal males”) and the rest display aggression (i.e. “virgin aggressive males”). To explore the molecular underpinnings of these biphasic paternal behavioral phenotypes of virgin male prairie voles, we investigate the transcriptome and DNA methylome in hippocampal dentate gyrus (DG) through RNAseq and RRBS, respectively. Within the four groups of adult animals (virgin aggressive males, virgin paternal males, fathers, mothers), we found the transcriptome was generally up-regulated in virgin aggressive males when compared to virgin paternal males. Interestingly many of these affected genes were similarly changed when virgin aggressive males are compared to fathers after pup exposure. For DNA methylation, we recognized comparable numbers of hypermethylated and hypomethylated CpG sites across the genome with similar genomic feature distributions when comparing aggressive and paternal virgin males. We then intersected the RNAseq and RRBS datasets and identified several significant relationships. Profiling gene ontology pathways, a number of cell signaling and neurogenesis related pathways are overrepresented. Taken together, we found widespread transcriptome and methylome changes in hippocampus that are selectively enriched within certain signaling pathways that may contribute to the paternal behavioral phenotype in male prairie voles.

Disclosures: N. Waddell: None. **J.M. Chitaman:** None. **Y. Liu:** None. **Z. Wang:** None. **J. Feng:** None.

Poster

P686. Motivation: Social Communication and Behavior Across Species

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P686.08

Topic: G.03. Motivation

Support: NIDA-IRP

Title: Waving through a window: a model of volitional social interaction in female mice

Authors: *F. HOLLOMAN[1], B. T. HOPE[1], Y. SHAHAM[1], M. VENNIRO[2], L. A. RAMSEY[1]; [1]IRP/NIDA/NIH, Baltimore, MD; [2]Anat. and Neurobio., Univ. of Maryland Sch. of Med., Baltimore, MD

Abstract: Background: Mouse models of social behavior fail to account for volitional aspects of social interaction and current neurobiological investigation of social behavior is performed almost exclusively using C57BL/6J mice, the background strain of most transgenic mice. Here, we introduce a mouse model of operant social self-administration and choice, using a custom-made apparatus. **Methods:** First, we trained adolescent and adult female C57BL/6J and CD1 mice to self-administer palatable food pellets and then to lever-press under increasing fixed-ratio response requirements for access to a female social partner. Next, we tested their motivation to seek social interaction using a progressive ratio reinforcement schedule, relapse to social seeking after social isolation, and choice between food versus social interaction. We also tested social conditioned place preference (CPP) in adult female CD1 and C57BL/6J mice. **Results:** Adolescent and adult female mice of both strains showed similar rates of food self-administration. In contrast, CD1 mice demonstrated significantly stronger social self-administration than C57BL/6J mice under both reinforcement schedules. CD1 but not C57BL/6J mice demonstrated robust social seeking after social isolation. In the choice task, CD1 mice strongly preferred social interaction, whereas C57 mice strongly preferred food. CD1 but not C57BL/6J mice demonstrated robust social CPP. The strain differences were age-independent. **Conclusion:** Our data demonstrate that CD1 mice are a better strain for studying female social reward learning. Our mouse operant social model provides a tool for research on neurobiological substrates of female social reward, and disruption of social reward psychiatric disorders, using mice-specific genetic tools.

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Poster

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Program #/Poster #: P686.09

Topic: G.03. Motivation

Support: CIHR Grant 102722

Title: The role of ghrelin in promoting the motivation for social interaction through AGRP neurons in mice

Authors: *A. ALFTIEH, B. AL-TAHIR, K. JOY, L. HYLAND, A. ABIZAID;
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Abstract: Ghrelin is a stomach-derived peptide that increases food intake by acting as an endogenous ligand for growth hormone secretagogue receptor (GHSR). GHSR is abundantly expressed in the brain with high expression found in agouti-related peptide (AGRP) and neuropeptide-Y (NPY) neurons within the hypothalamic arcuate (ARC) nucleus. Recent data suggests that, in addition to increasing food intake, ghrelin facilitates social interaction whereas ghrelin receptor antagonists increase social vigilance. In this present study, we investigated the effects of ghrelin on the motivation for social interaction in mice and the role of AGRP neurons in mediating these effects. Sixty-three male and female C57BL/6J mice were assigned to groups receiving intraperitoneal (IP) injections of saline or ghrelin (10ug of ghrelin in .1cc of isotonic saline) followed by injections of saline or the MC3/4 receptor agonist melanotan-II (1mg/kg of MTII in .1cc of isotonic saline) which competes with AGRP, an endogenous antagonist to the MC3/4 receptors. Thirty minutes later, mice were tested on a social interaction test to investigate how these drug treatments affected social behaviors. Results showed that, regardless of sex, ghrelin injected peripherally significantly decreased the latency to approach a conspecific and the frequency of corner observation, while increasing the total duration of social interaction. In contrast, IP injections of MTII alone resulted in a significant increase in the latency to approach a conspecific and the frequency of corner observations, while decreasing the total duration of social interaction. Mice that were given MTII and ghrelin responded like saline treated animals, and did not show ghrelin induced pro-social behaviors. Taken together, the results suggest that ghrelin promotes social interaction, and blocking the ability of AGRP from binding to its receptor attenuates these effects.

Disclosures: A. Alftieh: None. **B. Al-Tahir:** None. **K. Joy:** None. **L. Hyland:** None. **A. Abizaid:** None.

Poster

P686. Motivation: Social Communication and Behavior Across Species

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Program #/Poster #: P686.10

Topic: G.03. Motivation

Title: Comparison of the Behavioral Effects of Chronic Intranasal Administration and Intraperitoneal Injection of Oxytocin on Male and Female Mice

Authors: M. MEADOWS, H. HUDSON, L. PAUL, I. RAMOS, *T. ROGERS;
Middle Tennessee State Univ., Murfreesboro, TN

Abstract: Comparison of the Behavioral Effects of Chronic Intranasal Administration and Intraperitoneal Injection of Oxytocin on Male and Female Mice
AuthorsM. MEADOWS[1], H. HUDSON[1], L. PAUL[1], I. RAMOS[1], ***T. D. ROGERS**[1];[1]Middle Tennessee State Univ., Murfreesboro, TN**Disclosures T.D. Rogers:** None. **M. Meadows:** None. **H. Hudson:** None. **L. Paul:** None.
While the effect of oxytocin (OT) on social behavior and anxiety-like behavior is commonly studied in animal models, differing routes of administration between studies potentially limit generalizability. The current study compared the behavioral effects of chronic intranasal administration and chronic intraperitoneal injection of the same dose of OT in male and female mice. Adult C57BL/6J mice were chronically pretreated with saline or OT (12 ug) for 14 consecutive days. Mice received either intranasal (12 ml, 6 ml per nostril) or intraperitoneal (120 ml) administration. On day fourteen, mice were tested in the elevated plus maze (EPM) to determine differences in anxiety-like behaviors. Twenty-four hours after EPM, mice were tested in the three-chamber test, commonly used to test sociability and preference for social novelty. Twenty-four hours after the three-chamber test, mice were placed in an open field arena measuring 12”x12” with a sex-matched conspecific for ten minutes to measure social behavior in a free dyadic social interaction (FDSI). Analysis of the EPM behavior indicated a significant three-way interaction (drug x route x sex) in which OT administration reduced open arm exploration only in females administered OT via intraperitoneal injection suggesting increased general anxiety-like behavior in this group ($F(1, 57) = 14.42$, $p = 0.0004$). In the three-chamber, mice that received either OT or saline via intraperitoneal injection displayed less social sniffing ($F(1, 60) = 7.93$, $p = 0.007$) and less preference for social novelty ($F(1, 59) = 5.06$, $p = 0.028$). In the FDSI, a significant main effect for the type of drug administered indicated that OT increased social approach ($F(1, 58) = 7.49$, $p = 0.008$) and reduced social anxiety-like behaviors ($F(1, 58) = 40.96$, $p < 0.0001$) compared to saline. A main effect for route of administration indicated that mice receiving intraperitoneal injections displayed more social approach than mice receiving intranasal administration ($F(1, 58) = 7.95$, $p = 0.0066$). Our findings suggest that the route by which OT is administered may affect social and anxiety-like behaviors in mice. Further these differences may be sex specific. The findings highlight the need to identify potential limitations of generalizability between studies that use varying routes of administration of OT.

Disclosures: M. Meadows: None. **H. Hudson:** None. **L. Paul:** None. **I. Ramos:** None. **T. Rogers:** None.

Poster

P690. Human Emotion I

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Program #/Poster #: P690.01

Topic: G.04. Emotion

Support: Japan Society for the Promotion of Science Grant 18K03171

Title: An fMRI study examining the role of the extended amygdala in negative emotion and inhibitory control in second language learners.

Authors: M. SUGITA-MCEOWN[1], M. MAKUUCHI[2], T. NAOE[2], J. ELLINGER[3], ***K. MCEOWN**[1];
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Abstract: The current studyinvestigated the role of the extended amygdala in negative emotion andinhibitory control in second language learners. fMRI scans, using a 3T MRIscanner, were used to measure neurophysiological responses to neutral and negativeemotional linguistic stimuli presented in participants' native language (i.e.,Japanese) or in a second language (i.e., English) using a modified version ofthe emotional-linguistic Go-No Go task. Data were collected from 12 healthyyoung adults (9 female; mean age = 20.3 years) who were attending a privateuniversity in Japan and learning English as a second language. The right extendedamygdala (coordinates: 21, 6, -12), right amygdala (18, -6, -18), left extendedamygdala (-18, 6, -9) and left amygdala (-24, -7, -17) were selected as regionsof interest for the current study. The region of interest of each participantwas defined as a 6mm radius sphere centered at these coordinates. We found thatleft amygdala activation was significantly lower in the English neutral wordNo-Go condition compared to the Japanese neutral word No-Go condition. Furthermore,as predicted, right extended amygdala activation was significantly lower in theEnglish negative word No-Go condition compared to the Japanese negative word No-Gocondition. These findings suggest that brain regions (i.e., the extendedamygdala) associated with negative affect are significantly less active whenparticipants are performing a task that requires inhibitory control and are exposedto negative emotional linguistic stimuli in a second language compared to when theyperform an inhibitory control task and are exposed to negative emotional linguisticstimuli in their native language.

Disclosures: M. Sugita-McEown: None. **M. Makuuchi:** None. **T. Naoe:** None. **J. Ellinger:** None. **K. McEown:** None.

Poster

P690. Human Emotion I

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Program #/Poster #: P690.02

Topic: G.04. Emotion

Support: Start-up funds from the UCLA Psychology Department

Title: Multivoxel Patterns of Human Emotion Regulation Neurodevelopment

Authors: *J. GUASSI MOREIRA, A. MÉNDEZ LEAL, N. SARAGOSA-HARRIS, Y. WAIZMAN, E. NINOVA, J. SILVERS;
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Abstract: The acquisition of emotion regulation—a critical process implicated in mental health, achievement, and every day functioning—is an inherently developmental process that occurs over the course of decades. To date, much research has examined the neurodevelopmental bases of human emotion regulation using univariate methods. That is, this work predominantly focuses on using fMRI to chart the neurodevelopment of mean brain activity in individual regions of interest, or in whole-brain univariate contrasts. By comparison, relatively little work has leveraged multivoxel analytic techniques that are prevalent in other arenas of human neuroimaging. These techniques are powerful because they consider how *patterns* of brain activity—comprised of suites of voxels responses in fMRI—meaningfully encode information about psychological phenomena of interest. In this study, we leverage multivariate pattern analyses to better understand the neurodevelopment of emotion regulation. A sample of 50 youths (ages 10-22) completed four runs of an emotion regulation task while undergoing fMRI. On a trial-by-trial basis, youths were asked to either cognitively downregulate emotional responses to a series of negative stimuli, or to emotionally react to negative stimuli (as an emotional responding baseline). After data preprocessing, we examined how emotion regulation activity was encoded in multivoxel patterns using several different analyses. While controlling for age, we first investigated whether we could decode task conditions on the basis of multivoxel patterns. Next, we examined age-related effects neural representations of emotion regulation using both standard multivoxel pattern analysis techniques as well as feature encoding models. Finally, we examined whether an analytically derived neural signature of emotion regulation from our sample matched canonical signatures of adult regulation. While analyses remain ongoing, early results broadly suggest age appears to be meaningfully encoded in multivoxel representations of emotion regulation.

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Poster

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Topic: G.04. Emotion

Support: NSF Grant No.1908299
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UF AI Catalyst Fund
University of Florida Informatics Institute Graduate Fellowship

Title: A deep neural network model for emotion perception

Authors: *P. LIU[1], K. BO[3], L. CUI[1], Y. CHEN[1], C. T. TRAN[2], R. FANG[1], M. DING[1];
[1]Dept. of Biomed. Engin., [2]Dept. of Electrical and Computer Engin., Univ. of Florida, Gainesville, FL; [3]Dept. of Psychological and Brain Sci., Dartmouth Col., Hanover, NH

Abstract: Deep neural networks (DNNs) have shown to be excellent models of visual object recognition in the human and the nonhuman primate brain. Recent work has begun to explore the capability of DNNs as models of more complex cognitive constructs such as memory and emotion. We have developed a deep neural network model for perceiving emotions in affective images that includes both a visual cortex module and an amygdala/orbital frontal cortex (OFC) module. Specifically, the visual cortex module consisted of a VGG16 network pre-trained on ImageNet data, and the amygdala/OFC module consisted of three fully connected layers that receive input from the last fully connected layer of VGG16 as well as the early layers, with the latter mirroring the inferior longitudinal fasciculus (ILF) connection between the occipital and anterior temporal cortex. The model was trained to predict the valence from affective images (training data) and tested against the normative valence ratings of similar images (testing data). Four datasets were considered: the International Affective Picture System (IAPS), the Nencki Affective Picture System (NAPS), the dataset from In-Kwon Lee, and the static frames of the emotion video dataset from Alan S. Cowen. The activities of the amygdala/OFC module of the fully trained model were also compared with fMRI data recorded from human subjects viewing IAPS images using the representational similarity analysis (RSA). The results show that (1) the model was able to predict valence with high fidelity ($R>0.6$) for all four datasets, (2) the model's performance was considerably worse without the "ILF" mediated pathway, and (3) activities in the second-to-last and last layer of the amygdala/OFC module were comparable with activities in the human amygdala and OFC respectively. These results demonstrated that our DNN model has the capability to recognize the emotional content of affective images and can become a platform for formulating and testing neuroscientific hypotheses.

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Poster

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Program #/Poster #: P690.04

Topic: G.04. Emotion

Support: NIH Grant NIMH111640
NSF Career Award (Neta)

Title: Reappraisal-related downregulation of amygdala BOLD activation occurs only during explicit post-stimulus evaluation

Authors: *J. PIERCE[1], J. R. BLAIR[2], K. CLARK[1], M. NETA[1];
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Abstract: During cognitive reappraisal, an individual reinterprets the meaning of an emotional stimulus, for example, to make a disturbing image seem less threatening and thus regulate the intensity of their emotional response. Prefrontal cortex activity has been found to support reappraisal and is putatively thought to downregulate the amygdala response to these stimuli. The timing of these regulation-related responses during the course of a trial, however, remains poorly understood. In the current fMRI study, participants were instructed to view or reappraise negative images and then rate how negative they felt following the presentation of each image. The hemodynamic response function was estimated in 11 a priori regions of interest for the entire time course of the trial including image viewing and rating. An analysis of the reappraisal-related activity during early and late windows of the trial indicated that although some prefrontal regions (e.g., inferior frontal gyrus) had increased activation in the early window, others (e.g., middle frontal gyrus) had increased activation primarily in the late window. Notably, the amygdala showed no evidence of downregulation in the early window, but did show such evidence in the late window, which also correlated with a measure of behavioral reappraisal success. These results highlight the importance of examining the time course of neural responses during cognitive reappraisal, as different brain regions respond to different subunits of the trial, and this pattern of results provide some insight into the underlying cognitive and affective processes.

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Poster

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Program #/Poster #: P690.05

Topic: G.04. Emotion

Support: Kids Kicking Cancer
D. Dan and Betty Kahn Foundation

Title: Greater mindfulness is associated with lower internalizing, externalizing, and attention problems in elementary school children

Authors: *P. MECHNIKOV[1,2], B. BORG[2], H. MARUSAK[2];
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Abstract: Mindfulness-based interventions are increasingly implemented in school settings. However, empirical research on mindfulness-based interventions lag behind their widespread implementation. Here, we tested whether greater mindfulness is associated with lower internalizing, externalizing, and attention problems in elementary school children. This study included a total of 310 students (grades 2-4; 47% female, 68% Black American, 15% White, 17% Other) recruited from two public suburban schools with the same district and one nearby private school. Mindfulness was measured using the Child and Adolescent Mindfulness Measure. Internalizing (e.g., anxiety, depression), externalizing (e.g., conduct, oppositional defiance), and attention (e.g., ADHD) problems were measured by the Pediatric Symptom Checklist. Overall, greater mindfulness was associated with lower internalizing, externalizing, and attention problem behaviors. These results remained significant when controlling for gender, grade, and district. Overall boys reported more externalizing problems than girls and the association between mindfulness and externalizing symptoms was moderated by gender such that boys showed greater reductions in externalizing symptoms with higher mindfulness as compared to girls. There were no other interactive effects with gender, race/ethnicity, district, or grade. These results demonstrate the potential benefits of mindfulness for reducing internalizing, externalizing, and attention problem behaviors among diverse students from public and private schools at the elementary school level. Boys, in particular, may benefit from mindfulness-based school interventions to reduce externalizing problems. School-based programs that cultivate mindfulness may be beneficial for improving student health and outcome (e.g., academic performance).

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Poster

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Topic: G.04. Emotion

Support: H2020 / Marie Skłodowska-Curie-813234

Title: The functional connectome for emotional responses- an EEG analysis

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Abstract: The functional connectome for emotional responses- an EEG analysis
Background: Recently, the field of neuroimaging has been moving from "brain mapping" which focused on brain regions, to a view of the brain as a densely interconnected network. In this view, it is the "network code" of a particular function that is key, rather than any singular structure. The network code for critical functions such as emotional responses are currently being studied. In the study of emotions, there is also an ongoing discussion about the nature of emotions, and whether there are any classes of emotions that can be robustly identified across individual differences. Here, brain connectivity analyses to different emotional responses are thought of as particularly challenging. In this study, we report the results from a connectivity analysis using EEG to different emotional responses. **Method:** 57 participants exposed to videos with six types of emotional content, while their brain responses recorded using a 9-channel EEG (Fz, F3, F4, Cz, C3, C4, POz, P3, P4). The EEG data were pre-processed using band-pass Butterworth filter, re-referencing, removing bad epochs, and ICA. In analyzing the data, we used Imagery Coherence (Icoherence), to test for significant differences among emotions (sadness, happiness, neutral, anger, fear, and amusement). We applied an ANOVA test for 36 pairs of channels using different frequency ranges (Theta, Alpha, Beta, Gamma), to test which pairs of electrodes were discriminatory regarding their Icoherence values. The ANOVA table constructed via 57 observations for 6 types of emotion. In overall, 36 ANOVA were applied. A post-hoc pairwise analysis was run to test which types of emotions were different on the Icoherence value of that pair. **Result:** In the alpha band, there were 7 significant different values (p-value < 0.05); C3-Fz: AMU-HPY, F4-F3: HPY-NEU, P3-Fz: ANGR-HPY, P4-P0z: Fear-NEU, P4-C4: HPY-NEU, Fear-NEU, C4-Fz: HPY-NEU, and in beta band; F4-F3: HPY-NEU, C4-C3: Sad-NEU, F3-C4: AMU-NEU, P4-P3: AMU-HPY, AMU-NEU. There were no significant differences in theta and gamma. **Conclusion:** These results indicate that most of the unique features of functional brain connectivity during emotional responses occur in the alpha band. Moreover, these outcomes suggest that phase connectivity features can be discriminative for positive emotions (happy and amusement). This could mean that phase connectivity tracks both the arousal and valence level of emotion. However, since none of the values were significantly different among negative – negative comparison, this implies cross frequency features should be consider to produce better discriminatory features.

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Poster

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Program #/Poster #: P690.07

Topic: G.04. Emotion

Title: The Affective Neuroimaging Collaboratory (ANiC): Towards a better understanding of the neural architecture of affective processes

Authors: *M. KWON[1], S. VAN 'T HOF[2], R. BOTVINIK-NEZER[1], M. AMINI, C. BANGO, B. HUNT, E. MURPHY[1], D. CHRISTIANO[4], C. G. DAVEY[5], M. FULLANA[6,7], B. J. HARRISON[5], O. HARRISON[8,9,10], A. J. JAMIESON[5], S. KLEIN[11], B. D. KNUTSON[4], L. KOBAN[12], H. KOBER[14], T. B. LONSDORF[15], I. MARTÍNEZ-ZALACAÍN[16,17], C. MERZ[18], L. MORTAZAVI[4], K. PATTINSON[9], H. PLASSMANN[19,13], D. SCHILLER[20], A. J. SHACKMAN[21], D. SMALL[22], C. SORIANO-MAS[16,23], R. STARK[11,24], J. SUNG[25], B. VERVLIET[3], P. A. KRAGEL[26], L. VAN OUDENHOVE[27,28], T. D. WAGER[1];
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Giessen, Giessen, Germany; [25]Dept. of Psychiatry, Div. of Nutritional Psychiatry, Yale Univ. Sch. of Medicine; Modern Diet and Physiol. Res. Ctr., New Haven, CT; [26]Dept. of Psychology; Dept. of Psychiatry and Behavioral Sci., Emory Univ., Atlanta, GA; [27]Lab. for Brain-Gut Axis Studies, Translational Res. Ctr. for Gastrointestinal Disorders, Leuven, Belgium; [28]Leuven Brain Inst., Leuven, Belgium

Abstract: In spite of decades of research on the brain bases of affective and motivational processes, we still have much to learn about the organization of brain systems underlying them. Previous studies are limited in sample size and number of affective tasks. Although a number of meta-analyses have attempted to overcome these issues by aggregating findings across multiple studies, this effort has primarily been focused on finding reliable 'hot spots' in the brain using brain coordinates from published papers and thresholded results. Establishing the generalizability, sensitivity, and specificity of brain measures for different categories of affective processes, however, requires a more complete understanding of the underlying neural architecture informed by maps with information across all brain voxels at the individual level. This can be accomplished with collaboration across multiple laboratories by utilizing data from multiple studies. Here, we introduce the Affective Neuroimaging Collaboratory (ANiC), a large-scale initiative that provides a platform to aggregate and analyze individual subject-level data across affective and motivational tasks, studies, and research groups worldwide. Our goals are threefold: 1) curate a quality-controlled data resource; 2) make data available to the consortium and subsequently the public, with contributing researcher-determined access; and 3) work collaboratively to publish articles to advance our understanding of the organization of affect and motivation in the brain. Among a number of potential outputs, we are envisioning developing brain measures that are sensitive to, and specific for particular domains of affect and motivation; evaluating which ontological structures are best reflected in the brain; providing mechanistic brain targets for translational use; and contributing to the development of a biologically-grounded understanding of brain-based disorders. As proof of concept, we compiled data from 18 studies, across three domains (pain, cognitive control, negative emotion) and nine subdomains (three modalities per domain) with a total of 270 participants (15 sampled per study), and tested if single sub-regions in the insula integrate multimodal affective processes. We found that specific insular sub-regions respond to different types of affective domains and subdomains. These results challenge the concept of a single integrative center across types of affect in the insula and provide evidence for an organizational scheme grounded in sensory systems where each insular sub-region processes different affective information characterized by different sensory modalities.

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Poster

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Topic: G.04. Emotion

Support: NIH R01MH107513
NIH R01MH119430
NIH R01MH111629
NIH R37HD090153

Title: Face and Emotion-Sensing in right Temporal Parietal Junction: A Dyadic Complex for Emotion Contagion

Authors: *J. HIRSCH[1,2,3,4,5], A. BHATTACHARYA[6], X. ZHANG[1], J. A. NOAH[1]; [1]Psychiatry, [2]Neurosci., [3]Comparative Med., Yale Sch. of Med., New Haven, CT; [4]Haskins Labs., New Haven, CT; [5]Dept. of Med. Physics and Biomed. Engin., Univ. Col. London, New Haven, CT; [6]Yale Col., Yale Univ., New Haven, CT

Abstract: How does the human face convey emotion to another human? Although emotional contagion is a primitive and foundational feature of social interactions, the underlying mechanisms are not well understood. Here we apply a dyadic approach to investigate the mechanisms of emotional contagion using functional near infrared spectroscopy hyperscanning. One participant, "Video-Watcher", watched emotionally provocative silent videos and the other partner, "Face-Watcher", saw only the face of the Video-Watcher, Fig 1 A. Ratings of the emotional experience (dial rotation) were acquired simultaneously for both participants at the end of each 5 sec video. Facial recognition cameras used Open Face to record Action Units of facial expressions. Partners sat across a table from each other and alternated roles as "Video-or Face-Watcher". The library of videos included "neutrals" featuring landscapes, "adorables", featuring animal antics, and "creepy crawlies" featuring spiders, worms and states of decay. All were pre-tested for emotive properties. The "Video-Watcher" rated his/her emotional feeling for each video. The "face watcher" rated his/her emotional feeling for each expression of the "Video-Watcher's" face. The correlation of emotional ratings between the "Video and Face-Watchers" ($r = 0.60$) confirmed that the emotion was transmitted by facial expressions and taken as a measure of emotional contagion. Facial Action Units of the "Video Watcher's" face were taken as the stimuli for the "Face- Watcher", and the largest units, "Lip Corner Puller" and "Dimpler Buccinator" were combined and used as a regressor on the "Face-Watcher's" neural responses. A complex of right TPJ regions consisting of supramarginal gyrus, angular gyrus, and superior temporal gyrus) was found to be responsive to these dynamic emotional expressions ($p < 0.05$, FDR corrected peak voxel, Fig 1B) and consistent with an emotion-sensing function.

Findings suggest a dyadic model of emotion contagion and face sensing where brains and faces are tuned to send and receive emotional states via facial cues.

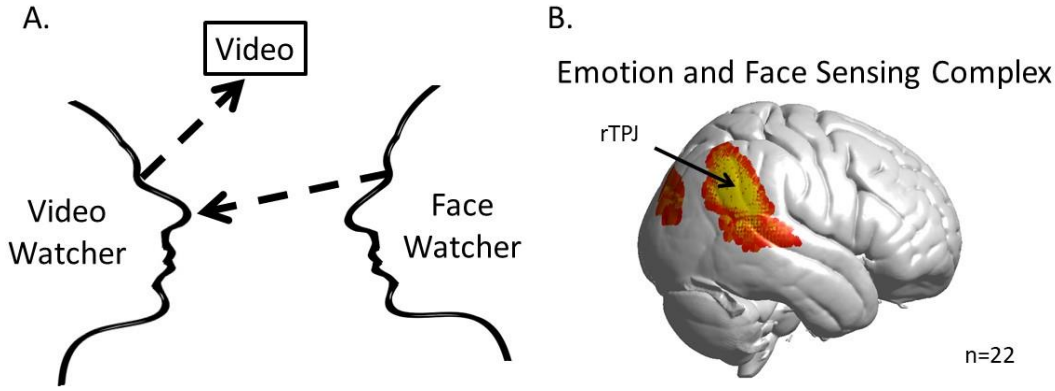


Fig 1. The "Face-Watcher's" brain activity was regressed with the "Video-Watcher's" Facial Action Units and reveals a rTPJ complex associated with emotion and face sensing.

Disclosures: J. Hirsch: None. **A. Bhattacharya:** None. **X. Zhang:** None. **J.A. Noah:** None.

Poster

P690. Human Emotion I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P690.09

Topic: G.04. Emotion

Support: MH112558

Title: Role of the Pulvinar in Affective Scene Processing

Authors: *L. CUI[1], Y. LIANG[1], K. BO[3], A. KEIL[2], M. DING[4];
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Abstract: The pulvinar is the largest nucleus of the thalamus. The proposed functions of the pulvinar range from working memory to attention to emotion. In this study we examined the role of the pulvinar in affective scene processing. Simultaneous EEG-fMRI data were recorded from

20 participants viewing pleasant (20), unpleasant (20), and neutral (20) scenes from the International Affective Picture System (IAPS). Analyzing single-trial BOLD responses using machine learning decoding techniques, we found that linear methods such as SVM were not able to detect the differences in neural representations of the three categories of affective scenes in the pulvinar, whereas a deep neural network (DNN) based model was able to do so with a decoding accuracy of 42%, which is significantly higher than the chance level of 33%. A weight map analysis revealed the particular importance of the medial pulvinar in contributing to the DNN decoding performance. Furthermore, fusing EEG-fMRI data via representational similarity analysis (RSA), we compared the temporal profiles of emotion-related activation in the amygdala and in the pulvinar and observed that emotion-related activation of the amygdala preceded that of the pulvinar. These results demonstrated that the pulvinar contributes to the processing of affective scenes and its distinct neural representations of the different categories of affective scenes may come from reentry from the amygdala.

Disclosures: L. Cui: None. **Y. Liang:** None. **K. Bo:** None. **A. Keil:** None. **M. Ding:** None.

Poster

P690. Human Emotion I

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Program #/Poster #: P690.10

Topic: G.04. Emotion

Support: NIH Grant R01DA046064
NIH Grant R01DA035484

Title: Neural architecture of negative affect: common and stimulus type-specific representations

Authors: *M. CEKO[1], P. A. KRAGEL[2], C.-W. WOO[3], M. LOPEZ-SOLA[4], T. D. WAGER[5]; [1]Inst. of Cognitive Sci., Boulder, CO; [2]Emory Univ., Atlanta, GA; [3]Ctr. for Neurosci. Imaging Res., Sungkyunkwan Univ., Suwon-Si, Korea, Republic of; [4]Univ. of Barcelona, Barcelona, Spain; [5]Psychology and Neurosci., Dartmouth Col., Hanover, NH

Abstract: How is negative affect encoded in the brain? Prior studies have provided evidence for a common neural representation and for distinct representations for different types of negative affect (i.e. induced by different stimulus types), but have largely focused on these accounts separately without direct comparisons. Here we combined a multimodal experimental paradigm and an analytic approach designed to test the hypothesis that brain representations of negative affect are conveyed both by common and stimulus type-specific representations. In Study 1 (N = 55 healthy participants) we collected fMRI responses to mechanical pain, thermal pain, aversive sounds, and aversive images of four intensities each, rated on a common aversiveness scale to facilitate direct comparisons. Brain models were developed using Partial Least Squares

Regression which allowed us to simultaneously predict common and stimulus type-specific effects. Brain models tracked negative affect and not general salience or arousal. Early sensory input areas largely predicted stimulus-type specific aversiveness, with insula and somatosensory cortex predicting pain, auditory cortex predicting auditory aversiveness and visual cortex predicting visual aversiveness. The common model of aversiveness was associated with a set of core affect areas, including medial and orbital prefrontal areas, amygdala, anterior insula and limbic forebrain. Some core affect areas, particularly the midcingulate cortex, anterior insula, amygdala and ventral striatum also served as 'hubs' integrating common and stimulus-type specific effects. We found different brain representations for negative affect induced by the two types of pain, with primary sensory cortex involved in mechanical pain and thalamus, caudate, insula and midcingulate involved in thermal pain. Aversiveness ratings in all modalities were explained by a mix of common and specific representations. Brain models generalized to new samples (Studies 2 - 4, N = 247). In conclusion, our results show that negative affect is organized in the brain as a combination of common and stimulus type-specific processes.

Disclosures: M. Ceko: None. **P.A. Kragel:** None. **C. Woo:** None. **M. Lopez-Sola:** None. **T.D. Wager:** None.

Poster

P703. Animal Models of Relevance to Stress, Anxiety, and Depression

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P703.01

Topic: G.05. Mood Disorders

Support: National Science Foundation Graduate Research Fellowship Program (DGE—1644869)
Columbia University RISE Award

Title: Bringing the lab to the field: stress-related behavior and ecology of wild urban rats

Authors: *C. K. LARDNER[1], K. A. KAM[1], J. Y. ZHANG[5], K. MAYRING FASANO[1], I. MOLLICONE[1], M. HUSSAIN[2], I. KYMISSIS[3], D. DUMITRIU[4];
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Abstract: Rodents are used ubiquitously in neuroscience research as preclinical models for stress-induced changes in behavior. However, inbred populations housed under laboratory conditions are meant to control for, not model, complex interactions between development, genetics, and environmental uncertainty, which ultimately foment long-term maladaptive behaviors. Alternatively, studying stress pathophysiology in wild Norway rats (*Rattus*

norvegicus) offers an opportunity to “bring the lab to the field” and close the translational gap that exists between our understanding of stress-induced behavioral changes in laboratory rodents and how these findings inform our ability to prevent and treat mental health disorders in human patients. To achieve this, we have employed a capture-mark-recapture (CMR) protocol and standard behavioral assays to characterize stress-related behaviors in wild urban rat populations in New York City over time. Initial behavioral tests were conducted on a pilot wild rat colony in which 8 individuals were captured (N = 3 males, N = 5 females) with a recapture rate of 25%. One juvenile female was captured up to 6 times over 3 weeks. Preliminary results demonstrated a high degree of neophobia that precludes comparing performance in the open field, light/dark box, and novel object preference tests to that of laboratory rat strains. In contrast, performance in the novelty-suppressed feeding task improved over consistent recapture events, suggesting the possibility that wild rat cognition and decision-making can be assayed by standard laboratory tests which utilize hedonic food and water satiety. In ongoing studies, long-term population monitoring and phenotyping of wild rat behavior and physiology will be carried out with the overall goal of establishing wild rat phenotypes associated with stress susceptibility and resiliency.

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Poster

P703. Animal Models of Relevance to Stress, Anxiety, and Depression

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P703.02

Topic: G.05. Mood Disorders

Support: NSF Grant DGE - 1644869

Title: Bringing the Lab to the Field: Wireless Telemetry Systems for Tracking and Monitoring Wild Urban Rats

Authors: *K. A. KAM[1], C. K. LARDNER[2], V. KUMAR[1], I. MOLLICONE[2], D. DUMITRIU[2], I. KYMISSIS[1];
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Abstract: In preclinical neuroscience research focusing on stress pathophysiology, a “translational gap” exists between our understanding of how chronic stress contributes to mental health disorders and our ability to treat them in humans. This could be due in part to an over reliance on modeling complex human behaviors in rodents without also collecting concurrent data on internal physiological states or external environmental contexts associated with observed behaviors. In order to address this gap between research and treatment, we are developing a

wireless, implantable telemetry and tracking system optimized for use in wild Norway rats (Rattus norvegicus). Renowned for their resilience to stressors, wild rats represent the ultimate ethologically-relevant model for neuroscience. Yet, our ability to monitor their ecology, behavior, and social interactions in a city setting is limited; GPS-based tracking in urban environments often fails due to poor signal transmission and physical removal of devices when attached externally. The technology we present here overcomes these challenges by being implantable and based on long-range wide area networks (LoRaWAN), a wireless communication protocol with much lower power consumption than GPS. Further, the LoRaWAN devices are coupled with a low-noise, multi-channel, configurable silicon integrated circuit capable of reading rat ultrasonic vocalizations and biopotentials such as EEG, ECG, and EMG. Additional developments include both a custom 915MHz LoRa antenna with an efficiency of 14.86%, and a -10dB bandwidth of 42.21 MHz and a an inductive wireless power transfer system capable of transmitting up to 40mW in standard implant depths. In this work, a discussion of the system's overall hardware design, implantation methodology, wireless power transfer efficiency, and tracking capabilities are presented. Through these technical advances, experiments assaying rodent behavior that were previously limited to strict laboratory conditions can be conducted outdoors in natural environments. In future studies, an implanted wild rat's location can be triangulated via LoRa signal, its interactions with conspecifics monitored in detail, and relevant physiological data then paired with these observations.

Disclosures: K.A. Kam: None. **C.K. Lardner:** None. **V. Kumar:** None. **I. Mollicone:** None. **D. Dumitriu:** None. **I. Kymissis:** None.

Poster

P703. Animal Models of Relevance to Stress, Anxiety, and Depression

Location: Virtual Only

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Program #/Poster #: P703.03

Topic: G.05. Mood Disorders

Support: TWU Research Enhancement Program Award (Na, E. S.)
TWU Startup Funds (Na, E. S.)

Title: Depressive-like phenotype in mice with Mecp2 knockout in pro-opiomelanocortin neurons

Authors: *A. BAIRD, P. FRAYRE, J. FRAYRE, K. PONCE-RUBIO, M. RUIZ, J. MORENO, E. NA;
Texas Woman's Univ., Denton, TX

Abstract: Mutations in methyl-CP-G binding Protein 2 (MeCP2) gene function are implicated in neurodevelopmental disorders such as Rett syndrome, *MECP2* duplication syndrome, and autism spectrum disorder, producing learning and memory deficits, as well as anxiety phenotypes. Past

research demonstrates that knockout (KO) of *Mecp2* in hypothalamic pro-opiomelanocortin (POMC) neurons produces an overweight phenotype after prolonged high fat diet exposure, but it is not yet known if KO of *Mecp2* in POMC neurons might also affect other behaviors related to POMC function such as depression. Given that obesity and depression are highly co-morbid and that POMC is the precursor peptide to the stress hormone, adrenocorticotropin hormone (ACTH), we hypothesized that knockout of *Mecp2* may alter depressive-like or anxiety-like behaviors as a result of alterations to POMC function. POMC-MeCP2 KO mice and WT controls were maintained on an obesogenic diet and then subjected to tests that assess depression and anxiety. Body weight and high fat (HF) food intakes were recorded for all mice. We demonstrate that younger POMC-MeCP2 KO mice (2-3 months of age) had higher body weight than controls despite comparable food intake. Additionally, younger POMC-MeCP2 KO mice spent significantly more time immobile in the forced swim test than controls, demonstrating behavioral despair. The aged cohort of POMC-MeCP2 KO mice (7-8 months of age) were not significantly different in body weight or immobility in the forced swim test from WT mice. These findings suggest that phenotypic implications of MeCP2 disruption may attenuate with age in mice. Interestingly, there was no difference in anxiety-like behavior between POMC-MeCP2 KO and WT mice in either age cohort thus the effects of POMC-specific MeCP2 knockout may only affect depressive-like behavior. Taken together, our data implicate POMC-specific loss-of-MeCP2 function may adversely impact depressive-like behaviors in mice which could recapitulate the depression observed in clinical populations of obesity.

Disclosures: A. Baird: None. **P. Frayre:** None. **J. Frayre:** None. **K. Ponce-Rubio:** None. **M. Ruiz:** None. **J. Moreno:** None. **E. Na:** None.

Poster

P703. Animal Models of Relevance to Stress, Anxiety, and Depression

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Topic: G.05. Mood Disorders

Support: VA merit award BX000559
VA merit award BX003512
NIMH R01 MH053851

Title: Stress alters cognitive biases in the ambiguous cue interpretation test

Authors: *A. STEPHENS[1], S. WAY[1], F. R. CARRENO[1,2], A. FRAZER[1,2,3], D. A. MORILAK[1,2,3];
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Abstract: Major depressive disorder (MDD) is a highly prevalent and debilitating disorder, the pathogenesis of which is not fully understood. Whereas the two core symptoms of MDD are depressed mood and anhedonia, it is also associated with significant cognitive symptoms, including a negative cognitive bias. Due to these cognitive biases, patients perceive, remember, or interpret information as being more negative than it actually is, which may account for their anhedonia. In order to study the neural mechanisms underlying cognitive biases, we utilized the ambiguous cue interpretation test which measures cognitive biases in rats. In this task, rats are trained to make distinct responses to two reference tones in order to receive either a sucrose reward or avoid an aversive air puff. During the test, rats are presented with ambiguous tones that are in between the frequencies of the two reference tones. The animal's response to the ambiguous tone is recorded and interpreted as their expectation of either a positive or negative outcome. Rats were trained over 1-2 months and tested prior to stress to determine their baseline performance. Multiple baseline tests were performed to ensure that repeated testing does not alter test performance. Then, the animals underwent two days of inescapable foot shock stress (IS), in which one session consists of sixty 15 second foot shocks (0.8 mA). We found that IS induced a negative cognitive bias in male rats, such that they increased their negative responses and decreased their positive responses when the ambiguous tone was played. However, IS did not alter female rats' cognitive biases in this task compared to their baseline performance. The effects of chronic unpredictable stress (CUS) were also tested. We found that three weeks of CUS in female rats did induce a significant negative bias in the ACI test. Having shown a stress-induced negative cognitive bias in this task, future studies will be aimed at investigating neural mechanisms underlying this bias and whether antidepressant treatments can reverse it.

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Poster

P703. Animal Models of Relevance to Stress, Anxiety, and Depression

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Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P703.05

Topic: G.05. Mood Disorders

Support: BUAP-CA-288

Title: Evaluation of sertraline effects in depressive-like behavior in high and low yawning rats

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Abstract: High levels of stress or chronic exposition have been linked to an increased susceptibility to developing anxiety and depression. The high- (HY) and low- yawning (LY) sublines have shown that they differ in the behavioral anxiety tests, being the LY more anxious than the HY. The aim of the present study was to assess the effect of sertraline (2.5 and 5 mg/Kg) on immobility and motor responses on the LY, HY and Sprague-Dawley (SD) rats in the forced swim test (FST). Adult male rats (n = 6 per dose) were housed in standard room conditions with free access to rodent pellets and purified water. All subjects (Ss) had two swim sessions in two consecutive days, the first session with a 15 min duration and 24 h later a second session of 5 min. The Ss received sertraline at 23.5, 5 and 1 h before the second test through three i.p. injections. We scored the following behaviors: immobility, swimming, climbing, and diving. Our results shown that at 2.5 mg/kg dose of sertraline immobility time significantly decrease in the HY rats with respect to LY and SD rats ($P< 0.05$). Only swimming was increased significantly in the HY rats ($P< 0.05$). In conclusion, these results show that HY rats are not only resilient, but also better responders to low doses of sertraline with a greater antidepressant effect compared to that obtained in LY and SD.

Disclosures: D. Bravo Durán: None. **J. Eguibar:** None. **C. Cortes:** None.

Poster

P703. Animal Models of Relevance to Stress, Anxiety, and Depression

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P703.06

Topic: G.05. Mood Disorders

Title: Meningealmast cell calcium activity in models of major depressive disorder

Authors: *E. SARNO, N. DUQUE-WILCKENS, W. JACKSON, A. MOESER, A. ROBISON, A. ROBISON, A. ROBISON, A. ROBISON, A. ROBISON, A. ROBISON, A. ROBISON; Michigan State Univ., East Lansing, MI

Abstract: About 20% of U.S. adults have suffered from Major Depressive Disorder (MDD) in their lifetime[1]. However, physicians are underprepared to treat this health crisis because current treatments fail in about one-third of MDD patients[1]. Over the past decade, preclinical and clinical evidence has shown that neuroinflammation is associated with and may be contributing to mood disorders, but scientists and clinicians require an improved understanding of brain immunity in order to develop new targets for depression treatment. Mast cells (MCs) are innate immune cells that orchestrate overall immune response, and they have an emerging role in mood regulation. Meningeal mast cells, found in the membranes surrounding the brain and spinal cord, sense the environment and release a variety of signaling molecules which direct the activity of blood vessels, glia, and neurons in the brain. Mice lacking mast cells display phenotypes associated with depression. Furthermore, MCs in mice exposed to stress have increased expression of the

gene *FosB* and contain higher levels of allergen-induced mediators and cytokines, including TNF-alpha and IL-6, which are elevated in patients with MDD[15]. *We hypothesize that psychological stress increases FosB expression in meningeal MCs, altering cell function, and, ultimately, driving behavioral changes associated with depression*. To address this, we have engineered a mouse line lacking *FosB* gene expression specifically in mast cells. We show here that mast cells lacking *FosB* have increased histamine and serotonin release, increased cytokine production, and altered vesicular morphology. Moreover, we present the very first GCAMP Ca^{2+} imaging of mast cells in *ex vivo* preparations of meninges as well as Ca^{2+} imaging of cultured mast cells to show that mast cells lacking *FosB* have increased Ca^{2+} signaling. These findings will be critical in understanding the role of MCs in MDD and other diseases and potentially lead to the identification of new therapies.

Disclosures: E. Sarno: None. **N. Duque-Wilckens:** None. **W. Jackson:** None. **A. Moeser:** None. **A. Robison:** None. **A. Robison:** None. **A. Robison:** None. **A. Robison:** None. **A. Robison:** None. **A. Robison:** None. **A. Robison:** None.

Poster

P703. Animal Models of Relevance to Stress, Anxiety, and Depression

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Program #/Poster #: P703.07

Topic: G.05. Mood Disorders

Title: The role of chronic caffeine administration on the attenuation of depressive behaviors in long evans rats

Authors: L. LANDAETA, D. ROEVER, C. PETERSON, S. PECK, D. G. EHLINGER, ***J. J. CORTRIGHT**;
Univ. of Wisconsin River Falls, River Falls, WI

Abstract: Caffeine, the most consumed psychoactive drug, appeals to populations with psychiatric histories due to its readily available nature and alertness exerting effects. Given the prevalence of psychiatric conditions such as major depressive disorder (MDD), in which symptoms such as lethargy are commonplace, a potential relationship between caffeine administration and attenuation of MDD behaviors has been suggested. In particular, in groups exposed repeatedly to stress, a frequent trigger of MDD, caffeine intake surges. This has led to an observed inverse correlation between caffeine intake and MDD behaviors. Clinically, the current state regarding caffeine administration as part of MDD treatment remains limited, but established. When administered in the presence of selective serotonin reuptake inhibitors (SSRIs), caffeine has shown to augment the therapeutic effects in human and animal models. However, the mechanism of action by which caffeine specifically exerts these effects remains unknown. The purpose of our study was to expand upon previous literature and to elucidate these

mechanisms by combining behavioral and morphological analysis. Subjects were exposed to 28 days of randomized stressors during early adulthood including forced swim, cage tilt, wet bedding, mild restraint and restriction of food and water. Control animals were housed in pairs, while animals exposed to randomized stress were housed in isolation. Following stress exposure, a subset of control and stress-exposed animals were orally administered caffeine (15 - 20 mg) twice daily for five weeks. Subjects were then tested for latency in a forced swim test and hot plate test, for motivation in a radial arm maze, for lethargy in an open field test, and for anhedonia using sugar pellets. Following testing, rats were deeply anesthetized, transcardially perfused with saline, and their brains were placed in a Golgi cox solution for subsequent dendritic spine density analysis in the medial prefrontal cortex (mPFC), nucleus accumbens (NAcc), hippocampus and basolateral amygdala (BAL). To date, an attenuation in latency in the forced swim test and hot plate test has been found in stress-exposed animals which had receive caffeine treatment compared to controls. Currently, the effects on depressive behaviors in other tests and dendritic spine density are being analyzed. Collectively, these findings hold significance in that they build on recent research that has aimed to link caffeine intake with the attenuation of depressive behaviors.

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Poster

P703. Animal Models of Relevance to Stress, Anxiety, and Depression

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VA BX003523-01A1
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Title: Role of prefrontal cortex CCR5 expression in social defeat stress

Authors: H.-Y. LIN[1], F. CATHOMAS[2], R. DURAND-DE CUTTOLI[3], C. GUEVARA[1], L. LI[5], K. CHAN[6], L. F. PARISE[7], F. EL GAAMOUCH[1], S. J. RUSSO[1], ***J. WANG**[8,4];
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Abstract: Stress-related psychiatric disorders affect many individuals worldwide with broad health implications. These disorders differ in the prevalence, symptom presentation and in

response to treatment between men and women. The chronic social defeat stress (CSDS) model has been widely used to study stress-related behavior changes and circuitry alteration in male mice. It was shown that in male mice peripheral inflammation, specifically IL-6, and neurovascular pathology contribute to stress susceptibility. Using a recently developed female CSDS model, we found that stress susceptibility significantly correlates with elevated plasma levels of C-C Motif Chemokine Ligand 5 (CCL5) and blocking peripheral CCL5 significantly attenuates stress susceptibility. RNA-seq analysis in the brain found that susceptible female mice had increased expression of CCR5, a CCL5 receptor, in the prefrontal cortex (PFC). In humans, higher plasma levels of CCL5 was also reported in women with major depressive disorder (MDD) and increased CCR5 expression in human female MDD subjects based on published human RNA-seq data. Neither CCL5 nor CCR5 changes were seen in male MDD subjects. These observations suggest that CCL5/CCR5 response in CSDS may recapitulate important and unique inflammatory features of female response to stress. CCR5 is a seven-transmembrane G protein-coupled receptor expressed in microglia, astrocytes and neurons in diverse brain regions. Ligand activation of CCR5 has been show to suppress adenylyl cyclase, activate PI3K/AKT and MAPK signaling and alter intracellular Ca2+ mobilization, all of which can influence synaptic function. Using RNAscope combined with immunohistochemistry we found microglia to be the main cell type in the PFC that expresses CCR5. Further analysis of microglia in the PFC found that compared to resilient or non-stressed mice, stress-susceptible mice had increased numbers of activated microglia, increased PSD95-positive microglia, and increased number of (micro)vessel-associated microglia. We also found that stress-susceptibility is associated with reduced expression of claudin 5, a tight junction protein indicative of blood-brain barrier (BBB) integrity. We are continuing investigating the interactions between peripheral and central CCL5/CCR5 signaling, microglia activity, neuronal/synaptic structure and function, and stress susceptibility.

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Poster

P703. Animal Models of Relevance to Stress, Anxiety, and Depression

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Topic: G.05. Mood Disorders

Support: 5F31MH114624
5R01MH072908

Title: Depression-related consequences of psychosocial stress is governed by bnst crf neurons

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Abstract: Chronic stress is a key risk factor for the development of Major Depressive Disorder (MDD), yet it is unknown how stress that is primarily psychological in nature is translated at the level of neurons to produce behavioral dysfunctions associated with MDD. The limbic forebrain region, Bed Nucleus of the Stria Terminalis (BNST), encodes the nature and chronicity of stress to organize behaviorally adaptive responses. Importantly, the Oval Nucleus of the BNST (BNSTov) contains an enriched population of CRF ($BNSTov^{CRF}$) neurons that are stress responsive and thereby may provide the neural link between CRF and depression. Employing a range of behavioral, molecular, and cell-type specific optogenetics and fiber photometry, we unveil a dimension to depression vulnerability that relies upon accumulated stress-exposure. In this study, we seek to elucidate CRF-related neural mechanisms that underlie depression onset that arise from unmitigated exposure to chronic stress. To elicit stress-related depressive behaviors, the Chronic Social Defeat Stress (CSDS) paradigm was administered. Mice underwent behavioral assays that solicit for a range of anxiety and depression related behaviors including social interaction (SI), sucrose preference, and elevated plus maze tests. To manipulate the activity of $BNSTov^{CRF}$ neurons, Crh-Cre mice were injected with viral constructs (DREADDs, opsins, calcium-indicators) and ferrule cannulae implanted. To record electrical activity of $BNSTov^{CRH}$ neurons, Crh-cre::TdTomato reporter mice were used and cell-attached electrophysiology was used to record the spontaneous firing rate. We have discovered a discrete time-window between 7 and 10 days of CSDS, where mice rapidly diverged from baseline behavior to resilient or susceptible phenotypes, unmasking a temporal dimension to stress-susceptibility. Importantly, electrophysiological and fiber photometric studies were conducted to correlate cell-type and circuit level neuroadaptive changes respectively with the behavioral observations. Cell-type specific chemogenetics reveal the necessity and sufficiency for BNSTov CRF neurons in promoting the development of depressive- and anxiety-like behaviors. Altogether, these findings provide evidence of a temporal domain to stress adaptation where alterations in CRF neurotransmission may shift the predisposition of mice toward either stress-induced depressive-like susceptible or resiliency phenotypes. This discrete period of neuroadaptation may present a window of opportunity for targeting molecular mechanisms with the possibility of preventing the onset of depression in stress-susceptible populations.

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Poster

P703. Animal Models of Relevance to Stress, Anxiety, and Depression

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P703.10

Topic: G.05. Mood Disorders

Support: MOST 107-2320-B-002-046-MY3

Title: Investigating the resting-state functional connectivity and microstructural alterations in the brain of Blm-s-knockout moue with depression- and anxiety-like behavior

Authors: S.-M. HUANG[1], W.-C. CHU[2], K.-H. CHO[1], L.-W. KUO[1], *P.-H. HUANG[2]; [1]Inst. of Biomed. Engin. and Nanomedicine, Natl. Hlth. Res. Inst., Miaoli, Taiwan; [2]Grad. Inst. of Pathology, Natl. Taiwan Univer, Taipei, Taiwan

Abstract: *Blm-s* (*Bcl-2*-like molecule, short form), a *Bcl-2* family gene, has been identified and shown to be a key role in regulating the apoptosis of postmitotic immature neurons during corticohistogenesis. How BLM-s expression affects the structural and functional brain network remains unexplored. The present study employed resting-state functional MRI (rs-fMRI) as well as diffusion MRI (dMRI) to investigate the influence of *Blm-s* deletion on brain network in a mouse model because mice with *Blm-s* null present depression- and anxiety-like behaviors. Rs-fMRI is a noninvasive neuroimaging modality in discovering brain functions, which characterizes the intrinsic brain networks under rest condition and can serve as a robust tool in exploring neurobiological disorders both in human and animal studies. dMRI monitors the property of water diffusion and provides the information of fiber orientation, which is valuable to the understanding of structural connectivity as well as local microstructural changes at specific brain regions and axonal pathways. *Blm-s*-knockout mice (KO, n=12) and age-matched wild type (WT, n=14) were scanned under dexmedetomidine anesthesia (0.2 mg/kg) on a 7T animal MRI system (Biospec, Bruker, Germany). Diffusion indices including fractional anisotropy (FA) and diffusivity were reconstructed using the tensor model. Rs-fMRI images were pre-processed and analyzed to assess the brain functional network. Multiple regions of interest were identified to characterize local property as well as the connectivity between each two specific brain regions. Results show that, comparing to WT mice group, KO mice group presents reduced connectivity in the following pairs of regions: left ventral dentate gyrus (vDG) - left caudate putamen (CPu), left insula - right CPu, left insula - left vDG, and right insula - right vDG. The amplitude of low frequency fluctuation (ALFF) was also evaluated to assess regional rs-fMRI activity. Reduced ALFF was found in bilateral dorsal dentate gyrus, while increased ALFF was found in the orbital cortex. From dMRI analysis, elevated FA and reduced radial diffusivity was found in the vDG of KO mice group. Our findings in MRI suggest that microstructures and local rs-fMRI activity in hippocampal area, particularly the vDG region, might be altered in *Blm-s*-null mice. As MRI can provide a neuroimaging insight in mesoscale, the underlying mechanism of the consequence after *Blm-s* deletion can be further uncovered by cytological approaches.

Disclosures: S. Huang: None. **W. Chu:** None. **K. Cho:** None. **L. Kuo:** None. **P. Huang:** None.

Poster

P703. Animal Models of Relevance to Stress, Anxiety, and Depression

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Program #/Poster #: P703.11

Topic: G.05. Mood Disorders

Support: CIHR Project Grant PJT#165852
Campbell Family Mental Health Research Institute

Title: Behavioral and Neurostructural Changes Associated with Chronic Amygdala Hyperactivation

Authors: *K. A. MISQUITTA[1], S. CODELUPPI[1], Y. BANSAL[1], T. D. PREVOT[1], D. F. NEWTON[2], J. ELLEGOOD[3], J. P. LERCH[4], T. TOMODA[1], E. SIBILLE[5], Y. S. NIKOLOVA[1], M. BANASR[6];
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Abstract: The amygdala (AMY) is a key brain region of the limbic system that plays a critical role in emotion processing and stress response. Functional magnetic resonance imaging (fMRI) studies identified abnormal amygdala (AMY) activation in psychiatric illnesses including major depressive disorder (MDD). Stress exposure is a major precipitating factor of MDD episodes which are associated with AMY hyperactivity. We also demonstrated that chronic stress exposure in mice increases AMY volume and synaptic strength which correlate with anxiety- and depressive-like behaviors. Moreover, preclinical studies using pharmacologic, opto- and chemogenetic approaches to activate AMY neurons have consistently demonstrated that acute AMY hyperactivation induces anxiety-like behaviors in mice. However, it remains unknown if chronic amygdala hyperactivation (cAH) is sufficient to induce chronic stress-like deficits or is a susceptibility factor for chronic stress-induced behavioural, volumetric and synaptic deficits. Using *designer receptor exclusively activated by designer drug (DREADD)* approach, basolateral amygdala (BLA) neurons of Camk2a-cre mice infected with AAV2-DOI-GqDREADD were activated with clozapine-N-oxide (in drink water for 5 weeks). Mice were then exposed to chronic restraint stress (CRS; 1X/day for 1hr) for 2 weeks. All mice were behaviorally assessed in the Phenotyper (PT), and sucrose consumption tests (SCT) each week and in the novelty supressed feeding (NSF) test once at the end of the experiment. Animals were then perfused for *ex vivo*-MRI and puncta density analysis. We found that mice with cAH displayed a progressive increase in baseline anxiety-like deficits in the PT test and slightly more marked deficits following CRS compared to controls, but not statistically different from animals subjected to CRS alone. Also, cAH did not exacerbate CRS effect in the NSF. No significant cAH effect was found in the SCT before or after CRS. MRI and synaptic analysis are currently underway. Our results suggest that cAH is sufficient to induce anxiety and may exacerbate CRS effects on

anxiety measures. However, cAH was not sufficient to induce depressive-like behavior and was not a vulnerability factor for stress-induced depressive-like behavior. In conclusion, our findings suggest that although AMY hyperactivity is associated with elevated anxiety, it does not appear to be a predictor or a biomarker of clinical depression.

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Poster

P703. Animal Models of Relevance to Stress, Anxiety, and Depression

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Program #/Poster #: P703.12

Topic: G.05. Mood Disorders

Support: NIDA, Grant# 1R15 DA044500-01A1
NIH-ENDURE R25-NS080268706

Title: The interaction of the endocannabinoid system and the serotonergic system and its effect in voluntary wheel running in depression-like behaviors in rats

Authors: *A. P. RAMOS-ROLÓN, W. NORZE, P. A. MUÑOZ-RODRÍGUEZ, A. I. RODRÍGUEZ-LEON, A. I. ACEVEDO-JETTER, V. M. SCHLEIER-ALBINO, K. M. TORRES-MONTERO, M. C. APONTE-REYES, C. S. MALDONADO-VLAAR;
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Abstract: Depression is one of the most prevalent mental health disorders in the United States. Studies have shown that the endocannabinoid system (eCB) is involved in the pathophysiology of depression. The cannabinoid-1 receptor (CB1) plays a role in depression-like behavior by modulating the serotonergic system. Other studies have revealed that aerobic exercise decreases depression-like behavior by upregulating serotonin (5-HT) release. However, it is still unknown if the eCB system is responsible for modulating the antidepressive properties of exercise through an alteration in 5-HT firing. This project aimed to observe if the manipulation of the CB1 receptor would enhance the therapeutic effect of aerobic exercise in depression. Male Sprague Dawley rats were submitted to an aerobic exercise protocol, while a group remained sedentary. Within the sedentary and exercise group, some rats received intraperitoneal injections of an agonist of the eCB receptors, (+)-WIN 55,212-2, while the others received the vehicle. The rats were divided into four groups: sedentary with the vehicle (Sed/Veh), sedentary with WIN55,212-2 (Sed/WIN), exercise with the vehicle (Ex/Veh), and exercise with WIN55,212-2 (Ex/WIN). We used the forced swim test (FST) paradigm as a depression model. The FST results showed that the Sed/Veh group had significantly less swimming and more floating than Sed/WIN and

Ex/Veh groups, but no the Ex/WIN group. These results suggest that exercise and (+)-WIN 55,212-2 have antidepressive properties, but both treatments simultaneously are not antidepressive. Further studies will be conducted to measure the expression of the CB1 receptor and 5-HT 1A receptors in the hippocampus and medial prefrontal cortex to better understand the neural circuits underlying the depression-like response. Studies like these are essential to better understand the potential molecular antidepressive mechanisms that could help develop new strategies to treat depression.

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Poster

P713. Preclinical Models: Post-Stress Trauma

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P713.01

Topic: G.07. Post-Traumatic Stress Disorder

Support: Staglin Center for Brain and Behavioral Health

Title: Cfos expression during stressors significant and non-significant enough to produce stress enhanced fear learning

Authors: *S. Y. MARKOWITZ, L. LAUGHLIN, M. S. FANSELOW;
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Abstract: Approximately 10% of people who experience a highly stressful event, or trauma, will go on to develop Post-Traumatic Stress Disorder (PTSD). As the majority of people who experience intense stress do not develop PTSD, an important question is what makes a stressful event “significant” enough to cause this disorder to manifest. To investigate this question we used an animal model of stress reactivity called Stress Enhanced Fear Learning (SEFL). Using this model we found that that not all stressors are significant enough to cause SEFL, even if both experiences cause comparable levels of fear. Specifically, rats that experienced 15 presentations (1s, 1mA) of Shock Stress showed future SEFL, while rats that experienced 15 presentations (1s, 120 dB) of Noise Stress did not. Additionally, only Shock Stress animals exhibited a panic-like response to stressor presentations. We concluded that shock is a Significant enough stressor to cause SEFL, while noise was Nonsignificant. The periaqueductal grey (PAG) is a key brain region for defensive responses. It is well established that activity in the PAG is both necessary and sufficient for both fear and panic responses toward external stressors. To understand how the neural signature of significant and nonsignificant stressors differ we examined cFos expression in the PAG following these two types of stressors. Rats were first exposed to either 15

presentations of Shock Stress or 15 presentations of Noise Stress during a 90 minute session. 30-45 minutes after the final stressor presentation, these experimental rats, as well as homecage controls, were perfused. Brain slices from central PAG stained for c-Fos expression showed a significant difference in activity in the lateral PAG (lPAG) and ventral-lateral PAG (vlPAG) between Shock Stress and Noise Stress animals. Thus, we have begun to characterize how activity in specific subregions of the PAG correlate with exposure to "Significant" and "Non Significant" stress, which in turn may better help us understand the underlying neural mechanisms of PTSD.

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Poster

P713. Preclinical Models: Post-Stress Trauma

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Program #/Poster #: P713.02

Topic: G.07. Post-Traumatic Stress Disorder

Support: NIH Grant RO1-MH115678

Title: Post-stress glucose treatment enhances performance on hippocampal-dependent tasks in both humans and rodents

Authors: *N. J. SMITH, D. B. WEATHERILL, M. S. FANSELOW;
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Abstract: Post-stress glucose treatment enhances performance on hippocampal-dependent tasks in both humans and rodents
A third of the US population will experience a traumatic event at some point in their life. Posttraumatic stress disorder (PTSD) is a severally debilitating mental disorder affecting up to 20% of that population. It is well established that increased serum levels of glucocorticoids following significant stress impair hippocampal glucose uptake and function, including contextual processing. Without proper hippocampal activity, contextual memory of trauma may not be properly encoded. This deficit in encoding may subsequently contribute to generalization of fear and intrusive memories, hallmark diagnostic criteria for PTSD. We have previously shown, in rats, consumption of glucose within a critical 3-hour window following a significant stressor rescues deficits caused by the stress. The present study provides evidence, in human trauma patients, that post-stress glucose consumption within a 3-hour window of the traumatic event similarly protects hippocampal function. Two main results from this experiment show: (1) glucose following stress benefitted the performance on a hippocampus-dependent task but not a hippocampus-independent task 3-hours, 5-7 days, and 60 days after trauma and (2) the post-stress glucose group recalled significantly more details of the trauma than the placebo group.

Because research on the mechanism(s) by which glucose deficits lead to impaired hippocampal function is presently scarce, we next followed up on our human data by probing performance on a hippocampal-dependent task in rats, using the Object in Place (OIP) task. Both male and female rats were first exposed to either 100 inescapable tail shocks or simple restraint. Animals were then given 24-hour ad libitum access to glucose or a non-caloric sweetener (stevia) before OIP acquisition and test phases, which were separated by a 2-h delay. The post-stress glucose group spent significantly more time investigating the objects in different locations compared to the objects in the same locations. Thus, we have begun to characterize a rodent model to further study the underlying mechanism(s) mediating a potential prophylactic effect of glucose on PTSD/PTSD-like symptom development.

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Poster

P713. Preclinical Models: Post-Stress Trauma

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Topic: G.07. Post-Traumatic Stress Disorder

Support: NIDA Grant DA044980
BSI Student Support Grant at CU Boulder

Title: Stressor controllability in Long Evans rats: Comparisons of strains, sex, time-of-day, and regional cFos activation patterns

Authors: *N. BOSNIAN[1], C. SINGH[1], J. GRIZZELL[2], M. SADDORIS[3];
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Abstract: The perception of control over the cessation of a string of acute tail shocks has been directly associated with various patterns of resilience to stress in adult, male Sprague Dawley (SD) rats. However, the expression of this behavioral phenomenon in adult, female SD rats is not as readily demonstrated. Here, we present results from a series of studies in our laboratory that demonstrate that the differential expression patterns of behavioral resilience in Sprague Dawley male vs female rats extends to Long Evans (LE) rats. Preliminary results show that following a standard stressor controllability paradigm (ES/IS), adult male LE rats having learned to control tailshock duration (ES) nonetheless fail to demonstrate the male SD-like patterns of behavioral resilience in the juvenile social exploration (JSE) and defensive burial tests (DBT) when stressed and tested during the light phase. Moreover, male LE rats in the ES condition are not statistically differentiable from male counterparts in the inescapable stress (IS) conditions. However, when the stress and behavioral paradigms were presented during the first two hours of the dark cycle,

ES male LE rats approach the social target significantly more than IS male LE rats. In a second study, we found that female LE rats in the ES condition that were stressed during the light cycle similarly approached the same-sexed juvenile in the JSE more than female IS counterparts. These patterns collectively support the notion that strain and sex differences in the expression of behavioral resilience following the stressor controllability paradigm may be linked with ethologically distinct patterns of behavior in a strain-specific manner. Finally, we will present region-specific differences in neuronal activation patterns (cFos) brought on by stressor controllability conditions in age-matched male and female LE and SD rats.

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Poster

P713. Preclinical Models: Post-Stress Trauma

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Program #/Poster #: P713.04

Topic: G.07. Post-Traumatic Stress Disorder

Support: NIDA Grant DA044980

Title: Social experience-dependent influences of stress resilience: Applying a novel paradigm for hierarchy-mediated differences in social stress responses

Authors: *Z. Q. MARSHALL[1], J. GRIZZELL[2], M. SADDORIS[3];
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Abstract: Stressor controllability paradigms demonstrate that rodents having learned to exert control over stressful stimuli display prefrontal cortical mediation of limbic system responses during traumatic stress exposure. Similarly, social experience acts upon these limbic circuits such that acquiring and maintaining a dominant social status promotes a stress resilient phenotype whereas social subordination promotes stress susceptibility. That said, most rodent models of social status-mediated stress resilience/susceptibility rely on heavy bouts of territorialized aggression or food-motivated states to drive social status formation and maintenance. Here, we present results of a novel use of the "Warm Spot" test. Adult, male, Long Evans rats ranging in age from early, middle, and late adulthood were age- and weight-matched into dyads and placed for 10 minutes daily for two weeks into a modified polycarbonate cage that was placed onto a bed of dry ice and contained a heated respite (ave: 36° C) that was large enough for one rat to escape the ice-cold floor (ave: -10° C). 24 hours later, each rat was placed one at a time into a similar cage with the respite removed for a 10-minute "no spot" stressor. Subsequently, rats were tested for social approach/avoid behaviors in either the juvenile social exploration or social interaction test. As in other paradigms, animals demonstrating social

dominance (e.g. greater total durations of warm-spot occupation over 14-day period) showed greater social approach behaviors relative to subordinate counterparts, however this effect was only seen in the social interaction test. We will also present results from preliminary studies demonstrating social status-dependent differences in fear extinction following this novel paradigm, as well as "no spot"-induced expression of cFos in vmPFC, amygdala (BLA and MeA), and dorsal raphe nucleus. Taken together, this paradigm presents a novel opportunity to ascertain social status-dependent differences in stress-induced behavior and neural processing patterns in a naturalistic scenario while minimally driving aggressive behavior and food/water-deprived physiological states.

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Poster

P713. Preclinical Models: Post-Stress Trauma

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Program #/Poster #: P713.05

Topic: G.07. Post-Traumatic Stress Disorder

Support: NIH Grant R15HL132322

Title: Enhanced negative feedback of the HPA axis and increased noradrenergic enzyme levels in females exposed to a predator-based psychosocial stress model of PTSD

Authors: *P. R. ZOLADZ[1], C. R. DEL VALLE[1], I. F. SMITH[1], C. S. GOODMAN[1], J. L. DODSON[1], K. M. ELMOUHAWESSE[1], C. D. KASLER[1], B. R. RORABAUGH[2]; [1]Psychology, Ohio Northern Univ., Ada, OH; [2]Pharmaceut. Sci., Marshall Univ., Huntington, WV

Abstract: People with post-traumatic stress disorder (PTSD) exhibit numerous physiological alterations, including lower baseline cortisol levels, enhanced negative feedback of the hypothalamus-pituitary-adrenal (HPA) axis, and increased noradrenergic activity. Male rats exposed to a predator-based psychosocial stress model of PTSD exhibit similar changes in HPA axis function and neurotransmission. Here, we examined whether similar effects would be observed in female rats exposed to this model. Adult female Sprague-Dawley rats were exposed to a cat on two occasions, separated by 10 days, in conjunction with daily social instability. Three weeks following the second cat exposure, we examined rats' anxiety-like behavior on an elevated plus maze (EPM), collected blood samples at baseline and following dexamethasone administration to quantify corticosterone levels, and extracted brains to quantify markers of noradrenergic activity in the hippocampus and prefrontal cortex (PFC). Stressed females did not exhibit heightened anxiety on the EPM, but, relative to controls, they displayed lower corticosterone levels and a greater suppression of corticosterone following dexamethasone

administration. Stressed females also exhibited increased markers of noradrenergic activity in the dorsal hippocampus and PFC. These findings suggest that this model of PTSD may be useful for studying mechanisms underlying trauma-induced changes in female physiology.

Disclosures: P.R. Zoladz: None. **C.R. Del Valle:** None. **I.F. Smith:** None. **C.S. Goodman:** None. **J.L. Dodson:** None. **K.M. Elmouhawesse:** None. **C.D. Kasler:** None. **B.R. Rorabaugh:** None.

Poster

P713. Preclinical Models: Post-Stress Trauma

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Program #/Poster #: P713.06

Topic: G.07. Post-Traumatic Stress Disorder

Support: EUROSTARS E! 113468 - DiSARM FEAR

Title: Development of a novel Post Traumatic Stress Disorder (PTSD) model in the rat

Authors: *J. E. FRIEDMAN, K. H. PARK, J. H. LEE, B. Y. LIM, H. PARK, A. LEE, P. J. SWEENEY, P. J. SWEENEY, L. C. PARK;
Naason Sci., Cheongju, Korea, Republic of

Abstract: PTSD is a serious condition that can lead to intense fear, anxiety and depression resulting in social distancing and other behavioral disorders. It has also been found that childhood exposure to highly stressful events increases the likelihood that the adult will be more prone to suffer from PTSD. 8.1M Europeans and 11.4M Americans are actively suffering from PTSD, severely reducing the quality of their lives. Current medication is unspecific, ineffective and has a large number of side effects (sedation, abuse potential, sexual dysfunction). As a result, many people continue to suffer from PTSD in spite of current treatment options, indicating a critical need for new treatments. A limiting factor in the development of treatment for PTSD is the lack of valid, translational models. Naason Science has been developing models of PTSD in the rat and the minipig based on a multi-faceted, prolonged stress paradigm, so as to test the efficacy of new compounds. Animals are stressed as juveniles in order to increase their susceptibility to PTSD as adults. We have used desipramine + methylphenidate or diazepam as controls in the rat. Juvenile SD rats, male and female, n=12/group, are exposed to different stressors over a 2-day period, comprised of restraint (2h), swimming (1m) and submersion (30s) in the Water Associated Zero Maze (WAZM) and queued electric shock (x5). This paradigm is repeated weekly for another 2 weeks. The behavior of the rats is recorded using our Home Cage Activity (HCA) monitoring system, recording the behavior of the mixed group housed animals 24/7. Behavior is automatically assessed using AI computerized systems. After being exposed to the 3-week PTSD paradigm, subjects are treated for 5 days and then exposed to the external track

of the WAZM and subsequently to queued fear conditioning (qFC). This is repeated after another 30 days. Upon terminating the study blood samples are collected. After 60 days, test subjects exhibited signs of anxiety. In the qFC test, normal (non-stressed) rats exhibited freezing for 42sec±17, rats exposed to the PTSD paradigm froze for 125sec±15. Diazepam (1, 3, 10mpk) was not effective in this model. Stressed rats treated with Methylphenidate (2.7mpk) and desipramine (10mpk) exhibited the same freezing time as normal rats (38sec±5). The cortisol levels of normal and treated rats were similar, at 461pg±39/ml, while that of the PTSD/vehicle treated rats was 684pg±66/ml. The model presented here is a simple, robust model of PTSD that should facilitate dissecting out various aspects of this disorder and provides a screening paradigm for new medical treatments.

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Poster

P713. Preclinical Models: Post-Stress Trauma

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Program #/Poster #: P713.07

Topic: G.07. Post-Traumatic Stress Disorder

Support: EUROSTARS E! 113468 - DiSARM FEAR

Title: Development of a novel Post Traumatic Stress Disorder (PTSD) model in the Jeju minipig

Authors: K. H. PARK[1], J. BYEON[1], T. H. KIM[1], S. C. LEE[2], Y. J. SON[2], P. J. SWEENEY[1], J. E. FRIEDMAN[1], *L. C. PARK[1];
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Abstract: PTSD is a serious condition that can lead to intense fear, anxiety and depression. Childhood exposure to highly stressful events has been found to increase the possibility that the adult will be more prone to suffer from PTSD. The percentage of those affected varies from country to country with 3.5% of Americans and 9.2% of Canadians actively suffering from PTSD. Current medication is not very effective and has a large number of side effects. As a result, many people continue to suffer from PTSD in spite of current treatment options, indicating a critical need for new treatments. One limiting factor in the development of treatment for PTSD is the lack of valid, translational models. Naason Science has been developing models of PTSD in the rat and in a larger animal-the minipig, based on multi-faceted, prolonged stress paradigms, so as to test the efficacy of new compounds. In addition, we expose the animals to stress as juveniles in order to increase their susceptibility to PTSD as adults. Pigs are intelligent animals that not only closely share physiology with humans, such as the cardiovascular system, but demonstrably show emotions, including fear. This is different from many wild animals that do not overtly express fear nor discomfort. This makes them viable candidates for behavioral studies. Jeju minipigs from the same litter, mixed gender, were used for this pilot study, n=3 per group, one group acted as a control for the second group that experienced stressors.
Piglets 1 week old were separated from their mothers for 2 hours/day until weaning. They were restrained in a net for 10 min/day and exposed to predator urine and sound. Both sets of animals appeared to thrive as they experienced similar weight gain. Animal behavior was subsequently tested 2 times/week in an open field from week 6, and for 20 hours in their home cage at week 7. Minipigs were recorded and analyzed using our digitized video recording and AI analysis system. PTSD pigs only moved 64% (55±13m) of the distance that their control littermates moved (86±14m) and over a shorter period of time (2790 vs 3078 seconds). Their speed was also significantly slower by approximately 30% (1.96±0.3cm/s vs. 2.78±0.3cm/s). PTSD pigs experienced longer periods of freezing, by about 55%. In their home cage, PTSD minipigs demonstrated significantly greater social distancing, maintaining 0.608±0.08m distance from their littermates as compared to 0.297±0.02m for the controls.
Whereas the data presented here is a pilot study, it demonstrates how early stress can affect simple behavior in juvenile pigs and can serve as a model system for the development of PTSD in large animals and possibly provide a translational model for humans.

Disclosures: K.H. Park: A. Employment/Salary (full or part-time):; FTE, Naason Science. **J. Byeon:** A. Employment/Salary (full or part-time):; FTE, Naason Science. **T.H. Kim:** A. Employment/Salary (full or part-time):; FTE, Naason Science. **S.C. Lee:** A. Employment/Salary (full or part-time):; FTE, Cronex, Inc. **Y.J. Son:** A. Employment/Salary (full or part-time):; FTE, Cronex, Inc. **P.J. Sweeney:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); ownership Interest, Naason Science. F. Consulting Fees (e.g., advisory boards); SAB, Naason Science. **J.E. Friedman:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Stock Ownership, Naason Science. F. Consulting Fees (e.g., advisory boards); SAB, Naason Science. **L.C. Park:** A. Employment/Salary (full or part-time):; FTE, Naason Science. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); ownership Interest, Naason Science.

Poster

P713. Preclinical Models: Post-Stress Trauma

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P713.08

Topic: G.07. Post-Traumatic Stress Disorder

Support: NIDA Grant DA044980

Title: Social status-mediated alterations of stress in a novel social conflict paradigm: comparing male and female Long Evans rats in a repeated, dyadic "Warm Spot" task

Authors: G. KUMAR[1], *J. GRIZZELL[1], M. SADDORIS[2];
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Abstract: Models of social dominance-mediated stress resilience in rodents have shown that dominant social statuses can promote resilience to traumatic stress when compared to subordinate counterparts. However, most models focus only on males due to the paucity of easily quantifiable aggressive behaviors in females. Here, we present results from two studies that compare male and female Long Evans rats that were (1) group-housed until adulthood or (2) socially isolated as juveniles. Upon adulthood, all animals were placed into a modified "warm spot" (WS) task daily for two weeks. For 10 minutes each day, age-, weight-, and sex-matched animals were dyadically paired and exposed to a cage that was placed onto a bed of dry ice but contained a small, warmed respite in a far corner to allow one animal at a time to escape the cold floor. Following the 14th day of pairing in the WS arena, each animal was placed alone in a similar arena but with the respite removed, which we called the "no spot" test and 1 hour afterward, tested in the social interaction test for socially avoidant behavior. 24 hours later, all animals were exposed to a standard cued fear conditioning paradigm and then tested for fear recall, fear extinction, and delayed fear extinction 1, 2, and 9 days afterward, respectively. Social dominance was determined by total WS occupation and greater WS durations were found to be highly correlated with other dominant status-related behaviors such as pushes, resists, attacks, and successful takes of the warm spot. Low WS durations were strongly correlated with yielding the WS to dyadic partner, particularly in females. While most dyads formed predictable hierarchies by day 3, females demonstrated greater WS sharing (i.e. co-occupation) and all animals that had been isolated as juveniles displayed more agonistic behaviors, reduced sharing of WS, and more exchanges of WS occupation. Preliminary results from the fear conditioning/extinction tests indicate that while all animals acquired fear memories at similar levels, socially subordinate males and females displayed greater freezing behavior during fear extinction training and recall, but this effect was attenuated by group housing status during

adolescence. Together, these results indicate that social status in male and female rats may be linked with behavioral markers of resilience.

Disclosures: G. Kumar: None. **J. Grizzell:** None. **M. Saddoris:** None.

Poster

P745. Molecular and Neural Mechanisms III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P745.01

Topic: G.09. Drugs of Abuse and Addiction

Title: Behavioral effects of the DA D1R antagonist, SCH-23390, in compulsive methamphetamine taking rats: molecular and biochemical neuroadaptations in the dorsal striatum after long-term withdrawal from methamphetamine self-administration

Authors: *S. JAYANTHI[1], B. LADENHEIM[1], M. T. MCCOY[1], P. SULLIVAN[2], D. S. GOLDSTEIN[2], J. L. CADET[1];
[1]Mol. Neuropsychiatry, NIDA-IRP, Baltimore, MD; [2]Clin. Neurocardiology Section CNP/DIR/NINDS/NIH, NINDS NIH, Bethesda, MD

Abstract: Methamphetamine (METH) use disorder (MUD) remains a persistent public health menace. Perturbations in striatal dopamine (DA) homeostasis are thought to underlie the behavioral and pathobiological consequences of METH exposure in humans and rodents. To identify potential consequences of METH use disorder, we have attempted to mimic DSM criteria for substance use disorders in rats. Towards that end, we trained rats to self-administer METH (9 hours session for 20 days) followed by exposing them to a footshock paradigm to represent adverse consequences. These adverse consequences led to the segregation of METH SA rats into two phenotypes: (*i*) shock-sensitive that drastically decreased their METH-intake and (*ii*) shock- resistant or compulsive drug takers that continued their METH intake despite footshocks. Thereafter, rats were injected intraperitoneally with the DA D1 receptor antagonist, SCH23390. SCH23390 caused substantial reduction of METH taking in a dose-dependent fashion. Stopping SCH23390 administration led to re-emergence of compulsive METH taking in the shock-resistant rats. Moreover, rats that showed compulsive METH taking exhibited greater incubation of METH seeking than non-compulsive rats at withdrawal day 7 and 30, suggesting differential neuroadaptations in the brains of these two phenotypes. Post-mortem analyses of dopamine metabolism revealed non-significant decreases (about 30%) in striatal DA levels in both resistant and sensitive rats. In contrast, striatal contents of the deaminated metabolites, DOPAL, DOPAC, and DOPET, were significantly increased in shock-sensitive rats, suggesting increased DA metabolism in the non-compulsive rats. VMAT2 and DAT protein levels were decreased in both shock-resistant and sensitive animals, suggesting that high levels of METH intake can cause DA terminal degeneration. Interestingly, members of the D2-type DA receptors,

DA D3R and DA D4R but not D2R, showed significant decreased protein expression in the dorsal striatum. These results indicate differential effects of METH SA on DA receptors located on post-synaptic neurons. RNA sequencing studies also revealed substantial differences in the expression of genes between METH-addicted and non-addicted rats. We identified 492 significantly differentially expressed genes that are involved in p13-AKT, cAMP and MAPK signaling pathways. Other differentially expressed genes participate in glutamatergic signaling. Future studies are needed to identify specific roles that genes of interest might play in compulsive METH taking or in suppressing METH self-administration.
Acknowledgement: This work is supported by DHHS/NIH/NIDA/IRP.

Disclosures: S. Jayanthi: None. **B. Ladenheim:** None. **M.T. McCoy:** None. **P. Sullivan:** None. **D.S. Goldstein:** None. **J.L. Cadet:** None.

Poster

P745. Molecular and Neural Mechanisms III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P745.02

Topic: G.09. Drugs of Abuse and Addiction

Title: Sex-and brain region-specific alterations in dopamine metabolism consequent to methamphetamine self-administration

Authors: *A. P. DAIWILE[1], P. SULLIVAN[2], S. JAYANTHI[1], D. S. GOLDSTEIN[2], J. L. CADET[1];
[1]Mol. Neuropsychiatry Res. Br., NIH, NIDA IRP, Baltimore, MD; [2]Autonomic Med. Section, NIH, NINDS IRP, Bethesda, MD

Abstract: Sex-and brain region-specific alterations in dopamine metabolism consequent to methamphetamine self-administration Atul P. Daiwile*[1], Patricia Sullivan[2], Subramaniam Jayanthi[1], David S Goldstein[2] and Jean Lud Cadet[11]Molecular Neuropsychiatry Research Branch, NIDA IRP, NIH, Baltimore, MD, USA. [2]Autonomic Medicine Section, NINDS IRP, NIH, Bethesda, MD, USA.Methamphetamine use disorder (MUD) is a neuropsychiatric disorder that inflicts significant health burdens throughout the world. Importantly, significant sex differences in use and relapse rates exist among human METH users. Some sex differences have also been reported in animal models of MUD. The biochemical bases for these behavioral differences remain to be determined. We began to address these issues by using a METH self-administration model to identify potential sex differences at biochemical levels. Rats were trained to self-administer METH (0.1 mg/kg/inj, i.v.) using two 3-h daily sessions for 20 days. Rats were then assessed for cue-induced drug seeking on withdrawal days (WD) 3 and 30. Rats were euthanized 24-h after WD30. Dorsal striatum (Dstr), nucleus accumbens (NAc) and prefrontal cortex (PFC) were dissected to measure catecholamine levels. We found that both

female and male rats increased their METH intake during the 4 weeks of the experiment and showed incubation of METH seeking during withdrawal. Male rats compared to female rats had higher levels of 3,4-dihydroxyphenylalanine (DOPA) in the Dstr and PFC; cys-dopamine (DA), a product of dopamine quinone, in the Dstr; and of 3,4-dihydroxyphenylethanol (DOPET) and 3,4-dihydroxyphenylacetic acid (DOPAC) in the NAc in the saline and METH groups. Females showed higher Cys-DOPA levels in the NAc; DA, DOPAL and DOPAC levels in the PFC; and of dihydroxyphenylglycol (DHPG), an oxidative deamination metabolite of norepinephrine (NE) in the Dstr of compared to male controls. Only male showed decreases in DOPA levels in the Dstr after METH SA. Increases were observed only in male low METH takers in the PFC whereas both sexes showed decreased DOPA levels in the NAc. Male high METH takers showed increased cys-DOPA levels in the NAc, whereas female high METH takers showed decreased cys-DOPA levels. Unexpectedly, male high METH takers, but not female rats, exhibited increased DA levels in the NAc. Interestingly, however, both sexes showed decreased levels of cys-DA, DOPAL, DOPET, and DOPAC in the NAc. Our findings further support the importance of understanding the basic sex-specific neurochemical effects of drugs during efforts to develop therapeutic agents against METH use disorder.**Acknowledgement:** This work is supported DHHS/ NIH/ NIDA/ IRP.

Disclosures: A.P. Daiwile: None. **P. Sullivan:** None. **S. Jayanthi:** None. **D.S. Goldstein:** None. **J.L. Cadet:** None.

Poster

P745. Molecular and Neural Mechanisms III

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Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P745.03

Topic: G.09. Drugs of Abuse and Addiction

Title: Sex-specific alterations in histone deacetylases following methamphetamine self-administration in the rat hippocampus

Authors: *M. T. MCCOY, A. P. DAIWILE, S. JAYANTHI, B. LADENHEIM, J. L. CADET; DHHS/NIH/NIDA/IRP, Baltimore, MD

Abstract: Methamphetamine use disorder (MUD) is a neuropsychiatric disorder that imposes significant health burdens throughout the world. Importantly, significant sex differences in use and relapse rates exist among human METH users. Sex differences in behaviors and biochemical changes have also been reported in preclinical models of MUD. Nevertheless, much remains to be done to clarify the epigenetic bases of METH-induced molecular differences identified in male and female rats. We have been using a METH self-administration model in order to try to further clarify the molecular substrates of behavioral differences between female and male rats. Rats were trained to self-administer METH (0.1 mg/kg/injection, i.v.) using two 3-hours daily

sessions (separated by a 30-minute off interval) for 20 days. Rats were then assessed for cue-induced drug seeking at 3 and 30 withdrawal days (WD). Rats were euthanized 24 hours after WD30. Hippocampi (HIP) were dissected and interrogated for the mRNA expression of histone deacetylases (HDACs). We observed that both female and male rats increased their METH intake during the 4 weeks of the experiment and showed incubation of METH seeking during withdrawal.
Quantitative PCR revealed that male rats had higher basal RNA levels of HDAC1, HDAC2, HDAC3, and HDAC4 in their HIP in comparison to female rats irrespective to treatment conditions. In addition, compared to controls female METH takers exhibited significant increased expression of HIP HDAC4 whereas there were no significant changes in HDAC1, HDAC2, and HDAC3 mRNA levels after exposure to METH SA and withdrawal. Male rats did not show any significant changes in the expression of any of the HDACs after METH SA and withdrawal.
These results identify further evidence for sexual dimorphic responses to METH. These observations support the notion that sex-specific therapeutic approaches to the treatment of METH use disorder may be necessary.
Acknowledgement: This work is supported Department of Health and Human Services/ NIH/ NIDA/ IRP.

Disclosures: M.T. McCoy: None. **A.P. Daiwile:** None. **S. Jayanthi:** None. **B. Ladenheim:** None. **J.L. Cadet:** None.

Poster

P745. Molecular and Neural Mechanisms III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P745.04

Topic: G.09. Drugs of Abuse and Addiction

Title: Footshock-induced abstinence from compulsive methamphetamine self-administration is accompanied by increased hippocampal expression of cannabinoid receptors (CB1 and CB2) in the rat

Authors: *J. L. CADET, R. PEESAPATI, S. JAYANTHI;
Mol. Neuropsychiatry, Natl. Inst. On Drug Abuse/ NIH, Baltimore, MD

Abstract: Methamphetamine (METH) use disorder (MUD) is characterized by compulsive and repeated drug intake despite negative life consequences. Large intake of METH in humans and animals is accompanied by dysfunctions in learning and memory processes. The endocannabinoid system (eCB) is known to modulate synaptic plasticity and cognitive functions. The eCB system has also been implicated in the manifestations of substance use disorders (SUDs). We therefore sought to identify potential changes in various enzymes and the receptors

(CB1 and CB2) that are members of the eCB system. Herein, we used a model of METH self-administration (SA) that includes a punishment phase (footshocks) that helps to separate rats into a compulsive METH phenotype (compulsive) that continues to take METH and a non-compulsive METH (abstinent) that suppressed or stopped taking METH in the presence of punishment. Animals were euthanized two hours after the last METH SA session and their hippocampi were used to measure mRNA levels of cannabinoid receptors (*CB/Cnr*), as well as synthetic (*Dagl-a, Dagl-b, Nape-pld*) and metabolizing (*Mgll*, *Faah*, *Ptgs2*) enzymes of the eCB cascade. Non-compulsive rats exhibited significant increased expression of CB1 and CB2 mRNA in the hippocampus. mRNA levels of the synthetic enzyme, *Dagl-a*, and of the metabolic enzymes, *Mgll, and Faah*, were also increased in the hippocampus. Non-compulsive rats also exhibited a significant decrease in Ptgs2 mRNA levels in the hippocampus. Taken together, these observations implicate the hippocampal endocannabinoid system in the suppression of METH intake in the presence of adverse consequences.
Acknowledgement: This work is supported by DHHS/NIH/NIDA/IRP.

Disclosures: J.L. Cadet: None. **R. Peesapati:** None. **S. Jayanthi:** None.

Poster

P745. Molecular and Neural Mechanisms III

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Program #/Poster #: P745.05

Topic: G.09. Drugs of Abuse and Addiction

Support: Korean Ministry of Education, NRF-2016R1A6A1A03011325
Korean Ministry of Education, 4th BK21 project I20SS7606145

Title: The effect of alpha-pinene on methamphetamine-induced conditioned place preference in C57BL/6 mice

Authors: *C. LEE[1], B.-K. DO[2], G. H. PARK[2], J.-H. JANG[1];
[1]Dept. of Pharmacol., Sch. of Medicine, Keimyung Univ., Daegu, Korea, Republic of; [2]Col. of Pharmacy, Kyungpook Natl. Univ., Daegu, Korea, Republic of

Abstract: Methamphetamine (METH) is a powerful neurotoxic psychostimulant drug affecting dopamine transporter (DAT) activity and leading to blockage of dopamine reuptake, which resulted in continuous excess extracellular dopamine levels between pre- and post-synaptic neurons. In this study, we have tried to elucidate a new molecular regulator involved in METH-induced reward and drug addiction. METH (1 mg/kg/day, *i.p.*) was injected to C57BL/6 mice for four alternative days and locomotor as well as conditioned place preference (CPP) tests were performed as behavior analysis. METH-administered group exhibited and increased sensitivity to place preference compared with saline-treated control group on CPP tests. METH significantly

decreased dopamine-related markers such as dopamine 2 receptor (D2R), and tyrosine hydroxylase (TH). Moreover, METH caused apoptotic cell death by induction of inflammation and oxidative stress. Furthermore we tried to investigate the effect of α-pinene (APN) (3 and 10 mg/kg, i.p) on METH-mediated CPP and elucidate underlying molecular mechanisms. METH-mediated place preference was reduced by pre-treatment of APN. APN restored the levels of D2R and TH in striatum of C57BL/6 mice. APN increased Bcl-2 to Bax ratio and conversely decreased the expression of Iba-1. METH-led to lipid peroxidation was effectively attenuated by APN via augmentation of antioxidant capacity. These findings suggest that APN may be considered as a promising therapeutic agent for METH-induced drug addiction and neuronal damages.

Disclosures: C. Lee: None. **B. Do:** None. **G.H. Park:** None. **J. Jang:** None.

Poster

P745. Molecular and Neural Mechanisms III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P745.06

Topic: G.09. Drugs of Abuse and Addiction

Support: Colombo Plan Drug Advisory Program

Title: Levamisole as a methamphetamine adulterant in mice: lethal, psychostimulant, and hematological effects

Authors: *Z. P. ESPINOSA-RIQUER[1], A. K. RUIZ-QUIÑONEZ[1], T. BROWNE, Jr.[2], S. L. CRUZ[1];
[1]CINVESTAV, IPN, Mexico City, Mexico; [2]Colombo Plan Secretariat. Drug Advisory Program, Colombo, Sri Lanka

Abstract: Levamisole (Lvm) is an anthelmintic drug commonly found as an adulterant of psychostimulant street drugs such as cocaine. Lvm addition to stimulant drugs seems to potentiate their effects compromise users' health and immunological responses. However, few preclinical studies have addressed the consequences of combining Lvm with psychostimulants. Here, we investigated the lethal, stimulant, and hematological effects of methamphetamine (Meth) combined with Lvm in male Swiss Webster mice. By using independent groups of animals (n=10), we conducted the following experiments: a) mice were i.p. injected with the lethal dose at 10% (LD_{10}) of Meth (5.6 mg/kg), Lvm (31.6 mg/kg), or Meth plus Lvm at the same doses; b) same as in a, but animals received mecamylamine (2.5 mg/kg) 20 min before Lvm or Lvm plus Meth; c) mice received 17.7 mg/kg Lvm twice daily for two weeks. In the third week, animals were divided to receive one of the following treatments: same Lvm treatment, or Lvm plus Meth (0.1 mg/kg; 3x/day until completing 19 administrations). Data were compared

with those obtained from animals treated with 0.1 mg/kg Meth alone (19 administrations) or saline solution (3 weeks). In addition, we evaluated psychostimulant effects by recording locomotion activity on the actimeter in response to 1 mg/kg Meth, alone or in combination with 17.7 mg/kg Lvm. In selected groups, we conducted automated hematological analyses. We found that Lvm increased Meth lethality and this effect was not prevented by 2.5 mg/kg mecamylamine. Lvm also prolonged the hyperlocomotion produced by Meth and worsen leucopenia. By itself, Lvm increased albumin and alanine aminotransferase but did not have additive effects when combined with Meth on other blood chemistry parameters. In conclusion, the addition of Lvm to Meth increases the risk of death and infections, and potentiates the stimulant effects, making them last longer.

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Poster

P745. Molecular and Neural Mechanisms III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P745.07

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH

Title: Development of CB_2 Receptor Selective Agonists for the Treatment of Stimulant Abuse Disorders

Authors: *A. T. ADAM[1], M. SHAMLOO[3], H. PARK[2], J. S. LIN[2], M. GREEN[2], A. JAHANGIR[2];
[1]Neurosurg., [2]BFNL, Stanford Univ., Palo Alto, CA; [3]Neurosurg., Stanford Univ. Sch. of Med., Palo Alto, CA

Abstract: As of 2019, 20.4 million Americans suffered from a substance use disorder. Stimulant abuse is a major contributor to the extension and severity of this social crisis with 1 million suffering from methamphetamine use disorder. Notably, this data is prior to the onset of the COVID-19 pandemic, which threatens a substantial increase in substance use disorders. This increase is anticipated for all age groups due to the stress and isolation caused by the pandemic. Unfortunately, there aren't any FDA-approved pharmacological treatment for stimulant use disorder. This challenge highlights an urgent need to develop non-opioid pharmacological interventions for stimulant addiction, particularly one that does not target the psychoactive opioid receptors. Therefore, it can be prescribed to patients for safe at home use without the risk of developing a dependency. The addiction effects elicited by this methamphetamine is believed to be mediated by cannabinoid receptors in the brain. The cannabinoid receptors 1 and 2 (CB_1R

and CB_2R, respectively) are part of the endocannabinoid system, which play crucial roles in modulating dopamine (DA) levels in the central nervous system (CNS). The inhibition of CB_2 receptor in the brain, in the ventral tegmental area, leads to a decrease in DA release in the nucleus accumbens. This creates new opportunities for targeting this pathway for therapeutic development. We hypothesize that activation of CB_2R is directly linked to the observed anti-addictive properties of the cannabinoids. Currently, our medicinal chemistry and pharmacology efforts focused on the following target profile for selection of the CB_2R agonist: 1) Potent CB_2R agonist; 2) High receptor selectivity (>100-fold over CB_1R and β-arrestin); 3) Good *in vitro* and *in vivo* pharmacokinetics; and 4) High CNS penetration. We have successfully identified a lead series of agonist compounds from extensive SAR, which display functional biased for cyclase over cAMP β-arrestin, highly selective for CB_2R with minimal activity in CB_1R. The lead candidates, CBS-136, CBS-139, CB-149, and CBS-159, are orally bioavailable, brain penetrable, and efficacious in reversing addictive behavior in mice model in nanomolar concentrations. We established 10-250-fold selectivity *in vitro* against closely related receptors, CB_1 and cyclase β-arrestin. We see this biological activity translating into hyperactivity studies in mice stimulated with D-amphetamine. Our goal is to optimize these leads into a preclinical/clinical candidate to treat stimulant use disorder.

Disclosures: A.T. Adam: None. **M. Shamloo:** None. **H. Park:** None. **J.S. Lin:** None. **M. Green:** None. **A. Jahangir:** None.

Poster

P745. Molecular and Neural Mechanisms III

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Program #/Poster #: P745.08

Topic: G.09. Drugs of Abuse and Addiction

Support: JSPS KAKENHI Grant Number JP18H01097
The Sasakawa Scientific Research Grant

Title: The relationship between risk-taking behaviors and methamphetamine-induced conditioned place preference in rats

Authors: *K. TAKAHASHI[1], Y. ICHITANI[1], K. YAMADA[2];
[1]Univ. of Tsukuba, Tsukuba, Japan; [2]Univ. Tsukuba, Tsukuba, Japan

Abstract: It is known that patients with gambling disorder and substance use disorder tend to exhibit maladaptive decision-making. We previously reported that risk-taking rats, which were assessed with a rat version of the Iowa gambling task (rGT) using a radial arm maze, were more likely to be impulsive and more vulnerable to drug dependence (Takahashi et al., 2020). In the present study, we assessed individual differences in risk-taking behavior using operant behavior

(lever press) and investigated the relationship between risk-taking behaviors and vulnerability to drug dependence using methamphetamine (MAP)-induced conditioned place preference (CPP). In the rGT, male Long-Evans rats (7-8 weeks old at the beginning) were trained to choose one of two choice levers (a low-risk/ low reward (L-L) lever and a high-risk/ high reward (H-H) lever). While the choice of the L-L lever stably resulted in frequent (87.5%) small rewards (a 45 mg pellet), the choice of the H-H lever resulted in either infrequent (12.5%) large rewards (four pellets) or intermediate (50 %) large rewards (four pellets) according to auditory cues (white noise or 3 kHz tone). The rGT continued until rats exhibited stable responses to each lever. Rats received 16 forced-choice trials and subsequently 20 free-choice trials in the gambling test. In the CPP, the conditioning sessions were conducted for six consecutive days, and rats received the pairing of MAP (0.5 mg/kg, i.p.) with the unpreferred compartment or saline with the other once a day. The pairing was alternately carried out three times each. During the preference test, rats were allowed to explore the chamber for 15 min, and the time spent in each compartment was recorded. The results demonstrated a significant positive correlation between the percentage of choice for the H-H lever, which resulted in infrequent (12.5%) large rewards, and the preference score for the MAP-paired compartment in the preference test. These findings suggest that risk-taking rats are more vulnerable to drug dependence. In addition, we investigated the effects of an acute MAP treatment on the H-H lever choice in ascending (12.5 % to 50 %) and descending (50 % to 12.5%) conditions). In the ascending condition, rats with acute MAP treatment kept a low percentage of choice for the H-H lever during the intermediate (50 %) large rewards trials, which was the most optimal choice. On the other hand, in the descending condition, they tended to keep a high percentage of choice for the H-H lever during the infrequent (12.5 %) large rewards trials. These results suggest that acute MAP treatment produced an obvious tendency toward response perseveration in the gambling tasks.

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Poster

P745. Molecular and Neural Mechanisms III

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Program #/Poster #: P745.09

Topic: G.09. Drugs of Abuse and Addiction

Support: MOST 107-2410-H-004-109-MY3

Title: Differential effects of ketamine isomers and amphetamine on locomotor sensitization and place conditioning in the rat

Authors: C.-Y. WANG, S.-F. CHEN, C.-C. CHAO, *R.-M. LIAO;
Natl. Cheng Chi Univ., Taipei, Taiwan

Abstract: Ketamine is repurposed in depression therapy, though the underlying neurobehavioral mechanisms remain elusive. One critical concern is the psychotropic effects of ketamine that could link to recreational drug use and resultant abuse. Previous studies frequently used an injectable ketamine solution testing its rewarding effects, whereas comparatively fewer studies have explored ketamine isomer(s) on this issue. We investigated the effects of RS (+/-)-ketamine (RS-KET) and S (-)-ketamine (S-KET) on a behavioral protocol combining conditioned place preference (CPP) and behavioral sensitization (BS) paradigms. Additionally, d-amphetamine (d-AMP) treatment was conducted as a positive control. In a 30-day test, the CPP was conducted by either the unbiased or the biased procedure; and, BS test were run across three phases including a pre-test and a post-test conducted respectively before and after the development phase (7 days of drug injection intermittently separated by saline injects). The drug and saline injections were respectively conducted in two different compartments as applied for the conditioning stage of CPP test. The dose given at the pre- and post-tests was lower (i.e., a half) than that administered in the development phase. For the results, in contrast to that d-AMP (1 mg/kg) significantly produced CPP, neither RS-KET (10 mg/kg) nor S-KET (10 mg/kg) produced CPP via the test with unbiased or biased procedure. However, a CPP was detected in the long-lasting effects test for S-KET treated rats run via biased conditioning procedure. BS appeared markedly to d-AMP treatment which was also profound in the long-lasting test. In RS-KET treated groups, rats run through either the biased or unbiased protocol displayed BS in the development phase and the long-lasting test. However, BS only appeared in the RS-KET treated rats in unbiased but not in biased group. S-KET treatment failed to form BS except that in unbiased group during the development phase. These data indicated that 1) KET isomers differ from d-AMP producing task-dependent effects of drug rewarding, 2) the CPP induced either KET isomers is subtle as compared with d-AMP, and 3) the degrees of BS to KET isomers are lower than d-AMP and varied in different test stages.

Disclosures: C. Wang: None. **S. Chen:** None. **C. Chao:** None. **R. Liao:** None.

Poster

P745. Molecular and Neural Mechanisms III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P745.10

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH DA029815

Title: Effects of extended-access methamphetamine self-administration on dopamine and cAMP-regulated phosphoprotein (DARPP-32) expression in rat perirhinal cortex and hippocampus

Authors: Y. CAM[1], L. K. CARRICA[3], S. R. WESTBROOK[3], J. M. GULLEY[3,4], *B. A. HEIDENREICH[1,2];
[1]Psychology, [2]Biol. Sci., Illinois State Univ., Normal, IL; [3]Psychology, [4]Neurosci. Program, Univ. of Illinois, Urbana-Champaign, IL

Abstract: Methamphetamine (METH) abuse is a major health concern associated with behavioral disturbances, impaired memory and cognitive deficits. Repeated METH self-administration in rats can produce behavioral tolerance or sensitization and numerous changes in the dopamine (DA) neurotransmitter system. Previously, we investigated the influence of age and sex on the effects of extended access self-administration (6h/day for 14 sessions) of METH or saccharin in Sprague-Dawley rats. Adolescent-onset rats showed a faster escalation of METH intake compared to their adult-onset counterparts, and showed a small decrease in performance on an object-in-place test of recognition memory during a 7-14 day abstinence period. Interestingly, there was no long-lasting effect of METH on dopamine D1 receptor protein levels in nucleus accumbens or prefrontal cortex after 21 days of abstinence.
Dopamine and cAMP-regulated phosphoprotein (DARPP-32) is produced by activation of DA D1 receptors and plays an important role in mediating the short- and potentially long-term effects of drugs of abuse, including dopaminergic stimulants. To further investigate the effects of METH self-administration on DA neurotransmission and the neural substrates of recognition memory, we performed immunohistochemistry for DARPP-32 on the hippocampus and perirhinal cortex of rats from our previous study. Forty micron thick cryostat sections were incubated in primary anti-DARPP-32 antibody, which was then visualized using the avidin-biotin-peroxidase method with 3,3'-diaminobenzadine as the chromogen. DARPP-32 expression in layers V and VI of the perirhinal cortex did not differ between rats that self-administered saccharin or METH. A two-way ANOVA showed a significant main effect ($p = .02$) of the region of the hippocampus examined; the dentate gyrus and CA1 region showed greater intensity of DARPP-32 immunostaining than CA3. There was no main effect of METH vs. saccharin self-administration, suggesting that METH did not cause persistent effects on hippocampal DARPP-32 levels. Together with our previous findings, our results suggest that the effects of METH self-administration on DA D1-mediated neurotransmission do not persist after a 21-day abstinence period.

Disclosures: Y. Cam: None. **L.K. Carrica:** None. **S.R. Westbrook:** None. **J.M. Gulley:** None. **B.A. Heidenreich:** None.

Poster

P765. Nicotine: Neural Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P765.01

Topic: G.09. Drugs of Abuse and Addiction

Support: IITP Grant (No. 2020 0 01450, Artificial Intelligence Convergence Research Center [Pusan National University]
BK21 Four, Korean Southeast Center for the 4th Industrial Revolution Leader Education
UNIST Internal grant (1.210046.01)
National Research Foundation of Korea (2018R1D1A1B07043582 to D.C.)

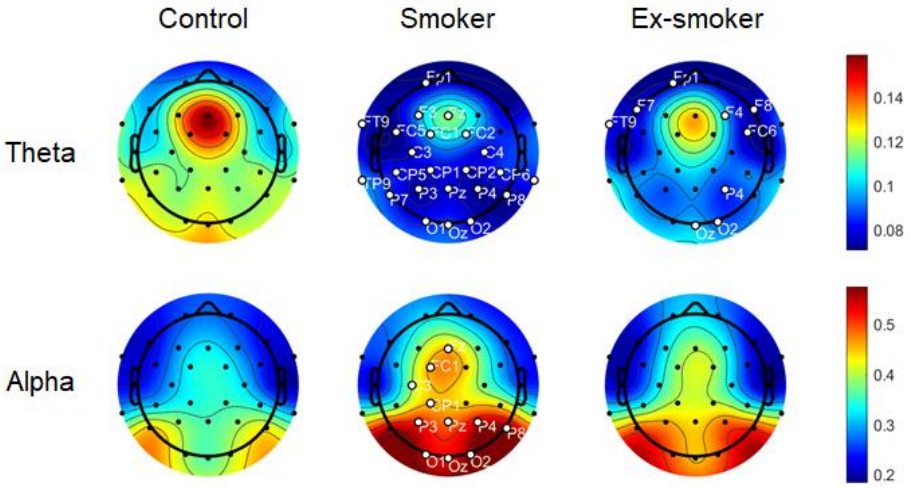
Title: Resting-state EEG in heavy chronic smokers, ex-smokers and non-smokers

Authors: Y. JEON[1], H. LEE[2], J. JEONG[1], C.-I. YOO[3], ***K. BAEK**[1], D. CHUNG[2];
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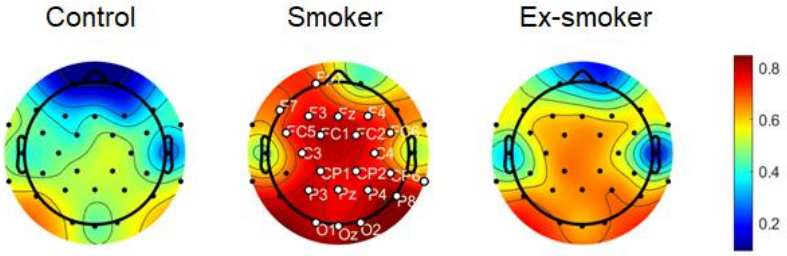
Abstract: Smoking is a severely chronic addiction. There have been several studies with smokers seeking for the EEG biomarkers, but it is not fully understood how the long-term use of cigarettes alters the resting state EEG and whether such changes can be restored after quitting. Here we examined the resting state EEG from 20 chronic heavy smokers (20 cigarettes per day for more than 20 years), 33 non-smokers and 23 ex-smokers with a long abstinence period (mean: 11.9 years). Resting state EEG was recorded using a cap with high density 31-electrodes using actiCHamp (Brain Products, Germany) for 3 minutes during the eyes-open and the eyes-closed conditions, respectively. EEG data was filtered with the off-line digital bandpass filter (1~50 Hz) after rejecting the artifacts. We compared the relative band power, EEG reactivity (relative change of band power from the eyes-closed to the eyes-open condition) and coherence. As shown in Fig.1, the smokers showed altered resting state EEG band power at the eye-closed condition: decrease in theta power and increase in alpha power when compared to the non-smokers ($p < 0.05$, FDR corrected). The ex-smokers showed decrease in theta power for the frontal and occipital regions but not increase in alpha power. Alpha reactivity, i.e. alpha desynchronization from the eyes-closed to the eyes-open condition, was much strongly manifested in the smokers compared to the non-smokers and the ex-smokers ($p < 0.05$, FDR corrected. See Fig. 1B). In the resting brain network (eyes-closed condition), the smoker exhibited significantly increased alpha coherence compared to the nonsmokers (across the whole brain) and the ex-smokers (primarily with the frontal areas). Increased coherence between left frontal/temporal and the occipital regions were also observed in the ex-smokers. These results suggest that chronic smoking may alter the resting state EEG activity and functional connectivity particularly in alpha band. Increase in alpha reactivity might be associated with changes in cholinergic influence from the basal nucleus of Maynert (Wan et al., Human Brain Map. 2019)

Figure 1. Resting state EEG characteristics in the non-smoking controls, the smokers and the ex-smokers. All $p < 0.05$ (FDR corrected)

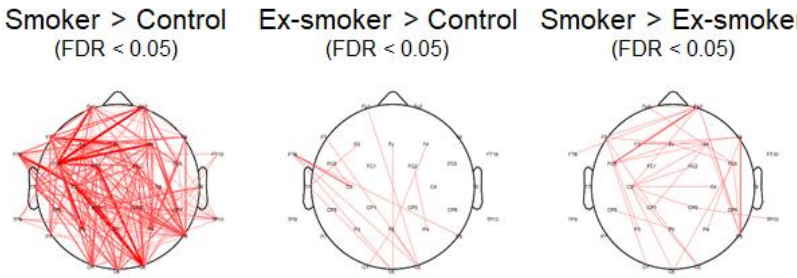
(A) Relative EEG (Eye-closed)



(B) Alpha Reactivity



(C) Coherence (Alpha band)



Disclosures: Y. Jeon: None. **H. Lee:** None. **J. Jeong:** None. **C. Yoo:** None. **K. Baek:** None. **D. Chung:** None.

Poster

P765. Nicotine: Neural Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P765.02

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant R01DA044760-01
NIH Grant P30CA14599
NIDA Grant T32DA043469

Title: Positron emission tomography (PET) in vivo imaging of 2-[^{18}F]-A85380 trapping in α4β2 receptor-containing acidic vesicles

Authors: C.-M. KAO[1], H. ZHANG[1], ***M. ZAMMIT**[1], N. HOLDERMAN[1], H.-M. TSAI[1], A. GOVIND[1], S. MITCHELL[1], M. BHUIYAN[1], R. FREIFELDER[1], X. ZHUANG[1], J. MUKHERJEE[2], C.-T. CHEN[1], W. GREEN[1];
[1]Univ. of Chicago, Chicago, IL; [2]Univ. of California, Irvine, CA

Abstract: 2-[^{18}F]-A85380 (2-FA) is a weak base PET radioligand that binds with high affinity to α4β2 subtype of nicotinic acetylcholine receptors (nAChRs). Previous work has identified that the smoking cessation drug varenicline (Chantix) becomes trapped in intracellular acidic vesicles containing α4β2Rs and is slowly released over time, whereas nicotine is not trapped. The purpose of this PET study in mice was to evaluate whether the slow in vivo kinetics of 2-FA stems from acidic vesicle ligand trapping, similar to varenicline. 180-minute dynamic 2-FA PET scans were performed on wild type (WT; 5M/5F), β2 nAChR knockout (KO; 8M/3F), acute nicotine-administered (AN; 5M/5F) and chloroquine diphosphate-treated wild type mice (CQWT; 3M/3F) in a β-cube preclinical microPET scanner. Mice were 3-10 months of age, and no sex differences were explored in this study. AN mice were administered 0.5 mg/kg body weight (BW) of nicotine IP 15 minutes before 2-FA injection. CQWT mice were administered 50 mg/kg BW/day of CQ IP for 3 days, with 2-FA PET performed 10 minutes after the final injection. CQ was administered to evaluate whether dissipating the pH gradient across α4β2R-containing acidic vesicles reduced 2-FA binding. Time activity curves were generated using standardized uptake values (SUVs) from the thalamus and midbrain normalized to the cerebellum. Using a cerebellum reference region, distribution volume ratios (DVRs) were determined for each ROI using Logan graphical analysis. Normalized time activity curves revealed reduced binding of 2-FA in the thalamus and midbrain of KO, AN and CQWT mice compared to WT mice. Significant differences in DVRs were observed between groups for the thalamus (ANOVA F(df)=37(3); p<.00001) and midbrain (59(3); p<.00001). Post hoc t-tests revealed that KO, AN, and CQWT mice had significantly reduced DVR compared to the WT mice (all p<.00001 adjusted using Tukey's HSD method). Uptake of 2-FA in KO mice was completely abolished, while AN mice revealed reduced specific binding of 2-FA, indicative of nicotine blocking at the binding sites. CQWT mice revealed significantly reduced uptake of 2-FA, which may indicate that pH neutralization from CQ administration is preventing 2-FA trapping within intracellular acidic vesicles. Together, these findings suggest that the slow in vivo kinetics of 2-FA are influenced by trapping in acidic vesicles containing α4β2Rs similar to our previous results in vitro with varenicline and epibatidine. Our findings highlight the utility of

PET to monitor α4β2R ligand trapping in vivo. All animal procedures were approved by the IACUC of the University of Chicago in accordance with NIH guidelines.

Disclosures: C. Kao: None. **H. Zhang:** None. **M. Zammit:** None. **N. Holderman:** None. **H. Tsai:** None. **A. Govind:** None. **S. Mitchell:** None. **M. Bhuiyan:** None. **R. Freifelder:** None. **X. Zhuang:** None. **J. Mukherjee:** None. **C. Chen:** None. **W. Green:** None.

Poster

P765. Nicotine: Neural Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P765.03

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH GM-123582
NIH DA043829
California TRDRP 27FT-0022
California TRDRP 27IP-0057
HHMI

Title: Genetically Encoded Biosensors Show Subcellular Pharmacokinetics of Nicotinic Ligands

Authors: *A. NICHOLS[1], Z. BLUMENFELD[1,3], L. LUEBBERT[1,4], C. FAN[1], P. M. BORDEN[5], A. E. M. BLOM[2], B. N. COHEN[1], J. S. MARVIN[5], C. KIM[1], A. K. MUTHUSAMY[2], H. J. KNOX[2], J. H. WANG[1], L. L. LOOGER[5], T. GALLAGHER[6], D. C. REES[1], D. A. DOUGHERTY[2], H. A. LESTER[1];
[1]Div. of Biol. and Biol. Engin., [2]Div. of Chem. and Chem. Engin., Caltech, Pasadena, CA; [3]Keck Sch. of Med., USC, Los angeles, CA; [4]Inst. of Biol., Leiden Univ., Leiden, Netherlands; [5]Janelia Res. Campus, HHMI, Ashburn, VA; [6]Sch. of Chem., Univ. of Bristol, Bristol, United Kingdom

Abstract: Nicotinic agonists play important roles in studying addiction, withdrawal, neuroprotection, cognitive behavior, and neural pharmacology. Previous work with nicotine showed that, in addition to actions on the plasma membrane such as receptor activation and desensitization, pharmacological chaperoning by nicotine begins intracellularly in organelles such as the endoplasmic reticulum (ER). Previous work also showed that nicotine and varenicline enter the ER. We now report genetically encoded fluorescent biosensors for the additional nicotinic agonists dianicline, cytisine, 10-fluorocytisine, and 9-bromo-10-ethylcytisine, which allow comparisons among subcellular pharmacokinetics of nicotinic drugs. Our iDrugSnFRs ("**i**ntensity-based **Drug**-**S**e**n**sing **F**luorescent **R**eporters") utilize mutated OpuBC, a monomeric bacterial periplasmic binding protein (PBP), which features (a) a binding site for amines including a cation-π box, and (b) ligand-induced "Venus flytrap" conformational change invoked by binding of target ligand. We inserted circularly permuted "superfolder" GFP

(cpGFP), flanked by several-residue linkers, near inter-domain hinge regions and applied directed evolution, including X-ray crystallography, to optimize sensing for each drug of interest. Each iDrugSnFR can detect its drug partner with responses of $\Delta F/F0 > 1$ at 1 μM. Using targeting and retention sequences, we directed the constructs to the ER or to the PM of clonal mammalian lines and cultured neurons. Live-cell video imaging (1 - 1000 s) shows that the kinetics of ER entry/exit differ among these nicotinic drugs by > 10-fold. The difference in these subcellular pharmacokinetic properties demonstrates how functional group addition to a parent compound can vastly alter its membrane permeability. With purified proteins in solutions, we examined the thermodynamics and kinetics of biosensor binding/unbinding to their respective drug partner. The biosensor-drug bindings are entropically driven to variable extents among the drug-iDrugSnFR pairs. The kinetics of binding showed several components, suggesting a four-state scheme of biosensor binding, which may extend to other Opu-BC based biosensors. iDrugSnFRs for exogenous molecules provide additional insights into aspects of nicotinic agonists, pharmacokinetics, organellar sequestration of drugs by acid trapping, protein trafficking, and upregulation—all crucial facets in the expanded understanding of "inside-out" neuropharmacology of neural drugs.

Disclosures: A. Nichols: None. **Z. Blumenfeld:** None. **L. Luebbert:** None. **C. Fan:** None. **P.M. Borden:** None. **A.E.M. Blom:** None. **B.N. Cohen:** None. **J.S. Marvin:** None. **C. Kim:** None. **A.K. Muthusamy:** None. **H.J. Knox:** None. **J.H. Wang:** None. **L.L. Looger:** None. **T. Gallagher:** None. **D.C. Rees:** None. **D.A. Dougherty:** None. **H.A. Lester:** None.

Poster

P765. Nicotine: Neural Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P765.04

Topic: G.09. Drugs of Abuse and Addiction

Support: Virginia Youth Tobacco Programs (VYTP/VCU)
Virginia Foundation for Healthy Youth

Title: Age and sex differences in the effects of nicotine on tyrosine hydroxylase protein levels and DARPP-32 phosphorylation at Ser130 in mice

Authors: T. L. PROFFITT[1], A. D. HUDSON[2], *K. J. FRYXELL[1];
[1]Sch. of Systems Biol., George Mason Univ., Manassas, VA; [2]Dept. of Reproductive Med., Univ. of California at San Diego, La Jolla, CA

Abstract: Nicotine and other drugs of abuse can cause dysregulation of the dopaminergic system. Tyrosine hydroxylase (TH) and Dopamine-and-cAMP-regulated phosphoprotein (DARPP-32) play important roles in dopamine synthesis, and dopamine signaling, respectively.

Nicotine acts directly on nicotinic acetylcholine receptors (nACHRs) in the brain and indirectly stimulates TH gene transcription, as well as stimulating dopamine release from mesocorticolimbic neurons. TH is the rate-limiting enzyme for the synthesis of catecholamines (including dopamine). Ca^{2+} currents that are stimulated by nAChR tend to increase TH gene expression. DARPP-32 integrates both glutamatergic and dopaminergic signaling via phosphorylation events at distinct sites within the DARPP-32 protein. In particular, nAChR signaling affects DARPP-32 (via glutamatergic and dopaminergic signaling), in part by stimulating protein phosphatase-2b (PP2b) activity, which indirectly activates casein kinase 1 (CK1); and also by stimulating inositol triphosphate (IP3) signaling, which increases CK1 gene expression. CK1 phosphorylates DARPP-32 at Ser130. In this study, TH protein levels and phosphorylation of DARPP-32 at Ser130 were measured in the ventral tegmentum (VT), ventral striatum (VS), and medial prefrontal cortex (mPFC) of adolescent and adult mice after subcutaneous nicotine injections. In the VT, we found a highly-significant nicotine-induced decrease in TH in adult males, but not in adult females. These findings were presumably influenced by the suppressive effect of androgens on tyrosine hydroxylase expression in the VT. DARPP-32 phospho-Ser130 decreased in the VT of both sexes of adolescent mice (but not adults) after repeated nicotine injections. This age-specific, nicotine-induced decrease in phospho-Ser-130 is likely to make adolescents more vulnerable to nicotine dependence. In the mPFC, nicotine injections decreased TH protein in adolescent females (prior to puberty), but increased TH protein in adult females, possibly due to the effects of estrogen levels on TH expression. In the mPFC, nicotine injections decreased DARPP-32 phospho-Ser130 in adult males, but increased it in adult females. Several precedents suggest that estrogen signaling may have intensified the nicotine-induced glutamatergic signal, and thus increased the net effects of nicotine on both TH and DARPP-32 phospho-Ser130 in adult female mice. If so, this may provide a novel basis for the vulnerability of females to nicotine dependence after puberty.

Disclosures: T.L. Proffitt: None. **A.D. Hudson:** None. **K.J. Fryxell:** None.

Poster

P765. Nicotine: Neural Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P765.05

Topic: G.09. Drugs of Abuse and Addiction

Support: TRDRP T30IP0931

Title: Lynx2 modulates neuronal activation in the VTA and the IPN

Authors: *M. BAUTISTA[1], J. P. FOWLER[2], C. D. FOWLER[3];
[1]Neurobio. and Behavior, Univ. Of California, Irvine, Irvine, CA; [2]Neurobio. and Behavior,

Univ. of California, Irvine, Irvine, CA; [3]Neurobio. and Behavior, Univ. of California Irvine, Irvine, CA

Abstract: Tobacco-use results in damage to several major organs in the body and remains the highest cause of preventable death and disease in the United States. However, currently available methods for nicotine cessation are only moderately efficacious, thereby creating a need for new therapeutics with endogenous allosteric modulators emerging as targets of interest for drug development. The lynx2 protein is a negative allosteric modulator of the nicotinic acetylcholine receptor (nAChR) and regulates cellular activity within the cholinergic system. Here, we used *lynx2* knockout mice and their wildtype littermates to explore the role of lynx2 in cellular activation, following acute nicotine administration. We quantified c-fos positive cells using unbiased stereology in the ventral tegmental area (VTA) and the interpeduncular nucleus (IPN), which are associated with nicotine reward and nicotine aversion, respectively. Our data show that Lynx2 knockout mice have altered cellular activation in both the VTA and IPN in comparison to their wildtype littermates following acute nicotine administration. Together, these findings demonstrate that Lynx2 contributes to mediating nicotine's actions in both the VTA and IPN, thereby illustrating its potential role in nicotine-related behaviors.
This work was supported by funding from the Tobacco-Related Disease Research Program (TRDRP T30IP0931 to CDF).

Disclosures: M. Bautista: None. **J.P. Fowler:** None. **C.D. Fowler:** None.

Poster

P765. Nicotine: Neural Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P765.06

Topic: G.08. Other Psychiatric Disorders

Support: 1R15DA046926-01A1

Title: An investigation of the associative rewarding aspects of nicotine in a novel heritable model of drug abuse vulnerability.

Authors: *L. D. PEETERS[1], L. J. WILLS[2], *S. E. TURNEY[1], L. P. BULLOCK[1], W. GILL[1], C. VIED[3], J. T. GASS[1], R. W. BROWN[1];
[1]Biomed. Sci., [2]Psychology, East Tennessee State Univ., Johnson City, TN; [3]Florida State University, Col. of Med., Tallahassee, FL

Abstract: Our laboratory established that neonatal treatment of the dopamine (DA)D_2-like receptor agonist quinpirole (NQ) to rats induces an increase in DAD_2 receptor sensitivity throughout the animal's lifetime, which has validity to a number of clinical conditions which

have high rates of substance abuse comorbidity. Based on this permanent change in DAD_2 sensitivity, we bred adult F0 generation NQ-treated male and female rats with their NQ-treated or neonatal saline (NS)-treated counterparts to determine whether increases in DAD_2 sensitivity were passed to the next generation. Results revealed that offspring of these animals also demonstrated increased DAD_2 signaling on both a yawning behavioral test, which is a behavioral profile of DAD_2 signaling, and produced changes in analyses of DAD_2 signaling proteins β-arrestin-2 and phospho-AKT consistent with increased DAD_2 activity. Based on these data, we analyzed genetic changes using RNASeq, and found that the F1 offspring of two NQ-treated rats demonstrated dramatic increases in cortisol synthesis and release, suggesting these animals demonstrate an enhanced sensitivity to stress, consistent with a model of drug abuse vulnerability. Therefore, we analyzed behavioral responses to nicotine on the conditioned place preference (CPP) task in these F1 offspring. Interestingly, F1 offspring of both NQ-treated rats or a male NQ- and female NS-treated pair demonstrated enhanced nicotine CPP in adolescence. Finally, we investigated two novel pharmacological targets to reduce enhanced adolescent nicotine CPP in F1 generation rats. DAD_2 receptors are known to form functional heteromers with several other classes of G protein-coupled receptors (GPCRs), including the adenosine A(2A) and metabotropic glutamate type 5 receptor (mGlu5). Stimulation of A(2A) or mGlu5 has been shown to reduce the affinity of the DAD_2 receptor for its endogenous agonist in striatal membrane preparations. Therefore, we administered the A(2A) agonist CGS 21680 (0.03 or 0.09 mg/kg) or the positive allosteric modulator to mGlu5, CDPPB (10 or 30 mg/kg) before nicotine during conditioning. Results revealed that both of these drugs alleviated enhanced CPP as well as nicotine CPP in F1 generation animals, and CDPPB alleviated nicotine CPP in controls. These results are the first to show that increased DAD_2 sensitivity is passed to the next generation, and this model is a viable heritable model of drug abuse vulnerability. In addition, we revealed two novel pharmacological treatment targets to reduce rewarding aspects of nicotine.

Disclosures: L.D. Peeters: None. **L.J. Wills:** None. **S.E. Turney:** None. **L.P. Bullock:** None. **W. Gill:** None. **C. Vied:** None. **J.T. Gass:** None. **R.W. Brown:** None.

Poster

P765. Nicotine: Neural Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P765.07

Topic: G.09. Drugs of Abuse and Addiction

Support: TRDRP

Title: Acute nicotine intake alters metabolism and brain circuitry in nicotine-dependent rats

Authors: *K. SHANKAR[1,2], O. GEORGE[2];
[1]The Scripps Res. Inst., La Jolla, CA; [2]UC San Diego Hlth., San Diego, CA

Abstract: Nicotine consumption in both human and animal studies has been strongly associated with changes in feeding-related behaviors and metabolism. The current dogma is that nicotine is an anorectic agent that decreases food intake and increases metabolism, leading to decreased body weight gain. However, we have found that acute intake of nicotine in rats through multiple administration models produces a short-term increase in feeding behavior, contrary to long-term anorexigenic effects of nicotine. To elucidate the mechanisms producing this phenomenon, we studied both changes in metabolic signaling as well as brain circuitry following acute nicotine intake. Adult male and female Wistar rats were subcutaneously implanted with nicotine osmotic minipumps and given acute subcutaneous nicotine injection at different timepoints. Plasma samples were collected from these animals at multiple timepoints for downstream metabolic analysis. Another group of animals was subjected to intravenous nicotine self-administration, and following completion of the paradigm, brains were collected for circuit analysis. As our primary behavioral results show differences between non-dependent and dependent animals, our studies focused on comparisons of acute nicotine intake in dependent *vs*. non-dependent rats. Our results challenge the notion that nicotine *per se* is an anorexigenic drug and suggest that nicotine's physiological dysregulation contributes to acute anti-anorectic effects that may lead to long-term anorexigenic effects of nicotine.

Disclosures: K. Shankar: None. **O. George:** None.

Poster

P765. Nicotine: Neural Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P765.08

Topic: G.09. Drugs of Abuse and Addiction

Support: R21 DA046184

Title: Laterodorsal tegmental nucleus mediates negative nicotine withdrawal affective symptoms

Authors: *A. MONICAL, D. S. MCGEHEE;
The Univ. of Chicago, Chicago, IL

Abstract: Tobacco dependence is the leading cause of preventable death worldwide. Understanding how nicotine withdrawal alters excitability of key brain areas associated with addiction is important for developing effective therapeutic interventions and identifying risk factors. Dopamine (DA) release from ventral tegmental area (VTA) projections to the nucleus accumbens (NAc) is important in the induction, progression, and maintenance of addiction. Suppression of VTA DA signaling is one consequence of withdrawal from chronic nicotine use. One of the key presynaptic regulators of VTA DA neuron activity is the laterodorsal tegmental nucleus (LDTg). Previous research has shown GABAergic drive from the interpeducular nucleus

(IPN) to the LDTg is important in the aversive effects of acute high-dose nicotine exposure. While it is known that nicotine withdrawal also activates IPN GABA neurons, the role of the projections from the IPN to the LDTg in nicotine withdrawal has not been tested. We hypothesize that suppression of LDTg projections to VTA mediates aspects of the aversive withdrawal symptoms. Using real-time preference testing with optogenetic inhibition of the GABAergic neurons synapsing within the LDTg, we observed a decrease in the negative affective state associated with nicotine withdrawal, as mice experiencing mecamylamine-precipitated nicotine withdrawal chose to spend more time in the environment paired with this inhibition. This preference persists over three days of testing, after which, animals were assessed for conditioned responses to the previously light-paired and unpaired chambers. Conditioned place testing revealed that preference was state-dependent, as the animals preferred the previously light-paired chamber only when they were experiencing nicotine withdrawal. In addition, we are exploring the impact of IPN GABAergic projections to LDTg in nicotine withdrawal-associated behaviors, including affective state and somatic signs. During optogenetic inhibition of IPN GABAergic terminals in LDTg, we observed a normalization of exploratory behavior that is typically reduced during the withdrawal state, as measured by open field test. As these tests were not associated with a change in locomotor behavior, this suggests that IPN projections to LDTg may contribute to withdrawal associated anxiety.

Disclosures: A. Monical: None. **D.S. McGehee:** None.

Poster

P765. Nicotine: Neural Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P765.09

Topic: G.09. Drugs of Abuse and Addiction

Support: NIDA R21

Title: The Role of the Central Extended Amygdala in Acute Nicotine Abstinence

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Abstract: Tobacco smoking is a leading cause of preventable disease, disability, and death. Relapse is common and existing cessation aids are far from curative for many smokers, underscoring the need to develop a deeper understanding of the underlying neurobiology. While the roots of nicotine addiction are clearly complex, an emerging body of research suggests that,

once established, negative reinforcement mechanisms (smoking to alleviate/avoid anxiety) play a central role in continued nicotine use and relapse. Indeed, heightened signs and symptoms of anxiety are a hallmark of nicotine withdrawal, manifesting in the laboratory as potentiated defensive responses to threat, particularly when threat is uncertain in timing, intensity, or likelihood. Although it is widely believed that withdrawal-related anxiety reflects sensitization of the central extended amygdala (EAc)—including the central nucleus of the amygdala (Ce) and bed nucleus of the stria terminalis (BST)—this hypothesis has yet to be rigorously addressed in humans. Here we used fMRI and the Maryland Threat Countdown paradigm to test this hypothesis in 78 daily tobacco smokers randomized to either smoke as usual or abstain for 24 hours prior to neuroimaging. A multiband pulse sequence, best-practice data processing techniques, spatially unsmoothed data, and anatomically defined Ce and BST regions-of-interest enabled unbiased testing with enhanced anatomical resolution. Results demonstrated that acute abstinence increased anxious distress (in-scanner ratings) and arousal (skin conductance) during the anticipation of aversive stimulation (noxious shock, visual, and auditory stimuli; ps<.01). Abstinence was also associated with heightened EAc activation during the anticipation of aversive stimulation with certain timing (p=.02). Other effects were not significant. These findings prompt further exploration on how acute nicotine withdrawal may interfere with orchestrating appropriate responses to threat and elucidate our understanding of how nicotine addiction may interact with anxiety responses to make smoking cessation challenging. Broadly, this work contributes to building a neurobiological framework of addiction processes in humans.

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Poster

P765. Nicotine: Neural Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P765.10

Topic: G.09. Drugs of Abuse and Addiction

Support: National Institute of Health Grants DA040626 and DA035942

Title: Nicotine Self-Administration Induces Plastic Changes to Nicotinic Receptors in Medial Habenula

Authors: *X.-T. JIN[1], B. R. TUCKER[2], R. M. DRENAN[2];
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Abstract: Nicotine Self-Administration Induces Plastic Changes to Nicotinic Receptors in Medial HabenulaXiao-Tao Jin , Brenton R Tucker , Ryan M Drenan

Chronic nicotine upregulates nicotinic acetylcholine receptors (nAChRs) throughout the brain, and reducing their activity may promote somatic and affective states that lead to nicotine seeking. nAChRs are functionally upregulated in animal models using passive nicotine administration, but whether/how it occurs in response to volitional nicotine intake is unknown. The distinction is critical, as drug self-administration (SA) can induce neurotransmission and cellular excitability changes that passive drug administration does not. In this study, we probed the question of whether medial habenula (MHb) nAChRs are functionally augmented by nicotine SA. Male rats were implanted with an indwelling jugular catheter and trained to nose poke for nicotine infusions. A saline SA group controlled for non-specific responding and nicotine-associated visual cues. Using patch-clamp whole-cell recordings and local application of acetylcholine, we observed robust functional enhancement of nAChRs in MHb neurons from rats with a history of nicotine SA. To determine whether upregulated receptors are generally enhanced or directed to specific cellular compartments, we imaged neurons during recordings using two-photon laser scanning microscopy (2PLSM). nAChR activity at the cell soma and on proximal and distal dendrites was examined by local nicotine uncaging using a photoactivatable nicotine (PA-Nic) probe and focal laser flash photolysis. Results from this experiment revealed strong nAChR enhancement at all examined cellular locations. Our study demonstrates nAChR functional enhancement by nicotine SA, confirming that volitional nicotine intake sensitizes cholinergic systems in the brain. This may be a critical plasticity change supporting nicotine addiction.

Disclosures: X. Jin: None. **B.R. Tucker:** None. **R.M. Drenan:** None.

Poster

P765. Nicotine: Neural Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P765.11

Topic: G.09. Drugs of Abuse and Addiction

Support: NIDA Grant DA046526
NIDA Grant DA045881

Title: Activation of accumbens microglia and adenosine A2a receptors contributes to nicotine self-administration and cue-induced-reinstatement

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Abstract: Neuroimmune mechanisms within the nucleus accumbens core (NAcore) are involved in nicotine seeking behavior. Further, microglia are activated following withdrawal from experimenter-delivered nicotine, and this is a critical neuroimmunological response in the

nicotine-induced cellular signaling cascade. Adenosine 2 receptors (A2aRs) regulate the proliferation and survival of microglia, and have been found to trigger microglial activation and inflammatory cytokine release. Systemically agonizing A2aRs effectively reduces ethanol self-administration and cocaine reinstatement, but these receptors have not been thoroughly studied for their role in nicotine-motivated behaviors. In the current studies, Long Evans male and female rats underwent nicotine self-administration (0.06 mg/kg/infusion) followed by extinction training in which rats were administered with A2a agonist (CGS 21680, 0.4 mg/kg i.p.), A2a antagonist (SCH 58261, 0.4 mg/kg i.p.), or vehicle. One cohort of animals were used to quantify NAcore microglia morphology following nicotine self-administration while a second cohort received A2a agonist/antagonist treatments through extinction and prior to cue-induced reinstatement. Results show altered morphology indicative of microglia activation, and an increase in A2a expression within the NAcore following chronic nicotine-self administration. Pretreatment with systemic injections of CGS 21680 resulted in the significant decrease of cue-induced nicotine seeking compared to SCH 58261-treated animals. These results indicate that A2a activation counteracts nicotine seeking behavior. Ongoing studies are determining if this is mediated by microglia.

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Poster

P778. Modeling and Behavior III

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Program #/Poster #: P778.01

Topic: H.03. Decision Making

Support: ARL W911NF-19
PIP 11220150100787CO
PICT 2018-02699

Title: cIBS: an ideal Bayesian observer for visual search in natural scenes

Authors: M. SCLAR[1], ***G. BUJIA**[1,2], S. VITA[1], G. RUARTE[1], F. TRAVI[1], G. SOLOVEY[2], J. KAMIENKOWSKI[1,3];
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Abstract: Visual search is a vital task that humans perform systematically on an everyday basis. In the last decades, thanks to the advance of deep neural networks, there was a large

development of models that predict the most likely gaze fixation locations when observing a scene (saliency maps). Today, one of the biggest challenges in the field is to go beyond these saliency maps to predict the exact sequence of fixations and tasks different from simple observation. In visual search tasks, Bayesian observers have been proposed to model the visual search behavior as an active sampling process. In this process, during each fixation, humans incorporate new information and update the probability of finding a target at every location. Here, we combine these approaches for visual search in natural images and propose a model to predict the whole scanpath. Our Ideal Bayesian Searcher (IBS) uses a saliency map as prior and computes the most likely next location given all the previous fixations, considering visual properties of the target and the scene. We collected eye-movement visual search data (N=57) in 134 natural indoor scenes and compared different variants of the model and its parameters. Firstly, different state-of-the-art saliency maps performed similarly to previous reports only on the first fixations. After the third fixation, their performance rapidly decayed to almost chance. This suggests that saliency maps are not able to capture fixations distributions when top-down information is critical. Secondly, we introduced different variants of the IBS model to deal with natural scenes, as the priors and measure of similarity between the target and the whole scene. We showed that IBS models using saliency-based priors produced scanpaths that were more similar to humans than using simpler priors, both in the percentage of target found as a function of the fixation rank and the scanpath similarity, reproducing the entire sequence of eye movements. Thirdly, the IBS model presented here behaves more similarly to humans than other state-of-the-art scanpath prediction models based on deep neural networks in the present dataset with scenes of interiors with multiple distractors. Overall, our results and others showed that the Bayesian observer is a natural framework to model the top-down processes, such as the integration of information and predictions, that influence visual search and also guide eye movements beyond the saliency models.

Disclosures: M. Sclar: None. **G. Bujia:** None. **S. Vita:** None. **G. Ruarte:** None. **F. Travi:** None. **G. Solovey:** None. **J. Kamienkowski:** None.

Poster

P778. Modeling and Behavior III

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Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P778.02

Topic: H.03. Decision Making

Title: Discovering the temporal correlation between physiological states and risk preferences

Authors: *Z.-Y. YAN[1], P. W. GLIMCHER[2];
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Abstract: Economics has traditionally viewed preferences, for example risk attitude, as immutable features of the chooser that lie outside of models. There is, however, some evidence that preferences are impacted by both biological and environmental variables. Both human and animal experiments have shown that internal metabolic states can impact risk preferences. (Symmonds et al., 2010; Levy, Thavikulwat & Glimcher 2013; Yamada et al., 2013). Less work has focused on predicting risk attitudes from combinations of behavioral, physiological, and environmental measurements. We longitudinally measured daily behavioral activities, emotional states, environmental variables and risk preferences at the within-subject level to determine of any of correlated with risk attitudes. Our goal was to identify state variables that regulate, and ultimately allow prediction of, the temporal dynamics of risk preferences. We recruited 123 subjects residing in the US using CraigsList and an NYU subject recruitment system for an 8-week experiment. Over the 8-weeks subjects completed between 2 and 3 behavioral instruments per day for a total of 112 behavioral instruments. We employed a mobile smartphone-based experimental platform (Linkt) for delivering daily behavioral instruments. Each participant was asked to complete a weekly time use diary and the following instruments at least once every week: a risky choice task (Levy et al. 2010), the self-report affect. In the time use diary, we asked subjects to fill out a timeline of everything they did on that day, and we categorized these activities into Sleep, Work, Home, Leisure, and Social times. We also gathered 13 personality inventories, one time each, throughout the study from the following domains: attitude toward uncertainty, temporal discounting tendency, impulsivity, and the level of psychological distresses to better understand the individual difference of the risk attitude. Here we present data showing how the amount of a particular activity spent last week correlates with risk attitude, as measured using a Principal Component Analysis (PCA) that allowed us to collapse our many risk-related measures to a single vector. We used a Granger-Causality-style approach to relate this PCA vector weekly behavioral assessments. We found that Sleep time can predict significant changes in risk attitude in the coming week. With more sleep time, our subjects became more risk-tolerant. In future work we will expand the scope of our analyses and the scope of the data we will gather from additional subjects. Our results, however, do suggest that changes in a simple bio-behavioral variable can influence risk attitude from week to week.

Disclosures: Z. Yan: None. **P.W. Glimcher:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Datacubed Health.

Poster

P778. Modeling and Behavior III

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Program #/Poster #: P778.03

Topic: H.03. Decision Making

Support: 365mc network

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Title: Identifying unique neurocognitive mechanisms of food addiction and binge eating using longitudinal study design and computational modeling

Authors: *M. KWON[1], J.-H. LEE[1], J. YANG[1], R. S. CHANG[1], W.-Y. AHN[1,2];
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Abstract: Strong evidence indicates that dietary restraint and excessive body image concerns are ironically associated with pathological overeating. Overeating is present in both food addiction and binge eating disorder (BED) but the role of dietary restraint and body image concerns in food addiction and BED still remains unclear. Furthermore, decision-making processes associated with food addiction and BED are yet to be identified. To address this gap, we designed a longitudinal study to investigate whether improved body image concerns by liposuction surgery would play a different role in food addiction and BED. We also investigated their decision-making under uncertainty using novel weight-related decision-making paradigms and computational modeling. In Study 1 (N=93), we validated the novel weight-related decision-making tasks in the community samples and found that measures from weight-related decision-making tasks are more related to body image concerns than those from tasks involving monetary decisions. In Study 2, we recruited obese individuals who registered for liposuction surgery (N = 54, body mass > 25), and longitudinally tracked their food addiction symptoms, binge eating behaviors, dietary restraint, body image concerns, and model parameters (e.g., loss aversion, risk-seeking, and ambiguity tolerance) before and after the surgery. The results showed that 1) changes in body image concerns are significantly associated with binge eating behavior but not with food addiction, and 2) binge eating was positively associated with risk-seeking in pursuing weight loss while food addiction showed the opposite pattern. Our results suggest differential decision-making mechanisms in food addiction and BED, providing novel insights into their constructs.

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Poster

P778. Modeling and Behavior III

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Program #/Poster #: P778.04

Topic: H.03. Decision Making

Support: NEI/NIH award R01 EY028811-01
NSF/NRT graduate training grant NSF-1449828

Title: A confirmation bias during the active seeking of new perceptual evidence across saccades

Authors: *A. CHATTORAJ[1], S. SHIVKUMAR[1], Y. S. RA[2], R. M. HAEFNER[1];
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Abstract: Human decision-making is often biased, and few biases are as ubiquitous as the confirmation bias (Nickerson 1998; Michel and Peters 2020). Two major components contribute to the confirmation bias: first, a biased seeking of new information to support one's belief, and second, a biased interpretation of the observed information in agreement with one's existing belief. In a previous study, we have shown under what circumstances a confirmation bias of the second kind arises as the result of approximate hierarchical inference, and shown it to induce a primacy effect during perceptual evidence integration (Lange et al. 2020). However, the seeking of new sensory evidence, most notably by eye-movements, has so far mostly been found to be close to optimal (Najemnik and Geisler 2005; Yang et al. 2016).
Here, we describe a new psychophysical task that requires collecting sensory information by making multiple saccades in the context of a two-choice coarse orientation discrimination task. Using a gaze-contingent display, our task allows us tight control over the information received by an observer both in the fovea and in the periphery. Importantly, it allows us to estimate the observer's subjective belief about the correct choice before each saccade. We find that eye-movements of 13/16 participants exhibit a confirmation bias in that they are more likely to saccade to a peripheral stimulus that is expected to provide new information in agreement with the current belief than to a stimulus of the opposite orientation. Interestingly, such a confirmatory strategy is also favored by an ideal observer who tries to maximize the gain in task-relevant information with every saccade. However, we also find that 9/13 observers are significantly more biased than the ideal observer. We show that this can be explained by a computational model that starts with a Bayesian active sensing strategy (Yang et al. 2016) but assumes that the computations are implemented approximately. Interestingly, the biased strategy arises when the part of the computations that only involves sensory information is performed with higher accuracy than the part that also involves task-specific (cognitive) information. Such a difference is compatible with previous observations on the difference in information capacity comparing sensory periphery and central processing, and suggestions that lower sensory areas act as a "high-resolution buffer" for higher-level computations (Lee and Mumford 2003). Fitting our model to trial-by-trial choices and eye-movements allows us to infer the degree of approximation, here measured in number of samples, for the cognitive component of each observer.

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Poster

P778. Modeling and Behavior III

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Program #/Poster #: P778.05

Topic: H.03. Decision Making

Support: Faculty of Business and Economics at the University of Melbourne.
Monash Business School

Title: Pupil size reflects computational complexity of decisions in humans

Authors: *E. A. BOWMAN[1], K. ROTARU[2], J. FRANCO[1], C. MURAWSKI[3];
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Abstract: This project examined how computational complexity and cognitive load affect pupil response during complex decisions.
73 participants aged between 18 and 35 years (mean age 22.7, 52 female, 21 male) completed 72 trials of the decision knapsack task. Before each trial, participants were required to memorise either a 1-digit or a 6-digit number displayed for 5 seconds. Each trial consisted of a 5- to 7-second grey fixation screen, followed by the knapsack decision task where participants were given up to 25 seconds to decide if a combination of any of the six items displayed on the screen could be found to satisfy a both minimum value and maximum weight constraint. After the knapsack decision screen, participants were required to enter the number they were asked to memorise at the start of the trial. Participants were paid according to the number of trials in which they both correctly recalled the memorised number and made the correct knapsack decision.
Knapsack task instances varied in computational complexity (as measured by a control parameter defined by the ratio of normalised capacity and normalised profit), but the number of satisfiable and unsatisfiable instances was the same. Gaze and pupil recordings were performed during the task, sampling at 300Hz with Tobii Pro Spectrum video eye trackers. Pupil recordings were de-blinked, interpolated, smoothed, and downsampled into 1000ms bins for modelling using Growth Curve Analysis (GCA).
Mixed-effects modelling of participant responses found a strong effect of instance complexity, and of satisfiability, on trial performance. However, only an inconsistent effect of memory load was found. Participants also spent significantly less time solving knapsack trials of lower instance complexity, and the relationship between instance complexity and time taken significantly predicted performance on a trial. Pupil diameter change was correlated with instance complexity. This difference in pupil response to trials of different instance complexity was detectable up to 15 seconds before the decision point at the end of the (maximum 25-second) trial, and was greatest in incorrect trials.
These results demonstrate that the computational complexity of a task is detected by the

participant, and reflected in the effort-related pupil response, well before the decision is signalled through behaviour.

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Poster

P778. Modeling and Behavior III

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Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P778.06

Topic: H.03. Decision Making

Support: NIH Grant U01NS122040

Title: Perceptual Decision Making in Tree Shrews

Authors: *C. LI[1], K. M. MCHANEY[2], P. B. SEDERBERG[1], J. CANG[2,1];
[1]Psychology, [2]Biol., Univ. of Virginia, Charlottesville, VA

Abstract: Studying decision making and its underlying neural mechanisms can benefit our understanding of how the brain computes goal-related variables with the information it receives. Recently, there has been growing interest in the role of time pressure on the decision-making process, the effects of which are often more profound in animals than in humans, likely complicating the interpretation of the underlying processes. To test the plausibility of different theories of decision-making, it is often required to combine behavioral data with high-resolution neural data and experimental manipulations that can most easily be acquired from animals. Here we present a new animal model - tree shrews (*Tupaia belangeri*) - for decision studies. Tree shrews are evolutionarily closer to primates and have a well-developed visual system that is more ideal for visual decision making tasks compared to mice or rats. In this study, we aim to 1) develop a visual decision paradigm for tree shrews, and 2) test different computational models of perceptual decision-making with and without processes that affect the decision based on the passage of time in each trial with the tree shrew behavioral data. We successfully trained 5 freely moving tree shrews (M=4, F=1) to perform a two-alternative-forced-choice (2AFC) contrast discrimination task, and collected both choice and reaction time (RT) data. All animals produced standard sigmoidal psychometric curves, showing increased proportion correct with higher contrast difference between the choices, along with a plateau effect at the lower levels of difficulty. The choice-RT data were then fitted with a variety of sequential sampling models with and without a timing mechanism that could interrupt evidence accumulation and force an immediate decision. Tests within a Bayesian framework demonstrated strong evidence in favor of models with the timing mechanism. This work establishes tree shrews as a new animal model for studying perceptual decision-making tasks, and also suggests tree shrews exhibit choice behavior similar to other animals, relying in part on a subjective expectation for trial durations to

guide when to make decisions. Future work will explore alternative mechanisms for the timing mechanism, including both urgency gating and collapsing bounds, as well as augmenting the analysis with physiology data to map the decision-making processes to different neural substrates.

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Poster

P778. Modeling and Behavior III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P778.07

Topic: H.03. Decision Making

Title: Simultaneous evidence evaluation over different timescales revealed in change point detection task with multiple alternatives

Authors: *T. STEVENSON, T. HANKS;
UC Davis, Davis, CA

Abstract: To interact appropriately with a dynamic environment, individuals must be able to detect when aspects of their environment change, however the space of potential changes with behavioral significance is often large. In this study, we used an auditory change point detection task to investigate how knowledge about the direction of potential changes affects subjects' abilities to detect when they occur. Nine human subjects listened to a series of clicks that were randomly generated from a Poisson process with an underlying generative rate that either increased or decreased at a random point in the trial, which subjects were instructed to detect. Prior to each click train, subjects were either informed of the direction of the change or not. From the behavioral data, we characterized the subjects' decision-making process using both model-free and model-based methods. When fitting the data to a sequential sampling model parameterized as an Ornstein-Uhlenbeck process, we found that subjects simultaneously applied distinct timescales of evidence evaluation to the same stream of stimuli when there were multiple potential changes. Additionally, subjects had worse task performance when unsure of the change direction, which can be explained by sub-optimal evaluation timescales and decision bounds in our behavioral model. When subjects were informed of the change direction, they were able to improve their performance by optimizing their decision bound on trials with an increase in click rate, or by optimizing both the timescale of evidence evaluation and the decision bound on trials with a decrease in click rate. One outstanding question of this work is whether this asymmetry in parameter tuning is robust to the behavioral model employed. Additionally, the traditional integration-to-bound behavioral model assumes that subjects respond when the estimated click rate reaches a learned criterion level, however it is also possible that subjects first estimate the baseline click rate on a trial-by-trial basis to compare with their estimate of the current click rate.

To investigate both of these questions, we are currently developing a new behavioral model that allows us to determine whether subjects used prior stimulus history to inform their decision on a per-trial basis. The results of this study establish three important capacities of information processing within any proposed neural mechanism of decision making: the ability to simultaneously employ multiple timescales of evidence evaluation, the ability to rapidly adjust those timescales to task demands, and the ability to modify the amount of information required to make a decision in the context of flexible timescales.

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Poster

P778. Modeling and Behavior III

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Program #/Poster #: P778.08

Topic: H.03. Decision Making

Title: Characterizing individual difference in decision-making strategy by normative solution from the Markov decision process for a non-stationary Bernoulli bandit task

Authors: S. KOTANI[1], K. NAKANISHI[1], S. HIROSE[1], Y. YAMASHITA[2], K. KATAHIRA[3], S. SUZUKI[4], *M. FUKUSHIMA[1];
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Abstract: There is growing interest in elucidating individual characteristics of computations ("computational phenotypes") underlying decision-making for complementing traditional trait and symptom measures. A normative solution in Markov decision process (MDP) corresponds to optimal behavior in a decision-making task under the assumption that the current choice influences the outcome in the distant future. Humans often deviate from the normative solution, and there is individual difference in the degree to which one deviates from the solution. Here we develop a method to describe the difference in individual decision strategy parametrically as deviations from the normative solution. In particular, we derived the normative solution for a three-armed bandit task with time-varying reward probabilities (a non-stationary Bernoulli bandit task). This task has been used to investigate the relationship between decision-making performance and psychiatric symptoms. Our MDP framework uses the Volatile Kalman Filter (VKF) to estimate the information state that is updated as the task progresses. The VKF is an extended Kalman filter algorithm appropriate for a volatile environment as in the bandit task. Thus, we obtained a normative solution from the utility for every state calculated by a value iterative algorithm. The parameters of a normative solution determine the behavioral

characteristics of the solution in the task. An example of such parameters is the discount parameter that controls the degree of explore-exploit trade-off. We confirmed that the discount parameter controlled the behavior as expected in our numerical simulation. The agent with a larger discount parameter value tends to choose an arm recently not selected (i.e., exploration). In contrast, the agent with a smaller value is more likely to stay choosing an arm rewarded recently (i.e., exploitation). Next, we use multiple normative solutions with different parameter settings as reference strategies to characterize individuals. For each reference strategy, we calculate the likelihood of each decision-making history on the bandit task so that the likelihoods collectively represent the decision-making tendency for each individual. This set of likelihoods could characterize the individual difference in performing the task. We plan to apply this to the data obtained from 1,900 non-diagnostic participants performing the bandit task (Suzuki et al. 2021). Our approach with the normative solutions has the potential to uncover a novel computational phenotype for decision-making in a broad range of normal and psychiatric populations.

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Poster

P787. Neural Mechanisms of Perception, Cognition, and Decision-Making I

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Program #/Poster #: P787.01

Topic: H.03. Decision Making

Support: Simons Collaboration on the Global Brain award 542961SPI
NIH Grant R01EY022930
McKnight Foundation Scholar award to MRC

Title: Neuronal mechanism for decision-making under dynamic belief states

Authors: *C. XUE, L. E. KRAMER, M. R. COHEN;
Dept. of Neurosci. and Ctr. for the Neural Basis of Cognition, Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Decisions in natural environments consist of two conceptually separable cognitive processes: flexibly adapting to the changes in the task-demands under a dynamic environment (task-switching), and performing the believed-relevant decision task (perceptual decision-making), both extensively studied individually for their impacts in science and translation. But

do our brains make decisions under dynamic and uncertain environments in these two separable steps? We trained macaque monkeys and artificial neural networks to make perceptual decisions while keeping track of implicitly evolving task demands at the same time. In contrast to their conceptual separability, we demonstrated inextricable neuronal interactions between the flexible belief about the task-demand (or task-belief) and perceptual decision-making. These specific neuronal interactions are associated with behavioral interactions between decision-making performance and task-switching strategy on a trial-by-trial basis, which is not predicted by artificial neural networks trained to perform the same task. Specifically, by analyzing task- and perception-related signals from simultaneously recorded neuronal populations in monkey cortical areas 7a and V1, we demonstrated that stronger task-belief is correlated with better perception, and in turn, response fluctuations in visual neurons affect the updating of task-beliefs. Meanwhile, the behavioral and neuronal effects of interaction are not observed with recurrent neural networks trained to perform this task. These results revealed a specific and inextricable link between the neuronal mechanisms for task-belief and perceptual decisions in the brain, which is not a necessary feature for any network trained to make decisions under dynamic environments. This suggests that studying task-belief and decision-making as an integrated system would reveal unknown neuronal constraints and lead to more holistic understanding of task-switching and decision-making in ways that treating either as a standalone function cannot. Our findings demonstrate how multiple brain areas interact to perform seemingly separable components of complex behavior and provides a path toward therapies for neuropsychiatric disorders that affect multiple brain systems.

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Poster

P787. Neural Mechanisms of Perception, Cognition, and Decision-Making I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P787.02

Topic: H.03. Decision Making

Support: SFI-HRB 19/US/3599

Title: In search of motor-independent evidence accumulation signals in monkey EEG

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Abstract: Neural signals that accumulate evidence for perceptual decisions have been identified in a variety of species and brain regions. Thus far, studies of non-human electrophysiology have

exclusively observed such signals in effector-selective brain regions that play a role in preparing the decision-reporting actions. In parallel, investigations of human electroencephalogram (EEG) data have identified a new class of decision signal over centro-parietal scalp sites (the Centro-Parietal Positivity, CPP) that has the unique characteristic of tracing evidence accumulation irrespective of the sensory or motor demands of the task. Such a signal has not yet been observed in non-human animals, raising the question of whether it is a uniquely human process or simply one that has been overlooked by intracranial investigations. In the present study, we collected EEG data from both human and macaque subjects while they performed the same auditory oddball task. We collected EEG data from 6 human participants, while they were asked to discriminate a frequency deviant tone among a number of identical standard tones. A response was considered as valid only if it was given between 100 and 700 ms after target onset. The frequency difference between each standard and target tone (dF) was varied to define 5 different levels of sensory difficulty. The stimulus onset asynchrony (SOA) was varied between 0.25 and 12 seconds to manipulate the short-term memory demands of the task. For our human participants, target detection probability increased with frequency separation (dF) and decreased with SOA. Human EEG data highlight a slowly building positive CPP component following target tones, peaking between 400ms and 700ms, that is absent following standard tones. Furthermore, there was a significant effect of dF on the amplitude reached by the CPP by the time of response. To test for the existence of a similar signal in monkey EEG, we conducted a re-analysis of 32-channel cranial EEG data collected from macaques performing the same auditory oddball task (Teichert 2017). The behavioral results indicate a rise in accuracy with dF and vice versa with SOA. In the EEG we observed a slowly building positivity over central electrodes rising 300ms after target tone presentation and peaking close to response execution, whose slope is modulated by dF, SOA and RT and reaches higher amplitudes for target than standard tones. This presents a potential monkey homolog of the CPP that further analysis will be needed to substantiate.

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Poster

P787. Neural Mechanisms of Perception, Cognition, and Decision-Making I

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Program #/Poster #: P787.03

Topic: H.03. Decision Making

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NIH-NINDS U19NS107609-03
DOD VBFF
NIH T32GM007281

Title: Neuronal encoding of rapid categorical decisions across the primate oculomotor network

Authors: *O. ZHU[1], V. SHIRHATTI[1], S. DAVID[1], S. CHANG[1], A. MEDOFF[1], A. A. SILVA[1], D. J. FREEDMAN[2];
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Abstract: Our ability to rapidly categorize incoming stimuli is an essential cognitive process that imbues our world with meaning and guides our behavior. Previous studies have examined the oculomotor system's role in categorical decisions, but few have explored this process when ongoing motor plans are rapidly modulated and updated by incoming sensory information. In this study, we designed an urgent saccade-based, motion categorization task that utilizes a compelled-saccade paradigm to systematically limit the viewing time of the motion stimuli. This task structure exploits the animal's natural variability in reaction times across trials to quantify the psychophysical impact of categorical information towards ongoing motor plans with millisecond resolution. During task performance, we recorded populations of neurons using up to six linear microelectrode arrays (Plexon V-Probes) simultaneously targeting the lateral intraparietal area (LIP), frontal eye field (FEF), and superior colliculus (SC). On average, each session yielded around 50 well-isolated single units from each area. We quantified category selectivity in each area using linear support vector machine (SVM) classifiers trained from population activity from each area in individual sessions and found that SVMs for each area achieved >90% decoding accuracies and, on average, exhibited latencies that were not significantly different from each other (~120ms after stimulus onset; $p>0.05$). In particular, SVMs for SC populations surprisingly achieved >95% decoding accuracy at a similar latency to LIP and FEF. We also quantified the saccade selectivity of each area with linear SVMs and found that all areas achieved >95% decoding accuracies, though FEF and SC exhibited slightly shorter latencies than LIP ($p<0.05$). The similar latencies and high decoding performance for each area support a distributed model of categorical decision making in which brain areas operate as a collective network rather than discrete modules for perception and motor control. In particular, our results demonstrate that SC is engaged in cognitive functions such as categorization and decision making beyond its traditionally understood role in saccade execution and orienting behaviors. Ongoing experiments will reversibly inactivate each individual area while simultaneously recording the other areas to investigate the causal relationships between brain areas during rapid categorical decision making.

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Poster

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Generalitat de Catalunya Grant AGAUR 2017 SGR 1565 to K.W
Spanish ministry of science and innovation PhD fellowship (PRE2018-083963) to C.V

Title: Neural network dynamics underlying the adjustment of temporal evidence weighting in perceptual decisions

Authors: *C. VIVAR[1,2], G. MOCHOL[1,2], A. J. LEVI[3], A. C. HUK[3], K. WIMMER[1,2];
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Abstract: During perceptual decision making, sensory information can be accumulated using distinct strategies; weighting some stimulus epochs more heavily than others. A recent study has shown that primates can flexibly adapt their temporal weighting strategy to the stimulus statistics (Levi et al., Eneuro 2018). Sensory stimuli with more information at the beginning of the trial produced early weighting, while stronger sensory evidence in later stimulus epochs caused a switch to a late weighting strategy.
To shed light on how this flexible adjustment can be mechanistically implemented at a neuronal level, we used a two-area firing rate model composed of a sensory and a decision circuit with bottom-up and top-down connectivity (Wimmer et al., Nat. Commun. 2015). We included the time-varying MT activity as a constraint to the sensory circuit as well as a time-varying modulation signal, ("choice commitment" signal), that altered the attractor dynamics of the decision circuit. This modulation signal affected the decision circuit in two ways. Firstly, it initiated the decision process by pushing the network into a competition regime. Secondly, it changed the decision dynamics by accelerating or delaying the choice, similar to an urgency signal.
The model could reproduce the experimentally observed primacy weighting for early and flat stimulus statistics and late weighting for late stimulus conditions. We reasoned that the modulation signal may be related to the subject's task engagement, which we measured as the time needed to execute a successful fixation at the start of the trial. Consistent with the model, we found that the subject's engagement was higher (faster fixation) in the early weighting condition and lower (slower fixation) for the late condition.
Preliminary analysis of neural data recorded from areas MT and LIP indicated that the neurons' pre-stimulus activity was correlated with task engagement, providing further evidence for the modulation signal.

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Poster

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Topic: H.03. Decision Making

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Wellcome Grant WT 204811
European Research Council Advanced Grant 293549

Title: Neuronal economic utility arising from the heterogeneous coding of reward magnitude and probability in orbitofrontal neurons, elicited through the continuity axiom of Expected Utility Theory.

Authors: *S. FERRARI-TONIOLO, W. SCHULTZ;
Univ. of Cambridge, Cambridge, United Kingdom

Abstract: Introduction. Utility is a fundamental economic concept which defines subjective values. Expected Utility Theory (EUT) describes choices as a utility-maximization process: we assign a numerical utility to each option and choose the option with the largest utility. Crucially, this "as if" framework (we make choices "as if" maximizing utility) makes no assumption on the underlying brain mechanism and on the existence of a neuronal utility signal, which remains undetermined. We investigated whether single neurons and neuronal populations in the macaque orbitofrontal cortex (OFC) encode a utility signal compatible with the axiomatic economic theory.
Methods. We first identified a quantitative utility measure based on the continuity axiom of EUT, a test for robustness of magnitude-probability integration into a scalar utility value, across multiple settings. This showed that utility maximization can reliably describe risky choices in macaques (Ferrari-Toniolo et al, 2021). We then recorded the activity of single OFC neurons using the same paradigm, to infer a corresponding neuronal utility measure. Both behavioral and neural procedures considered the non-linear contribution of magnitudes and probabilities to the formation of utility values, in line with modern economic theories (e.g. Prospect Theory).
Results. Within the neurons responding to both reward magnitude and probability, we found that: 1) Neurons complied with the continuity axiom, allowing us to define a neuronal utility measure; 2) Such neuronal measure corresponded with the behavioral one in a minority of neurons, while most units predicted risk attitudes significantly different from the behavioral ones; 3) A reliable utility code was reconstructed from this mixed population, which included cells with diverse non-linear magnitude and probability coding patterns.
Conclusions. Our data showed that, while many reward-modulated neurons encoded utility values incompatible with the animal's choices, a neuronal utility signal able to reliably describe

choices was reconstructed from a population of such neurons. The variability with which single units represented magnitude and probability non-linearly, resembling a distributional code, suggested a population scheme able to flexibly accommodate a broad pattern of risk attitudes. These results represent an insight into the process for value computation, proposing a mechanism for the neuronal counterpart of economic utility. The existence of a neuronal utility signal supports a neural implementation of the utility-maximization principle and provides a basis for understanding the origin of risk attitudes and choice biases.

Disclosures: S. Ferrari-Toniolo: None. **W. Schultz:** None.

Poster

P787. Neural Mechanisms of Perception, Cognition, and Decision-Making I

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Program #/Poster #: P787.06

Topic: H.03. Decision Making

Support: ZIA MH002928

Title: Examining neural dynamics of motivational decisions using large-scale array recordings in macaque prefrontal cortex

Authors: *S. WANG, R. FALCONE, B. J. RICHMOND, B. B. AVERBECK;
NIMH/NIH, Bethesda, MD

Abstract: Animals naturally balance the tradeoff between effort and reward. Some rewards are worth the effort while others are not. The more valuable the reward is, the more motivated the animal is to get it**.** In this work, we studied neural dynamics in prefrontal cortex that underlie motivational decisions on whether to accept or reject an offer. Offers were defined by the magnitude and delay-to-reward. We investigated population dynamics of prefrontal neurons using large-scale Utah-array recordings.
Two macaques were trained to either reject or accept (by either releasing or holding a bar) an instrumental trial (a sequential red-purple color discrimination). Accept or reject decisions were based on a visual cue that indicated the reward size (2, 4 or 6 drops) and delay-to-reward (1,5 or 10s). The monkeys almost always accepted the offer for some cues (high reward, short delay), and almost always rejected the offer for other cues (low reward, high delay). For other cues which signaled intermediate value offers, the monkeys had intermediate probabilities of rejecting vs accepting.
Prefrontal neurons encoded the cue information. Using a KNN based population decoding analysis, we were able to decode the 9 cues (3 reward sizes by 3 delay-to-reward times) at an accuracy of 50% (significantly above the chance level of 11.1%) using cross validation. The decoding accuracy peaked between 400ms and 700ms after cue onset. Using a linear SVM

classifier, we were able to predict monkey's choice to accept or reject at an accuracy of 80-85% from 500-1000ms after cue onset.
We further examined the neural dynamics of motivational decisions in a low dimensional neural subspace. We projected the neural dynamics onto the 1-dimensional space perpendicular to the separating hyperplane in the SVM classifier to maximize the information about choices. We investigated how the landscape of the neural manifold in this 1-dimensional space varied between different offers of reward sizes and delay-to-reward times. Preliminary data suggests that the manifolds have a steeper attractor basin following presentation of cues that have clear motivational preferences (high reward low delay, or low reward high delay), and have shallower attractor basins following cues that have ambiguous preferences. This suggests that the landscape of the neural manifolds shape how motivational decisions are made in macaque prefrontal cortices.

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Poster

P787. Neural Mechanisms of Perception, Cognition, and Decision-Making I

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Topic: H.03. Decision Making

Support: KAKENHI JP16H06567 (to M.M.)
JST CREST JPMJCR1853 (to M.M.)
KAKENHI JP19K16890 (to M.N.)

Title: Causal role of midbrain dopamine neurons and ventral striatum circuit in economic decision-making in macaque monkeys

Authors: *M. NEJIME[1], M. YUN[1], T. KAWAI[1], H. YAMADA[1], K.-I. INOUE[2], M. TAKADA[2], M. MATSUMOTO[1];
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Abstract: The brain reward system, consisting of midbrain dopamine (DA) neurons and their target (i.e., the ventral striatum (VS)), is involved in economic decision-making. However, how these structures cooperate with each other to make appropriate decisions remains poorly understood. To address this issue, we conducted single-unit recordings therefrom in macaque monkeys performing an economic decision-making task. The monkey was required to decide whether to choose an option based on its value. We analyzed how neuronal activity varied over time as the economic decision-making process progressed, and found that DA and VS neurons showed different activity dynamics. These neurons represented four decision-making variables,

the value of option (value), the value of the option that the monkey would choose (chosen value), the value of the option that the monkey would not choose (unchosen value), and whether to choose the option (choice). The DA neurons exhibited a short-latency phasic activity signaling these variables just after the option was offered, and their largest population represented the chosen value. By contrast, many of the VS neurons displayed a long-latency ramping activity signaling the choice. These results suggest that DA and VS neurons may be involved mainly in an earlier or a later stage, respectively, of the economic decision-making process. From the difference of their signal dynamics, we hypothesized that the DA signals in the VS will affect the choice behavior in the economic decision-making. To verify this hypothesis, we next examined the causal relationship between DA signals transmitted to the VS and monkey's choice behavior using an optogenetic technique applicable to the monkey brain. We optically stimulated axon terminals of DA neurons in the VS when the option was offered in the economic decision-making task. Of all stimulation sites, 12% produced a significant increase in the monkey's choice rate of the option, while none generated a significant decrease in the choice rate. Our findings have confirmed and expanded the current knowledge as to how DA and VS neurons regulate economic decision-making by highlighting their distinct signal dynamics and causal roles in monkey's choice behavior based on the signal interactions.

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Poster

P787. Neural Mechanisms of Perception, Cognition, and Decision-Making I

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Topic: H.03. Decision Making

Support: HHMI
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NIH F32MH115416

Title: Evidence accumulation and suppression of trial history-related influence by dorsomedial frontal cortex

Authors: *T. Z. LUO[1], A. G. BONDY[1], D. GUPTA[1], V. A. ELLIOTT[1], C. D. KOPEC[1], C. D. BRODY[2];
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Abstract: Perceptual decisions depend on multiple processes, including the gradual integration of noisy sensory evidence over time. A related process involves the incorporation of prior

knowledge of the environment into the present decision and depends on the integration, over a longer timescale, of past choices and their outcomes. In trial-based tasks in which the rewarded choice is solely determined by the present sensory stimulus, the influence of priors manifests in a history-dependent choice bias that can be observed even in expert subjects. It remains unclear how these two processes interact during the formation of perceptual choices. The dorsomedial frontal cortex (dmFC) has been implicated in decisions that depend on behavioral history and also in decisions that depend on sensory evidence. To understand the interaction between evidence integration and the influence of behavioral history, we studied the causal roles of dmFC in rats performing a task that requires the gradual accumulation of pulsatile auditory evidence. Neuronal populations in dmFC and interconnected frontal brain areas were recorded using Neuropixels probes, and neurons in dmFC were silenced using pharmacology, photoinactivation, and chronic lesion. Population activity in dmFC showed a robust representation of evidence. Compared to several frontal areas interconnected with dmFC, including medial prefrontal cortex, striatum, and primary motor cortex (M1), this evidence signal was stronger and emerged earlier in dmFC. Silencing of dmFC through different methods consistently decreased sensitivity to the sensory evidence while increasing bias to the rewarded option of the previous trial. Optogenetic silencing randomly interleaved across trials increased history-dependence on the same trial as the photoinactivation, but not on the subsequent trial. This indicates that the increase in history-related bias occurred before or during, but not after, choice formation. The behavioral consequences of dmFC inactivation can be reproduced in simulations of a bounded drift-diffusion process by biasing the initial point toward the rewarded option of the previous trial, but not by increasing sensory or accumulation noise. Consistent with these simulations, dmFC silencing resulted in an additive bias toward the previous rewarded option in the evidence signal in the neural activity of striatum and M1. These results indicate that during perceptual decisions, dmFC is causally involved in the gradual accumulation of evidence and also the suppression of history-dependent choice bias. A competition between evidence- and history-related signals may underpin the formation of perceptual choices.

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Poster

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HHMI

Title: A mechanistic account of how trial history biases give rise to lapses in perceptual decisions

Authors: *D. GUPTA[1], B. DEPASQUALE[1], C. D. KOPEC[1], C. D. BRODY[1,2];
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Abstract: Experienced subjects in perceptual tasks often deviate from the predictions of optimal decision theory, displaying several suboptimalities. Among the most pervasive of these are history biases or the inappropriate influence of past trials on perceptual evidence, and lapses or evidence independent errors. History biases and lapses are often assumed to arise from independent processes, since the former seems to interact with evidence while the latter does not. However, numerous recent observations suggest that they may share a common origin. For instance, the prevalence of history biases and lapses covaries (Gold ‘08, Busse ‘11), lapse trends are modulated by trial history (Scott ‘15, Ashwood ‘20, Pisupati ‘21) and their proposed neural substrates overlap (Erlich ‘15, Siniscalchi ‘19, Sindreu ‘21). What cognitive process could underlie the links between these suboptimalities? Here, we demonstrate that history biases and lapses can both emerge from normative accumulation of evidence under the incorrect belief that the environment is non-stationary. This corresponds to an accumulation to bound process whose initial state undergoes history dependent updates. While small deviations in the initial state result in largely additive biases to the evidence, larger deviations also decrease the effective sensitivity to evidence. Therefore trial to trial deviations in the initial state produce history-biased choices which have varying degrees of attenuation in evidence dependence, resembling the evidence independence of lapses. In this model, the parameters governing evidence accumulation and initial state updating impose constraints on the relative strengths of history biases and apparent lapses. We test this proposal by fitting a trial-by-trial model to a large dataset of 6.4M choices from 160 rats trained on an evidence accumulation task (Brunton ‘13). We show that the constraints posited by the model are obeyed in this dataset, despite the heterogeneity in history biases and lapse rates in this population. Finally, because variations in the initial state modulate the time to reach decision bounds, reaction times offer a strong test of our proposal. Hence we apply our model to rats trained on a similar task with reaction time reports and show that it explains detailed patterns of choices, reaction times and their history dependence. Our model offers a unified explanation for two common suboptimalities in perceptual decisions, and enhances our ability to predict the precise dynamics of decision variables both within and across trials. This mechanistic account has the potential to improve our understanding of neural correlates of the decision making process.

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Poster

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Topic: H.03. Decision Making

Support: Wellcome Trust (219880/Z/19/Z) to Elena Menichini
Wellcome Trust (562763) to Athena Akrami
Gatsby Charitable Foundation (562980) to Athena Akrami

Title: Sensory priors, choice and outcome history in service of optimal behavior in noisy environments

Authors: *E. MENICHINI, V. PEDROSA, V. PLATTNER, R. LOW, A. AKRAMI;
Univ. Col. London, London, United Kingdom

Abstract: While navigating the world, we constantly make decisions under uncertainty which can originate from ambiguous sensory inputs or unknown action outcomes. In face of noisy perceptions, we build and continuously update our priors and expectations about future events, in order to efficiently estimate sensory information. Via learned policies, agents then have to map the relevant sensory information onto appropriate behavioral responses in order to maximize reward. In a non-stationary environment where action values may change, the rules mapping stimuli to outcomes may not be fixed and exploratory behavior represents an optimal strategy to identify the most rewarded contingencies at any given context. How do animals integrate knowledge about the statistical context and action policy of the environment into an overall model of the world? In this work, we investigate how previous experience (sensory priors, choice and outcome history) can inform such models during decision-making in a perceptual categorization task. Combining behaviors in rats and humans with computational models, we show agents' sensitivity to history of sensory and action events as a mechanism to constantly update policies. By performing large-scale electrophysiological recordings, we will identify neuronal dynamics in key brain areas involved in learning and utilization of priors, in conjunction with policy updates.

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Poster

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Program #/Poster #: P787.11

Topic: H.03. Decision Making

Title: Neural correlates of an approximate prior in perceptual decision making

Authors: *B. GERÇEK[1], G. T. MEIJER[2], B. BENSON[3], R. SCHAEFFER[4], C. FINDLING[1], K. BANGA[5], C. LANGDON[6], N. J. MISKA[5], M. SCHARTNER[2], K. SOCHA[5], M. R. WHITEWAY[7], P. DAYAN[8], K. D. HARRIS[5], A. POUGET[1], T. INTERNATIONAL BRAIN LABORATORY[5];
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Abstract: To select and initiate appropriate actions in complex decision-making tasks requires information to be aggregated and exploited within and across trials. We know little about the brain mechanisms for computation and storage of across-trial information, such as prior probabilities of stimuli or rewarded actions. We examine neural correlates of this information using neural activity recorded in the international brain lab (IBL) task: 583 neuropixel penetrations covering 361 brain regions in the Allen atlas. In the IBL task, mice maximize rewards by exploiting a blockwise prior probability governing the appearance of stimuli. Behavioural modeling has revealed the (usually approximate) way in which the mice are sensitive to the identity of the block. We therefore use a combination of encoding and decoding to localize which brain regions contain either the best estimate of the stimulus prior taken from behavioral modeling or other related quantities such as the true Bayesian posterior over the block.For encoding, we use a Generalized Linear Model (GLM) with a linear link function to express neural activity as a function of numerous task-related variables such as stimulus onset time, contrast, licking, reward delivery, and wheel movement. We incorporate either the block probability or our approximate prior estimate into this model as well, and are examining the variance explained by each regressor.Our decoding approach includes the use of linear decoders, linear discriminant analysis, and a reduced-dimensional representation of the PSTH of neural activity by region to examine how neural activity changes as a function of block probability, stimulus side, or prior estimate and how information is tied together between successive trials.Combining these approaches, we hope to gain insight into the distribution of information about the prior across the brain, as well as clues as to where and how it is computed and updated across trials and how it is brought to bear upon within-trial information processing.Preliminary results indicate substantial variability in the encoding and decodability of prior information across all of: subjects, sessions and brain regions. Despite this, we identify a set of brain regions which still show information content beyond what we expect from said variability. Cortical (retrosplenial) and subcortical (MRN) areas offer some particular promise for investigation, with evidence for distinct sub-populations in the latter integrating prior and stimulus information across different timescales.

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Poster

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Support: Wellcome Trust (216324/Z/19/Z)
Simons Collaboration on the Global Brain (687458)
NIH BRAIN Initiative (F32 MH123010)

Title: Reproducibility of electrophysiological measurements during decision-making

Authors: N. BONACCHI[1], S. BRUIJNS[2], G. A. CHAPUIS[3], A. K. CHURCHLAND[4], M. FAULKNER[5], H. LEE[6], G. T. MEIJER[1], L. PANINSKI[7], N. ROTH[8], N. A. STEINMETZ[8], ***M. TAHERI**[4], S. J. WEST[9], O. WINTER[1], .. INTERNATIONAL BRAIN LABORATORY[10]; [1]Champalimaud Ctr. for the Unknown, Lisbon, Portugal; [2]Max Planck Inst. for Biol. Cybernetics, Tübingen, Germany; [3]Univ. de Genève, Geneva, Switzerland; [4]Univ. of California Los Angeles, Los Angeles, CA; [5]Univ. Col. London, London, United Kingdom; [6]Columbia Univ., New York City, NY; [7]Columbia Univ., New York, NY; [8]Univ. of Washington, Seattle, WA; [9]Sainsbury Wellcome Centre, UCL, London, United Kingdom; [10]The Intl. Brain Lab., London, United Kingdom

Abstract: Understanding whole-brain-scale electrophysiological recordings relies on the collective work of multiple labs. Because two labs recording from the same brain area often reach different conclusions, it is critical to quantify and control for features that decrease reproducibility. We addressed this by repeatedly inserting Neuropixels probes in the same location in the mouse brain, via a joint effort of 8 labs in 3 countries. Here, we report results for 66 total insertions. First, we screened probe insertion accuracy via standardized histological reconstruction of probe tracts. Mean displacement on the brain's surface was 398 μm from the planned site, and 532 μm from the experimenter's intended position (read from the micro-manipulator). Probe insertion angle showed small mean perturbations from the planned angle: 3.3° (coronal) and 5.7° (sagittal). As a result of these displacements, 10/66 insertions failed to target the repeated site, and were excluded. Next, for successful insertions, we compared response properties of recorded neurons across labs. We restricted the analysis to labs with at least 4 successful insertions. Firing rate and LFP power were reproducible across 4 labs: within-lab variability (though considerable) exceeded across-lab variability. Further, neural responses to task events were reproducible across labs up until movement initiation. In most brain regions, the observed variability in neural activity, within or among labs, did not arise from spatial position or lab ID. Finally, we trained a multi-task neural network to quantify variance in neural activity explained by task variables (e.g., stimulus onset time), behavioral variables (timing of licks/paw movements), and other variables (e.g., spatial location in the brain or the lab ID). The model extends common, simpler regression approaches by allowing nonlinear interactions between

variables. We found that within-lab random effects were comparable to between-lab random effects. Taken together, these results suggest that across-lab standardization of electrophysiological procedures can lead to reproducible results across labs. Failures of reproducibility are often driven by deviations from planned insertion locations, and may be ameliorated by refinements in stereotaxic procedures.

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Poster

P787. Neural Mechanisms of Perception, Cognition, and Decision-Making I

Location: Virtual Only

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Program #/Poster #: P787.13

Topic: H.03. Decision Making

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Title: Coordinated prefrontal activity during choice behavior revealed by intracranial recordings

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Abstract: Risky decision-making involves coordinated activity across multiple brain regions involved in evaluation and comparison of options, cognitive control, and action selection. Contrary to notions that assign compartmentalized and sequential roles for individual brain regions, the associated neural activity is likely represented across multiple brain regions in a parallel and distributed fashion (e.g., Hunt & Hayden, 2017; Hunt et al. 2018). Understanding these complex inter-regional dynamics requires simultaneous recordings across multiple brain regions with both high spatial and temporal resolution, which are limited in non-invasive studies of human cognition. In this study, we examine the invasive recordings of multiple brain areas implicated in reward behavior in a large dataset of human neurosurgical patients (n=20) using intracranial electroencephalography (iEEG) while they played a gambling game designed to probe value-based decisions. On each trial, participants chose between a certain reward and a risky gamble of varying win probability. Electrode coverage was determined clinically and therefore varied per subject, but consistently included orbitofrontal cortex (OFC), lateral prefrontal cortex (LPFC), pre- and post-central gyri, cingulate cortex (CC), amygdala, and hippocampus. We analyzed iEEG local field potential (LFP) data in six power bands across all

regions, at multiple time epochs. Here, we focused on the deliberation period (1s prior to choice) and examined power modulation across frequencies and regions by comparing pre-choice activity to a pre-stimulus baseline. We found significant task-related power modulations across all power bands: in general, lower frequency activity (delta, alpha, theta) decreased in frontal and limbic regions, and increased in pre- and post-central gyri and parietal regions. In contrast, higher frequencies (beta, gamma, and high gamma) were more likely to show both increases and decreases in power modulation across electrodes in LPFC, OFC and CC. We compared power across trials between safe bets and gambles and found that task-related modulations related to subsequent choice (gamble or safebet) existed in multiple regions and frequencies. Multivariate regression analyses relating powerband data to task regressors suggest a role of beta and high gamma across OFC, LPFC, and precentral gyrus in choice. These results describe critical components of neural activity underlying human decision-making and have broad implications for neuropsychiatric disorders that involve maladaptive decision-making such as in addiction and obsessive-compulsive disorders.

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Poster

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Title: Single-trial choice decoding from distributed intracranial recordings

Authors: *M. STICKLE[1], J. OVERTON[1], T. DEVESETTI[1], I. SAEZ[1,2], K. MOXON[1];
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Abstract: Risky decision-making involves coordinated activity across multiple brain regions for evaluation and comparison of options, cognitive control, and action selection. Intracranial recordings from distributed brain regions provide a high spatial and temporal resolution representation of inter-regional neuronal dynamics, and a unique substrate to carry out single-trial decoding of choice behavior. Machine learning tools have been applied to movement-related neural data to gain similar insight into brain dynamics controlling movement. Here, we compare the effectiveness of classification techniques to predict the decision outcome of neurosurgical patients undergoing invasive monitoring (intracranial electroencephalography, iEEG) while

performing a gambling task. In this task, patients chose between a safe bet (fixed, small amount of money) or a risky gamble of varying probabilities. Electrode coverage was determined clinically and therefore varied per subject, but consistently included orbitofrontal cortex (OFC), lateral prefrontal cortex (LPFC), cingulate cortex, and hippocampus. We analyzed iEEG local field potential (LFP) data in six power bands (delta, alpha, theta, beta, low gamma, and high gamma) across all regions, at multiple time epochs. Our aim was to construct a method for classifying subject's choice (gamble or safe bet) on a given trial using neural power features alone (time bin x frequency bands x regions) as inputs. We used Principal Component Analysis (PCA) to determine the main axis of variance of neural data, and found that the first principal component (PC) was a good representation of average power in each frequency band. Subsequently, we used a linear classifier (Euclidean distance classifier) to determine what neural features predicted overt trial-by-trial choice. Classification was carried out on both representations of neural activation: raw power and neural PCs. We found that the raw power classifier tended to outperform the PC classifier, obtaining upwards of 70% performance in some patients using a leave-one-out cross validation procedure. Finally, we examined which features of the analysis influenced variance observed in model performance across patients and found that behavioral performance as well as electrode count and localization had a significant impact on decoder performance. Specifically, higher decoding accuracy was achieved in patients with better behavioral performance and a higher number of electrodes in LPFC. These results demonstrate a promising approach for decoding of high-order cognitive states (i.e. choices) from multi-regional, multi-frequency invasive iEEG recordings.

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Poster

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Title: The effect on sensory statistics and serial dependency on sound categorization behaviour

Authors: *V. PEDROSA[1], L. ZHOU[2], E. MENICHINI[1], P. E. LATHAM[2], A. AKRAMI[1];
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Abstract: Animals integrate evidence to infer the statistics of sensory stimuli. The appropriate use of these statistics is essential to optimise rewards when making decisions. It is not fully understood, however, how stimulus statistics affect decision making in humans and animals. In particular, it is not clear how stimulus temporal and spatial dependency guide decision making. Here, we investigate how humans integrate sensory evidence and use previous experiences to inform their decision in a perceptual categorisation task. We analyse the decisions performed by subjects based on overall sensory statistics and preceding choices, outcomes, and stimuli. Depending on the overall statistics of sensory stimuli, subjects adapt their perceptual responses. To unravel the potential underlying mechanisms of this decision-making process, we then compare subjects' performances with computational simulations and behavioural outcomes in a similar task performed by rats. Computational models simulate normative agents behaving based on different first principles. Since sensory perception is inherently limited, our results indicate that the observed adaptation in animal psychometric curves leads to an improvement in performance conditioned on certain constraints. Our combined behavioural and computational analyses unravel further details of specific aspects of sensory information taken into consideration by humans and rats when performing decision-making tasks.

Disclosures: V. Pedrosa: None. **L. Zhou:** None. **E. Menichini:** None. **P.E. Latham:** None. **A. Akrami:** None.

Poster

P790. Lateral Prefrontal Cortex: Decision-Making and Reinforcement Learning

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Topic: H.03. Decision Making

Support: NIH U19NS107609-03
NIH EY019041
DOD VBFF

Title: Distinct causal contributions of FEF and LIP to rapid categorical decisions in primates

Authors: *V. SHIRHATTI, O. ZHU, S. DAVID, S. CHANG, A. MEDOFF, D. J. FREEDMAN;
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Abstract: The ability to make rapid categorical decisions about incoming sensory stimuli is a behavioral capacity of fundamental importance. To study the neuronal mechanisms spanning multiple primate brain areas underlying rapid categorical decisions, we designed a novel rapid visual motion categorization paradigm that compels the subject to make rapid decisions about the abstract category membership of a visual motion stimulus, and allows independent assessment of the visual, cognitive, and motor aspects of the monkey's decisions. The lateral intraparietal (LIP) and frontal eye field (FEF) cortical areas are involved in an array of functions including visual categorization, target selection, saccade planning and execution. To delineate their contributions to rapid categorical decisions we reversibly inactivated local populations in these regions with a $GABA^A$ agonist, muscimol, during a saccade-based categorization task. We used two different stimulus-target spatial configurations, such that either the category stimulus or one of the targets was placed in the inactivated visual field to separately test the impact of inactivation of visual and motor aspects of behavior. The category stimuli transitioned from 100% noise to fully coherent motion (in 1 out of 6 different motion directions) at a variable gap time, relative to presentation of the response target (go cue for the saccade response). The monkey was required to rapidly categorize the direction of motion under variable durations of coherent motion on each trial, referred to as the processing time (PT). The tachometric curve plots the relation between this PT and accuracy, and reveals how long the monkey needs to view the stimulus in order perform the task with reliable accuracy. Inactivating FEF caused a marked decrease in accuracy for both spatial configurations, for nearly all processing times and motion directions. Furthermore, FEF inactivation resulted in longer processing times and slower cognitive processing to reach peak performance across all conditions, as evidenced from the tachometric curves. In contrast, LIP inactivation impaired accuracy and produced longer processing times only when the motion stimulus, but not saccade target, was placed in the inactivated visual field. The deficits from LIP inactivation were stronger for more difficult stimuli (i.e. near the category boundary). Our results suggest a causal role for FEF across oculomotor and perceptual-cognitive components of rapid visual categorical decisions, whereas LIP appears to be more engaged in the cognitive evaluation of sensory stimuli rather than saccade plan during this task.

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Poster

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Topic: H.03. Decision Making

Support: Institute for Information & Communications Technology Promotion (IITP) grant funded by the Korea government (MSIT) (No.2019-0-01371, Development of brain-inspired AI with human-like intelligence)

Title: Goal and context embeddings of the lateral prefrontal cortex during reinforcement learning

Authors: *Y. SUNG, S. LEE;
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Abstract: Humans flexibly control their behavior to adapt to context changes in the environment. The inferior lateral prefrontal cortex (ilPFC) is known to guide goal-directed learning by choosing the optimal strategy for the given context. However, it is still unclear how the neural representations of goal and context reflect the context-dependent control in ilPFC. We investigated the neural embedding structure of goal and context during reinforcement learning and their effects on choice behavior. We performed multi-voxel pattern analysis on twenty participants' fMRI data collected with a sequential two-stage Markov decision task (Lee et al., 2014). Participants make two sequential choices to obtain a reward in the goal state. There are two types of contexts: goal condition (specific goal and flexible goal) and uncertainty condition (high uncertainty with state-transition probability (0.5, 0.5) and low uncertainty with probability (0.9, 0.1)). We estimated the representational dimensionality of neural embedding by averaging accuracies of all binary classifiers across the contexts and goal states (Bernardi et al., 2020). First, we found that the context dimensionality of the ilPFC is significantly higher than that of other brain areas, including the hippocampus, primary visual cortex, and ventral striatum (paired t-test, all pairwise comparison with $p<0.05$). To further explore the nature of the context embeddings, we subdivided the combination of contexts into two cases: linearly separable embedding, in which the goal and uncertainty are represented independently, and nonlinear mixed embedding, which represents their interactions. Unlike other brain areas, we found that the ilPFC represents both types of embeddings, demonstrating its capacity for multiplexed context representations. This nonlinearly multiplexed context representation might support ilPFC's flexible control in the complex environment. We also investigated the goal state embedding of brain regions. We found that the goal state representational dimensionality of the ilPFC is high (0.538, significantly higher than the chance level 0.5 with $p<1e-3$), comparable to that of the primary visual cortex (0.54). Notably, the ilPFC's goal state dimensionality is seemingly a unique predictor of task performance in terms of average reward (Pearson's correlation, $R=0.502$, $p<0.05$) and choice optimality ($R=0.641$, $p<0.01$). Our study elucidates the critical role of goal and context embeddings of the ilPFC in guiding goal-directed behavior.

Disclosures: Y. Sung: None. **S. Lee:** None.

Poster

P790. Lateral Prefrontal Cortex: Decision-Making and Reinforcement Learning

Location: Virtual Only

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Program #/Poster #: P790.03

Topic: H.03. Decision Making

Title: Neuronal encoding of ranked items in primate prefrontal cortex during different phases of transitive inference task

Authors: *S. RAMAWAT[1,2], V. MIONE[1], F. DI BELLO[1], P. PANI[1], S. FERRAINA[1], E. BRUNAMONTI[1];
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Abstract: Problem solving based on deduction using logical reasoning is one of the aspects of intelligence in humans and non-human primates. Provided with the knowledge that A > B and B > C, the subject is able to infer that A > C. This ability to link the partially overlapping information and extend it to deduce a novel relationship is the basis of a Transitive Inference (TI) capability. Previous neurophysiological studies have described the pattern of neuronal activity from the Prefrontal Cortex being modulated while an abstract mental schema of ranked items is accessed during the inferential reasoning test phase. However, the question of how the neuronal encoding of the individual items subtending this representation is shaped by the learning of higher reciprocal relationships is relatively unexplored. In this study, we aimed to answer this question by investigating the single-neuron activity recorded from the dorsolateral PFC of 2 monkeys that learned the rank ordered series of items as (A>B>C>D>E>F) for solving TI problems. Each session was organized around two consequential Learning phases, where the relationship between the adjacent items (e.g. A>B, B>C) of the series were first learned separately and then intermingled, and a Test phase where inferential pairs were included. In each of the experimental sessions, we first searched for a signature of the acquisition of the mental schema in the monkey's performance during Inference. This was behaviorally assessed by observing a symbolic distance effect in each session, which characterizes the comparisons between items with greater rank difference as easier, than the ones with smaller rank differences, subtending a comparison between their positions on the mental schema. Then, we studied the neuronal activity of 80 neurons recorded in these sessions while the monkeys fixated each item of the series to define a tuning curve of activity of these neurons during the Test Phase. The tuning activity from the same neurons was then observed during the different Learning phases of the task to study if and how the changed with the acquisition of the items rank order. We observed that the progression of the task from Learning to Inference led to significantly different activity patterns in 50% of the studied neurons. A greater majority (43%) exhibited different tuning preferences for the series items, while only about 7% cells mirrored the previous tuning preferences but with different magnitudes of activity. These results emphasize the involvement of PFC neurons in the learning phases of the TI task by reorganization of rank ordered activity.

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Poster

P790. Lateral Prefrontal Cortex: Decision-Making and Reinforcement Learning

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Topic: H.03. Decision Making

Support: NIH Grant 2R01NS086104-06
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Title: Self-control signals in dorsolateral prefrontal cortex of monkeys during a multi-effector temptation task

Authors: *J. G. ELSEY[1,2], V. STUPHORN[2,3];
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Abstract: Self-control is the ability to resist rewards that are tempting but lead to suboptimal or negative long-term outcomes. We developed a new temptation task that allows to identify states of low and high self-control. General self-control signals should be effector independent. To probe the generalizability of self-control signals, we trained a monkey on a temptation task where they were had to control either an eye or arm movement to indicate and maintain their choice.

To initiate a trial, the monkey fixated a central point and centered a joystick. The central point then changed into a horizontal or vertical bar cue indicating that the trial would require either an eye or an arm movement to indicate the preferred option. The eye- and arm-response trials were randomly interleaved. Following the cue fixation period, two annulus-shaped targets representing the smaller-sooner (SS) and larger-later (LL) options were presented. Each annulus had a colored section, whose length and color indicated the delay and amount of rewards, respectively. The monkeys made an eye or arm movement to one of the targets and maintained fixation/directional joystick position, until they received the indicated reward amount. On no-temptation trials, the unchosen target disappeared upon choice of either target. However, on temptation trials, the unchosen target did not disappear, and the monkeys were allowed to change their initial choice as long as the time of the chosen target did not run out. Since temptation trials were randomly interleaved and rare, the monkeys' initial choice functions were identical in both conditions and indicated their true preferences. Nevertheless, the monkeys sometimes switched targets in temptation trials. This typically involved a switch from an initial delayed LL target to an immediate SS target (LL-SS switch). The opposite switch (SS-LL switch) occurred almost never. These results indicate that an LL-SS switch is the result of a failure in resisting temptation of the immediate reward and not the correction of a mistake in the initial choice.

We found that monkeys were able to reliably switch between selecting options with either eye or

arm movements. Response times on arm movement trials were longer than eye movement trials. However, monkeys showed a greater willingness to delay reward on arm than on eye movement trials. This indicates that controlling eye position is more effortful than controlling arm position. To investigate the effector-dependence of the neural correlates of self-control, we will record single-unit activity from the dorsolateral prefrontal cortex while monkeys perform the multi-effector temptation task.

Disclosures: J.G. Elsey: None. **V. Stuphorn:** None.

Poster

P790. Lateral Prefrontal Cortex: Decision-Making and Reinforcement Learning

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Topic: H.03. Decision Making

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Title: Human and non-human primates show similar sampling and choice behavior during multi-attribute decision making

Authors: *Y.-P. YANG[1], A. SAMPSON[1], E. NIEBUR[1,2], V. STUPHORN[1,2];
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Abstract: In the real-world, we make many complex multi-attribute decisions that require the integration and comparison of multiple pieces of information. This complexity is thought to tax limited cognitive resources, since an agent can only pay attention to a limited set of information and then use it to dynamically construct preferences. Previous evidence supports the role of attention in constructing subjective value and guiding choice. However, the cognitive and neural processes that are at the heart of preference formation are still poorly understood. An animal model that allows the tracking of attention during the decision process is of critical importance. Here, we developed a novel multi-attribute decision-making task in both human and non-human primates. This task allows us to observe the focus of attention of subjects as they sequentially inspect the individual attributes (amount, probability) determining the value of reward options and select one of them. Two important aspects make it well suited to investigate dynamic preference formation of risky choices. First, our task is a free-viewing task. Thus, the subject's eye movements directly indicate which decision-related variable is attended, and in what order. Second, the information about the value of the two functionally different attributes is parametrically encoded using two different symbolic cues. The cue shape explicitly indicates the attribute types. We observed behavior in the task in both humans (n=34) and macaques (n=2). Monkeys choice behavior indicates that they understand the meaning of the attribute cues. They can integrate this information into a single subjective value estimate for each option and compare

the value of each option before choosing. Risk attitude of individual subjects can be explained by their relative weighing of the two attributes. On most trials, both humans and monkeys inspected all available information before making a choice, and first fully inspect one option, before inspecting the other. Late fixations are strongly correlated with choice. Our new attention-guided sequential sampling task will allow the study of option value computation and comparison in a primate model of multi-attribute choice.

Disclosures: Y. Yang: None. **A. Sampson:** None. **E. Niebur:** None. **V. Stuphorn:** None.

Poster

P790. Lateral Prefrontal Cortex: Decision-Making and Reinforcement Learning

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Program #/Poster #: P790.06

Topic: H.03. Decision Making

Title: Direct representation of a Bayesian posterior in the prearcuate gyrus

Authors: *J. A. CHARLTON, R. L. T. GORIS;
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Abstract: When sensory information is ambiguous, perceptual inference is guided by prior knowledge of statistical regularities of the environment, in accordance with Bayesian theory. The neural mechanisms that integrate sensory signals with context-specific priors likely reside in a network of association areas. However, the nature of the neural representation that supports Bayesian-like inference is not known. Ideal Bayesian decision-makers interpret the world on the basis of a posterior stimulus belief. There may exist neurons whose firing rate *directly* represents this statistic. Alternatively, Bayesian-like choice behavior may arise from an *indirect* neural representation of the posterior. To distinguish between these hypotheses, we used multi-electrode arrays to record neural population activity in the prearcuate gyrus (PAG) of a macaque monkey while the animal performed Bayesian-like perceptual inference in a 2-AFC discrimination task. The observer judged the orientation of a stimulus presented in the near-periphery and communicated its judgment by means of a saccadic eye movement towards one of two choice targets. The meaning of the response options was not derived from their spatial position (one was placed within the neurons' motor response field, the other on the opposite side of the fixation mark), but from a visual feature (one was oriented clockwise, the other counterclockwise), enabling us to distinguish the neural correlates of perceptual inference from those of action selection. The animal performed the task under two contexts, each associated with a different stimulus distribution, and under conditions of low and high stimulus uncertainty. His choice behavior exhibited the key signatures of Bayesian inference. To gain insight into the neural correlate of this behavior, we built a population decoder optimized to predict the monkey's perceptual choice and corresponding motor action. The resulting decision variable trajectories

exhibited an initial excursion along the perceptual dimension, followed by an excursion along the motor dimension. This suggests that choice formation takes place in an abstract perceptual space, and is then mapped onto a motor response. During the putative deliberation epoch, firing rate of individual PAG units appeared to directly represent a posterior stimulus estimate. Specifically, when stimulus uncertainty was low, firing rate only depended on stimulus orientation. But when stimulus uncertainty was high, firing rates were additionally modulated by task context. These results clarify the nature of the neural representation that supports Bayesian-like perceptual inference.

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Poster

P790. Lateral Prefrontal Cortex: Decision-Making and Reinforcement Learning

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Topic: H.03. Decision Making

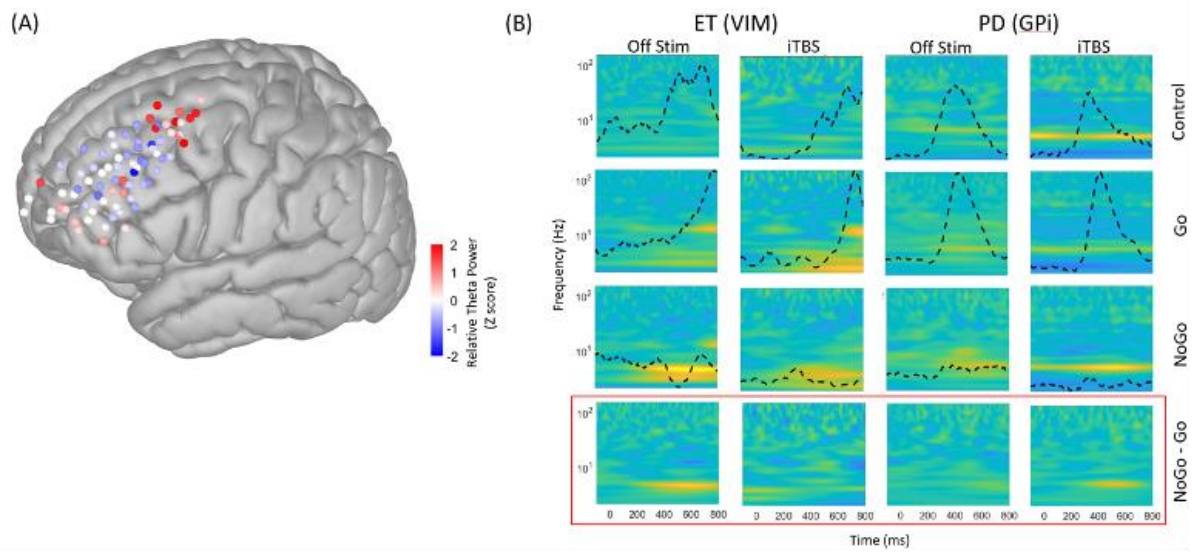
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Michael J. Fox Foundation: 15098 (HCW)

Title: Investigating prefrontal markers of action control and the effects of theta-burst deep brain stimulation in Parkinson's disease

Authors: *N. BENTLEY[1], Z. T. IRWIN[1], S. BLACK[1], A. ROLLER[1], C. L. GONZALEZ[2], K. M. VISSCHER[3], R. T. KNIGHT[4], B. L. GUTHRIE[1], H. C. WALKER[2];
[1]Neurosurg., [2]Neurol., [3]Neurobio., Univ. of Alabama at Birmingham, Birmingham, AL; [4]Dept. of Psychology, Helen Wills Neurosci. Inst., Univ. of California Berkeley, Berkeley, CA

Abstract: Dysfunction in the ability to control behavioral responses with changing environmental demands is a prominent feature in many neurodegenerative disorders such as Parkinson’s disease (PD). This dysfunction is associated with decreased local field potential (LFP) activity in brain areas underlying cognitive control. Our study aimed to investigate LFP changes in dorsolateral prefrontal cortex (DLPFC) during an action control task (Go/No-go). The task includes rest, a motor block consisting of simple button presses, and a cognitive block of 50% "No-go" trials. We investigated whether intermittent theta-burst deep brain stimulation (iTBS) is associated with increased theta power in the DLPFC and led to faster response times.We recorded LFPs from DLPFC using acute electrocorticography during awake deep brain stimulation surgery in PD (n=3) or essential tremor (n=2) participants. A 6-contact strip

electrode was inserted over DLPFC. We measured rest- and task-evoked LFP, then delivered iTBS, measuring changes in LFP and response times. Resting power spectra in both PD and ET participants demonstrated peaks both in the theta (3-8 Hz) and beta (13-30 Hz) range, with highest theta amplitudes in rostral and caudal DLPFC (Fig. 1A). iTBS increased cortical theta-power more when delivered in the globus pallidus interna in comparison to the subthalamic nucleus or to the ventral intermediate thalamic nucleus. Theta-power increased specifically during "No-go" trials requiring response inhibition for ET, but not PD, participants (Fig. 1B). However, delivery of iTBS enhanced theta-power in PD patients during "No-go" trials (Fig. 1B). Finally, we found a trend towards faster reaction times during task performance during iTBS compared to off stimulation. These exploratory preliminary results suggest that the Go/No-go task engages theta activity in the DLPFC, and iTBS may speed response times and increase theta power.



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Poster

P790. Lateral Prefrontal Cortex: Decision-Making and Reinforcement Learning

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Program #/Poster #: P790.08

Topic: H.03. Decision Making

Support: National Research Foundation (NRF) of Korea (NRF-2018R1C1B3007313 & NRF-2018R1A4A1025891)
Creative-Pioneering Researchers Program through Seoul National University

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Title: Anodal tDCS over the dorsolateral prefrontal cortex modulates the balance between Pavlovian and instrumental systems in the punishment domain

Authors: *H. KIM[1], J. HUR[1], M. KWON[1], S. KIM[1], Y. ZOH[1,3], W.-Y. AHN[1,2];
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Abstract: Literature suggests that the brain has multiple systems for decision-making, and the balance between Pavlovian and instrumental systems is critical for optimal decision-making. Pavlovian bias refers to a coupling of action and valence, for example, approach toward reward-predictive stimuli and avoidance of punishment-predictive stimuli, which is often against instrumental response. Recent neuroimaging studies have identified brain regions implicated with Pavlovian bias including the dorsolateral prefrontal cortex (dlPFC). However, it is unclear whether Pavlovian bias can be influenced by an exogenous method such as brain stimulation. Here, we investigated whether up-regulating the dlPFC using transcranial current direct stimulation (tDCS) would modulate Pavlovian bias. In our double-blind study, participants were assigned to either the anodal or the sham group and received stimulation over the right dlPFC for three successive days. On the last day, participants performed a reinforcement learning task called the orthogonalized Go-Nogo task which allows us to assess participants' degree of Pavlovian bias in both reward and punishment domains. We used computational modeling and hierarchical Bayesian analysis to estimate model parameters reflecting latent cognitive processes. Several competing computational models were tested using model comparison methods and we found that a model with separate Pavlovian bias parameters for reward and punishment domains showed the best model fit. The results showed that (1) when using a behavioral index of Pavlovian bias, the anodal group showed significantly reduced Pavlovian bias in the punishment domain, but not in the reward domain compared to the sham group. (2) The Pavlovian bias parameter in the punishment domain from computational modeling was also lower in the anodal group compared to the sham group, which is consistent with behavioral findings. In addition, the anodal group showed a lower go bias parameter than the sham group. These results remained significant even after controlling for age and sex. Together, these findings suggest that anodal tDCS may lead to behavioral suppression or change in Pavlovian bias and shed light on the neural basis of Pavlovian bias.

Disclosures: H. Kim: None. **J. Hur:** None. **M. Kwon:** None. **S. Kim:** None. **Y. Zoh:** None. **W. Ahn:** None.

Poster

P790. Lateral Prefrontal Cortex: Decision-Making and Reinforcement Learning

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P790.09

Topic: H.03. Decision Making

Support: MRC
Wellcome Trust
BBSRC

Title: Electrophysiological evidence for alternative value encoding in macaque frontopolar cortex

Authors: *M. AINSWORTH[1], J. M. GALEAZZI[1], C. PEDREIRA[1], F. A. MANSOURI[2], M. J. BUCKLEY[1];
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Abstract: Although the importance of prefrontal cortical regions in decision making is well established, the exact contribution of frontopolar cortex (FPC) has remained elusive. A number of recent studies have suggested that FPC is essential for representing the value of unchosen, alternative stimuli and rules with the cortex (Mansouri 2017). However, this hypothesis relies primarily or the neuroimaging in humans and combined behavioural (Boorman 2009) and lesion studies in non-human primates (NHPs) and there remains little electrophysiological evidence supporting the encoding of alternative/unchosen stimuli within the neuronal circuits in FPC.Here we use a combination of reinforcement learning (RL) models, local field potential recordings, and electrical micro-stimulation in two NHPs implanted chronically with Utah microelectrode arrays to explore local circuit activity in FPC whilst they performed a WCST analogue. In this task animals were required to match one of three targets with a sample stimuli. The correct target was determined by either shape or colour, on a block-by-block basis. To determine how animals learnt the correct rule we fit RL models to the animals choices. This modelling suggested that both animals attempted to estimate the value of both the current and the alternative rule throughout the task. Examination of LFP activity recorded from FPC revealed increased gamma frequency activity (60-120Hz) after animals made a choice, but not on presentation of sample stimuli or choice targets. This choice-aligned LFP activity corresponded both to whether animals received a reward, and to an RL derived value attached to the alternative rule. Electrical micro-stimulation of FPC did not impact the animals performance within a block. However it did decrease their ability to switch rules following a block change. These data directly link local circuit output from FPC, with the valuation of alternative choices and stimuli in decision making, and should motivate future research involving single unit recordings from FPC.

Disclosures: M. Ainsworth: None. **J.M. Galeazzi:** None. **C. Pedreira:** None. **F.A. Mansouri:** None. **M.J. Buckley:** None.

Poster

P806. Working Memory

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P806.01

Topic: H.01. Attention

Support: NSF BCS-1848440

Title: Stimulating Visual Cortex Reactivates Latent Visual Working Memories

Authors: *Z. XIE[1], J. SAMAHA[2], N. S. ROSE[1];
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Abstract: In this experiment we elucidate the neurocognitive processes that enable items to be held in working memory in different states of accessibility (actively- or passively-retained, or deleted). On each trial, healthy young-adult humans retained two line-orientations in WM, actively-retained the cued item and passively retained the uncued item until it was deleted when it was no longer relevant on the trial while delay-period transcranial magnetic stimulation (TMS) was applied to the retinotopically localized areas of visual cortex where the items were processed with simultaneously recorded EEG. Inverted encoding modeling (IEM) of the EEG data showed that TMS could reactivate both the actively- and passively-retained items, but TMS after the second retro-cue could no longer reactivate the item that was deleted. The findings are consistent with the sensorimotor recruitment hypothesis -- that features of items retained in WM are represented via “activity-silent” mechanisms in the sensory cortex.

Disclosures: Z. Xie: None. **J. Samaha:** None. **N.S. Rose:** None.

Poster

P806. Working Memory

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Topic: H.01. Attention

Support: Priority Program, SPP 1772 from the German Research Foundation (Deutsche Forschungsgemeinschaft, DFG), grant BO 649/22-1
Priority Program, SPP 1772 from the German Research Foundation (Deutsche Forschungsgemeinschaft, DFG), grant VO 1432/19-1
European Social Fund and the Free State of Saxony, grant 100342331

Title: Dual-task walking in older adults - the influence of cognitive task constraints on task interference

Authors: *M. MACK[1], R. STOJAN[1,2], O. L. BOCK[3,2], C. VOELCKER-REHAGE[1,2];
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Abstract: Performing a cognitive task while walking typically leads to task interference. These dual-task effects (DTEs) are particularly pronounced in older adults and occur in either the walking task or the cognitive task, or both. However, it is unclear how constraints of the cognitive task affect DTEs. Therefore, this study investigated how DTEs of the walking and the cognitive task differ when response intervals are fixed (Stroop task) versus self-paced (Serial 3's). We expected lower cognitive DTEs but higher walking DTEs with fixed compared to self-paced response intervals. Data of 38 healthy older adults (age: 68.64 ±4.09; 20 females) were analyzed. Participants completed a virtual dual-task walking scenario on a motorized treadmill with a fixed walking speed of 1 m/s. The scenario comprised two different dual-tasks (Walking + Stroop, Walking + Serial 3's) and three different single tasks (Walking, Stroop, Serial 3's). All tasks were carried out in a pseudorandomized order (5 trials per task, 30 s per trial), that was identic for all participants. Outcomes were relative DTEs in % for walking, calculated for Step Cadence and Step Time Variability, and relative DTEs in % for cognition, calculated for Response Speed and Accuracy. All DTEs were analyzed by Bonferroni corrected two-sided t-tests, comparing DTEs to zero. Further, Bonferroni corrected dependent t-tests were conducted to compare DTEs for Stroop and Serial 3's. Significant DTEs emerged for Step Cadence in Walking + Stroop ($t = 6.48$, $p < .001$), and for Response Speed in Walking + Stroop ($t = -3.46$, $p = .001$) and in Walking + Serial 3's ($t = 12.67$, $p < .001$). DTEs for Step Cadence were significantly higher ($t = 8.52$, $p < .001$) and DTEs for Response Speed were significantly lower ($t = 11.78$, $p < .001$) for Walking + Stroop compared to Walking + Serial 3's. No significant task differences were found for Step Time Variability ($t = 1.81$, $p = .078$) and Accuracy ($t = 1.06$, $p = .298$). Results suggest that task interference depends on task pacing. A fixed presentation mode (Stroop), seems to shift task priority towards the cognitive task, if compared to self-paced presentation mode (Serial 3's). As low postural variability might facilitate the achievement of the cognitive task, Step Time Variability may be not affected.

Disclosures: M. Mack: None. **R. Stojan:** None. **O.L. Bock:** None. **C. Voelcker-Rehage:** None.

Poster

P806. Working Memory

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Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P806.03

Topic: H.01. Attention

Support: JSPS Kakenhi 18H03500
JST COI JPMJCE1311

Title: Significant influence of task-irrelevant stimuli on task performance

Authors: *A. MATANI;
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Abstract: The Stroop, Stroop-like, and Simon effects are conflict effects where the congruency between two factors in a stimulus modulates response performances to the stimulus (e.g., reaction time (RT) and correct answer ratio). When analyzing data by repeated measures analysis of variance (rANOVA), significant effects are often observed besides the conflict effect between two factors, e.g., the main effect and interactions (primings) including the task-relevant stimulus. We performed a Stroop-like experiment, in which the stimulus was a left-or-right moving left-or-right pointing triangle and the task was assigned to the moving or pointing direction. Let the condition (2 levels: left or right) of the pointing and moving directions in the current trial be denoted by Pnt and Mov, respectively and those in the previous trial by pnt and mov, respectively. When task was on Pnt, the Pnt-Mov Stroop and Pnt-mov priming (often called negative priming) effects were strongly observed in RT ($p < 0.001$, $N = 47$). It is reasonable that one cannot make decision of left or right of Pnt before resolving conflicts including Pnt. However, the Mov-mov priming effect was weak but significant ($p < 0.05$) although Mov and mov were totally task-irrelevant. It may be interesting but we do not identify the specific reason so far. Such a task-irrelevant influence is sometimes found in emotional priming experiments, in which an emotional cue is given first and then an unemotional task is presented, and the emotional processing slows RTs. As a first reason, the Mov-mov priming may be due to the same kind of phenomenon. Second, the rANOVA ('r' is not important here) may have multiple problems to apply such RT analyses. The rANOVA employed a generalized linear model (Pnt×Mov×pnt×mov = 2×2×2×2): RT = aMeanRT + bPnt + cMov + ⋯ + dPnt*Mov + ePnt*mov + fMov*mov + ⋯, meaning that each brain process (e.g., to Pnt*Mov and Pnt*Mov) run serially, not parallel at all, because of the additions. If parallel, only the slowest process determines the RT. It has another problem; the model does not express that the task is assigned to Pnt and does treat all terms equally. We eliminated the invalid trials (e.g., wrong responses) from the rANOVA and it might have been an implicit reflection of the task. The model may be valid if there is one left-and-right integrator has four ports accepting the left-or-right signals from Pnt, Mov, pnt, and mov without knowing each port connection (Mov, pnt, and mov are inhibited in advance for correct response). As another problem, if the Pnt-Mov Stroop and Pnt-mov priming has an interaction, the Mov-mov priming is possibly observed as an apparent, or confounding phenomenon because of Pnt*Mov*Pnt*mov = Mov*mov.

Disclosures: A. Matani: None.

Poster

P806. Working Memory

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Program #/Poster #: P806.04

Topic: H.01. Attention

Support: NIH grant ROIMH087214
ONR grant N00014-12-1-0972

Title: Functional connectivity models track fluctuations in attention and working memory

Authors: *N. HAKIM[1], E. AWH[2], E. K. VOGEL[1], M. D. ROSENBERG[3];
[1]Univ. of Chicago, Chicago, IL; [2]Psychology, Univ. of Chicago Dept. of Radiology, Chicago, IL;
[3]Dept. of Psychology, Yale Univ., New Haven, CT

Abstract: Attention and working memory are deeply intertwined constructs that fluctuate synchronously - when attention lapses, participants are less likely to hold information in working memory (deBettencourt et al., 2019). At the same time, however, there is growing evidence to suggest that these two processes are neurally dissociable (Hakim et al., 2019). Here, we used fMRI functional connectivity to further delineate the relationship between attention and working memory. To do this, we re-analyzed a previously published dataset in which 8 participants each performed 2-6 runs of a spatial attention task and a working memory precision task using the same stimuli (Sprague & Serences, 2013). We then applied two open source pre-trained functional connectivity models to predict performance in each of these tasks. For each participant, we calculated functional connectivity matrices within each task by concatenating trial-wise BOLD activity from correct and incorrect trials separately. First, we applied an attention-based model that was trained to predict individual differences in performance on a gradual-onset performance task (gradCPT) and has been shown to track within-subject fluctuations in sustained attention (Rosenberg et al., 2020). We found that strength in this sustained attention network during the spatial attention task distinguished participants' correct vs. incorrect attention task trials. However, sustained attention network strength during the working memory precision task did not distinguish high vs. low working memory precision task trials. Next, we applied a working memory-based model defined to predict individual differences in 2-back task performance (Avery et al., 2020). Interestingly, strength in this working memory network during the working memory task distinguished high vs. low working memory precision task trials. However, this working memory network did not predict attention task performance. These results suggest that there is a double dissociation between attention and working memory in these two task contexts, and they provide further evidence that attention and working memory processes are neutrally dissociable.

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Poster

P806. Working Memory

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Program #/Poster #: P806.05

Topic: H.01. Attention

Support: National Science Foundation 1952050
National Science Foundation 2043740
National Science Foundation DGE-1746045
University of Chicago Research Computing Center

Title: Adult functional connectivity models differentially predict sustained attention and working memory in youth

Authors: *O. KARDAN[1], A. J. STIER[1], C. CARDENAS-INIGUEZ[1], J. C. PRUIN[1], K. E. SCHERTZ[1], Y. DENG[1], T. CHAMBERLAIN[1], W. J. MEREDITH[2], X. ZHANG[1], J. E. BOWMAN[1], T. LAKHTAKIA[1], L. TINDEL[1], E. AVERY[3], Q. LIN[3], K. YOO[3], M. M. CHUN[3], M. G. BERMAN[1], M. D. ROSENBERG[1];
[1]Univ. of Chicago, Chicago, IL; [2]UCLA, Los Angeles, CA; [3]Psychology, Yale Univ., New Haven, CT

Abstract: Sustained attention (SA) and working memory (WM) are central cognitive processes that vary between individuals and change over time. These abilities are impactful in development because of their consequences for life outcomes. Here, we characterize the functional brain architecture of SA and WM in children age 9-11 using models based on functional connectivity measured with functional magnetic resonance imaging. We analyzed data from the Adolescent Brain Cognitive Development (ABCD) Study dataset (*n*=1545) to ask whether connectome-based predictive models (CPMs) built to predict SA and WM in adults generalize to capture inter- and intra-individual differences in SA and WM task performance in youth.
We found that a predefined CPM of SA (Rosenberg et al., 2016) predicted individual differences (r=.18, p <.001) and block-to-block fluctuations (β=.07, p<.001) in children's performance on the 0-back task, an attentionally taxing low-WM-load task. A predefined CPM of WM (Avery et al., 2020), on the other hand, predicted individual differences (r=.14, p <.001) and block-to-block fluctuations (β=.04, p<.001) in performance on the 2-back task, an attentionally taxing high-WM-load task, in addition to the 0-back task (r=.15, p <.001).
We further assessed the degree to which the adult-defined models captured the functional architecture of SA and WM in youth in two ways. First, we compared the SA and WM models' predictive power to that of a CPM of general cognitive abilities defined in the ABCD sample itself. Although this sample-specific CPM outperformed the WM model for predicting 2-back performance, it did not outperform the SA model for predicting 0-back performance. Second, we tested whether the SA model showed worse predictive power in the ABCD sample that it did in the adult Human Connectome Project sample (HCP; *n*=754). We did not test the WM model because it was defined in HCP. We did not find evidence for reduced predictive power in youth relative to adults. Rather, the SA model showed comparable predictive power for inter- and intra-individual differences in 0-back task performance in the ABCD and HCP samples.
Finally, we evaluated the consequences of SA and working WM expression for long-term

memory in children and found that the WM model predicted children's later recognition memory for *n*-back task stimuli ($r = .12$, $p < .001$). Together these results demonstrate that functional connectivity models of SA and WM defined in adults generalize to youth. Whereas a network predicting SA specifically captures individual differences and fluctuations in attention in the developing brain, a network predicting WM model captures general aspects of memory and attention at this age.

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Poster

P806. Working Memory

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P806.06

Topic: H.01. Attention

Support: National Science Foundation 2043740

Title: Connectome stability as a marker of cognitive performance

Authors: *A. CORRIVEAU[1], K. R. YOO[2], Y. KWON[3], M. M. CHUN[2], M. D. ROSENBERG[1]; [1]Psychology, Univ. of Chicago, Chicago, IL; [2]Psychology, Yale Univ., New Haven, CT; [3]Psychology, Yale Univ., East Haven, CT

Abstract: Although sustained attention and working memory are essential to daily life, individual abilities vary. Whole-brain fMRI functional connectivity (FC) is a useful tool for building brain-based markers of cognition and has been used to identify network models of sustained attention [1] and working memory [2]. Further, features of an individual's FC pattern, or connectome, may provide additional predictive utility for cognition. In particular, connectome stability, or the similarity of an individual's FC pattern across repeated fMRI runs, increases across development and is related to social function [3,4]. The current study investigates whether connectome stability predicts sustained attention and working memory, critical cognitive abilities, in adults.

We identified three independent datasets that included repeated fMRI runs of working memory and/or sustained attention tasks. Sustained attention was assessed with the gradual-onset continuous performance task (gradCPT) in Datasets 1 and 2 [1,5]. Dataset 1 (*n*=25) contained 3 gradCPT runs collected in the same scan session and Dataset 2 (*n*=94) included 2 gradCPT runs collected ~2 weeks apart. Dataset 3 included 316 participants from the Human Connectome

Project sample [6,7]. We assessed attention using performance on 0-back blocks of two *n*-back task runs collected in the same scan session. Working memory was measured in Dataset 2 as performance on a visual short term memory task and in Dataset 3 as performance on 2-back blocks of the *n*-back task. Datasets also included other cognitive tasks (DS 2&3), movie viewing (DS 2), and rest runs (DS 1,2&3). We defined run-specific FC patterns using a 268-node brain atlas [8]. Connectome stability was defined as the Pearson correlation between vectorized connectomes and was calculated for all pairs of fMRI runs within a dataset.
Connectome stability was correlated with sustained attention and working memory performance in all three datasets (partial r_s value range = .16-.41, $p = 1.2*10^{-4}$-.09). Relationships were specific to stability within sustained attention and working memory task runs, respectively, and relied on stability of the whole connectome rather than specific brain networks. Furthermore, connectome stability improved the generalizability of behavioral prediction from previously published sustained attention and working memory connectome-based models [1,2]. These results demonstrate connectome stability's utility as a marker of cognitive performance.
[1] Rosenberg et al., 2016 [2] Avery et al., 2020 [3] Kaufmann et al., 2017 [4] Vanderwal et al., 2021 [5] Rosenberg et al., 2020 [6] Van Essen et al., 2013 [7] Yoo et al., 2020 [8] Shen et al., 2013

Disclosures: A. Corriveau: None. **K.R. Yoo:** None. **Y. Kwon:** None. **M.M. Chun:** None. **M.D. Rosenberg:** None.

Poster

P806. Working Memory

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P806.07

Topic: G.09. Drugs of Abuse and Addiction

Support: MUSC Internal Funding

Title: Drinking Frequency Modulates Functional Brain Activity in Chronic Alcohol Abusers During 2-Back Spatial Working-Memory Task

Authors: *G. WARNER[1], J. FLANAGAN[2], A. BENITEZ[3], K. BARLIS[4], J. RICHARDSON[4], S. STOMBERG-FIRESTEIN[4], N. BUSTOS[4], L. ANDREW[5], J. JOSEPH[4];
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Abstract: Chronic alcohol abuse is a serious public health issue that has recently been exacerbated as Americans sought refuge from the stress and isolation of the COVID-19 pandemic at the bottom of a glass of wine, beer, or liquor. Problematic drinking has been linked to a myriad of health issues and premature death. Neurobiologically, excessive drinking has been linked to widespread reductions in gray matter volume, aberrant functional activity, and consequently, impairments in a variety of cognitive domains. Spatial working-memory is one such domain with numerous studies reporting deficits in heavy drinking populations. To elucidate the relationship between spatial working-memory and chronic alcohol abuse 18 participants with Alcohol Use Disorder (AUD) and two non-AUD chronic heavy drinkers performed a spatial 2-back task during an fMRI scan. Individual-subject general linear modeling analysis was conducted to determine which voxels showed greater BOLD activity during correctly identified 2-back events (hits) compared to successful memory storage events (the trials two prior to a hit). A group level analysis was then conducted to determine which of the identified voxels correlated with drinking frequency (percentage of days where subjects drank as established by the Timeline Follow Back). This analysis revealed a cluster of 171 voxels primarily located in the right postcentral gyrus and edging into the right precentral gyrus which displayed a significant positive correlation ($r=0.715$, $p<0.001$) with drinking frequency. These results suggest that more frequent drinkers show higher BOLD activation in the right postcentral gyrus and right precentral gyrus more during successful memory storage than hits compared less frequent drinkers. This may indicate that frequent drinkers recruit these regions more heavily than infrequent drinkers in order in order to compensate for functional deficits resulting from frequent drinking.

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Poster

P808. Human Behavior, Disorders, and Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P808.01

Topic: H.06. Social Cognition

Support: NIMH Division of Intramural Research Grant ZIA MH002920

Title: Multimodal social and emotional information across the superior temporal sulcus

Authors: *J. M. DENNING, A. S. PERSICHETTI, A. MARTIN;
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Abstract: Humans are remarkably efficient and accurate at processing social and emotional information from multiple modalities. Other people's faces and voices are good cues to their identities and emotional states. Previous studies have shown that regions in the superior temporal sulcus (STS) represent social and emotional information while also being sensitive to auditory, visual or both modalities. However, the precise mapping of person and modality information, and how they overlap in the STS, is still not known. We scanned fifteen participants using high-resolution 7T fMRI with both univariate and representational similarity analyses (RSA) to ask how faces and voices are represented and integrated within the STS and in person-selective regions across the whole brain. During separate trials, participants either viewed images of faces or objects, listened to vocalizations or object sounds, or were presented with both visual and auditory stimuli at the same time. Specifically, the faces and voices are from nine individuals, each depicting three emotional states (laughing, neutral, and crying), while the objects and sounds are from nine inanimate objects that are either positive (e.g., champagne bottle), neutral (e.g., a broom), or negative (e.g., a gun). Presenting the stimuli in this way allowed us to dissociate neural representations of emotion and identity information related to people and objects that were either modality-specific or multimodal. Consistent with prior studies, our univariate results show that a known face-selective region in the posterior STS is sensitive to both auditory and visual modalities, while a region located in the middle of the STS exhibits a selective response to human vocalizations. In addition, RSA suggests that the vocalization-selective middle region of the STS represents emotional information in human vocalizations, while the face-selective posterior region shows a weaker preference towards emotions compared to identity information for both images and vocalizations. Furthermore, we found a general bias towards vocalizations and emotional information in the upper bank of the STS, and this effect is more prominent in the right hemisphere. Taken together, these results suggest that emotion information is represented in a category- and modality-specific format in the human STS.

Disclosures: J.M. Denning: None. **A.S. Persichetti:** None. **A. Martin:** None.

Poster

P808. Human Behavior, Disorders, and Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P808.02

Topic: H.06. Social Cognition

Support: NIH/NIA R01AG060054
NIH/NIA R01 AG070227
NIH 1UL1TR003098

Title: High socio-economic status is associated with low brain entropy

Authors: A. Q. WANG[1], ***Z. WANG**[2];
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Abstract: Background and hypothesis: Socio-economic status (SES) drives many aspects of human life. Those of a higher SES often seem to perform better and be more successful. To find whether this is underlined by any brain mechanism, we characterized brain activity used brain entropy (BEN) mapping and assessed its associations with SES. Low BEN suggests high coherence of brain activity, which has been shown to be related to better brain function and neurocognition. A recent study has shown that better educated people have lower BEN than their counterparts and the associations were predominantly located in the frontal-parietal brain networks. Since education is related to socio-economic status too, we **hypothesized** that BEN in the frontal-parietal regions is inversely correlated with SES as measured by personal income. **Methods**: Preprocessed resting state fMRI (rsfMRI) data were downloaded from 1081 young healthy subjects from the Human Connectome Project. BEN maps were calculated from the rsfMRI data using BENtbx (https://cfn.upenn.edu/~zewang/BENtbx.php). BEN vs income correlation was evaluated using simple regression implemented in SPM12. Age, sex, and education years were included as covariates. Statistical significance was defined by $p<0.001$. Multiple comparison correction was performed with the permutation-based cluster size correction method. The cluster size was >150 ($q<0.5$). The mediation effect of education on the relationship between income and BEN was assessed. **Results**: Income was significantly related to education and fluid intelligence. Significant inverse correlation between income and BEN was found in the executive control network and the default mode network. The relation between income and BEN in these regions was significantly mediated by education. Personal income did not mediate the correlation between education and BEN. **Conclusion**: Higher SES is associated with better organized brain activity (lower BEN) in the frontal-parietal circuit, which is known to be related to fluid intelligence and higher-order function. The social-economic effects of BEN are caused by education. Our data suggests that low fronto-parietal BEN may represent a brain mechanism for the profound SES effects on brain cognition or human life. Future study will be required to assess the mediation effects of family income.

Disclosures: A.Q. Wang: None. **Z. Wang:** None.

Poster

P808. Human Behavior, Disorders, and Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P808.03

Topic: H.06. Social Cognition

Support: CONACYT scholarship to M.D. 422636

Title: Intrinsic markers of the (dis)honesty

Authors: *M. DELGADO-HERRERA, B. I. ARCE LÓPEZ, M. GIORDANO;
Dept. de Neurobiología Conductual y Cognitiva, Inst. de Neurobiología, UNAM, Querétaro, Mexico

Abstract: Honesty can be defined as acting in accordance with the moral rules and social norms of a community and it includes giving up the opportunity to make a profit, for example, by refraining from cheating to obtain money. (Dis)honesty can be influenced by intrinsic factors, that is, cognitive, psychological, and emotional individual differences that characterize each person. The aim of this research was to assess whether intrinsic factors such as intelligence, gender, executive functions such as working memory and inhibitory control, social cognition skills (including empathy and theory of mind), and personality traits are linked to (dis)honesty. Fifty-six neurotypical young adults participated in this experiment. (Dis)honesty was evaluated with a novel "Honesty task" based on whether the participants were or not were tempted to: a) review the answers that the researcher had "accidentally" given them and b) misreporting his/her knowledge in order to pretend to know more. An honesty index was created based on a qualitative analysis of the behaviors shown by the participants during the "Honesty Task". The honesty index ranged from one to four, that is, from honest to dishonest. Number one of the honesty index included a combination of behaviors that denoted with certainty the participant's honest decision making; conversely, number four included the behaviors most representative of dishonesty. We found a high percentage of participants in number four of the honesty index, which corresponds to the most dishonest behaviors. The honest group had higher scores for Empathic Concern and Neuroticism than the dishonest group. We performed a cluster analysis based on the normalized intrinsic factors scores. Differentiation into four clusters provided the clearest delineation. The results of the cluster analysis replicated the results of the group analysis (honest vs. dishonest). Cluster 1 (the honest group) was made up of participants with high Neuroticism, Empathic concern and Personal Distress scores and with low Conscientiousness scores, and these scores are opposite in the participants who made up Cluster 4 (the dishonest group). In conclusion, although dishonesty is a highly complex behavior that can be influenced by a wide range of both, contextual and intrapersonal factors, this research suggests that prosocial behaviors such as empathic concern, theory of mind skills and personal distress are a cornerstone of (dis)honesty.

Disclosures: M. Delgado-Herrera: None. **B.I. Arce López:** None. **M. Giordano:** None.

Poster

P808. Human Behavior, Disorders, and Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P808.04

Topic: H.06. Social Cognition

Support: National Science Foundation SBE Postdoctoral Research Fellowship [Grant No. 1911783]
National Science Foundation [Grant No. SBE-1835239]

Title: Neural coordination between participants during naturalistic perception: Towards a marker of social dysfunction in schizophrenia

Authors: *E. C. BAEK, R. HYON, K. LOPEZ, M. A. PORTER, C. PARKINSON, E. A. REAVIS;
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Abstract: The neural factors that allow humans to forge social bonds more readily with some people than with others remain poorly understood. In particular, very little is known about how individual differences in brain activity contribute to chronic deficits in the formation and maintenance of social relationships in clinical disorders such as schizophrenia. Recent evidence from functional magnetic resonance imaging (fMRI) suggests that healthy individuals forge social ties more readily with people who process the world in a particularly similar way to them than they do with others. Such work has suggested that two people are more likely to befriend each other if particular regions of their brains respond similarly to naturalistic movies (e.g., regions of the default mode network [DMN]). One can quantify the response similarity for particular brain regions and pairs of participants by calculating inter-subject correlations (ISCs) of fMRI time series. Although it is known that ISCs can predict social-bond formation in specific pairs of people, it is not clear how ISCs relate to more general measures of social dysfunction (e.g., social disconnection). Further, the feasibility of ISC research has not been established in schizophrenia, although it may be a useful tool for understanding social dysfunction in the disorder. We collected fMRI data during naturalistic perception in 64 healthy adults. We also piloted the same paradigm in 6 stable adult outpatients with a diagnosis of schizophrenia. We assessed ISCs in a set of anatomically-defined cortical and subcortical regions that span the entire brain. Participants also provided measures of social disconnection. In each group, ISCs exhibited expected stimulus-driven patterns (e.g., high ISCs in visual and auditory cortex; low ISCs in motor and somatosensory cortex), providing evidence in support of data validity in both groups. In the healthy group, across several brain areas (including DMN regions), participants with high subjective and/or objective measures of social disconnection (i.e., loneliness or having few friends, respectively) tended to have lower ISCs than well-connected participants. ISCs of individuals in the schizophrenia pilot sample were very low for the same regions. Our findings suggest that individuals who process naturalistic stimuli in idiosyncratic ways may have more difficulty forming social ties. Additionally, the preliminary data in the schizophrenia group demonstrates the feasibility and promise of the method in a new clinical population. They also hint at the potential for this approach to shed light on the neural determinants of social dysfunction in serious mental illnesses.

Disclosures: E.C. Baek: None. **R. Hyon:** None. **K. Lopez:** None. **M.A. Porter:** None. **C. Parkinson:** None. **E.A. Reavis:** None.

Poster

P808. Human Behavior, Disorders, and Mechanisms I

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Program #/Poster #: P808.05

Topic: H.06. Social Cognition

Support: Young Academy Groningen

Title: Working memory load impairs tacit coordination and inter-brain EEG synchronization

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Abstract: Accomplishing shared goals with others often requires the ability to relate their actions to underlying mental states such as intentions and beliefs (Theory of Mind; ToM). Working memory load has previously been found to impair ToM processing in single-participant paradigms where participants are presented with static, pre-given social scenarios. However, ToM processing in more naturalistic, dynamic multi-participant scenarios remains relatively unexplored. “Hyperscanning” is a promising method for studying multi-participant coordination, in which brain activity is measured in multiple participants simultaneously during live interaction. Previous hyperscanning findings show greater inter-brain synchronization (IBS) in interacting participants compared with non-interacting participants, especially in regions previously implicated in ToM, and in alpha and theta frequency bands associated with attentional gating processes. These findings suggest that IBS may reflect a correspondence between the mental states of one participant and the (ongoing updating of) representation of those mental states by another participant (via ToM and requisite working memory resources). We sought to make this widely-held but untested interpretation of IBS more explicit by investigating the relationship between IBS and performance in a multi-participant ToM task. Participant dyads performed a computerized ToM task in which they were presented with novel images each trial and were instructed to each choose the same image, with the chosen images shown at the end of each trial. In addition, participants in the high-load condition performed one trial of an ongoing n-back task at the beginning of each ToM trial, requiring them to update and maintain numbers in working memory while performing the ToM task. Significantly lower P3 ERP amplitude at image onset in the high-load condition indicated that allocation of working memory resources was attenuated during ToM processing. Linear Mixed Models and Generalized Additive Models revealed ToM performance and IBS in a small number of channels significantly decreased in the high-load condition, and increased more slowly across trials, as compared with a low-load control condition. These results indicate that, expanding on traditional single-participant paradigms, ToM loads on working memory in a two-person coordination task, and that IBS is sensitive to changes in ToM induced by working memory load.

Disclosures: L.A. Newman: None. **M.K. van Vugt:** None.

Poster

P808. Human Behavior, Disorders, and Mechanisms I

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Program #/Poster #: P808.06

Topic: H.06. Social Cognition

Support: DFG Excellence Strategy - EXC 2002/1 "Science of Intelligence" – project number 390523135

Title: Am I speaking to a human, a robot, or a computer? Neural representations of task partners in communicative interactions with humans or artificial agents.

Authors: *D. PISCHEDDA[1], A. LANGE[1], M. KIRTAY[2], O. A. WUDARCZYK[3], R. ABDEL RAHMAN[3], V. V. HAFNER[2], A. K. KUHLEN[3], J.-D. HAYNES[1];
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Abstract: When people speak, they adapt their language to the specific conversational partner (e.g., Brennan & Hanna, 2009). Partner-adapted language production has been associated with the ability to take another person's perspective (i.e., mentalizing; Frith & Frith, 2006). This skill is supported by a brain network including ventromedial prefrontal cortex (VMPFC), precuneus, and temporoparietal junction (TPJ, Molenberghs et al., 2016). Partner-adapted speaking requires high flexibility. The brain supports flexible behavior by pre-activating the cortical structures needed to perform an upcoming task (e.g., Pischedda et al., 2017; Sakai & Passingham, 2003). These include ventrolateral prefrontal cortex (VLPF) and parietal cortex (e.g., Pischedda et al., 2017). A previous study (Kuhlen et al., 2017) showed that VMPFC and VLPFC encode information about the intention to address a conversational partner (a human confederate) as compared to having no conversational partner (to test a microphone). This suggests an involvement of mentalizing when addressing a human partner. As artificial agents are becoming frequent conversational partners, we explore, at the neural level, the difference between communicating with artificial agents and human partners (e.g., Rauchbauer et al., 2019). Specifically, we investigate partner-specific neural representations when preparing to address different task partners: a human, a Pepper robot, or a computer. Fifty participants (32 female) worked on a spatial task together with their task partners while undergoing functional magnetic resonance imaging. On each trial, the participant instructed one of the partners to perform an action and observed the outcome of the partner's action. Instructions were identical across partners. This paradigm allowed us (1) to identify neural representations of task partners prior to speaking and (2) to assess differences between these representations. We applied multivariate decoding methods (Haynes, 2015) to identify brain regions encoding information about different

communication partners. We show that both VMPFC and VLPFC, as well as precuneus and parietal cortex, encode information about the specific task partner that is addressed. Moreover, univariate analyses revealed higher activation in TPJ when talking to a human as compared to Pepper. These results suggest that while some areas of the mentalizing network are specifically activated when addressing a human partner, others respond to robots as well. Identifying which critical features maximize activation of the mentalizing network will allow for designing robots that can be accepted as interaction partners in everyday life.

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Poster

P808. Human Behavior, Disorders, and Mechanisms I

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Program #/Poster #: P808.07

Topic: H.06. Social Cognition

Support: Nova Southeastern University HPD Research Grant
Nova Southeastern University Quality of Life Grant

Title: Cognitive load and eye movement patterns during health literacy and executive functioning tasks

Authors: *G. CAVANAUGH[1], R. OWNBY[2], S. GRANT[2], M. ABREU[2], P. HARDIGAN[3], V. JOHNSON[4];
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Abstract: Introduction. Cognitive function decline is an expected consequence of aging; furthermore, risk factors, such as repeated exposure to stress, impact information processing and decision-making regardless of age. Many studies have been successful at evaluating cognitive processing in non-verbal individuals through eye-tracking measures, though to date, no studies have examined the impact of homelessness on executive functioning using eye-tracking measures. This study examines eye-movement and pupil diameter as an indicator of cognitive load during a computerized assessment of health literacy and executive function in two socioeconomic groups, proposing that individuals from disadvantaged backgrounds will demonstrate greater difficulty with executive functioning. **Methods.** The Montreal Cognitive Assessment (MoCA) and Fostering Literacy for Good Health Today (FLIGHT)/ Vive Desarollando Amplia Salud (VIDAS), a progressively more difficult health literacy tool, were used as the primary assessments; other assessments included grip strength and the Trail Making test. Participants were 85 adults (26-65 years old), 65 were homeless and compared to 20 persons

with stable housing. Participants completed assessments on a laptop mounted with a Tobii eye-tracker. Visualization and numerical data were exported from Tobii ProLab and were analyzed using SPSS v.27. **Results**. Mean baseline pupil diameter were 2.83mm ± 0.5329 (homeless) and 2.62mm ± 0.2366 (control). Both groups demonstrated significant pupil dilation from baseline while progressing through the tasks divided into 8 frames $F(8, 266156)= 1239.43$, $p = .000$. A visual analysis of the data suggests that the homeless group showed the greatest change, with more guessing events, more disorganized patterns of eye-movement, and more incorrect FLIGHT/VIDAS answers (mean score 11.34 vs 27.57). The homeless group's scattered eye-movement patterns were consistent with an increase in errors during the Trail Making Test, lower MoCA scores (M= 24.72 vs M= 27.1), and weaker grip strength. **Conclusions.** Results suggest that individuals who are homeless may have difficulty processing information from complex cognitive tasks. Furthermore, the homeless population may include individuals with unidentified and undocumented cognitive deficits that might prevent them from successfully processing information for better decision-making.

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Poster

P808. Human Behavior, Disorders, and Mechanisms I

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Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P808.08

Topic: H.06. Social Cognition

Support: Wellcome Trust 204702
ERC 819040

Title: Shared responsibility in collective decisions decreases the sense of agency in the human brain

Authors: *M. EL ZEIN[1], R. J. DOLAN[2], B. BAHRAMI[3];
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Abstract: Distributing responsibility among several individuals in social decisions helps minimize the burden of responsibility for the consequences of decisions. We investigated the neural correlates of this effect in a magnetoencephalography (MEG) study. Human participants performed a reward-based learning decision-making task in contexts where we parametrically modulated responsibility for decision outcomes and measured ratings of how responsible subjects felt after their decisions. Participants' subjective sense of responsibility decreased parametrically as a function of group size. An overall self-serving bias was evident in

participants consistently feeling more responsible for positive as compared to negative outcomes. Trial by trial regressions of MEG signals against responsibility contexts revealed both outcome-dependent and outcome-independent neural signatures that covaried with responsibility. Outcome processing was parametrically reduced with decreasing responsibility at 200ms post outcome onset, an effect centred on parietal and precentral brain regions. Prior to revealing an outcome, motor preparation for decisions at 500ms after stimulus onset was attenuated for social decisions. Interestingly, the boost in responsibility for positive outcomes was stronger in social contexts and associated with increased activity in regions related to social and reward processing, including orbito-frontal cortex and the superior temporal sulcus. Together, these results show that sharing responsibility with others reduces agency through an influence on pre-outcome motor preparation and post-outcome processing, while affording an opportunity to flexibly take credit for positive outcomes.

Disclosures: M. El Zein: None. **R.J. Dolan:** None. **B. Bahrami:** None.

Poster

P808. Human Behavior, Disorders, and Mechanisms I

Location: Virtual Only

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Program #/Poster #: P808.09

Topic: H.06. Social Cognition

Title: Paired creative idea generation and behavioral synchrony

Authors: *M. SLAYTON[1], A. RAGNHILDSTVEIT[2], N. RINCON[1], J. IBARRA[1], C. TAN[1], A. ADHIKARI[1], R. BEATY[3], J. SCHOOLER[4], T. WHEATLEY[5], P. WHITEHEAD[1], P. SELI[1]; [1]Dept. of Psychology & Neurosci., Duke Univ., Durham, NC; [2]Dept. of Psychology, Univ. of Utah, Salt Lake City, UT; [3]Dept. of Psychology, Pennsylvania State Univ., University Park, PA; [4]Dept. of Psychological and Brain Sci., Univ. of California, Santa Barbara, Santa Barbara, CA; [5]Dept. of Psychological and Brain Sci., Dartmouth Col., Hanover, NH

Abstract: The capacity to share creative ideas enables humans to communicate, work together, and produce novel solutions necessary for social development. This study aimed to examine how interpersonal neural synchrony affects creativity in dyadic pairs who are engaged in a brainstorming discussion. Dyads performed a modified alternate uses task (AUT) virtually over Zoom. We then assessed level of creativity (measured by creativity rank, ideas, and idea length) as well as interactivity (measured by speaker switching, pause, pause length, overlaps/interrupts, acknowledgements, and content) unrelated to the task. Each dyad completed four one-minute rounds, first responding to a conventional AUT prompt followed by prompts that built off the previous. Creativity was strongly correlated with off-task chatting ($r = 0.65$, $P = 0.02$) and verbal acknowledgment (e.g. ‘yeah’ or ‘uh-huh’) ($r = 0.52$, $P = 0.07$), and moderately correlated with speaker switching ($r = 0.41$, $P = 0.16$). These findings are consistent with previous research

indicating that group interaction improves creative performance among pairs, and further implicates underlying interactive mechanisms that may be responsible. Future work will explore temporal dynamics of creative cognition in dyads using EEG hyperscanning while participants complete a group AUT.

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Poster

P808. Human Behavior, Disorders, and Mechanisms I

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Topic: H.06. Social Cognition

Support: P50 NS022343/NS/NINDS NIH HHS/United States
NSF SL-CN grant 1640909

Title: Fixation duration and location differences between change blindness task phases for adolescents with ASD

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Abstract: The ability to detect and respond to changes in a visual scene is essential for functional orienting in our dynamic natural world. We are interested in how unique visual and social perceptual abilities affect the way people explore changing scenes. We used a change blindness task to characterize eye movements and fixations both in adolescents with an Autism Spectrum Disorder (ASD) and Typically Developing (TD) adolescents. Social and non-social changes were made in each of 16 natural scenes. These changes were balanced for low-level salience, eccentricity, and size. For each presentation, participants first freely viewed a static image for 6s (admire). Then, a modified image alternated with the original for 1s each with a 200ms black mask appearing between alternations to mitigate motion artifacts. Participants had up to 60s to identify the change (search). To encourage young participants, we simplified the task after 17s, decreasing the mask duration by 50ms after every other alternation.
We compared the gaze behavior and reaction time responses of ASD and TD participants. We observed that participants with ASD had significantly shorter fixation times than their TD peers in the search portion of each trial, but that this effect is weaker in the "admire" portion of each trial. We observed no between-group differences in search success or time to success. Similarly, we observed no consistent differences in speed or success of search for social or non-social

targets overall or by group. However, when the scene was divided into social versus non-social content regions, there was a consistent and large difference in fixation dwell time in both groups. The fixation dwell times for social content were longer during both the admire and search phase, but the effect was stronger in the search phase of the task. During the admire phase, both groups preferentially looked at the social content in the first second and the rest of the scene over the next 5s. For the first two alternations of the search phase, TD participants looked at social regions, but participants with ASD continued to search the scene. The tendency to look at the social regions gave TD participants a slight advantage in identifying social changes, at the cost of a slight advantage for participants with ASD for non-social changes.
These results reveal that the nature of the task has substantial effects on content-specific fixation times that are durable across groups. Also, while both groups indistinguishably preferred social content during free viewing, TD participants preferred to look at social regions during the search phase even in the absence of a task-related benefit.

Disclosures: J. Snider: None. **S. Hacker:** None. **S. Liu:** None. **L. Chukoskie:** None.

Poster

P808. Human Behavior, Disorders, and Mechanisms I

Location: Virtual Only

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Program #/Poster #: P808.11

Topic: H.06. Social Cognition

Title: Social media use experience escalates news sharing behavior: Behavioral Experiments on infodemics

Authors: *H. ONISHI, K. I. KOBAYASI;
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Abstract: In recent years, there has been a phenomenon called infodemic, which is the sharing and spreading of fear-inducing and untrue news, and it has become a major social problem, especially in the era of the Covid-19 pandemic. In other words, infodemics arise from the sharing of news. A factor involved in the active sharing of news is the experience of using social media. Social science research suggests that as people become more experienced in using social media, they are more willing to share the news. Considering neuroplasticity, it is conceivable that the more experience we have with social media, the more our neural circuits will be suited to it, however, it is not clear how this affects infodemics. We hypothesized that the experience of using social media to communicate one's opinions to the public was a factor in the infodemics. To replicate the subjects' actual use of social media, we conducted a behavioral experiment that mimics social media. Participants were presented with 60 different news articles and instructed to perform three actions on them: "Like", "Share", and "Comment". The stimuli were news related to Covid-19 and other topics, and these articles consisted of 20 positive news, 20 negative

news, and 20 neutral news. We divided our subjects into two groups: those who had at least one year of experience using social media to communicate their opinions to the public ("experienced group") and those who did not ("naïve group"). The experiment was divided into two phases: the first 30 sessions were conducted under the assumption that one's actions would be shown to the world (public condition), and the second 30 sessions were conducted under the assumption that one's actions would only be shown to one's friends (private condition). Our results showed that subjects expressed significantly more "like" for positive articles, which suggests that the use of social media was replicated in the experiment. In the experienced group, a significant increase in the number of comments to news about Covid-19 in the public condition trials was observed, compared to the condition of the private trials. This suggests that people with experience in using social media are publishing their own opinions, which may be the cause of the infodemics. Interestingly, in the experienced group, a significant increase in the number of “shares” of positive news in the public condition trials was observed, but not in negative news. From these results, we suggest that the experience of usage of social media affects the news sharing behavior, and it can act as a positive influence on the public, rather than causing the infodemic by promoting the negative news sharing.

Disclosures: H. Onishi: None. **K.I. Kobayasi:** None.

Poster

P808. Human Behavior, Disorders, and Mechanisms I

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Program #/Poster #: P808.12

Topic: H.06. Social Cognition

Support: Funding from University of Hamburg and DFG; SCHW1357/16-1

Title: Acute stress acts as a safeguard against social influence in risky decision-making

Authors: *L. KLUEN[1,2], M. BOGDANOV[1,3], L. SCHWABE[1];
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Abstract: Acute stress is well-known to affect decision-making under risk. However, previous work has mostly investigated this effect in individual subjects making isolated decisions, neglecting other potential sources of decision biases, such as the social context. Given that providing information about other people’s choices may nudge individuals’ decisions to conform with the observed behaviour, we here aimed to test how stress impacts the effect of social influence on decision-making behaviour.To this end, 86 participants underwent the Trier Social Stress Test (TSST) or a non-stressful control manipulation before they completed a social decision-making task, in which they were asked to choose between a risky and a safe gamble

option with different expected value propositions. During ‘information’ trials, information about how two ‘mock’ participants decided about the gambles was provided to the actual participant. In two thirds of the trials, this social information could be either expected value congruent (i.e., ‘mock’ participants chose the gamble with the higher expected value) or incongruent. Results revealed that participants’ choices in the control condition were indeed biased in the direction of the social information provided, whereas acute stress diminished the social influence on decision-making behaviour. This difference was most clearly pronounced at the beginning of the task. Interestingly, even when ‘mock’ participants chose the gamble with the lower expected value, participants in the control condition were also more likely to follow this behaviour and thus acting against their own interest. This was not seen in stressed participants as their choice behaviour did not differ between trials in which the social information was expected value conform or was not. Our findings provide the first insight into how stress may affect, i.e. reduce, the influence of social impact on risky decision-making behaviour. Possibly acting as a protective mechanism to reduce negative choice impact.

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Poster

P808. Human Behavior, Disorders, and Mechanisms I

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Program #/Poster #: P808.13

Topic: H.10. Human Learning and Cognition

Support: NSF Grant 1125596

Title: Dynamics of hidden brain states when people solve a verbal puzzle

Authors: *Y. YU[1], Y. OH[2], J. KOUNIOS[2], M. BEEMAN[1];
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Abstract: Dynamics of hidden brain states when people solve a verbal puzzle
When people try to solve a puzzle, they go through distinct steps: encoding the problem, retrieving the meaning, ideation, evaluation, etc. Those steps often occur recurrently and spontaneously in varying orders. To extract the discrete dynamics that can characterize such multi-stage solving processes, we applied an unsupervised machine learning method to continuously recorded EEG data. EEG was recorded for 39 participants when they attempted Compound Remote Associates problems (CRA, Bowden & Jung-Beeman, 2003). CRA problems are verbal puzzles that can be solved in either insight-leaning or analysis-leaning manner. Self reports for insight or analytic solving processes have been widely used to study neural substrates underlying those strategies (e.g., Becker et al., 2020; Kounios et al., 2006). We fitted a Hidden Markov Model to the time-frequency transformed EEG signals and decoded each trial as a time-

resolved state sequence. The model identified several hidden states where each state is characterized by a distinct, spectrally-resolved power topography. For example, one of the states features widespread activation in alpha-band frequency (8-9 HZ and 10-13 HZ) but neutral in other frequency bands, while some states feature localized theta-band activation. The probability of this state peaks before stimulus onset and decreases before response. Interestingly, this "alpha-dominated state" is more likely to be visited and maintained in a trial solved by insight than one by analysis, echoing the earlier findings on alpha power at posterior parietal electrodes before response (Jung-Beeman et al., 2004). Our analysis further suggested that the steeper decline in the probability of this state before response is associated with reporting a solution by insight, compared to a solution by analysis. Overall, the hidden brain states exhibit different transition patterns through various stages of a trial and can be related to behavioral outcomes. This paradigm provides a way to extract dynamic features that characterize creative problem solving and potentially provide a novel window into the nature of the underlying cognitive processes.

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Poster

P829. Cortical-Hippocampal Circuits in Human Memory

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Program #/Poster #: P829.01

Topic: H.07. Long-Term Memory

Support: NRF-2020R1A2C2007770
NRF-2020R1A4A1018714
NRF-2020M3E5D9079913

Title: Memory retention period-dependent human hippocampal processing during learning

Authors: *M. KWON[1], S.-H. LEE[1,2];
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Abstract: The capability to maintain important information for a long time is essential for survival. Here we investigate why some memories are forgotten in a few days while other memories remain for a month or more, although both types of memory are referred to as long-term memory. Given that the hippocampal activity during learning is correlated with subsequent memory performance, we focused on whether these memories are differentially processed in the hippocampus from learning stage. Using functional magnetic resonance imaging (fMRI) and trial-by-trial pattern similarity analysis, we tracked how hippocampal representations of associated information changed during repeated association learning. We found that the representations for associated stimuli in the hippocampus changed dynamically during learning

and that this dynamic representational change was related to the subsequent memory performance. Remarkably, while the degree of shared representations for pairs remembered for at least four weeks were increased and maintained during learning, the shared representations for pairs forgotten before four weeks were observed only temporarily. Furthermore, for long-lasting pairs but not for short-lived pairs, shared representations during learning were also observed in the ventromedial prefrontal cortex. These results suggest that there are two types of long-term memory with different degrees of maintenance and that which type is expressed depends on how information is processed in the hippocampus and prefrontal cortex during learning.

Disclosures: M. Kwon: None. **S. Lee:** None.

Poster

P829. Cortical-Hippocampal Circuits in Human Memory

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Program #/Poster #: P829.02

Topic: H.07. Long-Term Memory

Support: UK Medical Research Council G03000117
UK Medical Research Council G1002276

Title: The medial temporal lobe in developmental amnesia: structure and anatomo-functional correlations

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Abstract: BACKGROUND: Early-life episodes of hypoxia-ischemia can target the hippocampus and lead to developmental amnesia (DA). Patients with DA exhibit severe episodic memory impairment, but their semantic memory is largely preserved. It is not clear, however, whether the residual hippocampus plays a role in encoding and/or retrieval of new information, or the surrounding neocortical areas rescue hippocampal function after early injury.
AIMS: We aimed to investigate anatomo-functional correlations between cognitive performance and the volumes of (a) residual hippocampal subregions, and (b) parahippocampal gyrus in patients with DA.
METHODS: We used manual segmentation on MRI acquisitions to estimate the volume of three hippocampal subregions: uncus, CA-DG (including CA fields and dentate gyrus) and subicular complex; and the volume of three surrounding cortical areas: entorhinal, perirhinal and

parahippocampal cortices, in an exceptionally large cohort of 23 patients with DA and 32 controls of comparable age. The groups were assessed on: i) the Four Mountains Test, which provides measures of spatial perception and immediate spatial memory, and ii) the Doors and People Test, which provides equated measures of verbal and visual recognition and recall. RESULTS: In patients with DA, the level of CA-DG and subicular complex atrophy was about 40% while the atrophy of the uncus was moderate (−26%). In contrast, the entorhinal and perirhinal cortices showed only a mild (−5%) volume reduction in patients with DA and the volume of the parahippocampal cortex was not statistically different from controls'. Patients' spatial perception and spatial memory performance correlated negatively with the volume of the Subicular complex. Also, patients' recall, but not recognition memory performance, correlated negatively with the volume of the Uncus. We did not observe correlations between the volume of the entorhinal, perirhinal and parahippocampal cortices and either patients', or controls' performance.
CONCLUSIONS: Based on our findings, we propose that following early hippocampal damage preserved cortical areas surrounding the hippocampus rescue the encoding and storage of decontextualized memoranda depending on the extent of damage to the hippocampal subregions. Thus, the greater the damage, the more likely that the rhinal cortices rescue memory processes. However, irrespective of this rescue and the extent of damage, the critical hippocampal-dependent function of recall remains irreversibly compromised.

Disclosures: L.J. Chareyron: None. **W.K. Kling Chong:** None. **M. Mishkin:** None. **N. Burgess:** None. **R.C. Saunders:** None. **F. Vargha-Khadem:** None.

Poster

P829. Cortical-Hippocampal Circuits in Human Memory

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P829.03

Topic: H.07. Long-Term Memory

Support: NIH 1U19NS107609-01
NIH/NINDS U01-NS108916
NINDS R37NS31135

Title: Awake ripples enhance emotional memory encoding in the human brain

Authors: *H. ZHANG[1,2], I. SKELIN[2], S. MA[2], M. PAFF[3], M. A. YASSA[4,5,2], R. T. KNIGHT[6,7], J. J. LIN[1,2,4];
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Abstract: Multiple mechanisms have been proposed to support the prioritized encoding of emotional experiences, including neuromodulatory effects on plasticity and interplay between the hippocampus and the amygdala. Sharp-wave/ripples (SWRs) are transient hippocampal oscillations (80-150 Hz), associated with synchronous neural activation in the hippocampus and the amygdala and implicated in binding anatomically distributed memory traces. Moreover, behaviorally relevant reactivation of emotional memory occurs during awake SWRs (aSWR), and aSWR disruption immediately post-experience interferes with memory utilization. In this study, we tested the hypothesis that post-encoding aSWRs could facilitate better emotional memory discrimination, through the coordinated hippocampal-amygdala memory reinstatement. Using the simultaneous intracranial electroencephalographic (iEEG) recordings in the human hippocampus and the amygdala (n = 17, 20 electrodes respectively) in 7 presurgical epilepsy patients (4 females, age 33 +/- 16) performing an emotional memory encoding and discrimination task, we first show stimuli with higher arousal were associated with better discrimination memory ($p = 0.047$, logistic linear mixed-effect model). Next, we demonstrated that during the immediately post-encoding period (i.e., shortly after stimulus presentation and before the next stimulus), the number of aSWR is predictive of both stimulus arousal ($p = 0.03$) and probability of later discrimination ($p = 0.03$, Wilcoxon signed-rank test). Next, we implemented representational similarity analysis to quantify memory reinstatement for each trial. We observed that the post-encoding aSWR is associated with reinstatement in the hippocampus and the amygdala ($p < 0.05$, permutation test). Moreover, the reinstatement exhibits region-specific double dissociation. Specifically, amygdala reinstatement is associated with higher arousal ($p = 0.035$), whereas hippocampal reinstatement is linked to later correct discrimination ($p = 0.008$, cluster-based permutation test). Finally, we tested if the memory reinstatement occurs simultaneously in the amygdala and hippocampus, by computing the outer product of regional reinstatement traces for individual aSWR events. The aSWR-locked joint memory reinstatement during the post-encoding period was predictive of memory discrimination ($p < 0.05$, permutation test). Together, these findings suggest the aSWRs-mediated memory reinstatement in the amygdala and hippocampus as a putative mechanism accounting for better remembering emotional experiences.

Disclosures: H. Zhang: None. **I. Skelin:** None. **S. Ma:** None. **M. Paff:** None. **M.A. Yassa:** None. **R.T. Knight:** None. **J.J. Lin:** None.

Poster

P829. Cortical-Hippocampal Circuits in Human Memory

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P829.04

Topic: H.07. Long-Term Memory

Support: NIH 1RF1MH114277).

NSF 1554871

Title: Human visual cortex supports recognition memory for simple, visual stimuli.

Authors: *N. M. DE LA ROSA-RIVERA[1], R. A. COWELL[2];
[1]Neurosci. and Behavior, Univ. of Massachusetts Amherst, Amherst, MA; [2]Psychological and Brain Sci., Univ. of Massachusetts, Amherst, MA

Abstract: Most theories of memory assume that long-term, declarative memory relies primarily on the medial temporal lobes (MTL). In contrast, the Representational-Hierarchical (RH) Account - challenges this claim, replacing it with two assumptions: (1) the brain contains a hierarchy of stimulus representations, from simple features (lines, blobs) in visual cortex, to conjunctions of features (which make up objects) in the temporal lobe, to high-dimensional associative representations in hippocampus; and (2) any region in the hierarchy can support memory for the information that it represents. The RH Account thus predicts that long-term, declarative memory for simple, purely visual stimuli should recruit regions outside of the hippocampus, specifically, in visual cortex. In this study, participants studied a set of simple visual stimuli for 10-20 days via visual search training. Stimuli were built from conjunctions of shape and fill pattern. At test in the MR scanner, subjects saw Studied items, Recombination items (novel recombinations of features of studied items), and Novel items. Participants responded “Old”, “Recombo” or “New”. Preliminary fMRI analyses contrasting "New" vs. "Recombo" trials (a “feature memory” signal, because only the features of these stimuli differ in familiarity; the conjunctions are novel in both) show widespread recruitment across visual cortex. Analysis of the *distribution* of voxel activations to New, Recombo and Old stimuli suggest that patterns elicited by Old and Recombo stimuli have reduced variance: voxels highly active to New stimuli "drop out" once stimulus features are learned. This occurs more prominently in later visual regions (e.g., LO, TO), hinting that visual feature memory may be localized to mid-level visual cortex.

Disclosures: N.M. de la Rosa-Rivera: None. **R.A. Cowell:** None.

Poster

P829. Cortical-Hippocampal Circuits in Human Memory

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Program #/Poster #: P829.05

Topic: H.07. Long-Term Memory

Support: NSERC Discovery Grant RGPIN-2018-05770

Title: Perirhinal cortex automatically tracks recent and lifetime experience with objects regardless of decision relevance

Authors: *H. YANG, S. KOHLER;
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Abstract: A large body of evidence points to a critical role of the perirhinal cortex (PrC) in recognition memory, i.e. the ability to recognize prior occurrence of objects. Most of this work in humans has focused on recognition of recent occurrence in a study phase. Neurophysiological studies with non-human animals have shown that the PrC tracks not only recent but also cumulative long-term exposure. In prior research from our laboratory we have developed a paradigm that allows for probing of responses to cumulative prior exposure on both time scales. Using this paradigm with words denoting concrete objects, an fMRI study with healthy participants revealed that activity in PrC tracks judged degree of recent and lifetime exposure, in line with the non-human neurophysiological findings. In the current study, we asked whether this response occurs automatically or whether it is tied to a requirement to make memory decisions on the relevant time scale. In an initial study phase, healthy participants (N=30, age range 18-40, 7 males, all right-handed) provided animacy judgement about object concepts that were repeated at different frequencies. In the test phase, participants rated relative presentation frequency of the recently studied concepts, as well as cumulative lifetime familiarity of non-studied concepts. Cumulative lifetime familiarity of the previously studied concepts was probed in a third phase. We again found that left PrC tracks judged degree of recent and lifetime exposure. Critically, our results revealed that these responses were not tied to a requirement for memory decisions on the relevant time scale. They also occurred when participants judged prior exposure on the other time scale, and even when no judgments of prior occurrence were required at all (in the study phase). This pattern of findings suggests that PrC automatically tracks prior occurrence across multiple time scales, and that the specific evidence on which pertinent memory decisions rely is computed in cortical regions other than PrC.

Disclosures: H. Yang: None. **S. Kohler:** None.

Poster

P829. Cortical-Hippocampal Circuits in Human Memory

Location: Virtual Only

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Program #/Poster #: P829.06

Topic: H.03. Decision Making

Support: NIH R01MH100095

Title: Dissociations among three functional subnetworks in prefrontal cortex and resolution of the exploration/exploitation dilemma through interactions with hippocampus

Authors: *A. E. PAPALE[1], M. N. HALLQUIST[2], A. Y. DOMBROVSKI[1];
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Abstract: Interactions between hippocampus and ventromedial prefrontal cortex/orbitofrontal cortex (vmPFC/OFC) are thought to contribute to decision-making by optimizing future choice given past experience. The vmPFC has been shown to signal scalar value signals in neuroeconomic tasks and OFC has been broadly implicated in abstract state space representation. These structures facilitate rapid and adaptive transitions between exploration and exploitation. Prior work in our lab suggests that hippocampus helps to resolve the explore-exploit dilemma—posterior hippocampus encourages exploration through encoding of reward prediction errors and global value maximum locations, and anterior hippocampus facilitates exploitation strategies by encoding global uncertainty (value entropy) signals. However, the role of vmPFC/OFC, in association with hippocampus, in resolving the exploration-exploitation dilemma remains undetermined. Here, we investigate the role of fMRI signals in healthy human subjects from ventral prefrontal and orbitofrontal subnetworks (cognitive control, default mode network—DMN, and limbic network) on the clock task, a decision-making task where action values vary along a temporal interval with visuospatial cues. We used a reinforcement learning model that compresses value information based on the entropy of the value distribution (i.e., the agent selectively remembers preferred action values). We find that DMN activity correlates with negative entropy during exploitation, while limbic network activity correlates with the global value maximum location, suggesting a role in exploration. While control and limbic networks display positive correlation with reward prediction errors, strikingly, DMN displays a negative correlation. We further investigated how anterior and posterior hippocampus interact with these prefrontal networks in the context of exploration/exploitation. Preliminary findings suggest that anterior hippocampus activity post-reward is followed by a global increase in vmPFC/OFC activity. Posterior hippocampal activity following a negative entropy change also correlates with vmPFC/OFC activity. Both subdivisions of hippocampus convey information about entropy to the DMN and link the location of the value maximum to vmPFC/OFC activity. In summary, the DMN ventral prefrontal subnetwork is activated by hypothesis confirmation signals and receives hippocampal inputs that signal the spatial and temporal statistics of the environment.

Disclosures: A.E. Papale: None. **M.N. Hallquist:** None. **A.Y. Dombrovski:** None.

Poster

P829. Cortical-Hippocampal Circuits in Human Memory

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Program #/Poster #: P829.07

Topic: H.07. Long-Term Memory

Support: Wenner Gren Foundation
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European Brain Research Area
EU JPND

Title: Hippocampal Subfields Group progress update: The development of a valid, reliable, harmonized segmentation protocol for medial temporal lobe subregions

Authors: *V. CARR[1], D. BERRON[2], A. M. DAUGHERTY[4], R. LA JOIE[6], L. WISSE[3], K. M. AMUNTS[8], J. C. AUGUSTINACK[9], A. BAKKER[10], A. R. BENDER[11], M. BOCCARDI[12], M. BOCCHETTA[13], A. BURGGREN[14], M. CHAKRAVARTY[15], G. CHETELAT[16], R. DE FLORES[16], J. DEKRAKER[17], S.-L. DING[18], R. INSAUSTI[19], O. KEDO[8], S. MUELLER[7], N. OFEN[5], D. PALOMBO[20], N. RAZ[5], C. E. STARK[21], L. WANG[22], P. YUSHKEVICH[23], Q. YU[24], R. K. OLSEN[25];
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Abstract: Development, aging, and neurodegenerative pathologies are known to affect medial temporal lobe (MTL) subregions differently, selectively, and in a complex progression. Characterizing MTL subregional changes using high-resolution MRI may provide greater insight into these processes and better biomarkers for neurodegenerative disease. However, substantial differences in MTL subregion definition has hindered the ability to compare results across laboratories or draw robust conclusions. The Hippocampal Subfields Group (HSG) is an international organization seeking to address this issue by developing a histologically valid, reliable, and freely available segmentation protocol for high-resolution T2-weighted 3T MRI (http://www.hippocampalsubfields.com). Our workflow consists of four steps: 1) collecting histology samples labeled by multiple expert neuroanatomists to form a novel reference dataset to guide the development of the MRI segmentation protocol, 2) developing boundary definitions for each segment of the hippocampus (head, body, and tail) and MTL cortices on MRI, 3) assessing HSG community agreement with boundary rules using online questionnaires, and 4) testing reliability of the protocol definitions. We have developed a preliminary subfield segmentation protocol (i.e., completed steps 1-2) for the hippocampal head, body, and tail. With respect to the outer boundaries of the hippocampal body, i.e., boundaries of the hippocampus with neighboring regions, 29 labs reached consensus for all segmentation rules (step 3). We are

now assessing community agreement with segmentation rules for the inner boundaries of the hippocampal body, such as those between the cornu ammonis fields. Once consensus is reached, we will begin conducting reliability testing for both the inner and outer boundaries of the body (step 4). Similarly, we will assess community agreement with the head and tail segmentation rules, followed by reliability testing. Once completed, the harmonized protocol will significantly facilitate cross-study comparisons and provide increased insight into the structure and function of MTL subregions across the lifespan and in disease.

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Poster

P829. Cortical-Hippocampal Circuits in Human Memory

Location: Virtual Only

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Program #/Poster #: P829.08

Topic: H.07. Long-Term Memory

Support: National Key R&D Program of China (No. 2019YFA0709502)

Title: The effective connectivity of the human hippocampal memory system

Authors: *E. T. ROLLS[1], G. DECO[2], C.-C. HUANG[3], J. FENG[4];
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Abstract: The anatomical connections of the human hippocampus shown with tractography appear to be more extensive with many cortical areas than was previously known (Huang et al 2021), and this has implications for understanding the operation of human memory (Rolls 2021). Here this is developed with effective connectivity measurements based on the resting state BOLD signal which reveal the directionality and strength of the connectivity. The new asymmetric effective connectivity algorithm was developed from a previous approach (Deco et al 2019), and was used with a modified Human Connectome Project atlas (Glasser et al 2016). In 172 participants in the HCP imaged at 7T, it was found that a ventral ‘what’ hippocampal stream involves the temporal lobe cortex, perirhinal and parahippocampal TF cortex, and entorhinal cortex with generally stronger forward connectivity to the hippocampus than backwards. A

dorsal ‘where’ hippocampal stream connects parietal cortex with posterior and retrosplenial cingulate cortex, and with parahippocampal TH cortex, which in turn project to the presubiculum, which connects to the hippocampus. Thus some separate ‘what’ and ‘where’ streams can converge in the hippocampus, from which backprojections return to the sources. However, unlike the simple dual stream hippocampal model, there is some cross-connectivity between these systems before the hippocampus is reached; and the hippocampus has some effective connectivity with earlier stages of processing than the entorhinal cortex and presubiculum. For example, the human hippocampus receives effective connectivity from the posterior and retrosplenial cingulate cortex, and parietal cortex. This effective connectivity is consistent with the anatomical tractography, but goes beyond it by providing evidence about the directionality and strength of the connectivity. These findings provide the foundation for new concepts on the operation of the hippocampal memory system in humans, given that the connectivity allows for pre-hippocampal cortical areas to become specialized for particular functions, rather than primarily acting as serial gateways to and from the hippocampal system. A good example is the ventromedial visual area / TH system where spatial scenes are represented.

Deco G et al (2019). PNAS 116:18088-18097.
Glasser MF et al (2016). Nature 536:171-178.
Huang C-C, Rolls ET et al (2021) Extensive cortical connectivity of the human hippocampal memory system. Cerebral Cortex doi: 10.1093/cercor/bhab113.
Rolls ET (2021) Brain Computations: What and How. Oxford University Press: Oxford.

Disclosures: E.T. Rolls: None. **G. Deco:** None. **C. Huang:** None. **J. Feng:** None.

Poster

P829. Cortical-Hippocampal Circuits in Human Memory

Location: Virtual Only

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Topic: H.07. Long-Term Memory

Support: Neurtex Brain Research Institute Grant 19-02
National Institute of Child Health and Human Development Grant HD075800-05
National Institute of Neurological and Communicative Disorders and Stroke Grant DC009781
Medical Research Service of the Department of Veterans Affairs (51K6CX001644), Award CX000359
National Institute of Mental Health Grant 24600

Title: Two kinds of memory signals in neurons of the human hippocampus

Authors: *Z. URGOLITES[1], J. T. WIXTED[2], S. D. GOLDINGER[3], M. H. PAPESH[4], D. M. TREIMAN[5], L. R. SQUIRE[6,1], P. N. STEINMETZ[7];

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Abstract: Some recent work on the neural representation of episodic memory in the human hippocampus has identified a memory signal reflecting the generic status of test items (novel vs. repeated) whereas other work has identified another memory signal reflecting the repeated status of specific test items. Here, we investigated the co-existence and mnemonic functions of both episodic memory signals in the same experiment. We recorded single-unit activity from four brain regions (hippocampus, amygdala, anterior cingulate, and prefrontal cortex) of epilepsy patients as they completed a continuous recognition task. The generic signal was found in all four brain regions, whereas the item-specific memory signal was detected only in the hippocampus and reflected sparse coding. That is, for the item-specific signal, each hippocampal neuron responded strongly to a small fraction of repeated words, and each repeated word elicited strong responding in a small fraction of neurons. The fact that this neural code was sparse, pattern-separated, and limited to the hippocampus accords with longstanding computational models. We suggest that this item-specific episodic memory signal is fundamental, with the generic memory signal likely being derived from it.

Disclosures: Z. Urgolites: None. **J.T. Wixted:** None. **S.D. Goldinger:** None. **M.H. Papesh:** None. **D.M. Treiman:** None. **L.R. Squire:** None. **P.N. Steinmetz:** None.

Poster

P829. Cortical-Hippocampal Circuits in Human Memory

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P829.10

Topic: H.07. Long-Term Memory

Support: Neurtex Brain Research Institute

Title: Patterns of distributed neural firing code for perceptual and semantic features

Authors: *C. T. WEIDEMANN[1], P. N. STEINMETZ[2], J. JACOBS[1];
[1]Dept. of Biomed. Engin., Columbia Univ., New York, NY; [2]NeurTex Brain Res. Inst., Dallas, TX

Abstract: Intracranial recordings from neurosurgery patients have revealed that the spiking activity of individual neurons reliably predicts performance in episodic memory tasks. We investigated the extent to which the pattern of neural activity across multiple simultaneously recorded units revealed additional information about task performance. A total of 121 patients

with implanted micro-electrodes participated in at least one of 7 distinct tasks probing recognition memory and perceptual decisions. Across a total of 258 experimental sessions we identified 20,578 units which were predominantly located in medial temporal lobe structures. For each stimulus presentation in a given experimental session, we constructed a vector of neural firing from each unit's total number of spikes during the stimulus presentation. We quantified the similarity of the firing pattern associated with two stimulus presentations by calculating the cosine similarity between the corresponding vectors. The firing pattern across these units were similar for repeated items, but this similarity did not indicate whether participants would recognize the repetition. Additionally, we observed increased similarity of neural firing patterns for non-identical stimuli belonging to the same category, with higher similarity for stimulus pairs sharing a narrower category. The pattern of neural activity thus reflected the perceptual and semantic stimulus features rather than an episodic memory signal. These findings are consistent with a highly distributed neural code for perceptual and semantic stimulus features.

Disclosures: C.T. Weidemann: None. **P.N. Steinmetz:** None. **J. Jacobs:** None.

Poster

P868. Place Cells III

Location: Virtual Only

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Topic: H.09. Spatial Navigation

Support: JSPS Overseas Fellowship
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NIMH 1R01MH124867
NINDS 1U19NS104590
NINDS 1U01NS115530
NIH T32GM007367
NIMH F30MH125628

Title: Adaptive Stimulus Selection for Consolidation in the Hippocampus

Authors: *S. TERADA, J. O'HARE, T. GEILLER, Z. LIAO, B. VANCURA, A. LOSONCZY; Zuckerman Mind Brain Behavior Inst., Columbia Univ., New York, NY

Abstract: Associative memories guide behavioral adaptation by binding together outcome-predictive sensory stimuli. However, in a feature-rich environment, only a subset of stimuli may predict a desired outcome. How neural circuits enable behavioral adaptation by selectively and durably representing subsets of sensory stimuli pertinent to a specific outcome is not known. We investigated this feature selection process in the hippocampus during memory acquisition and subsequent consolidation. Two-photon calcium imaging of CA3 axonal projections to CA1

combined with simultaneous local field potential recordings revealed that CA3 projections encoding behaviorally-informative sensory stimuli were selectively recruited during the memory replay events that underlie hippocampal memory consolidation. These axonal projections formed sequential assemblies conjunctively linking sensory features to spatial location and thus reward proximity. By contrast, axons encoding uninformative, peripatetic sensory cues were notably suppressed during memory replay. Thus, while hippocampus comprehensively encodes real-time sensory environment, it implements a flexible filtering mechanism to maximize utility of memories destined for long-term storage. We propose utility-dependent recruitment of sensory experience during memory consolidation as a general coding principle for associative learning.

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Poster

P868. Place Cells III

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Program #/Poster #: P868.02

Topic: H.09. Spatial Navigation

Support: NIH 1R01MH124867
NIH 5U19NS104590
5F31NS110316-03

Title: Characterizing Direct Cortical Influences on Hippocampal region CA1 in Behaving Mice

Authors: *J. BOWLER, A. LOSONCZY;
Columbia Univ., New York, NY

Abstract: Characterizing Direct Cortical Influences on Hippocampal region CA1 in Behaving Mice
The hippocampus plays critical roles in navigation and episodic memory, with the most prominent neural correlate of these behaviors being “place cell” activity. It is widely assumed that afferent excitatory inputs to the hippocampus carry critical information required for generating and tuning these navigational and memory-related changes in hippocampal network dynamics. Two subregions of the Entorhinal Cortex(EC) have been particularly implicated in this process by providing distinct input streams to the hippocampus. Specifically, the medial EC (MEC) is thought to be involved primarily in processing spatial information, while the lateral EC (LEC) is thought to primarily process information related to individual items and how they have been arranged spatially or paired with a context. Despite these activity patterns having been well studied in the EC, a detailed understanding at the circuit level of the entorhinal influence on hippocampal population dynamics is still lacking. The direct projections from MEC and LEC to

CA1pyramidal cells (CA1PCs) offer a tractable bridge to this knowledge gap. Chronic, subcellular-resolution two-photon functional imaging of identified EC afferents during head-fixed virtual reality navigation and learning provides an opportunity for direct observation and characterization of the information transferred between the EC and CA1. These experiments test the hypothesis that CA1PCs receive spatial and sensory information from both MEC and LEC inputs which instruct CA1PC's population dynamics. Results indicate that it is possible to decode various navigationally relevant variables from the activity observed in the EC projections to CA1 including position, velocity and context. Increased activity is also observed in these afferents in response to task relevant salient cues as well as to goal proximity or anticipation. The direct EC to CA1 projection provides input to CA1PCs capable of evoking an abundance of both spatial and episodic representations, as observed in the hippocampus.

Disclosures: J. Bowler: None. **A. Losonczy:** None.

Poster

P868. Place Cells III

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Topic: H.09. Spatial Navigation

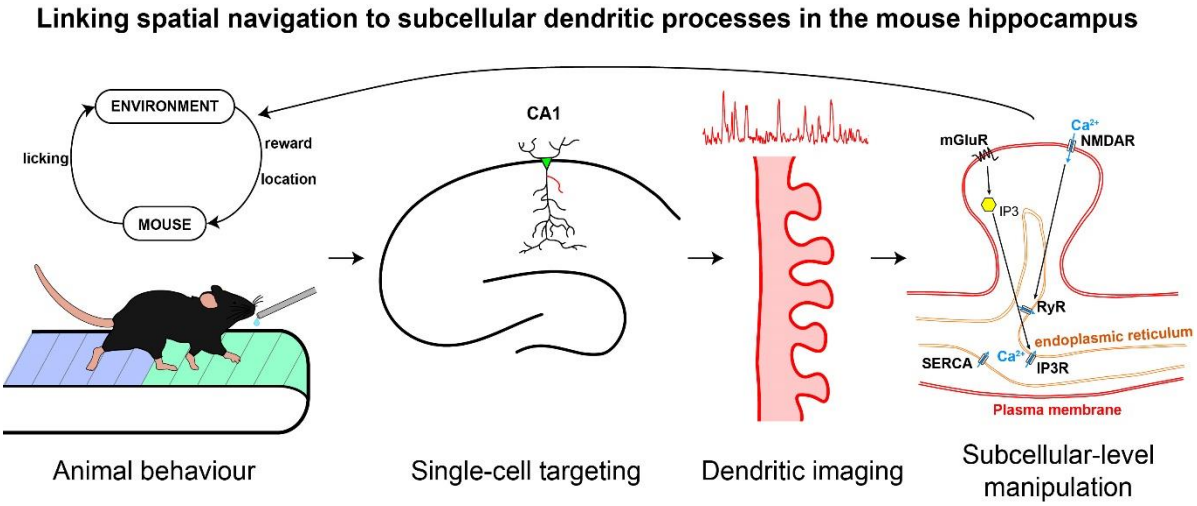
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Foundation Roger De Spoelberch

Title: Compartment-specific tuning of hippocampal dendritic feature selectivity by intracellular Ca^{2+} release

Authors: *J. K. O'HARE[1], Y. HIRABAYASHI[3], V. L. HEWITT[1], H. BLOCKUS[1], M. SZOBOSZLAY[1], S. V. ROLOTTI[2], T. C. GEILLER[1], A. NEGREAN[1], V. CHELUR[2], A. LOSONCZY[1], F. POLLEUX[1];
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Abstract: Dendritic Ca^{2+} is critical in determining how neurons respond and adapt to patterns of incoming excitation. The endoplasmic reticulum sequesters high concentrations of Ca^{2+} which it can release into the cytosol in response to strong synaptic activation; a phenomenon known as intracellular Ca^{2+} release (ICR). Given the centrality of Ca^{2+} to synaptic plasticity, these

feedforward release events have been suggested to shape and drive plasticity mechanisms underlying learning. Up to now, a number of methodological obstacles have prevented the investigation of ICR in mammalian neurons *in vivo*. By taking a combinatorial approach comprising single-cell electroporation of custom DNA cocktails, simultaneous *in vivo* imaging of dendritic and somatic activity during spatial navigation, optogenetic induction of spatial receptive fields, and an acute genetic approach to augment ICR itself, we were able to gain insight into the *in vivo* role of this subcellular process in pyramidal neurons of hippocampal area CA1. We found that ICR supports the establishment of dendritic feature selectivity and ultimately shapes dendritic integrative properties determining output-level receptive fields. This was particularly true of apical dendrites, which integrate distinct streams of information originating from a nonidentical set of afferent regions compared to basal dendrites. Thus, ICR, a subcellular postsynaptic process, acts cooperatively with circuit- and systems-level architecture *in vivo* to regulate behaviorally-relevant plasticity in a compartment-specific manner.



Disclosures: J.K. O'Hare: None. **Y. Hirabayashi:** None. **V.L. Hewitt:** None. **H. Blockus:** None. **M. Szoboszlay:** None. **S.V. Rolotti:** None. **T.C. Geiller:** None. **A. Negrean:** None. **V. Chelur:** None. **A. Losonczy:** None. **F. Polleux:** None.

Poster

P868. Place Cells III

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Topic: H.09. Spatial Navigation

Support: NIH Grant F31NS120783-01
Stanford Interdisciplinary Graduate Fellowship
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Columbia University Medical Scientist Training Program

Title: Hebbian plasticity of GABAergic synapses sufficient for consolidation of world structure by CA3 replay

Authors: *Z. LIAO[1], D. HADJIABADI[2], S. TERADA[1], I. SOLTESZ[3], A. LOSONCZY[1]; [1]Neurosci., Columbia Univ., New York, NY; [2]Stanford Univ., Palo Alto, CA; [3]Sch. of Med., Stanford Univ., Stanford, CA

Abstract: The replay of previously learned sequences during sharp-wave ripples at rest, a process required for memory consolidation, represents a defining computational feature of hippocampal population dynamics. However, the content of these replay episodes, and in particular whether specific experiences or an abstracted, utilitarian representation of the world are consolidated, remains controversial. Recent experimental work has shown that experience-specific representations are suppressed in sharp-wave ripples, while conserved, recurring representations are adaptively recruited. We propose a succinct but novel mechanism for this phenomenon: the reuse of the symmetric spike-time dependent plasticity rule (sSTDP), previously reported at CA3 excitatory synapses, to remodel inhibitory synapses as well. We show using three levels of modeling—random network, detailed biophysical, and abstract normative—that Hebbian plasticity of GABAergic synapses is sufficient to accurately reproduce the experimental findings. We make the key prediction that inhibitory plasticity is not only sufficient but necessary to functionally filter noisy observations in order to learn the latent statistical structure of the world. We further predict that ablating inhibitory plasticity leads to the disruption of replay and the emergence of pathological offline dynamics. Finally, we propose a number of experiments to validate these predictions in both physiological and pathological conditions. Our theoretical work here outlines a potential direct link between the synaptic and cognitive levels of memory consolidation, with implications for both normal learning and neurological disease.

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Poster

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Topic: H.09. Spatial Navigation

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Title: Behavioral state-dependent electrical signaling of neuronal dendrites

Authors: *A. NEGREAN[1], Z. LIAO[1], D. LI[1], C. YANG[1], N. E. MCCLAIN[1], D. HOLDER[1], G. ZHANG[2], S. W. EVANS[2], M. CHAVARHA[2], M. Z. LIN[2], A. LOSONCZY[1];
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Abstract: Excitation, inhibition and a variety of ion-channels interact at the level of dendrites to shape neuronal output. Studying this input-output transformation is a formidable technical challenge. In the awake behaving animal, sharp electrode and path-clamp based dendritic recordings are hampered by movement artifacts and development of optical means of measurement is in great need. Calcium imaging offers sufficient signal-to-noise ratio to measure neuronal output and suprathreshold dendritic activity, however, relying on depolarization, it provides only a limited view of dendritic excitability and its role in synaptic integration.
Recent advances in membrane potential imaging using ASAP-based genetically encoded voltage sensors demonstrate the feasibility to record somatic membrane potential changes in the awake behaving mouse [1]. Recent work from our group [2] showed that we can resolve suprathreshold dendritic activity in hippocampal CA1-area pyramidal neurons in awake mice running on a treadmill.
With the goal to understand how different behavioral states influence the electrical activity of dendrites, we further characterize ASAP3b sensor kinetics at 37C through a Markov-chain model and extend voltage measurements over the entire dendritic tree and across several electrophysiologically defined behavioral states: immobility with and without sharp-wave/ripples and locomotion-dependent theta oscillations. During locomotion on a cued belt we find multiple electrically place tuned compartments tiling the entire belt. During sharp wave/ripples, we find the cell body and the dendritic tree to be, on average, hyperpolarized, while local field potential theta-oscillations have a corresponding intracellular membrane potential oscillation distribution within the dendritic tree.
References
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1. V. Villette and M. Chavarha et.al. "Ultrafast Two-Photon Imaging of a High-Gain Voltage Indicator in Awake Behaving Mice", 2019 Cell.
2. SfN 2019, poster 336.12/BB65 "Optical measurement of dendritic membrane potential in the behaving mouse hippocampus".

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Poster

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Title: The functional architecture of glutamatergic release onto CA1 neurons

Authors: *J. R. CLIMER[1], M. D. ADOFF[2], H. DAVOUDI[1], J. S. MARVIN[3], L. L. LOOGER[3], D. A. DOMBECK[1];
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Abstract: Area CA1 of the hippocampus is necessary for spatial memory and hosts place cells, pyramidal neurons that fire as mice visit distinct locations in an environment. They are surrounded by silent and non-place cells, which may be place cells when the mouse is in a different context. Place cells receive inputs from spatial and non-spatial neurons in other regions, notably cells in hippocampal area CA3. In contrast to other regions containing neurons with well-defined receptive fields, there is no anatomical organization to the arrangement of the tuning of place cells. Thus, many models of place firing rely on random presynaptic inputs being amplified post-synaptically to generate selective firing. Advances in functional indicators and in vivo subcellular two-photon imaging provide means to examine the presynaptic inputs. Using resonant scanning two photon microscopy and the florescent glutamate sensor SF-iGluSnFR.A184S, we mapped glutamate release onto the dendrites of CA1 pyramidal neurons with micron scale resolution as mice navigated a linear track in virtual reality. By following these dendritic recordings with calcium imaging from the parent somas (using the red-shifted calcium indicator jRGECO1a), we determined how excitatory inputs contribute to the type and tuning of somatic output. Place cells receive more spatially tuned inputs than their non-place peers, and this input arrives preferentially in the receptive field of the neuron. This tuned input is anatomically organized along the dendrites, arriving in functional clusters along the dendrite. To examine the moment-by-moment transformation of inputs to the place cell outputs, we then imaged the dendrites and soma of neurons simultaneously. Overall, our results implicate increases in total excitatory input and co-activation of anatomically clustered synaptic input in place firing. Since they largely inherit their fields from upstream synaptic partners with similar fields, many CA1 place cells appear to be part of multi-brain-region cell assemblies forming representations of specific locations.

Disclosures: J.R. Climer: None. **M.D. Adoff:** None. **H. Davoudi:** None. **J.S. Marvin:** None. **L.L. Looger:** None. **D.A. Dombeck:** None.

Poster

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Department of Biotechnology through the DBT-IISc partnership program
Revati and Satya Nadham Atluri Chair Professorship
Kishore Vaigyanik Protsahan Yojana

Title: Spatial information transfer in hippocampal place-cell models depends on biophysical heterogeneities, trial-to-trial variability, and symmetry of place-field firing

Authors: *A. ROY, R. NARAYANAN;
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Abstract: The relationship between the feature-tuning curve and information transfer profile of individual neurons provides vital insights about neural encoding. However, the relationship between the spatial tuning curve and spatial information transfer of hippocampal place cells remains unexplored. Here, employing a stochastic search procedure spanning 12000 morphologically realistic models, we arrived at 127 conductance-based place-cell models that matched 22 intrinsic somato-dendritic measurements with their electrophysiological counterparts, and were endowed with sharp place-field tuning profiles (high peak firing rate and low spatial width). The parametric values exhibited neither clustering nor strong pairwise correlations. We introduced trial-to-trial variability (activity-dependent and activity-independent) in responses and computed model tuning curves and information transfer profiles, using stimulus-specific (SSI) and mutual (MI) information metrics, across locations within the place field. We found spatial information transfer to be heterogeneous across models, but to reduce consistently with increasing degrees of variability. Importantly, whereas reliable low-variability responses implied that maximal information transfer occurred at high-slope regions of the tuning curve, increase in variability resulted in maximal transfer occurring at the peak-firing location in a subset of models. Moreover, experience-dependent asymmetry in place-field firing introduced asymmetries in the information transfer computed through MI, but not SSI, and the impact of activity-dependent variability on information transfer was minimal compared to activity-independent variability. Biophysically, we unveiled a many-to-one relationship between different ion channels and information transfer. We also demonstrated critical roles for *N*-methyl-d-aspartate receptors, transient potassium and dendritic sodium channels in regulating information transfer via virtual knockout models (VKMs). Our results emphasize the need to account for trial-to-trial variability, tuning-curve shape and biological heterogeneities while assessing

information transfer, and demonstrate ion-channel degeneracy in the regulation of spatial information transfer.

Disclosures: A. Roy: None. **R. Narayanan:** None.

Poster

P872. Animal Navigation and Spatial Cognition

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Topic: H.09. Spatial Navigation

Support: University of Miami Academic Enhancement Research Fellowship
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Title: Motor-sensory integration punctuated by attentional-shifts as a key strategy to map navigational boundaries by zebrafish placed in a novel environment

Authors: *B. SANGHERA[1,2], M. GADDAM[2], J. S. KANWAL[2];
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Abstract: Movement is essential for gaining new information about one's environment, particularly in the relative absence of external sensory inputs. By integrating bouts of movement with a stream of sensory inputs, animals can begin to construct a spatial frame-of-reference within which to navigate. Here we tested whether time-segments of movement culminate in acquisition of allocentric cues to facilitate motor-sensory integration. Individual zebrafish (n = 9) were placed in a small (17 x 17 cm) tank with a smooth, curved and opaque surface. Video recordings were initiated immediately after fish placement and continued for 10 min. Machine-learning software packages for pose- and behavior-tracking (DeepLabCut, B-SOiD and VAME) elucidated up to 20 motifs that included forward glide, reorientation, and turns. Distance of animals from the center of the tank during exploratory swimming revealed that zebrafish spend more time along the edges of the tank during the first vs. the last minute. For mapping navigational boundaries, zebrafish employed a strategy of reorientations and forward glides terminating in two or more successive contacts with the tank wall followed by a sharp turn and free-swim back to the initial location. As the fish became familiar with its environment, bout frequencies of reorientations and their transitions to forward glides and sharp turns decreased by 26.6% and 20.8% (n = 7), respectively. The frequency of forward glides increased by 9.1%. Markov chain analysis revealed systematic transitions between these six swim patterns. Forward glides frequently terminated in contact with the tank wall followed by either reorientation or forward glide (transition probabilities of 22%, 22% and 58%, respectively). Reticular and reticulospinal neurons that control swim and turn movements in fish have complex touch-

sensitive receptive fields (Kanwal & Finger, 1997) and neurons in the lateral pallium encode the edges of the environment, head direction, swimming speed, and the swim velocity-vector (Vinepinsky et al., 2020). Moreover, fish can encode order for a series of spatially linked sensory inputs (de Perera, 2004). Together, these reports and our observations on zebrafish swim strategy suggest that fish are capable of employing a "go-and-touch" strategy to map navigational boundaries within a novel environment. In our working model, motor-sensory integration is coupled with bouts of sustained attention to create a neural representation of space. We hypothesize that attentional shifts (reorientations) triggered by the pallium and habenula may be critical for timely activation of brainstem mechanisms for action selection and specification.

Disclosures: B. Sanghera: None. **M. Gaddam:** None. **J.S. Kanwal:** None.

Poster

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Topic: H.09. Spatial Navigation

Support: Università di Padova - Fondi DOR

Title: Fruit flies use a spatial heuristic to navigate the environment

Authors: N. MEDA[1], G. M. MENTI[1], *A. MEGIGHIAN[1], M. A. ZORDAN[2];
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Abstract: Animals rely on multiple sensory information systems to make decisions. However, strategies that shorten decision-making time by taking into account more essential elements of information are preferred to strategies involving complex analyses. These "shortcuts to decision" are also termed "heuristics". It remains unclear whether animals can apply the exact same heuristics that humans employ. The identification of heuristic principles in species phylogenetically distant to humans would shed light on the evolutionary origin of speed-accuracy trade-offs and offer the possibility to investigate the brain representations of such trade-offs. During experiments on spatial learning, we observed that the search strategies of the invertebrate *Drosophila melanogaster*, the common fruit fly, resembled a spatial heuristic. Single fruit flies (adult males, aged 6-10 days, n = 40) were trained to find relief from a negative stimulation (optogenetic bitter taste) in a square virtual area close to a visual marker. Every time the visual marker changed position, the animal faced a cue conflict regarding where the following negative stimulation could be stopped: either in the spatial location where it was last experienced or in proximity to the new visual marker. Here we show that fruit flies learn the visual marker – relief association. However, their first, fast and frugal strategy to find relief relies

on the “Nearest Neighbour Rule” heuristic. That is, the fly visits the salient location closest to its current position to hopefully stop the negative stimulation. Only if this strategy proves unsuccessful, the animal then uses other learned associations to avoid bitter taste. The acknowledgment of a heuristic in *D. melanogaster* supports the view that invertebrates can leverage ‘economic’ principles when making choices, and that the existence of heuristics in evolution dates to at least 600 million years ago, the estimated time of metazoan divergence.

Disclosures: N. Meda: None. **G.M. Menti:** None. **A. Megighian:** None. **M.A. Zordan:** None.

Poster

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Topic: H.09. Spatial Navigation

Support: NSF Grant 17-537
NIH 5 T32 GM007276

Title: Spatial cognition in the cane toad (Rhinella marina)

Authors: *D. SHAYKEVICH, L. O'CONNELL;
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Abstract: The field of spatial cognition continues to grow increasingly robust with the characterization of spatial coding in the brains of species across different classes of animals. Since the initial description of place cells in the rodent hippocampus in the 1970’s, scientists have been able to study neurons with similar functions in other mammals, birds, and fish. However, how space is encoded in the amphibian brain is unknown, leaving an incomplete understanding of how the neural structures supporting spatial coding evolve. We are using the cane toad (Rhinella marina), a common species in South America and a successful invader in many other parts of the world, to identify the brain regions and neural signatures of spatial cognition. We conducted field-based translocation studies of cane toads within their native range in French Guiana and found that toads can navigate back to their home range after being moved 500 m from their home range, suggesting advanced navigational abilities. As our field studies with wild toads do not allow us to disentangle motivation and experience in contributing to navigational abilities, we are conducting spatial behavior assays in the laboratory and have found that half of the toads tested can learn to find a goal location based on allocentric visual cues within a four-arm maze. We are using immunohistochemical detection of pS6 as a proxy for neural activity to test the hypothesis that the medial pallium, a putative homolog of the mammalian hippocampus, has more active neurons in toads that have learned the correct exit from the maze compared to toads that did not meet learning criteria. Finally, the anuran brain has

large movements within the skull, and as their movement involves hopping and jumping, in vivo recordings of neural activity in freely moving frogs is challenging. Here, we report on the use of mesh electronics to record neural activity from the amphibian brain in a way that allows for large brain movements within the skull. We are currently testing if there are cells that fire with respect to spatial or environmental features. This technology that allows neural recordings from freely moving amphibianas will also open the doors for many other research programs studying amphibian behavior.

Disclosures: D. Shaykevich: None. **L. O'Connell:** None.

Poster

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Topic: H.09. Spatial Navigation

Support: NIH Grant R15DE027844-01

Title: Spatial learning and memory in the naked mole-rat (Heterocephalus glaber)

Authors: *N. HITE[1], D. K. SARKO[2];
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Abstract: The Hebb-Williams maze is a behavioral paradigm that is commonly used to test the learning and spatial working memory capabilities of animal models. Mouse and laboratory rat species have proven to be particularly adept at navigating such a task, particularly when visual cues and effectively motivating rewards are presented. The naked mole-rat, a less common but increasingly utilized animal model, is a subterranean rodent that presumably has well-developed spatial learning and memory skills in order to thrive within its natural habitat of extensive underground burrows and tunnel systems. However, the spatial learning and memory capabilities of this species remain to be tested. In addition, for a species with sensory adaptations that greatly diverge from those of rats and mice, it is necessary to develop a maze design and set of motivators that are more customized to naked mole-rats in order to facilitate their successful completion of the maze task and to more accurately elicit a true representation of their spatial learning and memory capabilities. Using animals from our laboratory colonies, housed in a vivarium setting that mimics the tunnel systems that naked mole-rats would experience in the wild, we developed a behavioral paradigm aimed to effectively assess the spatial working memory of naked mole-rats, including analysis of learning curves associated with maze performance and testing of memory retention in subsequent weeks. Using a modified Hebb-Williams maze, adult naked mole-rats underwent three days of training, consisting of five trials per day wherein they were allowed to freely explore the maze in search of the reward chamber.

Memory retention was then tested one day, one week, and one month following the last day of training. Performance was analyzed based on latency to the reward chamber and the number of errors made prior to reaching the reward chamber. Overall, the goal of this study was to establish a behavioral paradigm for maze navigation in naked mole-rats, including optimization of rewards and environmental motivators in order to best assess spatial learning and memory in this species. Future studies are aimed at investigating changes in learning and memory that result from sensory plasticity.

Disclosures: N. Hite: None. **D.K. Sarko:** None.

Poster

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OIST Graduate School

Title: Environmental and procedural aspects that influence navigational strategy usage in a dual solution plus maze

Authors: *L. ANDREOLI[1], J. R. WICKENS[3], K. Z. TANAKA[2];
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Abstract: Navigation is a fundamental behaviour for all animals, whether it is to find food, shelter, or a mate. It is thought to occur through three different processes: piloting, place strategy, and response strategy (Chersi, 2014 arXiv preprint arXiv). When animals follow a salient sensory cue which leads to a goal location, they are piloting. When the animal follows a flexible representation or a cognitive map of the space in which it is navigating, uses place strategy. Alternatively, animals use response strategy when they base their navigation in a sequence of actions to go to a specific place. It has been shown that both humans (Iaria et al, 2003 J Neurosci) and rodents (Packard et al, 1996 Neurobiol Learn Mem.) rely initially on place strategy and, after navigating several times in the same environment, start utilizing a more automatized response strategy. The dual-solution plus maze have been used to study the change in navigational strategy from place to response learning. In a plus-shaped maze, animals train to go from a South arm to West arm to get a reward. They can be probed after training from the opposite arm (North). Animals that undergo short training tend to use place strategy, and will

turn to the previously baited arm (West), making a different body turn than they did in training. After prolonged training, they tend to use response strategy, making the same body turn, and going to the previously non-baited arm (East). This project aims to elucidate environmental and training aspects that influence usage of place or response strategy. For that we trained animals in a protocol based on Wang et al. (2011 Neuron) in which they showed that majority of wild type mice displays place strategy with short training and response strategy for long training. We trained 11 mice (7 C57/BL6 and 4 pTRE-H2B-GFP) during short (5 sessions, 10 trials per session) and long training (10 sessions, 10 trials per session), to elicit place and response strategy, respectively. Although the animals learned the task (i.e., maintained performance above 70% for two consecutive days), all animals behaved as response learners during short training phase. We are currently testing which aspects (animal strain, number of trials per session, number of sessions, habituation process) can influence animals to behave as response learners after short training. With this work we hope to contribute to the understanding of how different training protocols and behavioural setups can influence usage of navigational strategy.

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Poster

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Title: TrkB antagonist ANA-12 decreases rats' use of place strategies in problem solving

Authors: *L. M. BUYNACK[1], J. A. WILLNER[2];
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Abstract: Brain-derived neurotrophic factor (BDNF) and its receptor, tropomyosin receptor kinase B (TrkB), are important not just for initial development of the nervous system, but also for synaptic plasticity in the adult nervous system. BDNF and TrkB play an important role in the cellular changes that mediate long-term potentiation (LTP) in hippocampus. Changes in BDNF and TrkB levels and expression in hippocampus are correlated with acquisition of place learning tasks that depend on this system. The present study examined the effects of ANA-12, a noncompetitive antagonist for TrkB, on rats' use of "place" and "response" strategies in an appetitive T-maze task that could be solved using either strategy. Male Long-Evans rats (75-120 days of age, N = 57) were initially handled, adapted to food restriction, and assigned to groups that received intraperitoneal injections of either 0.5 or 1.0 mg/kg ANA-12 in isotonic saline, or

saline alone 4 hours before each maze session. Rats were first adapted to the T-maze and allowed to explore and eat cereal reinforcers on the maze for two consecutive days (two 10 min sessions/day). The next day, the rats began reinforced training in which they were placed on the start arm of the maze and rewarded with a cereal reinforcer for choosing the goal arm opposite their initial side preference. The rats were trained to a criterion of 7 correct choices in 8 consecutive trials, with daily sessions of 20 training trials (1 min ITI). Training continued until a rat had reached criterion or received 4 days of training (80 trials), at which time it was dropped from the study. One minute after reaching criterion, the rat received a probe trial in which the maze was rotated 180° to pit place and response strategies against one another. The rat was placed on the maze and allowed 3 min to choose a goal arm; rats that went to the location where food was previously found were classified as using a place strategy, whereas those that made the same body turn as during training were classified as using a response strategy. Result show that administration of ANA-12 did not impair rats' ability to learn or perform the T-maze task; analysis of choice latencies and trials to criterion failed to reveal any significant between-groups differences during acquisition of the task. Significant differences were found on the probe trial, however, with ANA-12 decreasing rats' use of place strategies in a dose-dependent manner. These results support the idea that BDNF and TrkB are critical for place learning and memory.

Disclosures: L.M. Buynack: None. **J.A. Willner:** None.

Poster

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Title: Novel measures of Morris water maze performance that use vector field maps to assess accuracy, uncertainty, and intention of navigational searches

Authors: *M. PRABOD KUMAR[1], D. MEHROTRA[1], N. N[1], D. ALMEIDA-FILHO[2], Y.-S. LEE[4], A. J. SILVA[3], B. JAYAPRAKASH[5];
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Abstract: Most commonly used behavioural measures for testing learning and memory in the Morris water maze (MWM) involve comparisons of an animal's residence time in different quadrants of the pool. Such measures are limited in their ability to test different aspects of the animal's performance. Here, we describe novel measures of performance in the MWM that use vector fields to capture the motion of mice as well as their search pattern in the maze. Using these vector fields, we develop quantitative measures of performance that are intuitive and more sensitive than classical measures. First, we describe search patterns in terms of vector field properties and use these properties to define three metrics of spatial memory namely Spatial Accuracy, Uncertainty and, Intensity of Search. We demonstrate the usefulness of these measures using four different data sets including comparisons between different strains of mice, an analysis of two mouse models of Noonan syndrome (Ptpn11 D61G and Ptpn11 N308D/+), and a study of goal reversal training. Importantly, besides highlighting novel aspects of performance in this widely used spatial task, our measures were able to uncover previously undetected differences, including an animal model of Noonan syndrome, which we rescued with the mitogen activated protein kinase kinase (MEK) inhibitor SL327. Thus, our results show that our approach breaks down performance in the Morris water maze into sensitive measurable independent components that highlight differences in spatial learning and memory in the MWM that were undetected by conventional measures.

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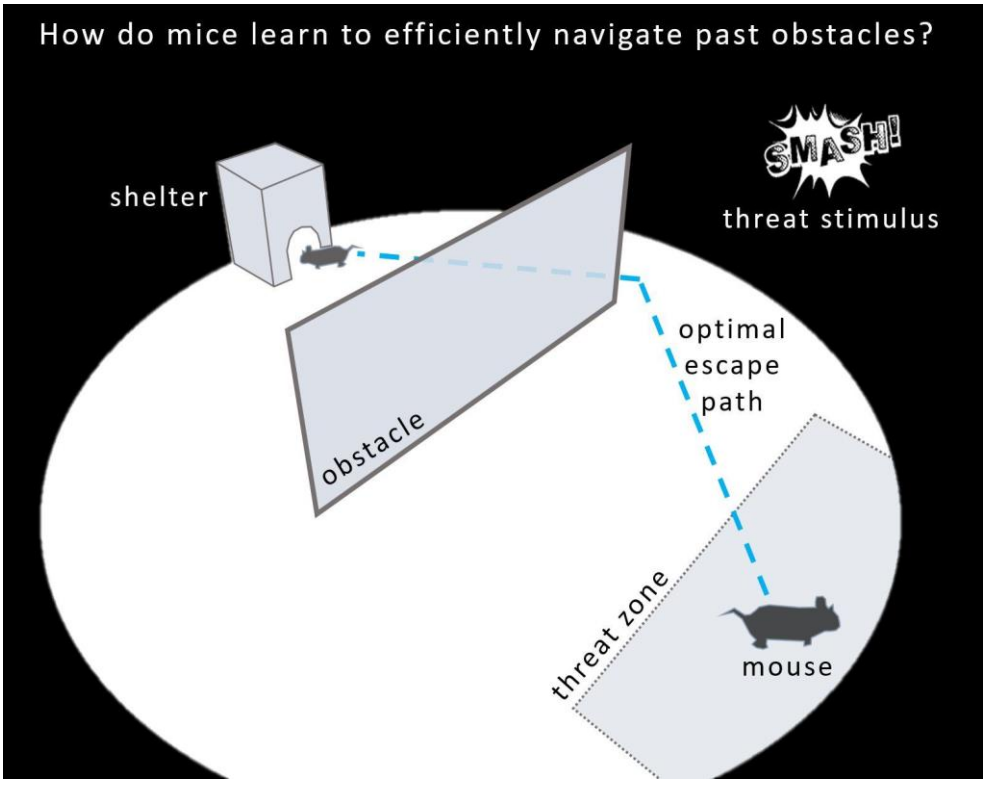
Program #/Poster #: P872.08

Topic: H.09. Spatial Navigation

Support: Wellcome Senior Research Fellowship 214352/Z/18/Z
Sainsbury Wellcome Centre Core Grant from the Gatsby Charitable Foundation and Wellcome 090843/F/09/Z
SWC PhD Programme

Title: Mice learn multi-step routes by memorizing subgoal locations

Authors: *P. SHAMASH, P. IORDANIDOU, T. BRANCO;
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Abstract:

The behavioral strategies that mammals use to learn multi-step routes are unknown. Results from maze-learning protocols and open-field environments have emphasized two possibilities: the cognitive map (model-based) and taxon (model-free) strategies. However, it is unknown how well this dichotomy explains the rapid learning profiles and non-random exploration patterns that characterize spatial learning in natural settings. Here we investigated how mice develop efficient escape routes to a shelter when the direct path is blocked by an obstacle. Capitalizing on the robustness and intrinsic motivation of threat-evoked escape, we examined spatial learning in four ways: analysis of exploration and escape routes; comparison of 14 different arena geometries; dynamic reconfiguration of the arena; and acute manipulation of neural activity during exploration. We found that mice at first escaped inefficiently, running toward the shelter and negotiating the obstacle using sensory cues. Within twenty minutes, they developed a subgoal strategy: escaping directly to the obstacle edge before heading to the shelter. Mice continued to escape in this manner even after the obstacle was removed, indicating use of spatial memory. However, neither the taxon nor cognitive map theory could explain how subgoals were memorized. Instead, mice used a hybrid approach. As with an internal map, they achieved all-or-none learning of allocentric locations; but similar to model-free behavior, subgoals inflexibly persisted even after shortcuts opened up, and learning occurred during ‘practice runs’ rather than investigation. To test the hypothesis that running to obstacle edges during exploration generates subgoals, we used a closed-loop optogenetic manipulation; interrupting these movements by stimulating premotor cortex successfully prevented subgoal learning. These results suggest that subgoal memorization - i.e., learning allocentric subgoal locations during instinctive practice runs - is itself a fundamental strategy by which rodents compute multi-step routes in new environments.

Disclosures: P. Shamash: None. **P. Iordanidou:** None. **T. Branco:** None.

Poster

P872. Animal Navigation and Spatial Cognition

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Program #/Poster #: P872.09

Topic: H.09. Spatial Navigation

Support: GAT3361
214352/Z/18/Z
090843/F/09/Z

Title: Innate heuristics and fast learning support flexible action selection during escape

Authors: *F. CLAUDI[1], D. CAMPAGNER[2], T. BRANCO[3];
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Abstract: When faced with imminent danger, animals must rapidly take defensive actions to reach safety. In species such as mice, knowledge about the spatial environment is used to select appropriate defensive actions, as well as to plan and execute efficient flights. Natural environments often present multiple possible routes to refuges and to maximize survival the choice between them must be rapid and accurate (i.e., selecting the option with highest adaptive value). Relying on extensive trial-and-error learning would be maladaptive because errors could be deadly. Escape path selection thus poses a challenging learning and decision-making problem, and how animals solve this problem is not well understood.
In this work we have investigated how mice choose escape routes to a shelter when there are multiple possible paths of varying length and geometry. Using a series of new behavioural assays together with high-resolution quantification of behaviour and theoretical modelling, we find that mice integrate both direct and path distance to shelter to compute route choice. Furthermore, mice display this choice strategy upon the first encounter with threat after only a few minutes of exploration in a novel environment. This shows that the action selection process leverages rapidly acquired spatial knowledge and uses innate heuristics to assign value to different options. The combination of learned and innate knowledge leads mice to preferentially select escape paths that minimize threat exposure with minimal learning requirements, providing an evolutionary advantage. Comparing the behaviour of mice with the performance of different classes of reinforcement learning agents shows that only model-based agents endowed with innate heuristics can accurately replicate the animals' choice behaviour. This suggests that sophisticated learning and decision-making strategies underly innate escape path selection behaviour in mice.
Our results provide an entry point for understanding the mechanisms behind the unequalled

learning abilities of the mammalian nervous system, and how the brain integrates acquired and innate knowledge to generate decisions. They may also inspire the development of more efficient learning algorithms for artificial agents.

Disclosures: F. Claudi: None. **D. Campagner:** None. **T. Branco:** None.

Poster

P872. Animal Navigation and Spatial Cognition

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Program #/Poster #: P872.10

Topic: H.09. Spatial Navigation

Support: LTAUSA19135

Title: Hippocampal signatures of avoidance of a moving object as revealed by immediate-early gene imaging

Authors: *J. SVOBODA, B. KRAJCOVIC, D. CERNOTOVA, N. AHUJA, S. KUBIK, A. STUCHLIK;
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Abstract: Animals can efficiently organize their spatial behavior with respect to other moving animals or objects but the underlying neural substrate is poorly understood. To model spatial interactions between an animal and moving object, we developed an aversive task in which a rat placed on a circular arena must avoid a 25cm distance from a small programmable robot to keep itself from a mild footshock. Our previous work showed that dorsal hippocampus plays a vital role in this process. Bilateral muscimol inactivations of the dorsal hippocampus impaired avoidance of a moving but not stable robot. In addition, unit recordings from CA3 found neurons responding to the mutual position of a rat and the robot. In the present experiment, we aimed at investigating the neural substrate in more detail using immediate-early gene imaging. Male Long Evans rats either underwent training to avoid a stable (S) or moving (M) robot in four 20-min daily sessions. On the last day, they either continued in this condition (SS group, MM group), or the condition changed (SM group, MS group). Control group remained in the cage on the last day (CG). Fifty minutes after the last session, rats were sacrificed and perfused. Brains were either stained for the presence of c-fos protein or the expression of other immediate-early genes, Homer1a and Arc, was determined using cellular compartment analysis of temporal activity by fluorescence in situ hybridization (catFISH). In all but the control group, we found increased c-fos activity in the dentate gyrus, followed by a less active CA1. Interestingly in CA3 and CA1, groups initially trained to avoid a mobile robot (MM, MS) displayed higher activity than groups trained with the stable robot (SS, SM), irrespective of the condition in the last day. These data

suggest that CA3 and CA1 plasticity may be related to experienced knowledge of the mobility of the robot. Detailed analysis of catFISH data is in progress.

Disclosures: J. Svoboda: None. **B. Krajcovic:** None. **D. Cernotova:** None. **N. Ahuja:** None. **S. Kubik:** None. **A. Stuchlik:** None.

Poster

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Program #/Poster #: P872.11

Topic: H.09. Spatial Navigation

Support: HHMI
NIH Grant NS107613
JSPS

Title: Orienting eye movements during REM sleep

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Abstract: REM sleep is accompanied by vivid visual dreams and characterized by rapid eye movements, from which this phase of sleep gets its acronym. It has been proposed that rapid eye movements during REM sleep represent orienting of the gaze in the virtual environment of dreams. While some behavioral studies in humans and animals support this hypothesis, other studies suggest that eye movements during REM sleep are simply the consequence of the random activation of the pontine nuclei. In order to address this controversy, we recorded from head direction (HD) cells in the anterodorsal thalamus (AD), a population of neurons whose activity reports the head direction of the animal. Previous work has shown that during REM sleep, the population activity of HD cells is similar to that during actual navigation, in that it maintains a coherent representation of HD, i.e. a “virtual HD”. Because fast eye movements are coupled to head movements during gaze shifts in awake freely moving animals, we hypothesized that during REM sleep rapid eye movements may predict changes in the virtual HD. To this end, we monitored eye movements during REM sleep while recording ensembles of HD cells from the AD. We trained a Bayesian decoder with the activity of HD cells to decode HD of mice navigating in the open field, and then applied the decoder to the spontaneous activity of HD cells during REM sleep to decode virtual HD. We focused our analysis on rapid eye movements occurring in isolation, i.e. separated by at least 400 ms intervals from preceding eye movements. We discovered that, during REM sleep, the direction of eye movements predicted the direction of the changes in virtual HD. Furthermore, the amplitude of rapid eye movements correlated with amplitude of changes in virtual HD. Importantly, such correlation between rapid eye movements

and virtual HD was similar to the relationship between fast saccade-like eye movements and HD representation in the AD of awake mice. This study provides the first physiological evidence that rapid eye movements represent gaze shifts in the virtual world during REM sleep.

Disclosures: Y. Senzai: None. **M. Scanziani:** None.

Poster

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Program #/Poster #: P872.12

Topic: H.09. Spatial Navigation

Support: NIH Grant NS053907

Title: The neural correlates of spatial disorientation in head direction cells

Authors: *R. M. GRIEVES, M. SHINDER, L. ROSOW, M. S. STEVEN, J. S. TAUBE; Dept. of Psychological and Brain Sci., Dartmouth Col., Hanover, NH

Abstract: Spatial disorientation is an experience that one does not easily forget. While the brain has likely evolved robust mechanisms to combat this affect, the neural underpinnings of these mechanisms are yet to be uncovered. To explore this, we monitored the activity of head-direction (HD) cells in the anterodorsal nucleus (ADN) of the thalamus under different conditions associated with disorientation: 1) while rats underwent uni- or bi-directional rotations in the light, or 2) while blindfolded, and 3) during passive restraint or while freely moving. We sought to determine if periods of presumed disorientation led to HD cell quiescence, random firing, or bursts of activity dissociated from HD. HD cells did not become quiescent in darkness or when animals were restrained suggesting that active locomotion and visual inputs are not necessary to drive HD cell activity. HD cells also continued to fire during rotations but average firing rates and burst frequency were reduced. Bursts during rotations were not associated with a particular HD, however, coherence remained high between co-recorded cells suggesting that the hypothesized attractor network remained intact but was decoupled from external stimuli. Peak firing rates and directionality (Rayleigh vector length) also decreased during rotation sessions, especially when rotations were fast, conducted in darkness, or while rats were passively restrained. Initially, HD cells maintained their directionality during rotations in darkness, however, with each revolution, tuning curves shifted in the direction of rotation, suggesting that the HD system was underestimating angular rotation. This result is consistent with the insensitivity of the vestibular system to accurately track constant unidirectional rotation. Lastly, after passive rotations in the dark, HD cells continued to fire in bursts and these bursts matched the frequency of rotation. This phenomenon is similar to post-rotational "nystagmus": the repetitive, oscillating movement of the eyes which occurs after sustained rotation and during

spatial disorientation. During rotations disorientation effects were reduced when visual cues were available, suggesting that while the generation of the HD signal is undoubtedly vestibular in origin, visual inputs provide a functional role in directional tuning. Thus, one of the ways the brain overcomes spatial disorientation is through multisensory integration of different inputs. These findings allow us to better understand not only the HD signal but also the systems of spatial orientation and navigation that subserve and depend upon it.

Disclosures: R.M. Grieves: None. **M. Shinder:** None. **L. Rosow:** None. **M.S. Steven:** None. **J.S. Taube:** None.

Poster

P872. Animal Navigation and Spatial Cognition

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Program #/Poster #: P872.13

Topic: H.09. Spatial Navigation

Support: St. Olaf CURI

Title: Anesthesia disrupts Head Direction Cell activity in freely-moving rats.

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Abstract: Head direction (HD) cells in the rat provide navigational information by increasing their firing rates when the rat's head points a specific direction in the horizontal plane. Some evidence exists to show that during sleep, HD cells stop firing with such directional specificity, but maintain their firing relationships to each other consistent with an attractor network hypothesis. In this study, we investigated the effects of Isoflurane and Nembutal anesthesia on HD cell activity to see if these properties would be consistent by recording from HD cells while rats were 1) unconscious on a rotating platform, and as they woke up and recovered from anesthesia (Isoflurane), and 2) as they went from an awake to an unconscious state under anesthesia (Nembutal). Results showed that under both forms of anesthesia, HD cells cease to fire directionally and began firing tonically in all directions as the rat was rotated. Additionally, HD cells' average and peak firing rates significantly decreased while under anesthesia. In the moments before the rats regained consciousness, HD cells showed a dramatic increase in firing rate. Most HD cells quickly recovered their original directional tuning following the rats' initial movement post-anesthesia in the presence of a polarizing cue card, but adopted a random firing direction in the absence of the cue. Cells were, however, less directional, on average, while recovering from Isoflurane than during the preceding baseline trial. We suggest this inconsistency could result from continued effects of isoflurane or short recovery sampling times. These results show, for the first time, how HD cells behave under anesthesia, and invite further

comparison with HD cell activity across other unconscious states. In addition, these results may be interpreted as supporting intrinsic attractor network dynamics in the HD cell network.

Disclosures: G. Muir: None. **I. Aarrestad:** None. **H. Cabasco:** None. **N. Weitermann:** None.

Poster

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Topic: H.09. Spatial Navigation

Support: NIH Grant R01
Klingenstein Simons Fellowship Awards in Neuroscience
MnDRIVE graduate fellowship in neuromodulation

Title: Prospective goal representation in monkey OFC during 3D virtual reality foraging

Authors: *M. WANG[1], B. Y. HAYDEN[2];
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Abstract: During natural foraging, animals exhibit goal-directed behavior to learn the structure of the environment and obtain reward. Prospective goal state representation and navigational planning are essential to this process. Although neural activities for route planning have been well-studied in hippocampus, the neural underpinnings for goal state encoding, which potentially informs route-planning and guide subsequent behavior, is still unknown. We hypothesized that goal simulation, i.e. reactivation of goal-related state in neural populations, is the key to use prospective goal representation to plan for subsequent action. We developed a 3D virtual reality foraging task to test this hypothesis. In this task, a jackpot reward is hidden at a random location in the virtual maze (goal location) for each session, while other maze configurations stay the same across all sessions. On each trial, the subject is teleported to another random location (start location) during the inter-trial interval. The subject then navigates through the virtual maze with a joystick to forage for the hidden jackpot reward. Subjects took significantly longer search time (from start to goal location) and less efficient travelled paths during pre-learning than during post-learning trials. We recorded large ensembles of OFC neurons (~55 units simultaneously/session) and found that: (1) OFC neurons showed mixed selectivity and encoded navigation and goal-related information, such as place, egocentric movement, allocentric moving direction, and relative relation between self and the goal. (2) Goal state representation was simulated right before the start of navigation and during navigation. (3) Goal-state simulation predicted subsequent goal-approaching behavior. (4) OFC goal state representation predicted learning. Together, we found that OFC showed neural signature of using goal simulation to guide behavior.

Disclosures: M. Wang: None. **B.Y. Hayden:** None.

Poster

P909. Biochemical and Molecular Techniques II

Location: Virtual Only

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Program #/Poster #: P909.01

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NRF-2017M3C7A1043841, National Research Foundation of Korea (NRF)

Title: DecoM: Molecule-specific nanoscale resolution imaging of cultured cells with scanning electron microscopy

Authors: *D.-H. SONG[1], C. SONG[1], J. CHUNG[3], E.-H. JANG[4,5], Y. HUR[2], E.-M. HUR[4,5], D. KIM[3], J.-B. CHANG[1];
[1]Dept. of materials science and engineering, [2]Biomed. research center (BMRC), Korea Advanced Inst. of Sci. and Technol., Daejeon, Korea, Republic of; [3]Dept. of chemistry, Hanyang Univ., Seoul, Korea, Republic of; [4]Dept. of neuroscience, college of veterinary medicine, [5]BK21 four future veterinary medicine leading education & research center, Seoul Natl. Univ., Seoul, Korea, Republic of

Abstract: Nanoscale imaging of biomolecules broadly facilitates biological findings. Various nanoscale imaging techniques for intracellular biomolecules in cells have been used and developed including super-resolution fluorescence microscopy (e.g. STED, STORM, and PALM), and electron microscopy accompanied by ultrathin sectioning or focused-ion beam (FIB) milling. However, current nanoscale imaging methods are still not widely used in typical biological laboratories, as high instrumental requirements, skilled personnel, or complicated sample preparation are needed. Herein, we demonstrate a new molecule-specific nanoscale imaging method based on scanning electron microscopy (SEM), termed DecoM (Development of silver on gold nanoparticles for correlative fluorescence and scanning electron Microscopy). In DecoM, secondary antibodies bearing both fluorophores and gold nanoparticles (AuNPs) are used to label target proteins, and in-situ silver development proceeds, followed by simple drying under ambient conditions. This simple sample procedure enables the imaging of labeled molecules with SEM at a resolution of 68 nm without any additional slicing technique, such as ultrathin sectioning using ultramicrotome or FIB milling. Multiplexed nanoscale imaging can be demonstrated by labeling two proteins with antibodies bearing different fluorophores and overlaying fluorescence microscopy and SEM images. Finally, we show DecoM to visualize the nanoscale changes in microtubules in cultured cells overexpressing microtubule-severing proteins, which are associated with multiple diseases. DecoM makes researchers who perform antibody staining routinely obtain molecule-specific nanoscale imaging in their study immediately.

Disclosures: C. Song: None. **J. Chung:** None. **E. Jang:** None. **Y. Hur:** None. **E. Hur:** None. **D. Kim:** None. **J. Chang:** None.

Poster

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Program #/Poster #: P909.02

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NRF-2017M3C7A1043841, National Research Foundation of Korea (NRF)
SRFC-IT1702-09, Samsung Research Funding & Incubation Center for Future Technology (SRFC)

Title: Picasso: ultra-multiplexed imaging of the brain through blind unmixing of spectrally overlapping fluorophores

Authors: *J. SEO[1], Y. SIM[3], J. KIM[2], H. KIM[1], I. CHO[1], Y.-G. YOON[2], J.-B. CHANG[1]; [1]Dept. of materials science and engineering, [2]Sch. of electrical engineering, Korea Advanced Inst. of Sci. and Technol., DAEJEON, Korea, Republic of; [3]Dept. of biomedical engineering, Sungkyunkwan Univ., Suwon, Korea, Republic of

Abstract: Fluorescence microscopy has been used as a useful tool for studying the spatial information, expression pattern, and molecular architecture of biomolecules in highly heterogeneous systems. However, its multiplexing capability is limited to 4-5 molecules due to the spectral overlap of fluorophores. To accomplish a much higher level in multiplexing with fluorescence microscopy, diverse techniques where spectrally overlapping fluorophores could be unmixed to their signals into each one have been developed. The reference-based unmixing (e.g., linear unmixing and spectral phasor) enables to unmix signals of spectrally overlapping fluorophores using preknowledge of fluorophores' emission spectra. However, particularly in highly organized specimens, precise measurements are challenging because emission spectra are possibly fluctuated by the degree of photobleaching, subregions of the specimen, and light scattering altered by the depth of the fluorophore. Therefore, the large volumetric imaging of more than 10 biomolecules has not been reported with reference-based unmixing. The non-reference-based unmixing (e.g., non-negative matrix factorization (NMF) and clustering-based unsupervised machine learning) has provided the unmixing without the preknowledge. NMF does not present a unique solution, so establishing human interventions and proper constraints are critical for multiplexed imaging. Moreover, NMF typically requires that the number of images (N) is sufficiently larger than the number of fluorophores (F) for reliable unmixing. Otherwise, the clustering-based unsupervised machine learning can achieve the unmixing, even if the N is fewer than the F. However, since it allocates each pixel into one of the multiple clusters results in zero-value in other clusters, it shows a low unmixing performance when

multiple biomolecules are spatially overlapped. Here, we propose PICASSO which can achieve more than 15-color multiplexed imaging without reference measurements through mutual information minimization. Even though PICASSO is a non-reference-based unmixing technique, it only requires that *N* is equal to *F*. Also, it presents a high unmixing performance even when it unmixes spatially overlapping biomolecules. We show diverse demonstrations of multiplexing *via* PICASSO including 3D and large area imaging, mRNA imaging, and FFPE sample imaging. We expect that PICASSO could be a helpful tool for mapping the spatial information or interactions of biomolecules and discovering new neuronal cell subtypes in a nervous system.

Disclosures: J. Seo: None. **Y. Sim:** None. **J. Kim:** None. **H. Kim:** None. **I. Cho:** None. **Y. Yoon:** None. **J. Chang:** None.

Poster

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Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NRF-2017M3C7A1043841, National Research Foundation of Korea(NRF)

Title: FRACTAL: Multiplexed signal amplification of immunofluorescence via cyclic antibody staining

Authors: *H.-M. YEON[1], Y. CHO[1], J. SEO[1], Y. SIM[2], J. CHUNG[3], C. PARK[1], C. PARK[2], D. KIM[3], J.-B. CHANG[1];
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Abstract: Amplification of immunofluorescent (IF) signal became an important issue in imaging field owing to its capability of high-resolution imaging of low-abundance target proteins and reducing imaging time for large-volume IF imaging. Also, simultaneous imaging of multiple markers is crucial for identifying the overall spatiotemporal distribution and relations of the target proteins. In this study, we demonstrate fluorescent signal amplification via cyclic staining of target molecules (FRACTAL), a technique that does not require custom optimization or repetitive activation and inactivation process for multiplexed imaging. FRACTAL only requires commercialized primary antibodies and secondary antibodies for IF signal amplification. We amplify the IF signals of cultured cells, clinical specimens, and tissue slices by more than nine-fold using only fluorophore-conjugated antibodies. We also show that cyclically applied secondary antibodies form uniform layers around a target protein using stochastic optical reconstruction microscopy (STORM) imaging. Theoretically, FRACTAL can be applied to the

multiplexed imaging if the cross-reactivity between the orthogonal secondary antibodies are eliminated. Here, we also demonstrate that simultaneous multiplexed signal amplification using FRACTAL is possible through simple antibody affinity purification process to prevent cross-reaction between the orthogonal secondary antibodies, using NHS-activated agarose gel spin column. The fluorescent signals of Lamin A/C, myelin basic protein (MBP), and glial fibrillary acidic protein (GFAP) in the mouse brain samples were amplified through simultaneous cyclic staining of purified secondary antibodies from six different host species, rabbit-donkey, rat-mouse, and chicken-goat pairs, respectively. We also show that multiplexed FRACTAL is compatible with protein-retention expansion microscopy (pro-ExM) technique and represents higher post-expansion fluorescent signal than that of the samples with conventional indirect IF staining. These results provide an effective shortcut for multiplexed signal amplified imaging of in-vivo target proteins of interest.

Disclosures: H. Yeon: None. **Y. Cho:** None. **J. Seo:** None. **Y. Sim:** None. **J. Chung:** None. **C. Park:** None. **C. Park:** None. **D. Kim:** None. **J. Chang:** None.

Poster

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Program #/Poster #: P909.04

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: National Research Foundation of Korea (NRF), NRF-2017M3C7A1043841
Samsung Research Funding & Incubation Center for Future Technology, SRFC-IT1702-09

Title: Expansion microscopy imaging of whole larval zebrafish

Authors: *J. SIM[1], C. PARK[1], I. CHO[1], K. MIN[3], J.-S. LEE[4], Y. CHONG[5], J. KIM[2], J. KANG[6], K. D. PIATKEVICH[7], E. E. JUNG[8], S.-K. KWON[9], Y.-G. YOON[2], E. S. BOYDEN[10], J.-B. CHANG[1];
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Abstract: An unbiased investigation of the entire organism at sub-cellular resolution is needed to understand neural systems from a systematic perspective[1]. The microbiota-gut-brain axis, for instance, full spatial coverage over the whole organism is essential for investigating different bidirectional communication paths between the central and the enteric nervous system. In contrast, a high spatial resolution is needed for elucidating the underlying molecular mechanisms of the effects of the microbiota on our brains and behavior[2]. Various imaging methods, including tissue clearing[3-5], computed tomography[6], and electron microscopy[7], have been utilized to visualize the anatomical structures of biological systems, but their resolution or throughput is not high enough for nanoscale imaging over multiple scales[6]. Here we present a whole-body super-resolution microscopy imaging technique, which can visualize anatomically relevant structures at a lateral resolution of 60-nm over the whole vertebrate model animal *via* non-specific labelling of proteins and expansion microscopy. Expansion microscopy is a super-resolution imaging technique of cultured cells and tissue slices and can achieve nanometer resolution with conventional microscopy through the physical expansion of specimens. Wholebody-ExM is based on expansion microscopy and functional-group-directed fluorescent labeling, which non-specifically labels proteins inside specimens with fluorophores without any target-specific labeling techniques. Using wholebody-ExM, we successfully visualized the major anatomical structures of vertebrates over the intact zebrafish larva and mouse embryo without any high-affinity antibodies, ultra-thin sectioning, and specialized instruments. We also demonstrated that wholebody-ExM retains the fluorescence signals of fluorescent proteins, and its resolution is high enough to visualize various structures that can be imaged only with electron microscopy. With these advantages, wholebody-ExM would be highly useful for studying transgenic lines showing anatomical abnormalities or studying the nanoscale neuro-morphological changes occurring in different brain developmental stages.

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Poster

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Program #/Poster #: P909.05

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: R01DC011099
R01MH120016

Title: Expansion microscopy of a single mixed synaptic contact reveals additional components of electrical synapses

Authors: *S. CARDENAS-GARCIA, S. IJAZ, A. E. PEREDA;
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Abstract: Auditory afferents terminate as "Club endings" on the teleost Mauthner cells that contain specializations for electrical and chemical transmission. In 4-6 dpf larval zebrafish, these terminals are easily identifiable by immunolabeling with anti-connexin (Cx) antibodies as oval contact areas of about 2 microns in diameter. However, their size does not allow for visualization of the spatial distribution of various synaptic proteins within each terminal. For this purpose, we applied expansion microscopy, which allowed us to reveal the presence of, and the spatial relationship between, multiple synaptic components by immunolabeling. By using the proExM protocol, we are able to expand the diameter by up to 5x the original size of these terminals. Expansion with Cx and GLUR2/3 antibodies revealed that, while multiple gap junction (GJ) plaques are distributed throughout the entire contact area, a much smaller number of glutamatergic synapses are restricted to the periphery. Antibodies for the fish homologs of the mammalian Cx36, Cx35 and Cx 34, showed that they co-localize at each punctum, as GJs in club ending terminals are formed by heterotypic channels made by these proteins. Strikingly, expansion also increased the distance between pre- and postsynaptic sites, exposing the lack of co-localization between labeling for the presynaptic Cx35 and ZO1, the latter of which is a scaffolding protein restricted to the postsynaptic hemiplaque. Electron microscopy of these terminals (Kimmel et al., 1981) showed the presence of adherens junctions in the vicinity of GJs. Therefore, we explored their incidence and spatial distribution within the terminal using N-cadherin and Beta catenin antibodies, in combination with Cx labeling. N-cadherin labeling was distributed throughout the contact area and was mutually exclusive with Cx labeled puncta, suggesting a possible functional relationship between these two proteins and structures. Thus, our results indicate that an electrical synapse might operate with multiple GJs and include the contribution of additional structural components such as adherens junctions.

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Poster

P909. Biochemical and Molecular Techniques II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P909.06

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: 21-BR-02-03
21-BR-02
NRF-2015M3C7A1029037
NRF-2017M3C7A1048092

Title: Development of matrisome isolation method in oil-based CASPERized mouse brains

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Abstract: Development of matrisome isolation method in oil-based CASPERized mouse brains Byung Geun Ha[1,§], Yu-Jin Jang[1,§], Sung-Jin Jeong[1, 2,*1]Research Group of Developmental Disorders and Rare Diseases, Korea Brain Research Institute (KBRI), Daegu 41062, Republic of Korea[2]Department of Brain and Cognitive Sciences, Daegu Gyeongbuk Institute of Science and Technology (DGIST), Daegu 42988, Republic of Korea
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Matrisome is defined as the ensemble of ECM components and ECM-associated proteins. Alterations of the matrisome reflect the development of various diseases. However, it has not been well studied due to its insolubility and complexity. Brain tissue has a higher cell density and weaker mechanical strength than other tissues in the human body. When removing cells using a general decellularization method to produce scaffolds and obtain ECM proteins, various problems must be considered because tissues are easily damaged. In this study, we performed decellularization in combination with polymerization to retain the brain shape and ECM components. We immersed mouse brains in oil for polymerization and decellularization via CASPER (Clinically and Experimentally Applicable Acellular Tissue Scaffold Production for Tissue Engineering and Regenerative Medicine) and then isolated matrisome using sequential matrisome preparation reagents (SMPRs), namely, RIPA, PNGase F, and concanavalin A. Adult mouse brains were well preserved with our decellularization method. Western blot and LC-MS/MS analyses revealed that core matrisome proteins, including collagen and laminin, were isolated efficiently from decellularized adult mouse brains using SMPRs. Our method will be helpful to obtain matrisome data and perform functional studies using adult mouse brains and other tissues.

Disclosures: B. Ha: None. **Y. Jang:** None. **S. Jeong:** None.

Poster

P909. Biochemical and Molecular Techniques II

Location: Virtual Only

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Program #/Poster #: P909.07

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NSERC
CONACYT

Title: A neurochemical atlas of the brain

Authors: *J. ARCHIBALD[1,4], E. L. MACMILLAN[2,5], J. L. K. KRAMER[3,4,6];
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Abstract: Neurological conditions are a growing concern and evidence-based treatments represent a research priority. Central to these are advanced neurophysiological techniques, which are used to examine neurological conditions including neurodegenerative (e.g., Alzheimer's) and psychiatric conditions (e.g., schizophrenia). These approaches focus on function in terms of electrophysiology and blood flow. Yet measurement of neurochemicals via proton-magnetic resonance spectroscopy (^{1}H-MRS) provides a unique opportunity to evaluate neurochemicals, colloquially termed *The Virtual Biopsy.* Mapping neurochemicals in the brain is desirable, to gain insight into biochemical processes in health and disease. The cingulate cortex is involved in cognition functions (e.g., attention, processing, reward, processing of somatic sensations). Thus, it has become a popular technique to examine behaviour, function and clinical conditions. The distribution, typical ranges and variability of neurochemicals have not been examined making the decision of where to collect the data from challenging. To address this, we have performed a metabolic map of the cingulate cortex at 3T. Timing of the scan, food intake and menstrual cycle were standardized across participants. Preliminary results from five healthy participants (1f, age= 22.7 ± 5.5) show a coefficient of variation (CV) of 22% creatine, 7.4% glutamate, 10.3% NAA, across the cingulate gyrus between subjects. The CV for each metabolite was calculated within subjects across the different locations on the cingulate gyrus and ranged from 15.1-29.7% for creatine, 4.2-9.7% for glutamate, and 3.3-18% for NAA. These findings will be examined in a larger sample size, further analysis will examine sex differences and cortical thickness, as well as other neurochemicals. As MRS moves towards becoming a standardized technique, answering these questions will provide an important basis for understanding the human brain’s biochemical environment.

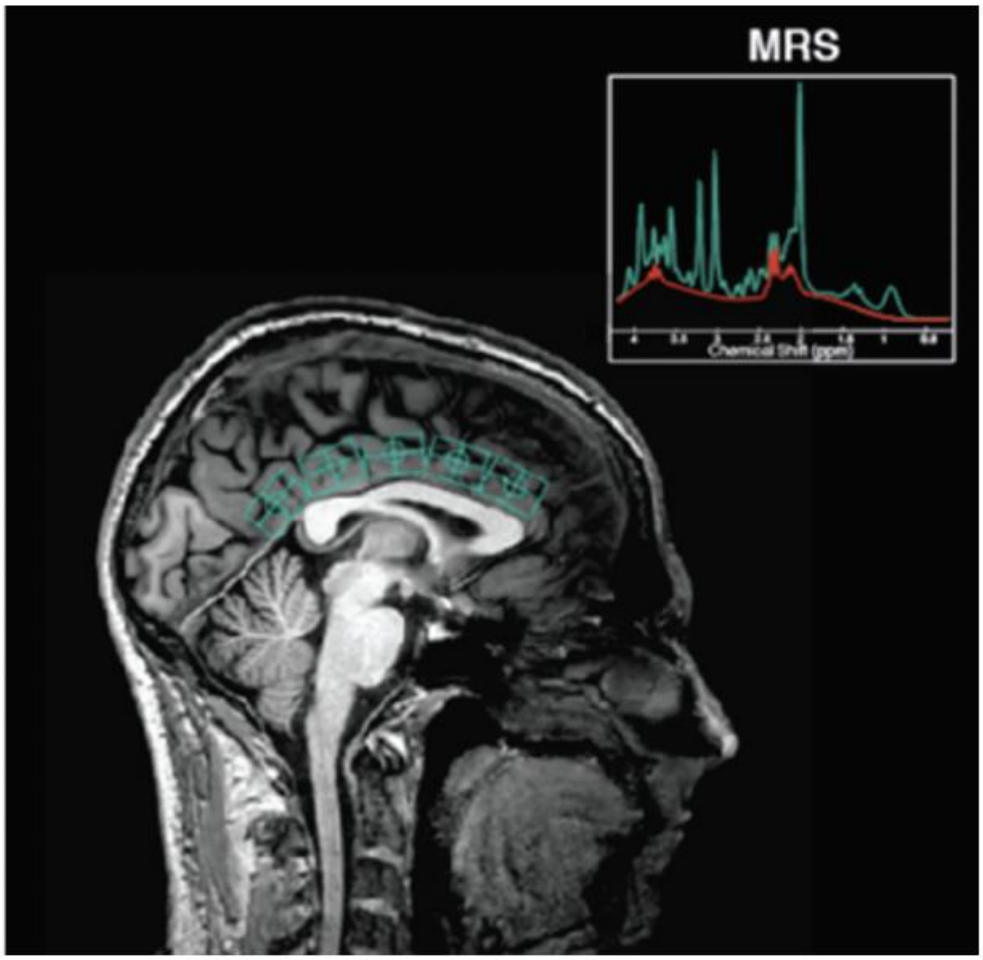


Figure- Cingulate gyrus neurochemical map

Disclosures: J. Archibald: None. **E.L. MacMillan:** A. Employment/Salary (full or part-time):; Philips Health Canada. **J.L.K. Kramer:** None.

Poster

P909. Biochemical and Molecular Techniques II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P909.08

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Title: Cerebrovascular Smooth Muscle Disease Causing ACTA2 Arginine 179 Pathogenic Variants disrupt Actin polymerization altering cell proliferation and migration.

Authors: *C. A. LUSK[1], C. L. LINO CARDENAS[2], S. K. MCCARTHY[1], C. G. BERGEN[1], M. E. LINDSAY[2], P. L. MUSOLINO[1];
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Abstract: The ACTA2 gene encodes for alpha smooth muscle actin isotope 2 (α-SMA) which is preferentially expressed in vascular and visceral smooth muscle cells (SMC). ACTA2 Arginine 179 (Arg179) pathogenic variants cause multi-systemic smooth muscle dysfunction and childhood onset of cerebrovascular disease. Narrowing of cerebral vessels appears to be at expense of intima hyperplasia (SMC proliferation) that leads to white matter and arterial ischemic injury. Previous in-silico 3D modeling has suggested that ACTA2 Arg179 variants (Arg179H and Arg179C) may alter the polymerization of actin from Globular Actin (G-Actin) to Filamentous Actin (F-Actin) but to date no cellular, molecular or functional studies in human cells have been done to determine the underlying mechanisms of intima hyperplasia in ACTA2 Arg179 mutant cells. Here, we characterized ex-vivo control and patient-derived myofibroblasts through G/F Actin ratio quantification via western blot, determined changes in cytoskeletal assembly by high resolution immunofluorescent microscopy, and assessed migration and proliferative function in wound healing assays. Mutant cells showed a significantly higher ratio of G-Actin to F-Actin and an abnormal response to transforming growth factor beta (TGF-β) compared to controls. Through immunocytochemistry, we confirmed disruption of stress fibers and F-Actin associated with the accumulation of G-Actin in the cytoplasm. Moreover, wound healing assays showed that mutant cells close the gap sooner than controls by increasing their cell numbers rather than their migratory capacity. Our findings suggest a dominant negative effect of mutant protein on actin polymerization that leads to decreased migration and compensatory proliferation of smooth muscle cells. These deficits may underlie the intima proliferation associated with cerebrovascular disease and ischemic injury seen in ACTA2 Arg179 pathogenic variants. This ex-vivo model provides a robust molecular and functional phenotype for studying the underlying cellular mechanisms of ACTA2 Arg179 mutations and provides an opportunity to test potential therapeutic interventions in the future.

Disclosures: C.A. Lusk: None. **C.L. Lino Cardenas:** None. **S.K. McCarthy:** None. **C.G. Bergen:** None. **M.E. Lindsay:** None. **P.L. Musolino:** None.

Poster

P909. Biochemical and Molecular Techniques II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P909.09

Topic: I.04. Physiological Methods

Support: NIH-NIGMS RISE R25GM061838
NSF-PIRE OISE-1545803

NIMHD 8G12-MD007600
NCCR 5R25GM061838-21
RCM1 Grant 2U54MD007600

Title: Alteration of H3K9-acetylation in the mesocorticolimbic system after the development of cocaine sensitization

Authors: *D. CONSUEGRA GARCIA[1], C. CALO-GUADALUPE[1], K. Y. BOSQUE CORDERO[2], R. VAZQUEZ-TORRES[1], M. S. BRODIE[3], C. JIMENEZ-RIVERA[1];
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Abstract: Addictive substances are known to induce changes in the brain's mesocorticolimbic system (MCL). The MCL is made of various structures which process pleasurable and reinforcing stimuli. Drugs of abuse cause long-term transcriptional changes, known to be produced by epigenetic modifications in the brain and contributing to the individual's vulnerability to develop addiction. Among the modifications that can change brain function by altering gene expression are histone post-translational modifications (PTMs). Histone acetylation, a type of PTM, is one of the most studied epigenetic modifications in the field of substance use disorder (SUD). One group of enzymes that control histone acetylation are histone deacetylases (HDACs), which regulate gene expression. Hyperpolarization activated cyclic nucleotide (HCN) channels are widely expressed in the MCL and are the molecular structure that is responsible for the expression of I_h current. Previous results from our laboratory demonstrated that cocaine behavioral sensitization, a non-contingent cocaine administration model, increased HCN2 subunit's expression in the ventral tegmental area (VTA), nucleus accumbens, prefrontal cortex (PFC) and hippocampus (HIP). We also demonstrated an I_h reduction in putative VTA DA neurons from cocaine-sensitized rats' concomitant to a decrease in surface expression of HCN2 subunits. Our preliminary data shows that I_h reduction can be reverted to baseline levels after in vitro incubation with an HDAC inhibitor (SAHA), indicating that histone acetylation is a major mechanism controlling HCN adaptation during cocaine sensitization. Our hypothesis is that acetylation profiles in other MCL structures are also altered by cocaine sensitization. Using the western blot technique, we investigated total protein concentration of H3K9-Ac, a histone modification marker, in MCL regions known to express I_h. Animals were sensitized for 7-days and 24-hours after the last cocaine injection were sacrificed and tissue punches of the PFC and HIP were collected. Densitometric analysis demonstrate that cocaine sensitized animals have a decrease H3K9-Ac expression in the PFC and HIP compared to saline controls. These results could indicate that cocaine sensitization causes alterations in histone modification in MCL structures that controls the expression of HCN and other neuroadaptations. Knowledge of epigenetic cocaine-induced changes in I_h within the MCL can ultimately identify new targets for the development of agents to treat SUD. Currently, CUT & TAG studies are being performed to view H3K9-Ac levels in gene promoters responsible for the modulation of HCN.

Disclosures: D. Consuegra Garcia: None. **C. Calo-Guadalupe:** None. **K.Y. Bosque Cordero:** None. **R. Vazquez-Torres:** None. **M.S. Brodie:** None. **C. Jimenez-Rivera:** None.

Poster

P920. Genomics, Proteomics, and Systems Biology

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P920.01

Topic: I.02. Systems Biology and Bioinformatics

Support: Mayo Foundation for Medical Education and Research
J. Willard and Alice S. Marriott Foundation
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NIGMS Center for Psychiatric Neuroscience COBRE grant P30 GM103328
NIMH grant R01 MH67996
Ulm Foundation
NIAAA grant AA018779

Title: Down-regulation of KCNQ3 in suicide: A post-mortem human brain proteomic study

Authors: *M. VELDIC[1], A. CABALLO-ARREOLA[2], M. HO[2], A. OZERDEM[2], A. B. CUELLAR-BARBOZA[3], M. U. KUCUKER[2], C. A. STOCKMEIER[4], G. RAJKOWSKA[4], M. A. FRYE[2], D.-S. CHOI[2];
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Abstract: Introduction
Global rates of suicide are estimated at >800,000 annually. The dorsolateral prefrontal cortex (DLPFC) has been an important area of interest in suicide research due to evidence of morphometric changes and neurotransmitter abnormalities.
Objectives
We aimed to identify proteomic profile and biological pathways in the DLPFC of suicide victims employing mass spectrometry-based non-biased untargeted proteomics.
Methods
DLPFC from age-matched male mood disorder suicide cases (n=5) and non-suicide cases (n=5) were compared. Proteome was subjected to trypsin digestion and analyzed by liquid chromatography coupled to tandem mass spectrometry. Protein identification and quantitation was achieved using MaxQuant and in-house software. Proteins that differed between groups at false discovery rate (FDR) corrected *p*-values (Benjamini-Hochberg-Yekutieli) <0.3 and absolute log2 fold change (FC) >0.4 were considered statistically significant and were subjected to pathway analysis by Qiagen Ingenuity software. Validation of two proteins of the top canonical pathway was performed using Western blot.
Results

In the suicide cases, 33 of the 5644 detected proteins showed significantly altered expression. The top differentially expressed proteins were potassium voltage-gated channel subfamily Q member 3 (KCNQ3) (log_2 FC=-0.481, p=2.10E-09, FDR<0.001), metallo-beta-lactamase domain-containing protein 1 (MBLAC1) (log_2 FC=-1.011, p=1.16E-06, FDR=0.002), and tripartite motif containing 36 (TRIM36) (log_2 FC=0.614, p=2.31E-06, FDR=0.003). The top canonical pathway enriched was the GABA-receptor signaling (p<0.001). We validated the downregulation of KCNQ3 by Western blot (p=0.045).

Conclusions

Here we report for the first time a reduction in the levels of KCNQ3 in the DLPFC of suicide victims. Future studies with larger sample and equal gender representation are needed.

Disclosures: M. Veldic: None. **A. Caballo-Arreola:** None. **M. Ho:** None. **A. Ozerdem:** None. **A.B. Cuellar-Barboza:** None. **M.U. Kucuker:** None. **C.A. Stockmeier:** None. **G. Rajkowska:** None. **M.A. Frye:** F. Consulting Fees (e.g., advisory boards); Janssen, Mitsubishi Tanabe Pharma Corporation, Myriad, Sunovion, Teva Pharmaceuticals. **D. Choi:** F. Consulting Fees (e.g., advisory boards); Peptron Inc..

Poster

P920. Genomics, Proteomics, and Systems Biology

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P920.02

Topic: I.02. Systems Biology and Bioinformatics

Support: F.8-762/A-762/2019/RS

Title: Blood based protein biomarkers for the diagnosis and differentiation of acute stroke using a discovery based SWATH-MS proteomics approach: Results from the Discovery phase

Authors: *S. MISRA[1], P. SINGH[5], D. BHALLA[1], P. TALWAR[1], R. CHAKRABORTY[5], M. NATH[1], S. SENGUPTA[5], P. KUMAR[1], A. KUMAR[1], P. AGGARWAL[2], A. K. SRIVASTAVA[1], A. K. PANDIT[1], D. MOHANIA[3], A. GULATI[4], K. PRASAD[1,6], D. VIBHA[1]; [1]Neurol., [2]Emergency Med., [3]Dr. R.P. Ctr., [4]Clin. Epidemiology Unit, All India Inst. of Med. Sci., New Delhi, India; [5]CSIR-Institute of Genomics and Integrative Biol., New Delhi, India; [6]Neurol., Rajendra Inst. of Med. Sci., Ranchi, India

Abstract: Background: Rapid diagnosis of stroke and its subtypes is essential in early stages. Due to unavailability of neuroimaging in developing nations and time-sensitive nature of revascularization therapies, blood biomarkers are required to aid clinical decision making. Recent developments in high-throughput proteomics approach have shown potential to discover biomarkers for diagnosing acute stroke.

Objective: To determine blood based protein biomarkers for the diagnosis & differentiation of

ischemic stroke (IS) from healthy controls and intracerebral hemorrhage (ICH) within 24 hours of symptom onset using a discovery based SWATH-MS proteomics approach.
Methods: An ongoing diagnostic test study conducted at Department of Neurology, All India Institute of Medical Sciences, New Delhi, India. In the discovery phase, serum samples were collected within 24 hours from acute stroke (IS & ICH) patients & healthy controls and were subjected to SWATH-MS based untargeted proteomics. A spectral ion library for serum proteins was generated by analysing SCX fractionated tryptic peptides in data dependent mode and UniprotKB database was searched using Proteinpilot™ software 5.0.1 for SWATH analysis. Peak processing and area extraction was performed using SWATH 2.0 in PeakView 2.2 software at 1% peptide false discovery rate (FDR). Significant proteins were selected using the following criteria: >1.5 foldchange for upregulated, <0.67 for downregulated, p-value<0.05 and confirmed/tentative selection using Boruta random forest. Protein-protein interactions and pathway analysis were conducted using STRING version 11. Statistical analysis was conducted in R version 3.6.2.
Results: Our study included 80 subjects; 40 total stroke cases (20 IS, 20 ICH) within 24 hours of symptom onset [mean age: 50.4(10.38)] and 40 age, sex, hypertension and diabetes matched healthy controls [mean age: 49.64(10.64)]. Total 251 proteins were identified in Proteinpilot™ search with 5% global FDR. Through SWATH analysis, we quantified 210 proteins. In IS cases, 17 proteins compared to ICH and 12 proteins compared to healthy controls were significantly differentially expressed. Sixteen proteins were significantly differentially expressed in total stroke compared to control subjects. The network for 13 proteins was highly connected showing molecular interactions with other proteins with high confidence.
Conclusion: Our study identified potential protein biomarker candidates for the diagnosis and differentiation of acute stroke. These biomarkers will further be validated in a large sample to determine their diagnostic potential in the ongoing validation phase of the study.

Disclosures: S. Misra: None. **P. Singh:** None. **D. Bhalla:** None. **P. Talwar:** None. **R. Chakraborty:** None. **M. Nath:** None. **S. Sengupta:** None. **P. Kumar:** None. **A. Kumar:** None. **P. Aggarwal:** None. **A.K. Srivastava:** None. **A.K. Pandit:** None. **D. Mohania:** None. **A. Gulati:** None. **K. Prasad:** None. **D. Vibha:** None.

Poster

P920. Genomics, Proteomics, and Systems Biology

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P920.03

Topic: I.02. Systems Biology and Bioinformatics

Support: NIH Grant P20GM103434
NIH Grant P20GM121299
NIH Grant 2U54GM104942

Title: Screening more vs. less mutagenic targets among 129 kinases of the druggable genome

Authors: I. MCKNIGHT[1], R. RAINES[1], H. WHITE[1], ***J. SHIM**[2];
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Abstract: Aim: Several factors have been reported to be associated with high mutation rates, including recombination rate, proximity to a telomere [F(i)], and high adenine and thymine (A+T) content [F(ii)]. Among these factors, we have previously shown that F(i) and F(ii) can explain some of the genetic mutations linked to neurological disorders, such as congenital hydrocephalus. Here, we analyzed 129 kinases in the Druggable Genome if they meet these two aforementioned factors. **Methods**: A list of understudied proteins has been reported by *the Illuminating the Druggable Genome Project* (IDG) of the NIH. Among 390 understudied proteins associated with cancer and neurological disorders, we tested how many previously known genes match with (i) proximity to telomeres or (ii) high A+T content in 129 kinases. The genomic information was extracted using a genome data viewer. **Results**: Out of the 129 genes of the understudied druggable kinases we examined, 74% (95 of 129) were of sufficient proximity to their telomeres at less than 50 million bases (Mb). Roughly 25% (32 of 129) of the kinases contained A+T content higher than 59%, which is the human genome-wide average. Furthermore, 106 of 129 kinases (82%) satisfied either telomere proximity (F(i)) or high A+T content (F(ii)), while 23 of 129 genes (18%) met neither of the two factors. Thus, these 23 kinases have a relatively lessened chance of deleterious mutations. **Conclusion**: Our initial results reveal that 129 understudied kinases proposed as druggable proteins by the IDG are highly susceptible to germline or somatic mutations. This suggests that 82% of the listed druggable kinases (n=129) are prone to high mutation rate due primarily to their proximity to telomeres and/or nucleotide compositions and that 23 of 129 kinases (18%) are relatively less mutagenic as druggable targets.

Disclosures: I. McKnight: None. **R. Raines:** None. **H. White:** None. **J. Shim:** None.

Poster

P920. Genomics, Proteomics, and Systems Biology

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Program #/Poster #: P920.04

Topic: I.02. Systems Biology and Bioinformatics

Support: NIMH R56MH113923
NSF CAREER Award 1846234
NINDS R01NS107539
NIMH R01MH117111
the Beckman Young Investigator Award
Searle Scholar Award

Rita Allen Foundation Scholar Award

Title: Genetically targeted proteomics identify compositional and phosphoproteome of corticostriatal axons across postnatal development

Authors: *V. DUMRONGPRECHACHAN[1,2], R. B. SALISBURY[3,4], L. BUTLER[1], M. L. MACDONALD[3,4], Y. KOZOROVITSKIY[1,2];
[1]Northwestern Univ., Evanston, IL; [2]Chem. of Life Processes Inst., Evanston, IL; [3]Univ. of Pittsburgh, Pittsburgh, PA; [4]Biomed. Mass Spectrometry Ctr., Pittsburgh, PA

Abstract: The basal ganglia (BG) process information from the cerebral cortex, thalamus, and limbic structures, essential for goal-directed behaviors including motor learning, action selection and habit formation. The striatum is the largest input to BG that receives diverse cortical and thalamic glutamatergic inputs. Innervating glutamatergic afferents form synapses onto dendritic spines of spiny projection neurons (SPNs), the output neurons of the striatum. Corticostriatal axon innervation begins in the embryonic stage and continues to develop postnatally in the rodent brain. Little is known about the molecular basis contributing to circuit formation including the innervating axon guidance systems, synapse formation, and their regulations. Abnormal development of striatal circuits has been implicated in the pathogenesis of many neurological disorders such as autism spectrum disorders (ASD) as well as Tourette's and obsessive-compulsive disorders. Studying the axonal proteomes of any neural system remains challenging. Axonal compartments are difficult to isolate. Suboptimal techniques such as fluorescent sorting of synaptosomes and axon growth cones require extensive tissue dissociation and result in loss of cell-type specific information. Here, we use a novel APEX2 reporter mouse line and proximity labeling proteomics to map compositional and phosphoproteome of corticostriatal axons across postnatal development.

Disclosures: V. Dumrongprechachan: None. **R.B. Salisbury:** None. **L. Butler:** None. **M.L. Macdonald:** None. **Y. Kozorovitskiy:** None.

Poster

P920. Genomics, Proteomics, and Systems Biology

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Program #/Poster #: P920.05

Topic: I.02. Systems Biology and Bioinformatics

Support: UM1HG009443

Title: Characterizing alternative splicing in the hippocampus and cortex of the ENCODE4 mouse postnatal time course using bulk and single-nucleus long-read RNA-seq

Authors: *F. REESE[1,2], E. REBBOAH[1,2], B. A. WILLIAMS[3], H. LIANG[1,2], M. GANTUZ[1,2], B. J. WOLD[3], A. MORTAZAVI[2,1];
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Abstract: Alternative isoforms that arise from alternative splicing as well as transcription start site (TSS) or transcription end site (TES) choice are known to play key roles during postnatal development especially in the brain, which is commonly known as the organ with the highest alternative splicing rates in humans. However, many of these isoform changes have been characterized using short-read sequencing techniques that were not comprehensive and did not probe the full length identity of each isoform. Furthermore, the cell-type specificity of these isoforms can be difficult to deconvolve from bulk or short-read single-cell data.
As a part of the 4th phase of the ENCODE Consortium, we collected hippocampus and cortex from male and female C57BL6J/Castaneus F1 hybrid mice at 8 postnatal timepoints (P4, P10, P14, P25, P36, P2mo, P8mo, P18-20mo). These samples were probed using multiple sequencing technologies within the consortium including DNase-seq, microRNA-seq, and RNA-seq. Notably, we sequenced these timepoints using bulk long-read RNA-seq as well as a form of single-cell long-read RNA-seq (LR-Split-seq). We are analyzing this full-length transcriptomic dataset in concert with the additional data modalities to characterize the landscape of alternative isoform usage as well as the regulatory influences such as haplotype, epigenetic state, and miRNA expression landscape that drive alternative isoform expression across tissues, timepoints, and cell types. We are also probing our long-read transcriptomes for genes that exhibit isoform, TSS, or TES switching across tissues, timepoints, and cell type. We are further querying our transcripts that have altered open reading frames or may be subject to nonsense-mediated decay with a specific focus on the genes that are found to display evidence of isoform switching.

Disclosures: F. Reese: None. **E. Rebboah:** None. **B.A. Williams:** None. **H. Liang:** None. **M. Gantuz:** None. **B.J. Wold:** None. **A. Mortazavi:** None.

Poster

P920. Genomics, Proteomics, and Systems Biology

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Program #/Poster #: P920.06

Topic: I.02. Systems Biology and Bioinformatics

Support: NIMH grant 1U24MH114827-01
Allen Institute

Title: Organization and access to BICCN data through the Brain Cell Data Center

Authors: *C. L. THOMPSON[1], T. S. MOLLENKOPF[2], K. BAKER[1], P. M. BAKER[3], A. E. BANDROWSKI[4], K. DEGATANO[6], T. FLISS[1], T. GILLESPIE[5], J. MATHEWS[1], F. KHAJOUEI[6], E. KIERNAN[6], K. KONWAR[6], P. RAY[1], N. SJOQUIST[1], S. MUFTI[1], J. GEE[7], P. YUSHKEVICH[7], M. E. MARTONE[5], T. TICKLE[6], L. NG[1], M. J. HAWRYLYCZ[8];
[1]Allen Inst. for Brain Sci., Seattle, WA; [2]Allen Inst., Seattle, WA; [3]Dept. of Biol. Structure, Univ. of Washington, Seattle, WA; [5]Neurosci., [4]UCSD, La Jolla, CA; [6]The Broad Inst. of MIT and Harvard, Cambridge, MA; [7]Univ. of Pennsylvania, Philadelphia, PA; [8]Modeling, Analysis, and Theory, Allen Inst. Brain Sci., Seattle, WA

Abstract: The BRAIN Initiative Cell Census Network (BICCN) is a consortium of laboratories generating high value data to profile various phenotypic aspects of cell types in mouse, human, and non-human primate brain with a goal to generate, map, and share brain cell data with research communities. The Brain Cell Data Center (BCDC) organizes the diverse data, tools, and knowledge and provides public access to data and multimodal classification of cell types. The BCDC, as part of its mission, focuses on ensuring that data outputs of the consortium are made accessible and strives towards the FAIR data principles (findable, accessible, informative and reusable).
BICCN Data Inventory. BICCN projects and data collections are served to the public in the BICCN Data Inventory. This resource allows the community to find relevant datasets for their research. Metadata regarding specimens profiled in this effort are staged in a public repo prior to presentation on in the Data Inventory. This resource also points to data archive locations for high resolution data and protocols.
Ontology development. The BCDC maintains an ontology for relevant fields for metadata collection, including specimen types, techniques, and other relevant fields. The BCDC is incorporating the cell type ontology being developed for the BICCN and neuron phenotypes.
Research Resource IDs (RRIDs). The consortium has encouraged BICCN participants to register their research resource identifiers relevant to the consortium to improve the identification and citability of resources and support reuse.
Data and resource documentation. Through a coordinated effort with data-generating centers, data archives and analysis tools, the BICCN generated a multimodal cell census and a cell atlas of the mammalian primary motor cortex. The teams synthesized a holistic framework to define the cell type composition of neural circuits. These datasets and others are described with detailed information about projects and data collections in a searchable format, with links to data repositories and other relevant tools in our new searchable Project Summary pages.
FAIR Data Pipelines. The BCDC works with consortia members to transition processing pipelines used in publications to cloud native implementations publicly available in Terra. These pipelines are installed in a cloud and ready to use with documentation, test data sets and outputs. The pipelines support community standards including GA4GH data standards, RRIDs, use of standard workflow languages, open licensing, open source code (public GitHub repositories) and deposition in Dockstore (a community resource for bioinformatics workflows).

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Poster

P920. Genomics, Proteomics, and Systems Biology

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P920.07

Topic: I.02. Systems Biology and Bioinformatics

Support: NIH T35DK074390
NIH OD010662

Title: Head to foot: the role of brain peptides in diabetic wound healing.

Authors: L. THURA[1], ***K. PHILIBERT**[3], X. SHAO[4], C. YANG[2], S. WU[1], M. J. GLUCKSMAN[4];
[1]Scholl Col. of Podiatric Medicine, Ctr. for Lower Extremity Ambulatory Res., [2]Midwest Proteome Ctr., Rosalind Franklin Univ. of Med. and Sci., North Chicago, IL; [3]Ctr. for Proteomics and Mol. Therapeutics., [4]Ctr. for Proteomics and Mol. Therapeut., Chicago Med. School/RFUMS, North Chicago, IL

Abstract: Diabetes mellitus is a metabolic disease accompanied by potentially debilitating complications creating major health problems for a significant population. Almost 20 million people worldwide develop diabetic foot ulcers (DFUs) annually that begin with peripheral neuropathy and susceptibility to wounds with accompanying inflammation. DFUs often exhibit impaired healing, placing patients at an increased risk of infection often progressing to lower limb amputation and increased morbidity and mortality. Biomarkers are utilized as an objective biologic measurement and evaluation of normal versus pathological biological processes. More specifically, we compare those diabetic patients who will heal with those who will not. Utilizing biochemical, genomic and proteomic approaches with clinical samples, we identify biomarkers of wound healing in DFU to allow improved clinical diagnosis, prognoses and targeted treatments by stratifying patient outcomes. We utilize a high throughput proteomics approach starting with patient serum from the wound site during debridement for the discovery phase. After immunodepletion of the 12 most common protein components in serum, there is a large-scale and high throughput separation by UPLC and mass spectrometry to quantify differences in patient groups. Lastly, the bioinformatics data handling, including meta-analyses of several of our studies is comprised of PEAKS data with transformation by Stata to gauge patterns in sampling or linkage in gene and protein pathways. Neuropeptides, such as neurotensin, and their enzymatic regulators, including neurolysin, as well as other proteins found in the nervous system have been implicated in the pathophysiology of diabetic foot wounds by virtue of neuropathic processes as well as the wound healing process from inflammation to migration/proliferation and

remodeling through wound closure. We enter the validation phase of identified biomarkers representing potential differences between diabetic healers and non-healers characterized by our multipronged workflow.

Disclosures: L. Thura: None. **K. Philibert:** None. **X. Shao:** None. **C. Yang:** None. **S. Wu:** None. **M.J. Glucksman:** None.

Poster

P920. Genomics, Proteomics, and Systems Biology

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Program #/Poster #: P920.08

Topic: I.02. Systems Biology and Bioinformatics

Support: Japan Agency for Medical Research and Development, AMED under Grant Number JP20dm0107140 to N.N.
the Ministry of Education, Culture, Sports, Science and Technology (MEXT) of Japan to N.N.(17H01564)

Title: Proteomic analysis of heat-stable proteins revealed an increased proportion of proteins with compositionally biased regions

Authors: *H. PARK[1], T. YAMANAKA[2], N. NUKINA[1];
[1]Lab. of Structural Neuropathology, Sch. of Brain Science, Doshisha Univ., Kyoto, Japan; [2]Dept. of Neurosci. of Dis., Brain Res. Institute, Niigata Univ., Niigata, Japan

Abstract: Intrinsically disordered proteins (IDPs) have been in the spotlight for their unique properties, such as their lack of secondary structures and low sequence complexity. Alpha-synuclein and tau are famous disease related IDPs with compositionally biased regions in their sequences, accumulating in the brains of patients with Parkinson disease and Alzheimer disease, respectively. Out of the various properties of known IDPs, their heat resistance in particular was what attracted our attention. We assumed that there exist many other unidentified proteins that are extremely resistant to heat, referred to as heat-stable proteins, which would also have low sequence complexity. In this study, we identified heat-stable proteins in the soluble fraction of mouse brains and found that most of the heat-stable proteins had low sequence complexity with biased amino acid composition. Found in abundance in human brains, IDPs can undergo different types of post-translational modifications (PTMs) leading to various human diseases. We then investigated the heat-stability and aggregation of phosphorylated synuclein and tau with different phosphorylation sites. We suggest that PTMs can be important factors that determine the heat-stability and aggregation property of a protein. Complete understanding of IDPs and their role in the pathogenesis of human diseases requires characterizing the features and properties of heat-stable proteins.

Disclosures: H. Park: None. **T. Yamanaka:** None. **N. Nukina:** None.

Poster

P920. Genomics, Proteomics, and Systems Biology

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Program #/Poster #: P920.09

Topic: I.02. Systems Biology and Bioinformatics

Support: AFOSR 20RHCOR04

Title: Anodal Transcranial Direct Current Stimulation Affects Rat Cortex and Hippocampal Protein Expression in an Intensity-Dependent Manner

Authors: *S. H. JUNG[1,2], S. HARSHMAN[1], C. HATCHER-SOLIS[1], J. MARTIN[1];
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Abstract: Prevailing evidence indicates transcranial direct current stimulation (tDCS) affects learning and memory in a region specific and intensity-dependent manner, but the regulatory mechanisms remain unclear. Previously, we provided evidence that tDCS enhances memory consolidation by modifying plasticity-associated proteins in the hippocampus and genes in the cortex. Here, we examined the impact of tDCS current intensity on proteomes in the rat cortex and hippocampus. A single session of anodal tDCS was applied for 30 minutes (near bregma 0-5 mm) at the current intensity of 0, 250 or 500uA, and the brain regions were collected after 2 hours (n = 5/group). Synaptoneurosomes were isolated and bottom-up liquid chromatography mass spectrometry was used to profile proteomics. The Proteome Discoverer software was used to search proteomic data with a target false discovery rate of 0.05. The normalized data of about 3000 proteins were analyzed. Functional enrichment analysis showed unique and similar pathways and functions were significantly enriched by tDCS when compared between cortex and hippocampus. Protein-protein interaction and Ingenuity pathway analyses resulted in significant pathways and functions when compared between different current intensities and different brain regions. Canonical pathway analysis showed that synaptogenesis signaling pathway was significantly affected by tDCS in hippocampus but not cortex: p values for 250uA intensity of anode and cathode polarities and 500uA intensity of anode and cathode polarities are 4.19E-12, 3.07E-04, 0.0026, & 0.023, respectively. However, no statistical significance was detected from the cortex. Neuritogenesis function was significantly affected by anodal 250uA in both hippocampal (p = 1.95E-16) and cortical (p = 1.12E-05) and only cathodal 250uA in hippocampal (p = 7.51E-09) synaptosomes. We detected overlapping proteins that were significantly modified between the two different current intensities and brain regions that were examined. For example, NUBP1 was significantly decreased by anodal 250uA and 500uA in both hippocampal (p = 0.0419 & 0.0391) and cortical (p = 0.0021 & 0.0216) synapses. However,

only cortical synapse showed a significant change in the NUBP1 expression by cathodal tDCS. In conclusion, our data show similar tDCS-induced alterations in cortical and hippocampal synaptosomes across 2 different current intensities: especially beneficial effects by the 250uA current intensity. Thus, our data provide supporting evidence that tDCS-induced enhancement of cognitive performance may be associated with tDCS-induced modifications in cortical and hippocampal synaptosomes.

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Poster

P920. Genomics, Proteomics, and Systems Biology

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Topic: I.02. Systems Biology and Bioinformatics

Support: OBI-NSTP Fellowship
ames S. McDonnell Foundation Understanding Human Cognition Scholar Award 220020467
NIH R01MH103517
HCA-A-1704-01747 Chan Zuckerberg Initiative

Title: Identifying epigenomic and transcriptomic alterations in human brain evolution at cellular resolution

Authors: *E. CAGLAYAN[1], F. AYHAN[2], T. M. PREUSS[4], S. YI[5], G. KONOPKA[3];
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Abstract: Human evolution is marked by an enlarged brain and increased cognitive ability. It is hypothesized that such traits must be genetically encoded, motivating decades of research to find human specific molecular traits linked to specific genetic modifications. Early approaches that compared humans to closely related species - primarily chimpanzees - showed that the majority of human specific molecular traits are likely caused by alterations in non-coding regions, leading to altered gene expression patterns. To understand both non-coding and gene expression patterns specific to humans at cellular resolution, we performed single-nuclei ATAC-seq (snATAC-seq) and single-nuclei RNA-seq (snRNA-seq) in human, chimpanzee and macaque adult cortex. This yielded ~73000 nuclei in snATAC-seq and ~150000 nuclei in snRNA-seq. We reproduced many previously identified neuronal subtypes and found certain neuronal subtypes to be more human specific than others indicating heterogeneity of human evolution at finer cellular resolution.

Through motif analysis on human specifically accessible regions, we were able to pinpoint human specific transcription factor activity for each cell type, including neuronal subtypes. We also found that human specific molecular features were more enriched in cognitive diseases compared to chimpanzees at the gene regulatory level and highlight certain cell types with increased disease enrichment. Taken together, our results demonstrate the epigenomic and transcriptomic features of human brain evolution and uncover their potential link with cognitive diseases at the cellular level.

Disclosures: E. Caglayan: None. **F. Ayhan:** None. **T.M. Preuss:** None. **S. Yi:** None. **G. Konopka:** None.

Poster

P925. Advances in Circuit Tracing Techniques

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Program #/Poster #: P925.01

Topic: I.03. Anatomical Methods

Support: MRC Grant MR/R01213X/1
NIH SPARC Program Grant 1OT2OD026545-01

Title: Tracing of cardiac, pulmonary and recurrent laryngeal fascicle groups in porcine vagus nerve to inform selective vagus nerve stimulation (SPARC)

Authors: *N. THOMPSON[1], E. RAVAGLI[1], S. MASTITSKAYA[1], F. IACOVIELLO[2], J. PERKINS[4], A. V. GOURINE[3], P. SHEARING[2], K. ARISTOVICH[1], D. HOLDER[1];
[1]Med. Physics and Biomed. Engin., [2]Chem. Engin., [3]Neuroscience, Physiol. and Pharmacol., Univ. Col. London, London, United Kingdom; [4]The Royal Vet. Col., Hatfield, United Kingdom

Abstract: Vagus nerve stimulation (VNS), a promising treatment for numerous conditions, often results in off-target effects due to the unknown fascicular organisation of the cervical vagus nerve and thus nonspecific nature of stimulation. In this work, we determine the fascicular anatomical representation of the recurrent laryngeal, cardiac and pulmonary branches of the vagus nerve at the mid-cervical level of VNS cuff placement in the vagus nerve in pigs using micro-computed tomography (microCT).
The left vagus nerves of four domestic female 60-70 kg pigs were dissected from the cervical region to the branches of the target organs, fixed in neutral buffered formalin, sectioned into 4 cm lengths with appropriate landmarks for registration left in place and soaked in Lugol's solution (1% total iodine) for five days for sufficient contrast staining. For scanning, the sections of nerves were blotted dry, and placed in a 3D printed mount within the microCT scanner (Nikon XT H 225, Nikon Metrology). A molybdenum target was used and the scanning parameters for all scans were set to the following: 7 μm voxel size, 35 kVp energy, 114 μA current, 4 W power,

0.25 fps in 4 s exposure time, and 3176 projections. Scans were reconstructed in CT Pro 3D (Nikon Metrology) and analysed in ImageJ. Fascicles of the three target organs/fascicle groupings were segmented and traced with Neurolucida 360 (MBF Bioscience) from the point of branching to the mid-cervical level of the vagus (20-25 cm total length per nerve).
Three distinct regions, with minimal overlap only between a small number of pulmonary and recurrent laryngeal fascicles (~5%), were identified within the cross-section of the cervical vagus nerves with the three identified fascicular groupings remaining mostly discrete at the cervical level (~30%, 30% and 4% of total vagus nerve area for pulmonary, recurrent laryngeal and cardiac fascicle groups, respectively). The cardiac fascicle(s) remained completely separate from the other fascicular groups. This confirmed an organotopic organisation of the cervical vagus nerve according to organ/function when looking at these three fascicular groupings. Further studies are required to identify the organisation of the fascicles carrying information to the other organs innervated by the vagus nerve. Knowledge of the functional anatomical organisation of the vagus nerve will enable accurate selective VNS ultimately allowing for the avoidance of off-target effects and improving its overall therapeutic efficacy. Work in progress is to extend the approach to delineate the functional anatomy at the cervical level of excised vagus nerves of humans.

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Poster

P925. Advances in Circuit Tracing Techniques

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Program #/Poster #: P925.02

Topic: I.03. Anatomical Methods

Support: NIH RO1-NS112104-01
NIGMS R25-GM078441

Title: Structural connectivity of the fore- and mid- brain in prairie voles

Authors: *K. R. GOSSMAN[1], B. DYKSTRA[1], K. TA[1], A. S. SMITH[2];
[2]Dept. of Pharmacol. & Toxicology, [1]Univ. of Kansas, Lawrence, KS

Abstract: Structural Connectivity of the Fore- and Mid- Brain in Prairie Voles
Kyle R. Gossman, Benjamin Dykstra, Kyle Ta, & Adam S. Smith.
Department of Pharmacology and Toxicology, University of Kansas, Lawrence, KS, USA
The socially monogamous prairie vole (Microtus ochrogaster) has been used for three decades to study the neurobiology of pair bonding. This has led to well-defined behavioral characterization

of partner affiliation and stranger aggression influences by particular neurochemicals and certain brain regions. However, limited knowledge is known about the overall neural circuity and interregional connections of vole brain. Neuroanatomical tracing methods remain fundamental for elucidating the complexity of brain circuits. Here in the present study, we used the highly gregarious prairie vole and cholera toxin subunit-B retrograde tracers (488nm, 555nm, 594nm, and 647nm) to map out the ipsilateral and contralateral fore- and mid- brain physical connections of the regions associated with the Social Decision-Making Network (SDMN) with the addition of the paraventricular nucleus of the hypothalamus (PVN) and the anterior cingulate cortex (ACC). In addition to limited work, these results will significantly expand our knowledge of the neural circuity of the vole brain and act as a guide for interregional connections for future research.

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Poster

P925. Advances in Circuit Tracing Techniques

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Program #/Poster #: P925.03

Topic: I.03. Anatomical Methods

Support: SNF CRSII5_177237

Title: Long range axonal projections of individual neurons in mouse layer 2/3 barrel cortex

Authors: *Y. LIU[1], G. FOUSTOUKOS[1], S. CROCHET[2], C. C. PETERSEN[3];
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Abstract: Neurons primarily communicate through release of neurotransmitter from presynaptic specialisations along their axonal arborisations. To understand the functional role of a specific neuron, it is thus important to know the structure of its axon. In particular, long-range axonal projections are thought to be important for communication between different brain regions. The mouse whisker system provides unique opportunities to investigate structure function relationships in the mammalian neocortex. The whisker primary somatosensory cortex (wS1) of mice contains a somatotopic map such that each whisker on the snout is individually represented by an anatomically-defined ‘barrel’. Each barrel column in wS1 primarily processes tactile sensory information from its homologous whisker. However, pyramidal neurons in wS1 signal information to many downstream targets, and even a single whisker deflection can evoke sensory signals across many brain areas. Here, in order to examine the structure of individual neurons, we used *in vivo* single-cell electroporation to express green fluorescent protein in layer 2/3 pyramidal neurons in wS1. After several days of expression, the mouse brains were fixed,

incubated in modified Cubic solutions, embedded in agarose, and then imaged using a two-photon tomographic microscope (Ragan et al., Nat Methods 2012). Semi-automated tracing of axons, allowed three-dimensional reconstruction of dendritic and axonal arborisations. Finally, registering the brain volume and the annotated morphology with the Allen Institute's Reference Atlas provided precise identification of long-range axonal targets (Han et al., Nature 2018). Although we were able to trace extensive axonal projections using this methodology, the signal-to-noise ratio was limited, likely giving rise to incomplete reconstructions. Our next goal is to obtain more complete axonal morphologies using iDISCO (Renier et al., Cell 2014), involving tissue clearing, immunolabelling and imaging with light sheet microscopy. (The first two authors contributed equally to this work)

Disclosures: Y. Liu: None. **G. Foustoukos:** None. **S. Crochet:** None. **C.C. Petersen:** None.

Poster

P925. Advances in Circuit Tracing Techniques

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Topic: I.03. Anatomical Methods

Support: NIH Grant R01MH118257
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UMN AIRP award
MnDrive Brain Conditions

Title: Posteromedial cortex projections to the striatum in the macaque

Authors: *M. MONKO, S. R. HEILBRONNER;
Neurosci., Univ. of Minnesota, Minneapolis, MN

Abstract: The Posteromedial Cortex (PMC), comprised of the posterior cingulate, retrosplenial cortex, and the precuneus, make up the posterior hub of the default mode network. Abnormal connectivity of the default mode network has been seen in a wide variety of psychiatric diseases. We are interested in how the interactions of the PMC with decision-making circuitry. To investigate this, we use a combination of tract tracing techniques and high-field resting-state fMRI in macaques. We examined the topography of the PMC to the striatum using bidirectional tract-tracers injected into the PMC. The resulting striatal terminal fields were charted and subsequently transformed into a 3D model registered to an MRI-generated reference space. These PMC-striatal cases were compared with other corticostriatal cases that have been implicated in decision-making. We also analyzed the functional connectivity of the PMC and striatum and compared this against our anatomical tracings. The PMC projects primarily, but not

exclusively, to the dorsomedial striatum, sharing striatal territory with prefrontal terminal fields involved in goal-directed decision making. These results contribute to growing literature about the involvement of the PMC in decision-making, as well as examining the relationship between anatomical and functional connectivity of corticostriatal circuitry.

Disclosures: M. Monko: None. **S.R. Heilbronner:** None.

Poster

P925. Advances in Circuit Tracing Techniques

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Topic: I.03. Anatomical Methods

Support: NIH Grant DA045284

Title: Implementation of a Novel Intersectional Viral Tracing Technique to Characterize Ventral Tegmental Area Dopamine Projection Neurons of the Olfactory Tubercle

Authors: *R. C. PAULY[1], J. PARK[1], R. V. BHIMANI[2], C. E. BASS[3];
[1]Biotechnical and Clin. Lab. Sci., [2]Neurosci., [3]Pharmacol. and Toxicology, Univ. at Buffalo, Buffalo, NY

Abstract: Ventral Tegmental Area (VTA)-dopamine(DA) innervation of the ventral striatum, the nucleus accumbens (NAc), and olfactory tubercle (OT), is known to play a critical role in reward/aversion processes as well as drug addiction. While the majority of studies have focused on the anatomical connections between the VTA and NAc, very little is known about VTA-DA projections to the OT despite it receiving some of the densest DA in the brain. Furthermore, recent reports have highlighted that similar to the NAc, the OT is compartmentalized into different subdivisions that have distinct roles in reward learning and addiction. In this study, we integrated a novel intersectional viral targeting approach in wild-type rats to restrict Red Fluorescent Protein (RFP) transgene expression to VTA-DA-OT projection neurons to selectively determine the density and distribution of VTA-DA projection neurons to the medial (m) and lateral (l) OT. This approach has distinct advantages over traditional tracing techniques by allowing for selective determination of projections based on neuronal phenotype. Expression of effector virus infused in the VTA encoding RFP in a Double Inverted Open (DIO) reading frame is dependent upon co-administration of retrograde targeting virus infused in the OT encoding a Cre recombinase, floxing the RFP sequence after selectively transfecting VTA-DA cell bodies. Thus RFP expression is confined to VTA-DA-OT neurons quantitatively measured through immunohistochemistry. Viral selectivity was also confirmed through immunofluorescence verification of TH and RFP co-expression in the VTA. Our findings highlight dense VTA-DA innervation of the *m*OT relative to the *l*OT through quantitative

measurements of VTA projection neurons. Through anatomical and neurochemical evidence, our data display that dense VTA-DA innervation of the *m*OT plays a key role in encoding reward and addiction.

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Poster

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Program #/Poster #: P925.06

Topic: I.03. Anatomical Methods

Support: AMED (JP20bm0204001, JP19bm0204001 and JP18bk0104017)

Title: Collateral network of corticospinal neurons projecting to thoracic/lumbar spinal cord mapped by light-sheet microscopy and retrograde AAV vector

Authors: *T. LU[1], M. SHINOZAKI[1], N. NAGOSHI[2], M. NAKAMURA[2], H. OKANO[1];
[1]Dept. of Physiol., [2]Dept. of Orthopaedic Surgery, Keio Univ. Sch. of Med., Tokyo, Japan

Abstract: Thoracic spinal cord injury (SCI) in adult mice leads to damage of hindlimb function. Our previous studies have achieved certainly hindlimb function recovery in subacute phase of 10th thoracic (T10) SCI models by using induced pluripotent stem cell-derived neural stem/progenitor cells (iPSC-NS/PCs) therapy. However, neural circuits of the supraspinal neurons projecting to rostral and caudal sites of T10 level in intact rodents are still unclear, due to lacking effective neural tracing technology and global connectivity of brain-spinal inputs. Here, we found the distinct location of descending pathway (corticospinal tracts, rubrospinal tracts, and reticulospinal tracts) projecting to the 9th thoracic (T9) and lumbar enlargement (T13) in intact mice using retrograde AAV vector. Mapping of these neurons revealed the broader cortex area of corticospinal neurons (CSNs) projecting to T9 compared to T13. Interestingly, we noticed some double-labeled CSNs from T9 and T13, suggesting that they projected both to the lower thoracic spinal cord and to the lumbar enlargement. AAV2-Retro is known in targeting projection neurons through uptake at axonal terminals and retrograde transport of their payload to the cellular nuclei. However, little is known about axon collateral network of CSNs projecting to precise spinal segment. Using FLEX system, we revealed complex axon collateral network of labeled CSNs project to T9/T13 distributed in brain and spinal cord. Using tissue clearing and light-sheet microscopy we presented the global brain-spinal map of axon collaterals originating from these CSNs. Together, these findings revealed a new neuroanatomic insight in the collateral network of the descending pathway project to thoracic and lumbar enlargement, which would provide a new therapeutic targeting on functional recovery of hindlimbs after SCI.

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Poster

P925. Advances in Circuit Tracing Techniques

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Topic: I.03. Anatomical Methods

Support: NIH Grant R01NS083848
HHMI

Title: Monosynaptic anterograde circuit tracing with vesicular stomatitis virus (VSV) vectors

Authors: *X. MA[1], C. L. CEPKO[2];
[1]Harvard Med. Sch., Boston, MA; [2]Harvard Med. Sch/HHMI, Boston, MA

Abstract: A map of the connectivity patterns of the nervous system is fundamental to our understanding of the principles underlying normal function, as well as disorders. Previously, we developed the vesicular stomatitis virus (VSV) as a polysynaptic anterograde circuit tracer in a wide range of organisms (Beier et al., 2011; Mundell et al., 2015). For greater interpretability, we wished to develop monosynaptic anterograde VSV tracers (VSVdG). To achieve this, we have needed to overcome several challenges, including toxicity of the VSV glycoprotein (G), toxicity of viral replication, and innate immune system inhibition of VSV replication. First, to alleviate G protein toxicity, G is delivered by an AAV, and its expression is controlled by by Cre or Flp encoded by VSVdG. This prevents the fusion-active G protein from killing the AAV infected cells before trans-synaptic spread occurs. Second, we use a VSV vector encoding a mutant VSV matrix protein (Md) to avoid host cell transcription shut off, usually carried out by wild type M. Although this eliminates some of the toxicity of viral replication, the VSVdG-Md loses the ability to block the expression of interferons and several other cytokines, which inhibit VSV replication. Therefore, the third adaptation is to deliver a gene that encodes an inhibitor of cytokine signaling, encoded by another AAV, which is delivered at the same time as AAV-VSVG. Fourth, an immune-deficient strain of mice was found to be essential for efficient transsynaptic spread. Combining these four adaptations, the efficiency of this strategy was evaluated in the circuitry of the basal ganglia of mice. AAV and VSVdG were injected into the striatum, and VSVdG was observed to spread efficiently to a distant anterograde target, the substantia nigra (SN).

Disclosures: X. Ma: None. **C.L. Cepko:** None.

Poster

P925. Advances in Circuit Tracing Techniques

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Program #/Poster #: P925.08

Topic: I.03. Anatomical Methods

Title: Cell-type specific viruses transduce oxytocin and vasopressin neurons more effectively than ubiquitous promoters in mice and rats

Authors: *A. LEITHEAD[1], K. THIRTAMARA RAJAMANI[1], M. BARBIER[1], M. KIM[1], M. PERUGGIA[1], A. LEFEVRE[2], L. BARTECZKO[2], V. GRINEVICH[2], H. HARONY-NICOLAS[1]; [1]Icahn Sch. of Med. At Mount Sinai, New York, NY; [2]German Cancer Res. Ctr., Heidelberg, Germany

Abstract: Recent technological advances in viral vectors and the development of transgenic rodent models have allowed for targeted gene expression to study the functions of specific cell populations and brain circuits. Studies concerning the hypothalamic oxytocin (OXT) and arginine-vasopressin (AVP) systems have begun to utilize these advanced methods to further characterize their roles across a range of physiological and behavioral functions. However, the efficacy of widely used viral constructs has not yet been systematically evaluated in these neural populations. Here, we injected viral vectors of different promoter and serotype combinations into the paraventricular nucleus of the hypothalamus in rats and mice then performed immunohistochemistry to determine specificity. We found that (1) ubiquitous promoters were not, on their own, effective at driving expression of a down-stream fluorescent protein in OXT or AVP neurons, (2) the OXT promoter efficiently drove gene expression in OXT neurons and this efficiency was attributable to the promoter and not the viral serotype, and (3) using a Cre recombinase-dependent system, in combination with a virus that expresses Cre recombinase under the control of OXT promoter, significantly improved the efficiency of viral transduction in OXT neurons. Finally, we demonstrate the utility of the OXT promoter for conducting circuit tracing studies to identify inputs to OXT neurons in mice by using a novel modified rabies virus system with specificity to OXT neurons. We conclude that extreme caution is needed when employing non-neuron-specific viral approaches/promoters to study neural populations within the hypothalamus.

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Poster

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Topic: I.03. Anatomical Methods

Support: RF1MH123213

Title: Find partners: Transgenic tools for trans-synaptic tracing

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Abstract: Techniques for directional tracing of neural circuits will propel our understanding of the organization and function of the brain. We adapted trans-Tango, a powerful genetic approach for anterograde circuit tracing in Drosophila, to a vertebrate nervous system, that of zebrafish. trans-Tango involves a synthetic signaling pathway introduced into all neurons. Specificity comes from genetically defined neurons presenting a tethered ligand at the synapse. Binding of ligand to a G protein-coupled receptor activates the pathway in post-synaptic neurons, resulting in recruitment of an arrestin-protease fusion protein, site-specific proteolysis of the receptor, and release of the QF transcription factor allowing its translocation to the nucleus. There QF binds to upstream regulatory sequences (QUAS) to drive expression of genes such as fluorescent reporters, which label and identify post-synaptic cells. The trans-Tango components were modified for zebrafish by placing receptor and arrestin fusion protein constructs downstream of the elavl3 promoter in Tol2 transposition vectors. Ligand variants differing in their pre-synaptic targeting sequences were also cloned into Tol2 plasmids under control of the Gal4 transcription factor. To test functionality, plasmids encoding ligand, receptor and arrestin, as well as Tol2 transposase RNA, were co-injected into 1-cell stage embryos, the progeny of a Gal4 driver that promotes strong expression of Tg(UAS:GFP) in the hindbrain mated with the Tg(QUAS:mApple-CAAX) reporter. Confocal imaging revealed GFP labeled axon terminals closely apposed to mApple labeled neurons, suggesting that trans-Tango is effective in transient expression assays. With such assays, we determined the optimal ligand for activating the receptor in post-synaptic neurons without off-target labeling. We are currently characterizing trans-synaptic labeling in stable transgenic lines with verified expression of trans-Tango components. We are also using optogenetics and calcium imaging to confirm that identified connections are functional synapses and developing a related configuration for retrograde circuit tracing. This work lays the foundation for mapping neural connectivity in vivo for both invertebrate and vertebrate nervous systems.

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Poster

P925. Advances in Circuit Tracing Techniques

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Topic: I.03. Anatomical Methods

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IARPA MICrONS D16PC0008

Title: Axonal Barseq, Novel Technique for High-Throughput Mapping Single-Cell Projections in Situ

Authors: *L. YUAN, X. CHEN, A. M. ZADOR;
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Abstract: Neurons are heterogenous and assembled into complicated circuits in the brain. To understand the underlying structure of circuits, it is essential to understand the long-range connectivity of neurons. However, neuronal processes are thin and intertwined, which presents a technical challenge for reconstructing long-range axons optically for large numbers of neurons. We previously developed BARseq to map long-range projections with high throughput and cellular resolution (Chen et al, 2019, Cell, 179: 772-786). In BARseq, each neuron is labeled with a unique RNA barcode that replicates and fills up the neuron. Barcodes in somas are sequenced *in situ* and matched to barcodes in potential projection target areas to reveal the projections. The target areas, however, are resolved by manual dissection, thus limiting the ability of BARseq to resolve projections to fine divisions in brain areas. Here we overcome this limitation by axonal BARseq, which uses *in situ* sequencing to resolve the barcodes in the axons. As a proof of principle, we mapped the projections of thousands of neurons in 2 mm area of mouse primary auditory cortex. We identified major cell types using projection targets and axonal trajectory. Moreover, axonal BARseq identified two clusters of intratelencephalic neurons with different soma and axonal lamina patterns. Thus, axonal BARseq is a promising technique for high-throughput mapping of long-range projections at high spatial resolution and single cell resolution.

Disclosures: L. Yuan: None. **X. Chen:** None. **A.M. Zador:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Cajal Neuroscience.

Poster

P925. Advances in Circuit Tracing Techniques

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Topic: I.03. Anatomical Methods

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Title: A computational platform for extracting and reconstructing images of single neurons in 3D from multi-colour labelled populations in iDISCO cleared brains

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Abstract: A fundamental aim in biology is to describe the morphology and connections of individual cells within tissue ensembles. Multi-reporter labelling technologies, such as Brainbow, apply stochastic expression of several fluorescent proteins to allow visualization of cell populations with single-cell resolution. However, extending this approach to generating objective quantification of anatomical features is more difficult and has to a large extent awaited the development of analysis platforms capable of extracting images of single cells in 3D. The main challenge of Brainbow method is the currently unavoidable variation in recorded fluorescence intensity (‘pseudo-colour variability’) within individual neurons making it challenging to trace and reconstruct individual cells with currently available tools. We present a computational platform - BRIAN (BraInbow Analysis of individual Neurons) that allows objective identification of voxels clusters, and their subsequent extraction to produce 3D images of single neurons. We show that BRIAN allows multi-reporter samples to be analysed using existing tracing technology (Filament Tracer, Imaris), to efficiently recreate single neurons. We show the successful application of BRIAN to retinal tissue by reconstructing individual cells (intrinsically photosensitive retinal ganglion cells, ipRGCs, and depolarizing ON bipolar cells) in densely labelled tissue with sufficient anatomical resolution for sub-type classification. Moreover, we demonstrate the successful application of BRIAN in Brainbow labelled brain

tissue to describe dendritic projections in the suprachiasmatic nucleus (SCN). Finally, we combine Brainbow and iDISCO tissue-clearing with immunohistochemistry to image large volumes of a mouse brain (1/4 mm^3), and again demonstrate successful application of BRIAN to trace individual axonal projections over large brain volumes.

Disclosures: N. Milosavljevic: None. **C.A. Procyk:** None. **J. Rogers:** None. **E. Zindy:** None. **R.J. Lucas:** None.

Poster

P925. Advances in Circuit Tracing Techniques

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Topic: I.03. Anatomical Methods

Support: F32DK123879
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Title: Whole brain imaging of neuronal projections using viral-mediated transfection and HaloTag bioorthogonal labeling

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Abstract: Axonal projections of specific neuronal populations define the anatomical and functional characteristics of a circuit. Typically, confocal microscopy of thin tissue sections has been used to visualize terminal fields. However, this approach only allows for a handful of selected tissue sections to be analyzed. Imaging axonal projections in whole brains using tissue clearing and light sheet microscopy would allow for better spatial and temporal resolution of intact neural circuits. The use of genetically encoded fluorescent proteins to label cell bodies in cleared brains exhibits bright labeling and excellent signal to noise ratios. However, capturing synaptic terminals is significantly more challenging. Using synthetic dyes that are brighter and more resistant to photobleaching compared to traditional fluorophores may offer a solution. Thus, we decided to use a bio-orthogonal approach to label regionally and genetically restricted synaptic terminals using click chemistry. To do this, HaloTag, a self-labeling protein, was tagged to synaptophysin and loaded into a viral construct (AAVDJ.hSyn.DIO.Synaptophysin:HaloTag). The bed nucleus of the stria terminalis (BST) was targeted in Corticotropin Releasing Factor (CRF)-Cre or Melanocortin 3 Receptor (MC3R)-Cre mice. The synthetic dyes, JF669-HTL or

JF635-HTL dye were dissolved in paraformaldehyde solution used for perfusion (669 and 635) or retro-orbitally injected (669) the day prior to tissue collection. These dyes readily labeled synaptophysin-HaloTag in specific brain regions via both delivery methods, and axonal projections were easily identified even at low magnification in whole brain tissue. As expected, prominent synaptic terminal labeling was observed in several distinct regions of the amygdala and hypothalamus that are known to be targeted by CRF and MC3R BST neuronal populations. Altogether, we demonstrate the utility of these tools and this approach to visualize axonal projections of relevant cell populations in an unbiased manner in intact, whole brain tissue.

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Poster

P954. Network Models: Experimentation I

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Program #/Poster #: P954.01

Topic: I.06. Computation, Modeling, and Simulation

Support: NIH Diversity Supplement Grant 3R44AG047722-05S1
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Title: Modular organization of the Medial-Frontal Parietal Network in cognitively normal older adults and its relationship with episodic memory

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Abstract: Normal aging entails highly variable cognitive, structural, and functional changes that could lead to pathological decline, such as that caused by Alzheimer's Disease. Episodic memory (EM), a cognitive function that critically depends on hippocampal-cortical interactions, is especially vulnerable to brain aging. The hippocampus is considered part of the Medial-Frontal Parietal Network (M-FPN, also known as the Default Mode Network), which is a functional brain network not only associated with EM processes, but also known to show functional connectivity (fc) alterations in normal and pathological aging as shown by resting state functional connectivity (rs-fc) magnetic resonance imaging. Leading neuroimaging frameworks for the M-FPN propose it to be comprised of three functionally distinct subnetworks. The *Schaefer parcellation* (Schaefer et al., 2018), further divides these three subnetworks into a set of

non-overlapping sections or parcels. However, because the M-FPN displays heterogeneous aging effects, whether the parcellations remain in the same functional groupings in cognitively normal older adults is not well characterized. The aim of this research is to understand the functional organization of the M-FPN in normal aging through modularity, a graph theory metric that can determine how regions or parcellations of a network are segregated into non-overlapping groups or modules. Specifically, we first developed data-driven modules (using the Schaefer parcels of the M-FPN) derived from rs-fc of cognitively normal older adults (OA; N=148, mean age=66.9±5.8) and used the normalized variation of information (VIn) as a metric of module dissimilarity. Results revealed a significant difference between the M-FPN modular organization of individuals from the OA sample and the Schaefer M-FPN parcellation (VIn mean=0.463±0.04, $t(147)=126$, $p<0.001$), suggesting normal aging influences the modular structure of the M-FPN. However, there were also significant differences between the M-FPN modular organization of individuals and that of the group average. Follow-up analyses will test the stability of the modular organization of the M-FPN in an independent OA group (N=71, mean age=71.5±4.7) and test whether individual differences in modular structure are related to EM performance. This project helps characterize the organization and stability of M-FPN subnetworks and their parcellations, as well as the relevance to EM, in cognitively normal older adults.

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Poster

P954. Network Models: Experimentation I

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Program #/Poster #: P954.02

Topic: I.06. Computation, Modeling, and Simulation

Support: National Institute of Neurological Disorders and Stroke. NS095094-01A1
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Title: A control-theoretical system for modulating hippocampal gamma oscillations using stimulation of the posterior cingulate cortex

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Abstract: Closed-loop stimulation for targeted modulation of brain signals has emerged as a promising strategy for episodic memory restoration. In parallel, closed-loop neuromodulation strategies have been applied to treat brain conditions including drug-resistant depression, Parkinson's Disease, and epilepsy. In this study, we seek to apply control theoretical principles to achieve closed loop modulation of hippocampal oscillatory activity. We focus on hippocampal gamma power, a signal with an established association for episodic memory processing, which may be a promising 'biomarker' for the modulation of memory performance. To develop a closed-loop stimulation paradigm that effectively modulates hippocampal gamma power, we use a novel data-set in which open-loop stimulation was applied to the posterior cingulate cortex and hippocampal gamma power was recorded during the encoding of episodic memories. The dataset was used to design and evaluate a linear quadratic integral (LQI) servo-controller in order to determine its viability for in-vivo use. In our testbed, we demonstrate that applying an LQI servo controller based on an autoregressive with exogenous input (ARX) plant model achieves effective control of hippocampal gamma power in 15 out of 18 experimental subjects who exhibited a gamma power increase related to stimulation. In contrast, we also implement a linear quadratic regulator (LQR) controller based on a linear state-space model (LSSM) which utilizes multi-channel gamma activities from the posterior cingulate cortex (PCC). We demonstrate that we are able to modulate gamma power using stimulation thresholds that are physiologically safe and on time scales that are reasonable for application in a clinical system, along with the ability to achieve temporal modulation of the control signal during item memory encoding. We outline further experimentation to test our proposed system and compare our findings to emerging closed-loop neuromodulation strategies.

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Poster

P954. Network Models: Experimentation I

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Title: Machine learning approach to predict EEG sleep stages and transitions

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Abstract: Motivation: Conventional sleep staging is interpreted by sleep technologists with visual inspection, which is a time-consuming task. Therefore, experts have tried to develop a perfect automatic sleep staging system, but currently, none of them can meet the clinical use standards set by the American Academy of Sleep Medicine (AASM). The transition epoch is defined as a epoch of 30 seconds of EEG signal in which there are multiple sleep staging features such as arousing, which is more difficult to be staged accurately by machine learning in the past, and although the clinical sleep technologist can make judgments according to the relevant criteria set by AASM, different results may be obtained between sleep technologists, resulting in lower inter-rater reliability. Therefore, the goal of this research is to develop an automated Electroencephalography (EEG) sleep staging algorithm to improve the overall sleep staging and transition accuracy by using the anterior and posterior EEG signals to assist in judgment. **Methods:**This research used 8444 Polysomnography (PSG) data provided by the Sleep Heart Health Study, with a mean age of 66.2 years, including healthy subjects and Obstructive sleep apnea patients. We use C3 and C4 two EEG channels in the 10-20 system, with a sampling rate of 125 Hz. Since the EEG frequency is lower during sleep, we only use data with a frequency of 0.5~30 Hz. All data were preprocessed into sleep stages as defined by the AASM, and three 30-second EEG signals were grouped together as one set and Fourier transformed to calculate a total of 90 seconds of features. By using the first and last EEG signals, we can predict the sleep staging of the middle epoch more accurately during the training phase of the machine learning, and use the first two epochs of sleep staging prediction results to assist in determining the staging of the third epoch of the signal in a time-ordered manner. **Results:**Since N1 only accounts for 5.7% of the total sleep time, and the probability of continuous N1 after Markov chain analysis is only 51.53%, which is much lower than that of other stages, we use downsampling to achieve a balanced data. At present, the accuracy rate can reach 67.3% for one continuous set of the same stage. **Conclusion:**The current research has achieved good accuracy, which shows that the use of longer time data such as 90 seconds can effectively improve the model interpretation. Therefore, it would be a prospective research if more different preprocessing or other methods of extracting features could be done in the future.

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Poster

P954. Network Models: Experimentation I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P954.04

Topic: I.06. Computation, Modeling, and Simulation

Support: JSPS KAKENHI Grant Number JP19K12145

Title: Estimating the degree of a driver's distraction based on functional connectivity analysis: An fNIRS study

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Abstract: Objective
Distracted driving is a significant cause of traffic injuries. One cause of distracted driving is mind-wandering, shifting the driver’s thoughts away from what they should concentrate on. The objective of our study is to predict the degree of the driver's distraction based on brain activity. We measured the driver's brain activity during real-vehicle driving using functional near-infrared spectroscopy (fNIRS). Moreover, distracted driving behavior was predicted from preceding brain activity using connectome-based predictive modeling.
Participants and Materials
Twelve young male adults (aged 23.4 +/- 1.3 years) participated in this study. They were asked to drive a compact single-seat electric vehicle along an oval track course while preserving the vehicle speed to 20 km/h for 15 minutes. Beep tones were given randomly with intervals of 20-40 s. The participants were asked to respond to the beep tones immediately by stepping on the braking pedal until the vehicle speed was decreased to less than 10 km/h. fNIRS (NIRSport2, NIRx Medical Technologies, LLC.) with 16 sources x 14 detector probes and 16 short distance detector probes was used to measure OxyHb concentration changes during driving. Driving behavioral measures such as braking/accelerator pedal positions and vehicle speed were recorded and synchronized with fNIRS data using the Lab Streaming Layer library (https://labstreaminglayer.readthedocs.io).
Methods
The functional connectivity matrix was calculated using the OxyHb time series of a defined interval before presenting each beep tone. The brake reaction time (BRT) was calculated as the time between when the beep tone was presented until the driver started to step on the braking pedal, and used as the behavioral measure of distracted driving. We constructed a regression model where the BRT was used as an objective variable and a vectorized functional connectivity matrix was utilized as an explanatory variable. The model was built for each participant, and its accuracy was evaluated.
Results
We found positive correlations between predicted and actual BRTs for 6 out of 12 participants (p < 0.05). Discriminative connections were extracted for each model but no common connections could be identified for all the models. This result indicates that each participant had a different pattern of functional connectivity associated with his distracted state. These results suggest that distracted driving can be predicted from brain activity patterns for each individual.

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Poster

P954. Network Models: Experimentation I

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Program #/Poster #: P954.05

Topic: I.06. Computation, Modeling, and Simulation

Support: M37 MH066078
NSF NCS-FO 1835209

Title: Precision Individualized Brain Models Directly Estimated From M/EEG

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Abstract: Current efforts in large-scale neuroscience have led to the collection of sophisticated datasets measuring single-subject brains and behavior from multiple points of inquiry. However, integrating these facets into a concise description of individual brains has proven vexing. These efforts have been particularly stymied in two fronts: describing brain mechanisms across multiple spatial/temporal scales. Unifying these relationships in a formal modeling framework remains a key challenge for neuroscience and particularly human neuroimaging. Generating unified descriptions requires the development of brain models which can interrogate multiple scales of brain activity. However, such needs have not yet been met due to the challenges of high-dimensional optimization and the necessary tradeoffs between spatial (fMRI) and temporal (M/EEG) resolution for high-coverage methods. In the current work we present a new technique for estimating detailed brain (neural-mass) models using single-subject MEG data from the Human Connectome Project (HCP minimal pre-processing) and headmodels derived from MRI data. We estimated neural mass-style models directly from each subject's MEG sensor-level timeseries using a novel optimization algorithm: the Backpropagated Kalman Filter. This technique enables the parameterization of multiple neural populations (inhibitory and excitatory cells) and their connections across the cortical surface (100 parcels) directly from single-subject data. We demonstrate that these models are highly reliable in estimating both excitatory-to-excitatory and excitatory-to-inhibitory long-distance connections for each subject. Models accurately forecast future brain activity timeseries and predict long-term statistics such as correlation patterns and spectral power.

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Poster

P954. Network Models: Experimentation I

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Program #/Poster #: P954.06

Topic: I.06. Computation, Modeling, and Simulation

Support: ERC-2018-SyG
ONR-N00014-18-1-2633
ONR-N00014-20-1-2074
FNS n°100014_178814-1

Title: Investigating the reliability of connectome-based predictive models across fMRI runs and datasets

Authors: *F. JOËSSEL[1], M. D. ROSENBERG[2], D. BAVELIER[1];
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Abstract: There is growing interest in building models that predict cognitive performance from functional connectivity (FC) data measured with fMRI. Although such connectome-based models can achieve high prediction accuracy and even generalize across independent participants and datasets [1-3], recent work reports that the reliability of feature weights is often low [4,5]. In addition, although large datasets of fMRI data are becoming increasingly common, constraints inherent to MRI data acquisition limit sample sizes for many experiments. We thus aimed to investigate how the number of participants and amount of data available per participant in single-lab samples impact the accuracy of connectome-based models and to characterize the feature-weight reliability of these models.
To this end, we analyzed data from four datasets (*N*s= 25, 62, 25, 49) collected at two scan sites. In each dataset, participants performed multiple runs of a sustained attention task, the gradual-onset continuous performance task (gradCPT). We built connectome-based predictive models (CPMs) [6] using leave-two-out cross-validation to predict participants' gradCPT performance from FC measured during the gradCPT. Specifically, in each dataset, we trained models using data from subsets of participants and task runs, and asked whether they generalized to predict gradCPT performance in novel participants and datasets. We also assessed the feature-weight similarity of models trained in the same and different datasets.
Our results show inconsistent accuracy of models trained on different datasets and different runs from the same dataset. For example, when models were trained on N=20, the mean correlation between predicted and observed behavioral scores was r=.78, 0.29, 0.01, and 0.11 in the four datasets, respectively . Looking at session-to-session variability in one dataset, the predictive accuracy for was r=.16 for one run and r=.4 for another. In addition, the correlation between the feature weights of models trained on different datasets does not exceed 0.3, even when models trained on one dataset successfully generalize to the other. The similarity of feature weights between CPMs trained on different runs in the same dataset range from .4-.7. These results suggest that CPMs can generalize across participants and datasets despite variable feature

weights, and compel further investigation into the relationships between sample size, amount of data per individual, prediction accuracy, and feature-weight reliability.
[1] Lee et al. (2021)
[2] Rosenberg et al. (2016)
[3] Kucyi et al. (2021)
[4] Marek et al. (2021)
[5] Tian et al. (2020)
[6] Shen et al. (2017)

Disclosures: F. Joëssel: None. **M.D. Rosenberg:** None. **D. Bavelier:** None.

Poster

P954. Network Models: Experimentation I

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Program #/Poster #: P954.07

Topic: I.06. Computation, Modeling, and Simulation

Support: NIDCD-R01DC012947-06A1
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ARO-URAP supplement
NIH U24EB028998
NSF 1904444-1042C

Title: Biophysically-detailed multiscale model of macaque auditory thalamocortical circuits

Authors: *E. GRIFFITH[1], S. DURA-BERNAL[2], A. BARCZAK[3], M. N. O'CONNELL[3], T. M. MCGINNIS[4], W. W. LYTTON[6], P. LAKATOS[5], S. A. NEYMOTIN[7];
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Abstract: We used the NEURON simulator with NetPyNE to develop a biophysically-detailed model of the macaque auditory thalamocortical system, simulating a cortical column of 2000um depth and 200um diameter, with over 12k neurons and 30M synapses. Using this model, we reproduced oscillatory patterns observed in spontaneous electrophysiology data recorded from awake, nonhuman primate (NHP) primary auditory cortex (A1). Neuron classes, densities, biophysics, and connectivity were derived from experimental data. The A1 model included 6 cortical layers, 36 excitatory and inhibitory neuron populations (e.g. intratelencephalic cells in L2-6, neurogliaform cells in L1), and layer-specific reciprocal connectivity between A1 and medial geniculate body (MGB). The thalamic model included the MGB and the thalamic

reticular nucleus (TRN). The MGB contained locally-projecting thalamic interneurons, along with core and matrix populations of thalamocortical neurons, each with distinct projection patterns to different layers of A1. TRN included thalamic reticular neurons to inhibit MGB. Outputs from cochlear nerve and inferior colliculus (IC) were simulated using phenomenological models that captured the physiological transformations occurring in these structures in response to auditory stimuli. This allowed us to use any arbitrary sound as input to the model, including the stimuli presented experimentally to awake macaques, facilitating our ability to match model to primate.
We used evolutionary algorithms to tune several high-level network parameters (e.g. synaptic weight gains) in order to generate experimentally constrained firing rates in all 43 neuron populations. Each simulated second required approximately 15 minutes on 96 supercomputer cores. We ran 100 simultaneous simulations (9600 cores) for each generation of the evolutionary optimization.
We validated the model by comparing in vivo and in silico electrophysiology data using local field potential, current source density (CSD), and direct comparisons of individual cortical oscillation events. We found a similar distribution of oscillation events in different frequency bands and favorable comparisons between the shape of the individual oscillation event waveforms occurring within these frequency bands. Preliminary comparisons between model and NHP data in response to click and speech stimuli have also begun. Using CSD and network model manipulations, we will develop predictions regarding the mechanistic origins of these spatiotemporal oscillatory patterns, and test these predictions in vivo using deep brain electrical microstimulation and pharmacological manipulations.

Disclosures: E. Griffith: None. **S. Dura-Bernal:** None. **A. Barczak:** None. **M.N. O'Connell:** None. **T.M. McGinnis:** None. **W.W. Lytton:** None. **P. Lakatos:** None. **S.A. Neymotin:** None.

Poster

P969. Data Analysis and Statistics: Human Data I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P969.01

Topic: I.07. Data Analysis and Statistics

Support: NRF-2018R1A2B2005687
IITP-No. 2017-0-00451
"HPC Support" Project, supported by the 'Ministry of Science and ICT' and NIPA.
'Artificial intelligence industrial convergence cluster development project' funded by the Ministry of Science and ICT(MSIT, Korea)&Gwangju Metropolitan City.

Title: Towards Intuitive BCI - Feasibility Study of Decoding Visual Imagery EEG using Deep Learning

Authors: *S. LEE, S. JANG, S. C. JUN;
Sch. of Electrical Engin. and Computer Sci. (EECS), Gwangju Inst. of Sci. and Technol., Gwangju, Korea, Republic of

Abstract: In brain computer interface (BCI) systems, motor imagery (MI) has the advantage in that the user can control right or left movements (two degrees of freedom) actively. On the other hand, studies on decoding visual imaginary EEG data have been recently more attention due to the development of deep learning. The BCI through visual imagery has great potential in increasing the degree of freedom of manipulation. In this work, we analyzed the visual imagery EEG using the deep learning and explored the possibility that it can be useful for BCI. EEG data were acquired by 32-channel BioSemi Active 2. Twenty healthy male and female university students participated in the experiment. Each participant performed up to 10 sessions. Three categories (objects, digits, shapes) of visual images were presented for 2 seconds and participants were instructed to imagine the presented image for 4 seconds. One session consisted of 5 runs and each run consisted of 36 trials. A total of 180 visual imagery trials was collected in one session. EEG data were filtered using a 0.5-50Hz bandpass filter, and artifacts such as eye blinking were removed through the Independent Component Analysis (ICA) technique. Time frequency map for time window of 3 seconds long (1 second before onset and 2 seconds after imagery) was calculated within 0~30Hz, and then this time-frequency map was used as an input for deep learning. The pretrained MobileNet V2 model was used for classification, and the classification accuracy was calculated through 5-fold cross validation. GradCam, one of the explainable AI techniques, was used to extract features from each category. We found the location of major activations in each category by averaging over trials of each category. The averaged classification accuracy (three categories and 5-fold) was 57.2%. As a result of the GradCam, it was found that the activation was notably high at 10-20Hz between 0-1 seconds after the imagery onset. This trend was similar in all three categories, but some difference in activation among categories was observed in 10-13 Hz from 500 ms after onset. In particular, in the comparison of object and digit categories, there was relatively big difference in activation at low beta (about 20-25Hz) after 1 second of imagery onset. We performed visual imagery EEG experiments on three categories. Through deep learning, 57.2% of classification accuracy was obtained. In addition, activation in the alpha wave during imagery was observed as a notable feature through an explainable AI technique. From this work, we believe that visual imagery may have potential in BCI system, which is a more intuitive BCI paradigm.

Disclosures: S. Lee: None. **S. Jang:** None. **S.C. Jun:** None.

Poster

P969. Data Analysis and Statistics: Human Data I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P969.02

Topic: I.07. Data Analysis and Statistics

Support: NIH ZIA-MH002781
NIH ZIA-MH002782

Title: Effects of preprocessing on connectome fingerprinting in a sample of anxious and healthy youth

Authors: *G. RINGLEIN, A. WINKLER, D. PINE, A. ZUGMAN;
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Abstract: Resting state fMRI connectivity has been found stable and sufficiently unique to identify subjects between sessions, a process known as connectome fingerprinting (CF). While simple ("full") correlation between regions is commonly used, partial correlations that take into account all other regions allow the study of direct connections. Motion correction is another area of ambiguity; there are many available methods, each with unknown effects on CF; regression of motion parameters ("6RP") and ICA-AROMA are popular approaches. Here we investigate how prediction accuracy is affected by type of correlation (full vs. partial) and motion correction (6RP vs. ICA-AROMA). Participants (n=84) were subjects with diagnosis of anxiety (n=56; F/M/unknown=33/22/1; age (mean±sd)=13.9±2.8) and controls (n=28; F/M=15/13; age =13.7±2.4). Each subject had two resting state fMRI scans (days between scans =126±42). Groups did not differ significantly in sex, age, or time between scans (p &gt 0.05). Functional connectivity was assessed between each pair of the regions from the Schaefer atlas (200 parcels) and 16 subcortical regions from FreeSurfer. We attempted to identify each individual by comparing their connectivity matrix at one time point with all individuals' matrices at the other time point. Thus, each preprocessing choice led to two accuracies: baseline to follow-up (b-f) and follow-up to baseline (f-b), for the 4 combinations of type of correlation and motion correction, producing 8 accuracies. For all analyses, accuracy was significantly better than chance.Results were as follows: partial correlation using motion regression (6RP) was the most accurate (b-f: 63 correct subjects, f-b: 64), followed by full correlation with ICA-AROMA (b-f: 55, f-b: 52), partial correlation with ICA-AROMA (b-f: 42, f-b: 46), and partial correlation with ICA-AROMA (b-f: 22, f-b: 23). In the full correlation analyses, there were significantly higher means of motion among the incorrect participants across both scans, except in f-b using 6RP motion regression. There was no significant association between motion when using partial correlations. We also computed the differential power (DP) of each edge, a measure of how much that edge contributes in identifying subjects. The edges with highest DP were inconsistent between the 4 analyses, with only 7 edges being in the top 5% for all analyses. These edges involve regions in the control network, default mode network, and temporal parietal network. The variability in accuracy and DP of edges between different kinds of analysis highlights the need for careful consideration of preprocessing steps before CF analysis.

Disclosures: G. Ringlein: None. **A. Winkler:** None. **D. Pine:** None. **A. Zugman:** None.

Poster

P969. Data Analysis and Statistics: Human Data I

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Program #/Poster #: P969.03

Topic: I.07. Data Analysis and Statistics

Support: NIMH T32-MH115866
NIH UL1TR000114
NIDA P30 DA048742-01A1

Title: Functional hubs in the executive and attentional resting-state networks as revealed with causal discovery directed connectivity analysis

Authors: *E. RAWLS[1], E. KUMMERFELD[2], B. A. MUELLER[1], A. ZILVERSTAND[3]; [1]Psychiatry and Behavioral Sci., [2]Inst. for Hlth. Informatics, [3]Psychiatry, Univ. of Minnesota, Minneapolis, MN

Abstract: Background: Brain network analysis has paid particular attention to so-called "hubs", highly-connected brain regions that facilitate efficient information transfer. Previous evidence for functional and anatomical hubs of the brain has been restricted to undirected networks, despite increasing recognition that understanding causal interactions between brain regions is crucial to the development of targeted interventions for psychiatric populations. Here, we introduce a scalable method for calculating directed connectivity from fMRI: GANGO (Greedy Adjacencies and Non-Gaussian Orientations), and use this to investigate the causal hubs of the human cortex. Methods: Using a large resting-state dataset (Human Connectome Project, n = 442 unrelated subjects; 56% female), we generated sparse whole-cortex directed connectivity graphs on a per-subject basis using the GANGO method. GANGO initially defines directly connected brain regions using a scalable greedy search that maximizes model fit (Bayesian Information Criteria), then orients edges using non-Gaussianities in the BOLD signal distribution. To summarize the directed connectivity structure of the generated graphs, we characterized the hub structure of these graphs using analyses of inter-module connectivity (participation coefficient), centrality metrics (degree centrality, betweenness centrality), and virtual lesion analyses. Results: Across a variety of analyses, our directed connectivity networks revealed causal hub roles of frontoparietal, dorsal attention, and cingulo-opercular networks. Inter-network connectivity analyses clarified the functional roles of these networks: the dorsal attention network derived its high centrality from receiving information from sensory networks and sending information to cognitive networks, while the frontoparietal network derived its high centrality from facilitating communication between higher cognitive networks (cingulo-opercular, default mode, dorsal attention, ventral attention). Conclusion: Here, we present a method for generating a sparse "causal brain connectome" that highlights the roles of attentional and executive networks as critical causal hubs for brain network integration. In the future, this may guide the development of targeted brain-based interventions for psychiatric conditions directed at these causal hubs.

Disclosures: E. Rawls: None. **E. Kummerfeld:** None. **B.A. Mueller:** None. **A. Zilverstand:** None.

Poster

P969. Data Analysis and Statistics: Human Data I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 8:45 AM - 9:45 AM

Program #/Poster #: P969.04

Topic: I.07. Data Analysis and Statistics

Title: Canonical Spectral Responses: Using large-scale group analysis of ECoG electrode spectral profiles to investigate the spectral organization of the brain.

Authors: *Z. FREUDENBURG[1], M. P. BRANCO[2], M. VANSTEENSEL[3], E. J. AARNOUTSE[4], N. F. RAMSEY[5];
[1]UMC Brain Center, Univ. Med. Center, Utrecht, Utrecht, Netherlands; [2]UMC Utrecht Brain Ctr., [3]UMC Brain Center, Univ. Med. Ctr. Utrecht, Utrecht, Netherlands; [4]Neurol. and Neurosurg., [5]UMC Brain Center,University Med. Ctr. Utrecht, Utrecht, Netherlands

Abstract: Neuroscience has generated varied maps of the human brain ranging from historic atlases based on cortical tissue composition [Brodmann 1909] to atlases that integrate structural and functional data, such as Human Connectome Project [Glasser et al. 2016]. Spectral features in human brain electrophysiology have been extensively studied and have been shown to be universal to human brain function to the extent that they are often used in clinical diagnostic practice and online closed-loop direct cognitive control of a computer. Yet, a spectral atlas of the brain has not been established. A spectral atlas can reveal brain region-specific fluctuations in power across frequencies, induced by changes in activity during task performance. Mu frequencies in motor cortex for instance (around 10 Hz), are known to selectively diminish in power during movement. Frequency-specific fluctuations as such indicate region-specific preferences for specific frequencies, possibly revealing neurophysiological mechanisms. , Scalp EEG recordings, the main modality used to study spectral features, are of limited use for spectral mapping due to limited sensitivity and spatial selectivity. Electrocorticography (ECoG), does allow for mapping of electrophysiological signals with high topographical precision, since each electrode measures only from cortical tissue in direct contact. ECoG has the added benefit of being sensitive to frequencies (50-300 Hz) closely linked to increased neuronal spiking and fMRI BOLD signal increases [Nir et al. 2007, Goense et al. 2008]. Due to the clinical setting in which ECoG data is collected, however, the cortical coverage in individual subjects is partial and varied. A spectral atlas thus requires data from a large number of patients, and techniques that can integrate results across subjects. In this work we leverage a large data set (57 subjects, 4418 total electrodes) and use clustering to group electrodes according to spectral profiles. ECoG was recorded with standard clinical grids during epileptic focus localization. Each electrode was

anatomically localized using MRI and CT scans [Branco et al. 2016], assigned a a spectral variance profile using irregular-resampling auto-spectral analysis [Wen and Liu 2016], and. then clustered using Affinity Propagation [Frey et al. 2007] into 9 groups of spectrally similar electrodes. While a recent paper [Wanga et al. 2021] used il-EEG to determine spectral peaks for anatomically defined regions, to our knowledge it is novel to uses spectral content to map cortical areas. This atlas can help to better interpret EEG recordings and be used to associate localized spectral information with other modalities.

Disclosures: Z. Freudenburg: None. **M.P. Branco:** None. **M. Vansteensel:** None. **E.J. Aarnoutse:** None. **N.F. Ramsey:** None.

Poster

P969. Data Analysis and Statistics: Human Data I

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Program #/Poster #: P969.05

Topic: I.07. Data Analysis and Statistics

Support: NIMH Grant F30MH122136
NIMH Grant K01MH107763
NIMH Grant R01MH120293
NIGMS Grant T32GM008444

Title: Identification and characterization of a simultaneous multi-slice (multiband) fMRI signal artifact in simultaneously acquired slices

Authors: *P. N. TUBIOLO[1,3], J. C. WILLIAMS[1,3], J. R. LUCENO[3], J. X. VAN SNELLENBERG[1,2,3];
[1]Biomed. Engin., [2]Psychology, Stony Brook Univ., Stony Brook, NY; [3]Psychiatry, Renaissance Sch. of Med. at Stony Brook Univ., Stony Brook, NY

Abstract: Multiband (MB) functional Magnetic Resonance Imaging (fMRI) achieves improved spatiotemporal resolution in clinical neuroimaging and cognitive neuroscience research, and is used in public datasets such as the Human Connectome Project (HCP). However, MB fMRI datasets may exhibit novel characteristics absent in legacy datasets. The aims of this study are to: (a) report the existence of a previously uncharacterized signal artifact across all MB fMRI datasets examined; (b) propose a regression-based technique to remove this artifact; and (c) provide a preliminary analysis of the isolated artifact. Unprocessed MB RS-fMRI data was utilized from a Siemens Prisma 3T scanner (n = 10, 4 runs/subject, MB acceleration factor (MBF) = 6, Repetition Time (TR) = 800 ms; one phantom run with TR = 1010 ms was analyzed separately), GE MR750 3T scanner (n = 10, 4 runs/subject, MBF= 6, TR = 850 ms), and the HCP 500 subjects release (n = 25, 4 runs/subject, Siemens Skyra 3T scanner, MBF= 8, TR = 720

ms). A shared artifact across simultaneously-acquired (SA+) slices was quantified as the difference in Pearson correlation between the average timeseries in each slice for SA+ and non-simultaneously acquired (SA-) slices, averaged over all slices in a volume. The isolated artifact time series was estimated separately for each slice as the residuals of a multiple linear regression modeling the average signal in each set of SA+ slices, but orthogonal to the global signal across SA- slices and estimated motion parameters. The original voxel-wise timeseries was then orthogonalized with respect to the estimated artifact signal for the corresponding slice to obtain corrected data. In addition, the estimated artifact timeseries was used to determine autocorrelation as a function of lag number. For each dataset, the power spectral density (PSD) was determined and colored noise characteristics were estimated using a linear regression of log-log transformed PSD with a lower bound of -2 log(Hz). Significantly elevated correlations between SA+ slices over SA- slices were observed in all datasets, indicating a shared signal source between SA+ slices (paired t-tests, all $p < 10^{-5}$). Correlations between SA+ slices were reduced after artifact removal in all datasets (paired t-tests, all $p < 10^{-5}$). Artifact signal was present in phantom data, suggesting a non-neural source. Power spectral analyses and autocorrelation calculations suggest that the artifact is largely composed of colored noise with a PSD proportional to frequency$^{-\alpha}$ where $1 \leq \alpha \leq 2$. We expect that removal of this artifact will improve neural activity estimates in MB fMRI studies.

Disclosures: P.N. Tubiolo: None. **J.C. Williams:** None. **J.R. Luceno:** None. **J.X. Van Snellenberg:** None.

Poster

P969. Data Analysis and Statistics: Human Data I

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Program #/Poster #: P969.06

Topic: I.07. Data Analysis and Statistics

Support: Fondecyt 1210069

Title: Detection of Gaussianity using envelopes

Authors: *V. M. HIDALGO[1], J. MPODOZIS[1], J. C. LETELIER[1], J. DIAZ[2];
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Abstract: Assessing Gaussianity is a classic problem in the field of signal analysis, and a variety of methods have been devised to infer Gaussianity in time series. These methods span ideas both from the time and frequency domains. Time domain approaches rely on goodness-of-fit tests to assess experimental amplitude distributions, while frequency domain methods are spectral-based tests. In general, these two classes of methods present different drawbacks and in practice are

difficult to use to settle the question of Gaussianity in time series. Here we introduce a different solution by using the envelope of a signal. In effect, the envelope of zero-mean Gaussian noise is distributed according to the Rayleigh distribution, which has the special property that the ratio between its standard deviation (σ) and mean (μ), known as the coefficient of variation (CV=σ/μ), is constant and equal to $\sqrt{(4-\pi)/\pi} \approx 0.523$ (Diaz et al. 2007, DOI:10.1523/JNEUROSCI.4512-06.2007). Thus, when the envelope of a given signal is considered as a random variable, its CV (called the coefficient ofvariation of the envelope, CVE) can be used as a fingerprint of Gaussianity. Previously, we have used this tool to study the Gaussianity of a) the peripheral waves in the vertebrate olfactory epithelium (Diaz et al. 2007, DOI:10.1523/JNEUROSCI.4512-06.2007), b) the rat EEG during sleep (Diaz etal,DOI:10.1016/j.neuroimage.2018.01.063), and c) the human alpha rhythm (Hidalgo et al. 2021,DOI:10.1101/2021.03.31.437785). Here we summarize the mathematics and computer methods behind using the CVE as a fingerprint of Gaussianity. In particular, we present how to perform Gaussianity detection in a step-by-step fashion for experimental data, paying attention to the use of the Hilbert transform, and how this procedure can be employed to classify behavioral states. Furthermore, we use our method to assess the degree of coupling between coupled oscillators in the Kuramoto model. Additionally, through the use of the Fourier transform phase randomization, we reveal the interconnections among CVE, Gaussianity and signal amplitude modulation profiles. Moreover, the CVE method allows the similarities and differences between neural signals (M/EEG, LFP, and fMRI) and Gaussian noise to be studied within a solid mathematical framework that could also be used to study neural synchronization.

Disclosures: V.M. Hidalgo: None. **J. Mpodozis:** None. **J.C. Letelier:** None. **J. Diaz:** None.

Poster

P969. Data Analysis and Statistics: Human Data I

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Program #/Poster #: P969.07

Topic: I.07. Data Analysis and Statistics

Support: The Facial Pain Foundation

Title: Imaging Trigeminal Neuralgia Pain: Insights from Combining AI and Granger Causality

Authors: *Y. LIANG[1], Q. ZHAO[1], J. K. NEUBERT[2], M. DING[1];
[1]Biomed. Engin., [2]Orthodontics, Univ. of Florida, Gainesville, FL

Abstract: Trigeminal neuralgia (TN) is a severe and disabling chronic facial pain condition. It is considered one of the most severe pain conditions, often called the “suicide disease,” but presently available therapies for TN are mostly ineffective or inadequate. Both the unpredictable response to treatment and variability in long-term clinical outcomes in TN strongly suggest that a

range of peripheral and central mechanisms underlying TN pain remain to be understood. This study aims to achieve two objectives. Objective 1 is to identify the regions in the brain that underlie TN pain. Objective 2 is to characterize how these regions interact to produce TN pain perception. Functional MRI (fMRI) data were recorded from TN patients (N=57) while they rated their pain levels in real time using a tracking ball. Analytically, recognizing the limitations of traditional data analysis techniques (e.g., fMRI activation analysis), we turned to AI-inspired technology. In particular, a deep neural network (DNN) was designed and implemented, which was able to take fMRI data as input features to predict the moment-to-moment pain levels with high accuracy (75%). Importantly, from the DNN model, a set of brain regions, including the thalamus and the dorsal anterior cingulate cortex (dACC), were derived as signature centers of TN pain. Applying Granger causality analysis to fMRI data from the thalamus and dACC, we further showed that thalamus->dACC causal influence, but not dACC->thalamus causal influence, predicted daily experienced pain levels (R=0.55), consistent with the known physiology that the thalamus is a key brain structure in linking nociceptive sensory input to pain perception and dACC is a key brain structure for perceiving pain.

Disclosures: Y. Liang: None. **Q. Zhao:** None. **J.K. Neubert:** None. **M. Ding:** None.

Poster

P969. Data Analysis and Statistics: Human Data I

Location: Virtual Only

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Program #/Poster #: P969.08

Topic: I.07. Data Analysis and Statistics

Support: LBNL LDRD

Title: Ecog high gamma is a band-limited signal

Authors: *J. A. LIVEZEY[1], A. HWANG[2], M. E. DOUGHERTY[3], K. E. BOUCHARD[4];
[1]E O Lawrence Berkeley Natl. Lab., Berkeley, CA; [2]Mathematical, Computat. and Systems Biol., Univ. of California, Irvine, Irvine, CA; [3]Univ. of California, San Francisco, San Francisco, CA; [4]Lawrence Berkeley Natl. Lab., Berkeley, CA

Abstract: A common problem in neuroscience is how to decompose noisy, multi-source signals measured in experiments (e.g., local field potentials, EEG) into biophysically interpretable components. Analysis of cortical surface electrical potentials (CSEPs) measured using ECoG arrays typifies this problem as CSEPs are believed to be composed of broadband and bandlimited signals with differing biophysical origins. The high frequency range (>70 Hz) has been shown to be a highly informative signal for encoding and decoding behaviors and stimuli. However, little work has been done to search for substructure in this range.
Here, in both rats and humans, we show that CSEPs are composed of band-limited high

frequency components. We first show evidence from rat microECoG that the high frequency band of cortical surface electrical potentials is composed of multiple distinct frequency components. In human CSEPs, it has previously been shown that at slow timescales, variations in the high gamma amplitude can be explained by a modulating power-law component. However, previous studies do not address whether variation in the high gamma amplitude can be explained by a modulating power-law component at fast behavioral timescales. Additionally, previous studies have not differentiated the structure of high gamma in electrodes which are engaged in the task from those which are not engaged. To answer these questions, we propose and use a robust regression model that can account for shoulders in the spectrum to show that at fast timescales and on single trials during speech production, the variation in high gamma amplitude cannot be explained by a modulating power-law component, i.e., it is band-limited. Furthermore, we show that the structure that cannot be explained by a power-law component contains a majority of the information that can be used for decoding produced speech. Together these results show that there are multiple, band-limited sources of high frequency power including the high gamma amplitude which may have different biophysical origins. Our work points to specific future modeling steps required to better understand the biophysical origin of different components of CSEPs.

Disclosures: J.A. Livezey: None. **A. Hwang:** None. **M.E. Dougherty:** None. **K.E. Bouchard:** None.

Poster

P969. Data Analysis and Statistics: Human Data I

Location: Virtual Only

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Program #/Poster #: P969.09

Topic: I.07. Data Analysis and Statistics

Support: NIH Grant R01 GM111293
Department of Anesthesiology, University of Michigan Medical School, Ann Arbor, Michigan

Title: Dynamic reconfiguration of frequency-specific cortical coactivation patterns during psychedelic and anesthetized states induced by ketamine

Authors: *D. LI[1], G. A. MASHOUR[2];
[1]Ctr. for Consciousness Science; Dept. of Anesthesiol., [2]Ctr. for Consciousness Science; Dept. of Anesthesiology; Neurosci. Grad. Program; Dept. of Pharmacol, Univ. of Michigan, Ann Arbor, MI

Abstract: Ketamine is a unique drug that, depending on its dose, has psychedelic and anesthetic properties. We recently demonstrated the dose-dependent effect of ketamine on spatiotemporal

complexity as assessed by Lempel-Ziv complexity in spontaneous scalp EEG signals. Lempel-Ziv complexity reduces the spatiotemporal pattern to a unidimensional measure and assesses the local signal diversity on a short timescale of a few seconds, thus providing a simple and powerful way of indexing the ketamine-induced altered states of consciousness. However, how the different dosing of ketamine modulates the complex spatial dynamics over a longer time scale has not yet been explored. Furthermore, recent neuroimaging studies have demonstrated that spontaneous brain activity exhibits rich spatiotemporal structure that can be characterized as the exploration of a repertoire of spatially distributed patterns that recur over time, which may be a manifestation of covert cognitive information processing and reflect the information capacity of the brain that is thought to be central to consciousness. Here we investigated the dynamic organization of spontaneous cortical activity during wakefulness and during altered states of consciousness induced by different doses of ketamine. Through k-means cluster analysis on the envelope data of source-localized EEG signals, we identified a set of recurring states that represent frequency-specific cortical coactivation patterns, with the spectral and spatial properties resembling those derived from hidden Markov modeling analysis of spontaneous MEG signals at rest. Importantly, we found the effect of ketamine on individual brain states in terms of the fractional occupancy and transition probabilities, and ketamine anesthesia tends to shift the configuration toward states with low spatial complexity. Furthermore, we demonstrated a dose-dependent effect of ketamine on the richness of the dynamical repertoire by quantifying the temporal dynamics of the occurrence and transitions of brain states. We found that subanesthetic ketamine is associated with a richer repertoire, while anesthetic ketamine induces dynamic changes in the brain state organization, with the repertoire richness evolving from a reduced level to one comparable to that during normal wakefulness. These results provide a full description of ketamine's modulations of the dynamic repertoire of spontaneous cortical activity and advances understanding of the neurophysiological mechanism of ketamine in terms of the spatial, temporal, and spectral structures of the underlying whole-brain dynamics.

Disclosures: D. Li: None. **G.A. Mashour:** None.

Poster

P011. Postnatal Neurogenesis: Environmental and Pharmacological Regulation II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P011.01

Topic: A.02. Postnatal Neurogenesis

Support: Cariverona

Title: Enriched environment increases immature neurons in meninges by TrkB signaling

Authors: S. ZORZIN[1], A. CORSI[1], A. AMENTA[2], S. DOLCI[1], F. CIARPELLA[1], E. BOTTANI[1], G. MALPELI[1], G. FUMAGALLI[1], C. CHIAMULERA[1], F. BIFARI[2], *I.

DECIMO[1];
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Abstract: Neural precursors (NPs) present in the hippocampus can be modulated by several neurogenic stimuli including environmental enrichment (EE), which acts through BDNF-TrkB signaling. We have recently identified NPs in meninges, however menigneal niche response to pro-neurogenic stimuli has never been investigated. To this aim, we analyzed the effects of 1 week EE treatment on NP distribution in mouse brain meninges. Following the neurogenic stimulus, although we did not detect modification of meningeal cell number and proliferation, we observed, in meninges, an increased number of β3-Tubulin+ immature neuronal cells. Lineage-tracing experiment confirmed that EE-induced β3-Tubulin+ immature neuronal cells present in meninges originated from GLAST+ radial glia cells. To investigate the molecular mechanism involved in this response, we studied the BDNF-TrkB interaction. Treatment with ANA-12, a TrkB non-competitive inhibitor, abolished the EE-induced increase of β3-Tubulin+ immature neuronal cells in meninges. Overall these data showed, for the first time, that the meningeal niche responded to neurogenic stimuli by increasing the immature neuronal population through TrkB-mediated signaling.This work opened new perspectives in the study of the neurogenic plasticity induced by EE.

Disclosures: S. Zorzin: None. **A. Corsi:** None. **A. Amenta:** None. **S. Dolci:** None. **F. Ciarpella:** None. **E. Bottani:** None. **G. Malpeli:** None. **G. Fumagalli:** None. **C. Chiamulera:** None. **F. Bifari:** None. **I. Decimo:** None.

Poster

P011. Postnatal Neurogenesis: Environmental and Pharmacological Regulation II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P011.02

Topic: A.02. Postnatal Neurogenesis

Support: Harpur Faculty Research Grant
Binghamton University Department of Psychology

Title: The long-term effects of prenatal methadone exposure on adult learning and memory and related hippocampal function

Authors: *M. E. GAMBLE, R. MARFATIA, M. R. DIAZ;
Psychology, Binghamton Univ., Binghamton, NY

Abstract: Prenatal opioid exposures lead to extensive cognitive and emotion-regulation problems in offspring, persisting at least through school-age. Surprisingly, this body of literature has only recently begun to track exposed children into adulthood. One opioid maintenance

treatment typically used for opioid use disorder is methadone, which has been approved for use in pregnant women for several decades. Importantly, however, the impacts of prenatal methadone exposure (PME), particularly on offspring as they progress into adulthood, has not been extensively examined. This is alarming, as methadone continues to be prescribed as a standard of care for use in pregnant women with opioid use disorder, without a complete understanding of the impacts on the exposed child. In recent years, animal models have shown several cognitive consequences as a result of PME, including evidence of hippocampal dysfunction and related behavioral deficits in exposed offspring. The present work aims to examine the persistent nature of these hippocampal and cognitive deficits resulting from PME. Pregnant Sprague Dawley rats received subcutaneous methadone or water injections twice daily from gestational days 3 -20. Following postnatal day 70, male and female offspring were behaviorally tested for several learning and memory tasks. Additionally, mRNA for markers of neurogenesis was quantified from the dorsal hippocampus and whole-cell patch-clamp electrophysiological recordings of dentate granule cells were performed. Results indicate that adult offspring show significant impairments in novel object recognition and preliminary data indicate some potential deficits in spatial learning and retention, particularly in females. Additionally, we observed a pattern of decreased mRNA expression in males for four markers associated with adult hippocampal neurogenesis, one mechanism that plays a major role in these learning and memory behaviors. Finally, preliminary electrophysiology data point toward decreased excitability of the dentate granule cells in both male and female adult exposed offspring. Overall, this work supports the continued investigation of the long-term effects of prenatal methadone exposure on adult male and female learning and memory, as well as promotes further exploration of adult hippocampal neurogenesis as a potential neural mechanism impacted by this exposure.

Disclosures: M.E. Gamble: None. **R. Marfatia:** None. **M.R. Diaz:** None.

Poster

P011. Postnatal Neurogenesis: Environmental and Pharmacological Regulation II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P011.03

Topic: A.02. Postnatal Neurogenesis

Support: Stony Brook University

Title: Fear conditioning and new cells in the amydala in turtles.

Authors: *A. POWERS[1], C. LAU[2], Z. LEE[2], J. WELK[2], J. DENARO[2], C. GRANADOS[2], F. SERRAINO[2];
[2]Psychology, [1]Stony Brook Univ., Stony Brook, NY

Abstract: The amygdala in mammals is importantly involved in fear conditioning. We studied fear conditioning in turtles in order to understand the evolution of the amygdala and its role in fear. Turtles are closely related to the stem amniotes that gave rise to both mammals and reptiles. Although fear conditioning has been demonstrated in many different species, it had not been shown in turtles prior to this study. We developed a paradigm to study fear conditioning in painted turtles (*Chrysemys picta*), in order to study the brain changes that accompany such learning. For the experimental group (n=4), we gave 10 trials per day, pairing a 5-s red light conditioned stimulus with a 1.0 mA footshock (0.5 s, scrambled through a grid floor, overlapping with the end of the red light). The intertrial interval was variable, averaging 2 min. We compared the behavior of turtles in this paradigm to that of two control groups, one given unpaired presentations of light and shock, the other given light alone and no shock (n=4 in each group). Because we did not know what behavior would indicate learning, we measured movement, climbing, pulling in the limbs and/or head, and location in the chamber, comparing the behavior during the red light to the behavior in the 5 s before the red light came on. The only behavior that changed reliably was movement: turtles in the experimental group learned to stop moving when the light came on.After 9 days of training, we ran all groups on extinction (light alone for all groups) for 6 days. The experimental group showed a reversal of their freezing behavior during extinction.All animals were given 9 injections of BrdU (50 mg/kg) over a three-week period, starting one week before conditioning. Four weeks after beginning BrdU injections, all animals were euthanized, and immunohistochemistry for BrdU was done. We counted the number of new cells in the caudal half of the telencephalon, the region that contains the amygdala, because we expected that that region would be involved in fear learning. The experimental group had significantly more new cells in the caudal telencephalon than the two control groups, which did not differ from each other. The presence of new cells in the amygdalar areas when turtles learn conditioned fear suggests that this area is activated by such learning. In mammals, no or very few new cells are produced in the amygdala during fear learning. Nevertheless, these findings demonstrate the involvement of the amygdala in fear learning in turtles and suggest that the amygdala played a role in learning fear in the common ancestors of mammals and reptiles, the stem amniotes.

Disclosures: A. Powers: None. **C. Lau:** None. **Z. Lee:** None. **J. Welk:** None. **J. Denaro:** None. **C. Granados:** None. **F. Serraino:** None.

Poster

P011. Postnatal Neurogenesis: Environmental and Pharmacological Regulation II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P011.04

Topic: A.02. Postnatal Neurogenesis

Support: NIH Grant K08MH122893

NIH Grant R01 MH068542
Hope for Depression Research Foundation

Title: A novel neurogenic compound, Ro6871135, improves pattern separation

Authors: *K. TEGANG[1], W.-L. CHANG[1], R. JAGASIA[2], J. WICHMANN[3], M. SAXE[4], R. HEN[1];
[1]Psychiatry, Columbia Univ/New York State Psychiatric Inst., New York, NY; [2]Neurosci. and Rare Dis. Discovery & Translational Area, [3]Therapeut. Modalities, Roche Innovation Ctr. Basel, Basel, Switzerland; [4]Biogen, Cambridge, MA

Abstract: Background: Prior work from our group has demonstrated that increasing adult hippocampal neurogenesis in mice was sufficient to improve pattern separation, a process in which the dentate gyrus transforms similar experiences or events into discrete, non-overlapping representations. In humans, individuals with panic disorder and posttraumatic stress disorder display poor performance in pattern separation tasks, and enhancing adult hippocampal neurogenesis is a potential therapeutic strategy for treating anxiety disorders. In this study, we determine the behavioral effects of the chronic administration of a novel pro-neurogenic compound, Ro6871135, on pattern separation in a contextual fear discrimination task.Methods: 8-week-old c57BL/6J male mice were administered 7.5 mg/kg of Ro6871135 compound by daily oral gavage. After 21 days of treatment with the vehicle or the compound, mice were behaviorally tested in a contextual fear discrimination task. On the days of behavioral testing, animals were subjected to gavage after cessation of behavioral testing for that day to avoid any possible acute effects of the drug treatment. Results: Using a two-way ANOVA, we found that vehicle and Ro6871135-treated mice showed elevated and indistinguishable levels of freezing in "Context A" 24 hours following training in the same context. Both groups displayed negligible levels of freezing behavior in a distinct "Context C" which differed in several features from Context A (and from Context B). The Ro6871135-treated mice successfully discriminated between Context A and Context B by the fifth day of exposure to both contexts while no significant interaction of Day and Context was found in vehicle-treated mice (n = 8 mice per group). Conclusion: We observed that chronic treatment with Ro6871135, a pro-neurogenic compound, improved performance on a contextual fear discrimination task without having a notable effect on baseline contextual fear conditioning. These results suggest that pharmacological stimulation of adult neurogenesis could be a promising direction for the development of novel therapeutics for disorders characterized by impaired pattern separation.

Disclosures: K. Tegang: None. **W. Chang:** None. **R. Jagasia:** A. Employment/Salary (full or part-time):; Roche. **J. Wichmann:** A. Employment/Salary (full or part-time):; Roche. **M. Saxe:** A. Employment/Salary (full or part-time):; Biogen. **R. Hen:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); drug was received free of charge from Roche.

Poster

P011. Postnatal Neurogenesis: Environmental and Pharmacological Regulation II

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P011.05

Topic: A.02. Postnatal Neurogenesis

Support: Ministry of Education of Singapore (MOE2015-T2-2-035, MOE2017-T3-1-002)
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Human Frontiers Science Programme

Title: A neurogenic microenvironment defined by excitatory-inhibitory neuronal circuits in adult dentate gyrus

Authors: M. UEMURA[1], S. BLANKVOORT[1], L. YUAN[2], L. F. COBAR ZELAYA[2], K. LIT[2], ***A. TASHIRO**[2,1];
[1]Kavli Inst. for Systems Neurosci., Norwegian Univ. of Sci. and Technol., Trondheim, Norway; [2]Nanyang Technological Univ., Singapore, Singapore

Abstract: Adult neurogenesis in the dentate gyrus plays a role in adaptive brain functions such as memory formation. Adding new neurons to a specific locus of a neural circuit with functional needs is an efficient way to achieve such an adaptive function. However, it is unknown whether neurogenesis is linked to local functional demands potentially specified by the activity of neuronal circuits. By examining the distribution of neurogenesis and different types of neuronal activity in the dentate gyrus of freely moving adult rats, we found that neurogenesis is positionally associated with active excitatory neurons, some of which show place-cell activity, but is positionally dissociated from a type of interneurons with high burst tendency. Our finding suggests that the behaviorally relevant activity of excitatory-inhibitory neuronal circuits can define a microenvironment stimulating/inhibiting neurogenesis. Such local regulation of neurogenesis may contribute to strategic recruitment of new neurons to modify functionally relevant neural circuits.

Disclosures: M. Uemura: None. **S. Blankvoort:** None. **L. Yuan:** None. **L.F. Cobar Zelaya:** None. **K. Lit:** None. **A. Tashiro:** None.

Poster

P011. Postnatal Neurogenesis: Environmental and Pharmacological Regulation II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P011.06

Topic: A.02. Postnatal Neurogenesis

Support: NIH Grant 21AMH002784

Title: A novel role for adult neurogenesis in a two-arm bandit reversal learning task

Authors: *K. B. HUNTZICKER[1,2], R.-M. KARLSSON[2], H. A. CAMERON[2];
[1]Neurosci., Brown Univ., Providence, RI; [2]Natl. Inst. of Mental Hlth. (NIMH), NIH, Bethesda, MD

Abstract: Neurogenesis in the mammalian hippocampus persists throughout adulthood, but the function of newly born hippocampal cells remains largely unknown. In this study, we employed a pharmacogenetic method of neurogenesis ablation to determine the effects of adult neurogenesis on decision-making behavior. Rats expressing herpes simplex thymidine kinase (HSV-TK) under control of the human glial fibrillary acidic protein (GFAP) promoter were fed an antiviral drug in adulthood to induce complete ablation of hippocampal neurogenesis. This method is highly specific, inhibiting neurogenesis only upon drug administration and affecting only neural progenitors. Treated transgenic (TK) rats do not exhibit deficits in many standard learning tasks. However, our lab has previously found that treated transgenic (TK) rats show differential responses to ambiguous threat cues when compared against wild-type controls. To test whether adult neurogenesis also underlies behavioral responses to uncertainty under non-threatening conditions, we trained rats on an operant probabilistic learning task. During this task, rats learn that one lever produces a food reward 80 percent of the time, and the other, 20 percent. This type of probabilistic learning – and subsequent reversal learning when lever outcomes are switched – provides rats with many instances of ambiguous feedback. For example, a previously lucrative lever failing to deliver a reward could represent either a reversal or the 20 percent chance that a correct lever does not produce a reward. By using outcome likelihoods that are probabilistic rather than deterministic, this paradigm is more translatable to human learning studies and more representative of situations in nature. We found that in one version of this task, male and female TK rats exhibited higher win-stay ratios and earned more rewards than wild-type controls, suggesting that TK rats may employ different strategies than WTs when seeking rewards under ambiguous conditions. Ongoing studies investigate whether these differences persist when reversal frequencies or reward probabilities are altered. Together, these findings suggest a role for adult neurogenesis in probabilistic decision-making and, more generally, in behavioral responses to situational uncertainty.

Disclosures: K.B. Huntzicker: None. **R. Karlsson:** None. **H.A. Cameron:** None.

Poster

P011. Postnatal Neurogenesis: Environmental and Pharmacological Regulation II

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Program #/Poster #: P011.07

Topic: A.02. Postnatal Neurogenesis

Support: NIH Grant ZIAMH002784

Title: Adult Hippocampal Neurogenesis Regulates Behavior in a Platform Mediated Avoidance Task

Authors: *A. SWIERCZ, H. MARTIN, R.-M. KARLSSON, H. A. CAMERON;
NIMH, NIH, Bethesda, MD

Abstract: Approach-avoidance conflicts occur when an animal is presented with stimuli associated with both positive and negative outcomes. These conflicts are resolved in part by the hippocampus, which acts to modulate the inhibition of prepotent behaviors. In the present study, we examined the role of neurogenesis in an approach-avoidance conflict, with the aim of determining whether adult-born neurons influence behavior when animals have the option to either avoid footshocks, obtain rewards, or attempt to do both. We examined the behavior of rats lacking adult hippocampal neurogenesis in a platform mediated avoidance (PMA) task (Bravo-Rivera et al., 2021). In the PMA task, animals can choose to avoid each footshock, but avoidance comes at the cost of being unable to lever-press for sucrose rewards. The simultaneous presentation of an auditory conditioned stimulus (CS) that reliably predicts a footshock, and a light cue signaling reward availability, creates an approach-avoidance conflict where the animal must balance threat avoidance with food-seeking behavior. Neurogenesis was eliminated by 8 weeks of treatment with valganciclovir in male transgenic Long Evans rats expressing the herpes simplex virus thymidine kinase (TK) under control of the glial fibrillary acidic protein (GFAP) promoter. Ablation of adult-born neurons was followed by three phases of behavioral training. During the avoidance training phase, animals learned to avoid CS-associated footshocks by stepping onto a small platform in the corner of the operant chamber. In reward training, sucrose pellets were dispensed each time a lever was pressed, but only when a light in the chamber was illuminated. During conflict training, auditory and light cues were presented simultaneously over the course of 16 days. The total time spent on the platform during CS presentation was similar between genotypes throughout avoidance training. Both groups also learned to increase lever pressing in response to the light during reward training. In the conflict phase, however, TK animals spent less time on the platform than wild-type controls. The TK group also pressed for rewards at a much higher rate and exhibited less freezing behavior in response to the cues. The loss of new neurons was associated with decreased avoidance and increased lever pressing during the presentation of conflicting stimuli, suggesting that neurogenesis plays an important role in regulating approach and avoidance behavior during high-conflict situations.

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Poster

P011. Postnatal Neurogenesis: Environmental and Pharmacological Regulation II

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Program #/Poster #: P011.08

Topic: A.02. Postnatal Neurogenesis

Support: NIH Grant ZIAMH002784

Title: The role of adult hippocampal neurogenesis in an approach-avoidance conflict task

Authors: *N. FREEDGOOD, R.-M. KARLSSON, H. A. CAMERON;
NIMH, Natl. Inst. of Hlth. (NIH), Bethesda, MD

Abstract: The hippocampus plays an essential role in behavioral inhibition and risk assessment. Previous studies using a non-spatial conditioned cue-based conflict task have shown that inactivation of the ventral hippocampus in rats suppresses avoidance and/or potentiates approach tendencies in a motivational conflict situation (Schumacher, Hippocampus 2015). Given these results, we sought to investigate whether animals lacking adult hippocampal neurogenesis also display impaired cue avoidance and/or approach in an approach-avoidance conflict test. Immature neurons were eliminated from the dentate gyrus of the hippocampus using transgenic rats that express herpes simplex virus thymidine kinase under the GFAP promoter and subsequent oral treatment with valganciclovir. The approach-avoidance conflict training and task utilized two or three arms of an 8 arm-radial arm maze. Bars of different colors and textures were placed along the lengths of each arm and served as cues for the unconditioned stimulus in each arm: appetitive (food reward), aversive (foot shock), and neutral (no stimulus). After all animals were trained to learn the association between the bars and the unconditioned stimuli, they were tested for arm preference to determine whether they had learned the bar cues associated with each stimulus. Both transgenic and wild-type control rats spent more time in the appetitive arm than the neutral arm, and more time in the neutral arm than the aversive arm, with no difference across genotypes, indicating learning of the cue associations was not impaired in the transgenic animals. The rats were then tested on an approach-avoidance conflict test, in which both appetitive and aversive bar cues were placed in one arm, while neutral cues remained in a second arm. Results of the conflict test show that the transgenic rats spent significantly more time in the conflict arm compared to wild-type rats. Overall, the transgenic rats spent more time in the conflict arm compared to the neutral arm, while the wild-type control rats spent more time in the neutral arm compared to the conflict arm. These results show that rats lacking adult hippocampal neurogenesis display suppressed avoidance and/or potentiated approach tendencies compared to controls in a conflict test, but not in the absence of conflict.

Disclosures: N. Freedgood: None. **R. Karlsson:** None. **H.A. Cameron:** None.

Poster

P025. Development: Mechanisms of Dendrite Growth

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P025.01

Topic: A.05. Axon and Dendrite Development

Title: Morpho-electric changes during the postnatal maturation of human supragranular pyramidal neurons

Authors: *M. B. VERHOOG[1], T. A. CHATIZA[1], R. M. HATTINGH[1], J. M. N. ENSLIN[2,3], R. L. MELVILL[4], A. A. FIGAJI[2,3], J. V. RAIMONDO[1];
[1]Human Biol., [2]Div. of Neurosurg., Univ. of Cape Town, Cape Town, South Africa; [3]Red Cross War Mem. Children's Hosp., Cape Town, South Africa; [4]Mediclinic Constantiaberg, Cape Town, South Africa

Abstract: Adult human pyramidal neurons possess unique morphological and physiological features which strongly impact their information processing capabilities, but little is known about what postnatal developmental stages these neurons go through to reach their mature form. Here, we make use of a rare opportunity to obtain live pediatric brain tissue resected during epilepsy surgery from patients in all stages of childhood. A pipeline involving multi-patch electrophysiological recordings, 3D dendritic reconstruction, high-resolution confocal imaging of dendritic spines, and computational modelling was used to systematically characterize human supragranular pyramidal neurons in temporal and frontal cortex from infancy to adulthood (n=25 patients, age range 0.8-40 yrs.). We find that, at 1.5 years of age, human pyramidal neurons are already ~70% of their adult size in terms of the extent of their apical and basal dendritic arborizations. Total dendritic length and number of branch points increase both as a function of depth from pia, as in adults, and as a function of age; these neurons appear to undergo a tightly correlated growth in size and complexity towards adulthood. Developmental changes were also observed in physiological properties, including resting membrane potential, input resistance, action potential kinetics and firing behavior. To explore the impact of these developmental changes on dendritic signal propagation, neural reconstructions of representative neurons for different age groups were used to build detailed compartmental models in the NEURON simulation environment, which were tuned to reproduce electrophysiological data recorded from the same neurons. Developmental changes in spine density were accounted for by adjusting the models' surface area using scale factors derived from dendritic spine imaging. Simulating synaptic activation of randomly distributed spines, we observe greater attenuation of synaptic potentials during passive propagation in pediatric neurons compared to adults, mainly as a result of increased spine density. Together, these findings show that human supragranular neurons undergo various morpho-electric changes during the protracted postnatal maturation of the human brain, which are likely to impact their function.

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Poster

P025. Development: Mechanisms of Dendrite Growth

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P025.02

Topic: A.05. Axon and Dendrite Development

Title: Impact of human gut microbiota on dendritic complexity in neural circuits regulating anxiety and social behavior

Authors: *R. ROYCHOUDHURY[1], H. DUBEY[2], A. WHITE[2], S. LIU[2], R. C. KNICKMEYER[3];
[1]Col. of Osteo. Med., [3]Pediatrics, [2]Michigan State Univ., East Lansing, MI

Abstract: The ability of the gut microbiota to regulate brain development and behavior is increasinglyrecognized. For example, germ-free mice exhibit altered anxiety-like and social behaviors in avariety of paradigms as well as altered dendritic morphology in relevant brain regions such as theamygdala. However, the clinical relevance of these germ-free studies has been challenged as themajority of human development occurs in the presence of complex microbial communities. Toaddress this limitation and determine whether variation in human microbiomes influencesdevelopment of the neural circuits regulating anxiety and social behavior, we introducedmicrobiomes from typically developing infants into pregnant germ-free mice and evaluateddendritic morphology in the prefrontal cortex and amygdala of their adult offspring. We chose tofocus on infant microbiomes because infancy is a critical period in the establishment of the gutmicrobiome and in brain development. Furthermore, our team recently reported associationsbetween features of the human infant gut microbiome and amygdala and medial prefrontal cortexvolumes. One infant inoculum was characterized by high levels of *Bacteroides,* which is oftenconsidered anxiogenic, while the other was characterized by high levels of *Bifidobacterium,*which is often considered anxiolytic. Dendritic length and volume of pyramidal neurons weresignificantly greater in the prefrontal cortex of humanized *Bif* mice (N=4) compared tohumanized *Bac* mice (N=2), $p < 0.05$, suggesting that microbiomes with a high relativeabundance of *Bifidobacterium* may promote dendritogenesis of pyramidal neurons in this region.Specific-pathogen free mice (N=4) appeared more similar to the *Bac* mice, but did not differsignificantly from either group. No differences in dendritic length and volume were observed inaspiny inhibitory neurons in either the amygdala or nucleus accumbens. These findings suggestthat differences in the composition of the human gut microbiome influence development of theprefrontal cortex, which may influence social information processing and emotional regulation.However, results must be considered preliminary given the small sample size.

Disclosures: R. Roychoudhury: None. **H. Dubey:** None. **A. White:** None. **S. Liu:** None. **R.C. Knickmeyer:** None.

Poster

P025. Development: Mechanisms of Dendrite Growth

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Program #/Poster #: P025.03

Topic: A.05. Axon and Dendrite Development

Support: MEXT KAKENHI JP20H03346
MEXT KAKENHI JP16H06459
MEXT KAKENHI JP21K18245

Title: High spatiotemporal-resolution imaging of dendritic refinement dynamics of barrel cortex layer 4 neurons in neonates

Authors: *L. WANG[1,2], S. NAKAZAWA[1,3], H. MIZUNO[4], T. IWASATO[1,2];
[1]Natl. Inst. of Genet., Mishima, Japan; [2]Dept. of Genet., The Grad. Univ. for Advanced Studies (SOKENDAI), Mishima, Japan; [3]Dept. of Basic Neurosciences, Univ. of Geneva, Geneva, Switzerland; [4]Intl. Res. Ctr. for Med. Sci. (IRCMS), Kumamoto Univ., Kumamoto, Japan

Abstract: The precise dendritic projection, which is an important component of cortical circuits, is established during early postnatal stages in an activity-dependent manner. The layer 4 (L4) spiny stellate (SS) neuron in the rodent barrel cortex is a good model to study dendritic refinement mechanisms. In adulthood, these neurons expand their basal dendrites predominantly in a single barrel and receive thalamic inputs specifically from thalamocortical axons (TCAs) transmitting tactile information from a single whisker. This characteristic dendritic projection pattern of SS neurons, which is critical for the proper sensory processing, is established during the first postnatal week under the strong influence of the thalamocortical inputs. We previously applied a two-photon time-lapse in vivo imaging approach to the analyses of dendritic refinement of SS neurons in neonatal mice (Mizuno et al., Neuron 2014; Nakazawa et al., Nature Commun. 2018). In the current study, to investigate the detailed dynamics of SS neuron dendritic refinement, we performed a shorter interval imaging. Barrel cortex L4 excitatory neurons were labeled sparsely using the in utero electroporation-based Supernova (Mizuno et al., 2014; Luo et al., Sci. Rep. 2016). TCA-GFP mice (Mizuno et al., 2014) were used to visualize the barrel map in vivo. We performed in vivo imaging of the same L4 neurons at postnatal day 4 with 1-hour interval for 8 hours. To visualize neuronal morphologies more clearly, we here used a membrane-bound RFP (mRFP) instead of a regular RFP. The regular RFP localizes primarily in the soma and often hampers the detailed analysis of dendritic morphologies. The usage of the mRFP dramatically improved the spatial resolution of the visualization of dendritic morphologies, particularly those of proximal dendrites. We found several interesting transient

morphological features in dendrites. We are currently quantifying the aspects of dendritic refinement dynamics of SS neurons as well as those of star pyramid (SP) neurons. L4 excitatory neurons are composed of SS and SP neurons, and SS neurons acquire specific dendritic orientation through dendritic refinement during development, while SP neurons do not. Thus, the comparison of dendritic dynamics between SS and SP neurons may reveal the dendritic refinement events that are critical for the formation of precise neuronal circuits. In particular, the dynamics of short dendritic trees, which are a key to dendritic refinement (Nakazawa et al., 2018), will be an important focus of our analyses. We will discuss more of our recent analysis in the presentation.

Disclosures: L. Wang: None. **S. Nakazawa:** None. **H. Mizuno:** None. **T. Iwasato:** None.

Poster

P025. Development: Mechanisms of Dendrite Growth

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Program #/Poster #: P025.04

Topic: A.05. Axon and Dendrite Development

Support: This work is supported by a Discovery Grant from the Natural Sciences and Engineering Research Council of Canada (NSERC) (JFW).

Title: Thioredoxin mediates neuronal differentiation in primary cultured mouse cerebral cortical neurons

Authors: M. LLANES-CUESTA, H. TAN, *J.-F. WANG;
Univ. of Manitoba, Winnipeg, MB, Canada

Abstract: Thioredoxin (Trx), as an oxidoreductase, can reverse protein cysteine thiol oxidative modifications, which plays a major role in cellular redox balance and defense against oxidative stress. Trx is maintained in a reduced state by Trx reductase (TrxR). Trx at a reduced state can bind to apoptosis signaling kinase 1 (ASK1) through its thiol group, which inhibits ASK1 and c-Jun N-terminal kinase (JNK)-mediated apoptosis. Recent evidence also showed that Trx overexpression increased the brain derived neurotrophic factor (BDNF) mRNA and protein levels. Because activation of ASK1 leads to neurodegeneration, while BDNF regulates neuronal differentiation, synapsis formation and survival through activation of transcription factor cAMP response element-binding protein (CREB), Trx may regulate neuronal differentiation and degeneration processes. The aim of the present study is to understand the role of Trx in neuronal differentiation, synapsis formation and survival. Primary cortical neurons were isolated from mouse fetus with 17-18 days gestation and cultured in neurobasal medium with N21 supplement from 1 to 32 days in vitro (DIV). Using western blot, we found that Trx, TrxR and phosphor-CREB protein levels increased during neuronal differentiation and maturation from 1 to 15 DIV.

Conversely, phosphor-ASK1 and phosphor-JNK protein levels are elevated during neurodegeneration after 22 DIV. Immunocytochemistry analysis showed that treatment with Trx inhibitor PX12 at 2 μM significantly reduced the number and length of primary dendrites and dendritic branches. Furthermore, treatment with 2 μM of PX12 reduced protein levels of synaptic proteins PSD95, syntaxin, SNAP25, vGlut1 and synaptophysin. Finally, treatment with 2 μM of PX12 reduced cell viability and phosphor-CREB levels while increasing phosphor-ASK1 and phosphor-JNK protein levels. Our findings suggest that Trx regulates neuronal differentiation and survival through activation of CREB and inhibition of ASK1-apoptotic pathway.

Disclosures: M. Llanes-Cuesta: None. **H. Tan:** None. **J. Wang:** None.

Poster

P025. Development: Mechanisms of Dendrite Growth

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P025.05

Topic: A.05. Axon and Dendrite Development

Title: Role of gamma-C4 Protocadherin in survival and self-avoidance in the mouse retina

Authors: *C. MCLEOD, G. R. NITSCHKE, A. M. GARRETT;
Pharmacol., Wayne State Univ., Detroit, MI

Abstract: Cell adhesion molecules (CAMs) are diverse cell surface molecules that are involved in an array of developmental functions, including neural circuit formation. One family of CAMs, the γ-Protocadherins, are a family of 22 protein isoforms hypothesized to play a role in defining neuronal identity. These proteins are capable of forming promiscuous multimers in cis, which then interact homophilically in trans resulting in thousands of potential recognition units between cell membranes. The specific cohort of γ-Pcdh isoforms expressed in each neuron varies at the single cell level and may endow the cell with a unique identity, thus mediating the process by which cells find their partners during neurodevelopment. The γ-Pcdhs regulate many essential processes during development of the CNS such as: neural cell survival, the formation of synapses, dendritic self-avoidance, and the formation of dendritic arbors. How exactly the γ-Pcdhs mediate these processes is not well understood. Recently one isoform, γC4, been identified as essential for postnatal viability and neuronal survival in mice, where mice lacking this isoform (C4KO) die soon after birth. This study seeks to understand the unique function and mechanism of the γC4 isoform in the processes of neuronal survival and self-avoidance in the mouse retina. We **hypothesize** that: 1) the γC4 isoform is the only necessary and sufficient isoform for promotion of neuronal survival in the retina due to its unique protein sequence. 2) γC4 alone cannot promote self-avoidance in starburst amacrine cells of the retina, also as a result of unique protein sequence. 3) γC4 has specific binding partners which promote neuronal survival. The **methods** used to investigate these hypotheses include the use of reduced diversity mouse lines

(γC4 knockout as well as γC4 only mice) to ask whether γC4's role in these two processes is truly unique among isoforms, and how that role is mediated. Preliminary **results** indicate loss of γC4 leads to decreased density of amacrine and retinal ganglion cell types, and that the presence of only γC4 in starburst amacrine cells results in disrupted self-avoidance. We **conclude** that by using the above outlined methods we will elucidate the function of and mechanism by which γC4 affects survival and self-avoidance in the retina.

Disclosures: C. McLeod: None. **G.R. Nitschke:** None. **A.M. Garrett:** None.

Poster

P025. Development: Mechanisms of Dendrite Growth

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P025.06

Topic: A.05. Axon and Dendrite Development

Support: NIH Grant R01 DC015903
NIH Grant R01 DC017482
NIH Grant K01 DC018852

Title: A hierarchical sequence dictates circuit formation during cochlear development.

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Abstract: Hearing by the mammalian cochlea involves two types of sensory hair cells, each innervated by a distinct pair of afferent and efferent neuron types. Inner hair cells (IHCs), which transmit the bulk of sensory input to the brain, are innervated by type I afferents, which are in turn innervated axodendritically by lateral olivocochlear efferents (LOCs). Outer hair cells (OHCs), which fine-tune and amplify the auditory response, are directly (axosomatically) innervated by both type II afferents and medial olivocochlear efferents (MOCs). This simple arrangement of 2 sensory cells + 4 neuron types offers an ideal model system to explore neural circuit assembly during development.
In addition to differing innervation, IHCs and OHCs localize to distinct positions (inner and outer compartments) and are surrounded by different types of supporting cells. We used *Insm1* mouse mutants, in which IHCs appear amidst OHCs in the outer compartment (OC), to determine effects of cell type, environment and location on innervation. We find that type I afferents innervate IHCs regardless of position or environment, as long as they do not encounter a repelling OHC. Type II afferents can innervate both OHCs and IHCs, but only if they are not contacted by a type I afferent.

OCs of *Insm1* mutants, with OHCs and IHCs, were innervated by MOCs, not LOCs, indicating that terminal location, not HC type, determines the type of innervating efferent. However, whether the efferents terminate axodendritically (on afferents) or axosomatically (on HCs) was determined not by location nor type of HC, but by the type of innervating afferent. When OC-IHCs were innervated by type I afferents, these were contacted axodendritically by efferents; when OC-IHCs were innervated by type II afferents, they were also innervated axosomatically by efferents; and when type II afferent innervation was lost, efferent innervation disappeared. We deduce a sequential hierarchy for cochlear circuit formation. (1) IHCs attract, and OHCs repel, type I afferents; (2) type I afferents prevent innervation by type II afferents, so that these can only innervate the remaining HCs; (3) the type of afferent then instructs if and how efferents innervate: axodendritically on type I afferents, asxosomatically on HCs, or not at all.

Disclosures: J.L. Webber: None. **J.C. Clancy:** None. **Y. Zhou:** None. **N. Yraola:** None. **K. Homma:** None. **J. Garcia-Anoveros:** None.

Poster

P025. Development: Mechanisms of Dendrite Growth

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P025.07

Topic: A.05. Axon and Dendrite Development

Support: Whitcome Fellowship
NIH T32-NS048004
Helen Hay Whitney Foundation
HHMI

Title: Temporal genetic program controls global wiring of post-mitotic neurons

Authors: *Y. LIN, S. JAIN, Y. Z. KURMANGALIYEV, S. A. LOCASCIO, P. MIRSHAHIDI, B. PARRINGTON, S. L. ZIPURSKY;
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Abstract: Brain development requires highly precise temporal regulation of gene expression. Though the temporal program of early neuron differentiation has been described in various contexts, how time is controlled in post-mitotic neurons remains unclear. In previous studies we demonstrated that developing neurons express many cell recognition molecules in neuron-type and temporal-specific ways. We speculate that these combinatorial patterns of cell surface gene expression play a central role in determining specific neural connections. Here we sought to identify the genetic programs regulating these expression patterns. We discovered that the ecdysone receptor induces a synchronous cascade of transcription factors in all differentiating neurons during synaptogenesis. Disruption of ecdysone signaling caused wiring defects in many

different neurons, including altering patterns of dendritic arborization and patterns of synaptic connections. Manipulation of different temporally expressed transcription factors in this cascade also yielded specific defects corresponding to the order of their expression during development. By performing single cell sequencing on genetically perturbed neurons, we identified downstream genes of this transcription factor cascade. The downstream genes include a common set of genes for general synapse formation and function, and a cell-type-dependent set for specific cell recognition during brain wiring. The divergence of cell-type-dependent targets of the global temporal program is regulated by cell-type-specific transcription factors. Taken together, this work identifies a synchronous temporal control of multiple aspects of wiring. We speculate that this global temporal program coordinates development across neurons to choreograph specific steps in circuits assembly.

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Poster

P025. Development: Mechanisms of Dendrite Growth

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P025.08

Topic: A.05. Axon and Dendrite Development

Support: Institutional Development Award (IDeA) from the National Institute of General Medical Sciences of the National Institutes of Health under grant number P20GM103423.
NSF RUI Grant 1753730

Title: Characterization of potential growth factor-like proteins in the embryonic and adult cricket, Gryllus bimaculatus

Authors: A. L. BELL[1], W. BULHAN[2], T. N. AL MUSAWI[2], L. S. MIRANDA PORTILLO[2], A. W. RUBENSTEIN[2], F. WANG[2], L. L. LEDWIDGE[2], ***H. HORCH**[2];
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Abstract: Although large scale structural plasticity in adult nervous systems is rare, some organisms maintain the ability to make compensatory structural changes into adulthood, offering the potential for insights into the mechanisms underlying structural plasticity in mature nervous systems. The cricket, *Gryllus bimaculatus*, exhibits unique and robust deafferentation-induced anatomical reorganization in the auditory system. In the cricket, two auditory organs on each forelimb send auditory afferents via nerve 5 to the prothoracic ganglion (PTG), where auditory afferents synapse onto mirror-image pairs of auditory neurons, notably Ascending Neurons 1 and 2 (AN1 and AN2). These cells receive auditory input only from the ipsilateral ear because

auditory system dendritic and axonal arbors are restricted by the midline. Upon unilateral loss of an ear, the deafferented ipsilateral AN1 and 2 dendrites sprout across the midline of the PTG, forming functional, frequency-specific synapses with contralateral auditory afferents. We have completed RNAseq experiments on control and deafferented PTGs. GO Term analysis of signalling pathways differentially expressed after deafferentation identified several growth factor-like candidates, including the Toll receptors and the Protein Yellows. Here we characterize several Toll receptors, their Späetzle ligands, and several Protein Yellow family members in the cricket. Generally, genes encoding these proteins are well-conserved in *Gryllus*, though there is an apparent loss of several genes, such as Späetzle-2, and Protein Yellow-c, -g, and -k. We also find evidence for duplication and divergence of Protein Yellows and Toll receptors. We are currently assessing the expression of several of these candidates in the nervous system of embryos and in the adult PTG and brain using *in situ* hybridization. Further characterization of splice variants and expression patterns will inform future experiments in which we explore the function of these growth factor-like proteins in the development, maintenance, and plasticity of the nervous system in the cricket.

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Poster

P025. Development: Mechanisms of Dendrite Growth

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P025.09

Topic: A.05. Axon and Dendrite Development

Title: Reelin's regulation of translation during neuronal development

Authors: *G. LAGANI, U. BEFFERT, A. HO;
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Abstract: Reelin is a large secreted glycoprotein with a broad spatiotemporal expression profile in the developing and adult mammalian brain. Reelin plays several important roles during brain development, including regulating neuronal migration, stimulating dendritogenesis, promoting dendritic spine maturation, and enhancing long-term potentiation (LTP) at mature hippocampal synapses. Canonically, Reelin binds to the receptors VLDLR and ApoER2, leading to phosphorylation of the Disabled 1 (Dab1) adaptor protein and stimulation of the PI3K/Akt/mTOR pathway. Since the mTOR pathway is a known regulator of protein translation, we hypothesize that Reelin stimulates specific translational dynamics during different phases of neuronal development. To address this, murine primary neuronal cultures were treated with Reelin or control conditioned media for 20 min at 3, 7, and 14 days *in vitro* (DIV). Neuronal

lysate was collected and subjected to SDS-PAGE and immunoblotted for total Akt, mTOR, and S6K1, as well as phosphorylated Akt-S473, mTOR-S2448, and S6K1-T389. Reelin treatment led to increased phosphorylation of both Akt-S473 and S6K1-T389 without changes in total Akt or S6K1 expression. Reelin-induced phosphorylation of Akt and S6K1 was most robust at DIV3 and DIV7 with an attenuated response at DIV14. In addition to immunoblotting, we immunostained DIV7 neuron cultures for Akt-S473 or S6K1-T389. Cultures were also immunostained with MAP2 or tau to visualize neuronal morphology. Total fluorescence intensity was used to measure the level of Akt-S473 and S6K1-T389 phosphorylation. To examine Reelin's role in stimulating translation, we used puromycin metabolic labeling. Primary neurons were treated with Reelin or control media followed by puromycin-labeling to tag nascent peptides. A puromycin specific antibody was used to visualize incorporation of newly translated peptides. Preliminary experiments show an increase in puromycin fluorescence intensity in response to Reelin, suggesting Reelin increases translational activity. Based on these results, we demonstrate Reelin to be a potent activator of the PI3K/Akt/mTOR pathway in early developmental time points and a positive regulator of translation.

Disclosures: G. Lagani: None. **U. Beffert:** None. **A. Ho:** None.

Poster

P034. Neural Circuit Maturation and Dysfunction

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P034.01

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: The convergence technology development program for bionic arm through the National Research Foundation of Korea (NRF) funded by the Korea government (MSIT) (2014M3C1B2048632)
Basic Science Research (2019R1I1A2A01060115) through the National research Foundation of Korea (NRF) funded by the Ministry of Science, ICT & Future Planning

Title: Successful functional recovery after peripheral nerve implantation to the targeted muscle in rats with nerve injury

Authors: *J. KWON[1,2], J. LEE[1,2], Y. YOON[3], J. KIM[2,4];
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Abstract: In the prolonged pathologic status of peripheral nerve injury, formation of traumatic neuroma inhibits appropriate functional restoration and produces painful condition. Because they cannot make spontaneous recovery without surgical intervention, a variety of surgical attempts have been tried. Especially, targeted muscle reinnervation (TMR) known as a prominent microsurgery of transferring donor nerve to an alternate target muscle for preventing primary neuroma and providing environments for axon sprouts to proper direction. However, it has still limited clear improvement in functional recovery and versatile application because targeted muscle should be innervated by other nerves.In this study, we investigated if the implantation of injured peripheral nerve to adjacent muscle has effect on sensory and motor recovery even 12 days after targeted muscle had been denervated. For this, the tibial nerve was transected and transferred to the adjacent part of gastrocnemius muscle at the different time intervals after denervation (immediately or 12 days). Behavioral test for mechanical sensitivity was quantified by using paw withdrawal threshold (PWT) test and tibial nerve function index (TFI) was analyzed for confirming change of paw deformity. 8 months after nerve implantation, ventral part of spinal cord was analyzed for synaptic change by immunofluorescence with synaptophysin. To verify functional re-innervation of targeted muscle, morphological formation of neuromuscular junction (NMJ) was investigated by labeling acetylcholine receptor (AChR) and electromyography (EMG) was performed at the targeted muscle at 8~12 weeks after surgery. After nerve implantation, PWT was significantly increased and muscle atrophy was less progressed in all the nerve implantation group regardless of denervation time interval. Expression of synaptophysin showed no difference with nerve implantation and normal group while control group was decreased. The structure of pretzel-like AChR cluster was observed and the results of EMG implicate that targeted muscle have recovered function following re-innervation although not as much as normal muscle.The present results suggest that denervated muscle can be re-innervated by implanted nerve even after formation of traumatic neuroma. These sensory and functional outcomes may provide experimental foundation of clinical application to maximize the functional efficacy of surgical reconstruction for peripheral nerve regeneration and reinnervation.

Disclosures: J. Kwon: None. **J. Lee:** None. **Y. Yoon:** None. **J. Kim:** None.

Poster

P034. Neural Circuit Maturation and Dysfunction

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P034.02

Topic: A.06. Synaptogenesis and Activity-Dependent Development

Support: NIH Grant R00 MH110665
The Mall Family Foundation

Title: Knockout of the receptor tyrosine kinase Kit impairs feed-forward inhibition in the mouse cerebellum

Authors: T. ZAMAN, A. M. STAFFORD, Q. A. ALSABIA, D. VOGT, ***M. R. WILLIAMS**; Pediatrics & Human Develop., Michigan State Univ., Grand Rapids, MI

Abstract: The receptor tyrosine kinase Kit is enriched in discrete populations of neurons in the mouse and human brain. The endogenous activator of Kit, the sheddable transmembrane protein Kit Ligand, is enriched in populations of neurons known to synapse with those expressing Kit. This complementary pattern of expression has suggested that Kit Ligand-Kit interactions may mediate some essential aspects of synapse biology. Consistent with this idea, in mice and humans, mutation of Kit Ligand or Kit is associated with abnormalities of neuronal function or with neurodevelopmental disorders. We have therefor sought to use the mouse as a model organism to explicitly test the central hypothesis that Kit Ligand-Kit interactions influence synaptogenesis. Within mice and humans, Kit is highly enriched in GABAergic Molecular Layer Interneurons (MLIs) of the cerebellar cortex, while Kit Ligand is highly enriched in Purkinje Cells (PCs), which MLIs migrate to and synapse upon. We have recently reported that depletion of Kit Ligand from PCs decreases GABAergic events recorded in PCs, suggesting that the decreased activation of Kit in MLIs reduces their functional synaptogenesis upon PCs. To test this model, we generated a conditional knockout allele of Kit (having exon 4 floxed), and crossed these mice to Pax2-Cre mice, which enables early developmental ablation of Kit from MLIs (and other cell types). In Pax2-Cre mediated Kit KO animals, we observed a significant reduction in the frequency (but not the amplitude) of spontaneous GABAergic currents recorded in PCs, as compared to control littermates. This result is consistent with a reduction in the functional number of MLI-PC synapses. These results support the model that trans-synaptic activation of Kit by Kit Ligand may be a mechanism for the development or maintenance of central synapses.

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Poster

P034. Neural Circuit Maturation and Dysfunction

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P034.03

Topic: B.06. Synaptic Transmission

Support: 1R01-NS116051-01A1
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1P20GM109089-01A1

Title: Nsg1 and nsg2 knockout mice display divergent behavioral phenotypes

Authors: A. J. ZIMMERMAN[1], P. CHANDER[2], R. AUSTIN[1], *J. WEICK[1];
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Abstract: Members of the neuron-specific gene family (NSG1/NEEP21, NSG2/HMP19 and NSG3/Calcyon) are small endosomal proteins that have been implicated in the trafficking of multiple neuronal proteins including L1/NgCAM, β-APP, and neurotensin receptors. In addition, they are best characterized for their critical roles in the regulation of synaptic development and plasticity via modulation of AMPAR trafficking through endolysosomal pathways. NSG1 is principally thought to promote recycling of AMPARs to the plasma membrane during long-term potentiation, while NSG2 appears to promote heightened synaptic strength during basal activity. However, little is known about the functional requirement of NSG family members and whether they are involved in particular forms of motor, affective, and/or cognitive behavior. To test this, we performed a battery of behavioral assessments on adult mice with constitutive loss of NSG1 or NSG2. Interestingly, opposing phenotypes were found between NSG1 and NSG2 KOs on several behavioral modalities, with a general tendency for NSG1 KOs to perform slightly worse than wild-type controls and for NSG2 KOs to show improvement over wild-type animals. NSG1 KO mice displayed subtle motor deficits in gait as manifest by alterations in paw-surface contact. They also display heightened anxiety on elevated mazes, hyperactivity during subjective daytime, and no change in hippocampal-dependent cognition. In contrast, mice lacking NSG2 showed no motor impairments, demonstrated *reduced* activity during wake phases, and *enhanced* hippocampal-based learning and memory. These contrasting results between NSG1 and NSG2 KOs suggest that they have disparate roles in excitatory synaptic function, a conclusion supported by previous molecular studies. Additionally, they suggest that NSG1 and NSG2 function have different circuit-specific roles in synaptic maintenance of excitatory neurotransmission in adult animals. This occurs despite largely overlapping expression patterns during development and in adults as demonstrated by whole-mount immunohistochemical staining. However, the maintenance of greater amounts of NSG2 expression in adults likely contributes to stronger phenotypes in loss-of-function mutants.

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Poster

P034. Neural Circuit Maturation and Dysfunction

Location: Virtual Only

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Program #/Poster #: P034.04

Topic: B.06. Synaptic Transmission

Support: DoD Grant W81XWH-09-1-0088
DoD Grant W81XWH-21-1-0356

Title: Role of Serotonin Signaling on Synaptic Plasticity in Tuberous Sclerosis Complex

Authors: R. SRINIVAS[1], R. KIRCHNER[2], W. FRANCESCONI[1], F. BERTON[1], ***A. YOSHII**[1]; [1]Dept. of Anat. and Cell Biol., Univ. of Illinois at Chicago, Col. of Med., Chicago, IL; [2]Biostatistics, Harvard Chan Sch. of Publ. Hlth., Boston, MA

Abstract: Tuberous sclerosis complex (TSC) is an autosomal dominant genetic disorder caused by mutations in *TSC1* or *TSC2*. While it is a multi-organ disease, neurological symptoms are prominent and include epilepsy, intellectual disabilities, and autistic behaviors. Dysregulated synapse formation and plasticity are likely to play a significant role in the pathophysiology of TSC. The TSC-1 and -2 proteins form a complex that downregulates the mammalian target of rapamycin (mTOR) pathway, and a gene mutation results in mTOR overactivation. mTOR is a critical regulator of protein synthesis, autophagy, cellular metabolism, and synaptic plasticity. Consequently, suppression of mTOR by rapamycin or its derivatives is one way to correct the pathophysiology of TSC. However, the mTOR pathway has multiple roles, and rapamycin neither specifically nor completely cures all neurological symptoms. A greater understanding of dysregulated synaptic function will enable identifying specific therapeutic targets for epilepsy and cognitive disabilities in TSC. To identify the cause of abnormal synaptic activities, we examined a pan-neuronal *Tsc1*$^{-/-}$ mouse. An unbiased analysis with RNA-seq revealed an excess of transcripts associated with the serotonin signaling pathway. The amount of protein was also higher, and its intracellular distribution was altered in pyramidal cells of the *TSC1*$^{-/-}$ cortex. We used calcium imaging and found that *Tsc1*$^{-/-}$ cultured cortical neurons had frequent Ca^{2+} bursts throughout the soma and dendrites that depended on the serotonin pathway, while wild-type neurons showed random and scattered transients mostly within dendritic spines. Furthermore, daily injection of the serotonin pathway antagonist rescued the premature death of the mutant pups. Remarkably, we also found that the developmental subunit switch of the NMDA-type glutamate receptor was aberrant in the pan-neuronal *Tsc1*$^{-/-}$ mouse cortex, and the serotonin pathway inhibition corrected perturbation of the GluN2B-to-2A subunit switch. Finally, *in vivo* calcium imaging using two-photon microscopy showed hypersynchrony of pyramidal cells in the *Tsc1*$^{-/-}$ mouse cortex. We will present the effect of the serotonin pathway antagonism. Our results indicate that intervening in the serotonergic pathway corrects synaptic dysregulations caused by excessive mTOR pathway activity and sets the stage for developing a new therapeutic approach.

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Poster

P034. Neural Circuit Maturation and Dysfunction

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Program #/Poster #: P034.05

Topic: B.06. Synaptic Transmission

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Mr. Charif Souki
The Joan and Stanford Alexander Endowed Chair for Neuropsychiatric Genetics
Philip R. Dodge Young Inverstigator Award from the Child Neurology Society

Title: Dysfunction of D2 dopamine receptor expressing neurons underlies manic-like behaviors in mice modeling SHANK3 duplication disorder

Authors: *J. HOLDER[1], K. PANG[2], W. WANG[2], K. HAN[3], T. ESTIPHAN[4], F. F. WAGNER[5], M. WEIWER[6], K. JAUNARAJS[7], D. G. STANDAERT[8], B. VUOCOLO[2], Z. LIU[2], H. Y. ZOGHBI[9];
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Abstract: While the contributions of some genes to neuropsychiatric disorders are clear, the downstream neuronal effects are poorly understood. Over-expression of *SHANK3*, which encodes a key synaptic protein, causes neuropsychiatric phenotypes in humans and manic-like behavior in mice providing an opportunity to interrogate the role of SHANK3 in a subset of neurons that might underlie the manic-like behavior. Herein, we describe Shank3's critical role in D2 dopamine receptor (D2dr) neurons and show that *Shank3* overexpression causes increased synaptic neurotransmission in D2dr, but not D1dr, expressing striatal medium spiny neurons. Either pharmacologic D2dr inhibition or genetic normalization of Shank3 abundance in D2-neurons ameliorates manic-like behaviors. Integrating bioinformatic analyses of Shank3's striatal interactome, D1 and D2 dopamine receptor binding proteins, and single-cell RNA-seq datasets, we demonstrate a functional relationship between Shank3 and the D2dr—but not the D1dr. Thus, while *Shank3* is over-expressed in both D1 and D2 dopamine receptor expressing striatal neurons, D2 neuronal dysfunction causes manic-like behaviors.

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Poster

P034. Neural Circuit Maturation and Dysfunction

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Program #/Poster #: P034.06

Topic: B.06. Synaptic Transmission

Support: NIH Grant R01 NS

Title: Mechanism and function of glutamate/GABA cotransmission in entopeduncular nucleus projections to the lateral habenula

Authors: *S. KIM[1], M. L. WALLACE[2], M. EL-RIFAI[1], A. R. KNUDSEN[1], B. L. SABATINI[1];
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Abstract: Many mammalian neurons release multiple neurotransmitters to activate diverse classes of ionotropic receptors on their postsynaptic targets. Previously, our lab has identified a subtype of entopeduncular (EP) nucleus neurons that project to the lateral habenula (LHb) releasing both excitatory and inhibitory neurotransmitters, glutamate and GABA, respectively. It remained unclear if these opposing neurotransmitters are packaged into the same or segregated pools of synaptic vesicles. Furthermore, function of glutamate/GABA cotransmission in behavior is not well understood.
First, we demonstrate a novel method combining electrophysiology, spatially-patterned optogenetics, and computational modeling designed to analyze the mechanism of glutamate/GABA corelease. We find that the properties of postsynaptic currents elicited in LHb neurons by optogenetic activation of EP terminals are only consistent with co-packaging of glutamate and GABA into individual vesicles.
Second, we perform fiber photometry to record bulk calcium signals from the glutamate/GABA coreleasing EP neurons during punishment-based learning. Our results describe response pattern of these neurons and have implications for how glutamate/GABA cotransmission may contribute towards aversive cue-outcome association.

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Poster

P034. Neural Circuit Maturation and Dysfunction

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P034.07

Topic: D.01. Sensory Disorders

Title: Aberrant visual responses in the SynGAP1+/- mouse model of autism spectrum disorder

Authors: *C. A. MARTIN[1], E. T. ANDERSON[2], K. F. K. EJENDAL[1], T. L. KINZER-URSEM[1], M. C. DADARLAT[1];
[1]Biomed. Engin., Purdue Univ., West Lafayette, IN; [2]IU Sch. of Med., Indianopolis, IN

Abstract: Autism spectrum disorder (ASD) is strongly associated with impairments in social function, including understanding complex and subtle social stimuli, which may arise in part from deficits in visual processing. Autism is a serious form of intellectual disability that can result from genetic mutations such as haploinsufficiency and loss of function mutations of the *SYNGAP1* gene. This gene codes for a synaptic GTPase-activating protein that is enriched in excitatory synapses and acts as part of a pathway regulating synaptic plasticity. Heterozygous *Syngap1*+/- mice, with reduced levels of SynGAP1 protein, exhibit an ASD-like behavioral phenotype with partial deficits in synaptic plasticity and an early-shifted developmental schedule, resulting in early closure of sensory critical periods and reduced experience-dependent plasticity that may interfere with normal development of the visual system. Furthermore, *Syngap1*+/- mice have decreased cortical gray matter brain volume throughout visual areas. However, hierarchical visual processing has not been previously well characterized in this mouse model. To address this gap, we used 64- and 128-channel microelectrode arrays to record neural activity in the primary visual cortex of four mice (140 neurons from 2 *Syngap1*+/- mice and 70 neurons from 2 wild-type littermate controls), ages between 2-6 months. Neural activity was recorded in response to visual stimuli to assess receptive field size, orientation tuning curves, and overall neural activity levels. *Syngap1*+/- mice had lower overall firing rates than WT controls (3.8 vs. 5.6 Hz, $p < 0.05$), but had significantly larger receptive fields (*Syngap1*+/- 450.5 visual degrees, wild-type 261.1 visual degrees, $p < 0.01$). For both genotypes, a fraction of neurons had no distinct receptive fields with 67.9% of *Syngap1*+/- and 25.7% of wild-type neurons demonstrating these properties. Furthermore, neurons from both groups showed clear orientation tuning curves in response to drifting oriented gratings. Overall *Syngap1*+/- mice had lower orientation selectivity values (0.29 (±0.19) vs. 0.40 (±0.32), $p < 0.01$), and similar circular variance (0.89 (±0.11) vs. 0.85 (±0.16), $p > 0.05$) – a parameter that estimates tuning width in a model-free manner. Our results support the hypothesis that the early maturation of excitatory synapses in *Syngap1*+/- mice impairs normal refinement of neural responses to visual stimuli during development, leading to deficits in normal visual processing in adult animals.

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Poster

P092. Metabotropic Receptors: Intracellular Signaling

Location: Virtual Only

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Program #/Poster #: P092.01

Topic: B.03. G-Protein Coupled Receptors

Support: Indian Institute of Science Education and Research Mohali.

Title: Norbin regulates group I metabotropic glutamate receptor (mGluR) trafficking and fine-tunes mGluR-triggered AMPAR endocytosis through its interaction with protein kinase A (PKA) in mouse hippocampal neurons

Authors: *P. OJHA, S. PAL, S. BHATTACHARYYA;
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Abstract: Synapses are dynamic formations within the neuronal architecture, they tend to strengthen and weaken depending on various neuronal responses. There are multiple proteins within the synaptic density for relaying information as well as for regulating "plasticity". One such group of proteins which are highly enriched in postsynaptic terminals and are essential for the regulation of synaptic plasticity are group I metabotropic glutamate receptors (mGluRs). Trafficking of mGluRs is known to modulate signaling and therefore synaptic plasticity by regulating the endocytosis of α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid receptors (AMPARs). Thus, it has to be tightly regulated by several postsynaptic density proteins in conjunction with other intracellular signalling molecules. In this study, we identify that Norbin is a scaffolding protein that regulates group I mGluR trafficking and mGluR-mediated synaptic plasticity. Using primary hippocampal neurons from P0 C57BL/6 mice of both sexes and differential antibody labelling of surface and internalized receptors to study receptor trafficking at single cell resolution, we report that Norbin is essential for the ligand-mediated internalization of group I mGluRs. We have used a "molecular replacement strategy" to knockdown endogenous Norbin and to study the effect of various heterologous constructs of Norbin within the same cell. We find that Norbin associates with protein kinase A (PKA) through its N-terminus and anchors mGluR5 through its C-terminus, both of which are necessary for ligand-mediated mGluR5 endocytosis. A point mutation (NorA687G) at the C-terminus of Norbin inhibits the binding of Norbin to mGluR5 and blocks mGluR5 endocytosis. We also identify a novel PKA-dependent modulation of the mGluR-AMPAR axis. Norbin acts as a central mediator for recruiting PKA to AMPARs upon the activation of group I mGluRs which is essential specifically for the mGluR-triggered AMPAR endocytosis, a cellular correlate of mGluR-mediated synaptic plasticity. Both knockdown of endogenous Norbin and deletion of the PKA-binding peptides from Norbin drastically inhibit the mGluR-mediated AMPAR endocytosis. Our findings confirm that Norbin has a dual role in the hippocampus,
(a) as a regulator of group I mGluRs and (b) as an A-kinase anchoring protein (AKAP) that recruits PKA and modulates mGluR-mediated AMPAR endocytosis. Thus, we have unraveled a previously uncharacterized role of Norbin in ligand-induced receptor endocytosis that can have clinical relevance in various neuropsychiatric disorders like schizophrenia, epilepsy etc., where Norbin and mGluR5 have together been implicated.

Disclosures: P. Ojha: None. **S. Pal:** None. **S. Bhattacharyya:** None.

Poster

P092. Metabotropic Receptors: Intracellular Signaling

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P092.02

Topic: B.03. G-Protein Coupled Receptors

Support: Intramural Research Program of NICHD,NIH

Title: Neurotrophic factor-α1 interaction with serotonin receptor 5HTR1E protects against neurotoxicity via β-arrestin/ERK-BCL2 signaling

Authors: V. K. SHARMA[1], X. YANG[1], S.-K. KIM[2], A. MAFI[2], D. S. SANCHEZ[3], L. XIAO[1], A. INOUE[4], W. A. GODDARD, III[2], ***Y. LOH**[1];
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Abstract: Neurotrophic factor-α1/Carboxypeptidase E (NFα1/CPE) is secreted by neurons and endocrine cells where it acts as a prohormone processing enzyme intracellularly and as a neurotrophic factor, extracellularly. Previously we showed that extracellular treatment of hippocampal neurons with NFα1/CPE protected them from oxidative stress-induced cell death through ERK/BCL2 signaling. Based on these results, we hypothesized that NFα1/CPE activation of the ERk/BCL2 pathway to mediate neuroprotection should involve interaction with receptors at the cell membrane. To search for the receptor, we carried out high throughput screening of a library of 324 orphan GPCRs using NFα1/CPE as ligand. The screen identified 5-hydroxytryptamine receptor 1E (5HTR1E), a serotonin receptor, as a binding partner (or receptor), which was confirmed by co-immunoprecipitation and pull down experiments with NFα1/CPE. Radioligand binding studies in HEK293-5HTR1E overexpressing cells showed that recombinant ^{125}I NFα1/CPE bound to 5HTR1E with high affinity (Kd = 13.82 nM). Molecular modeling studies predicted that NFα1/CPE interacted with 5HTR1E, not at the serotonin binding pocket, but via the extracellular loop (ECL)2 of 5HTR1E, forming 3 stable salt bridges with specific amino acid residues in NFα1/CPE. To identify the mechanism underlying the 5HTR1E-NFα1/CPE mediated ERK phosphorylation, we used HEK293 cells stably expressing 5HTR1E. First we tested Gi and Gq protein inhibitors, but found no effect, suggesting that NFα1/CPE stimulated ERK activation was independent of G proteins. We then explored the involvement of β-arrestin, an alternative coupling mechanism with GPCRs for ERK activation. Studies in 5HTR1E expressing-HEK293-β-arrestin KO cells revealed that NFα1/CPE-5HTR1E complex was unable to induce ERK signaling in the absence of β-arrestin proteins. Furthermore, molecular docking studies revealed a very strong coupling of β-arrestin to intracellular loop (ICL) 2 and ICL3 of 5HTR1E. Using LDH cytotoxicity assay we then demonstrated that NF-α1/CPE was able to protect HEK-293-5HTR1E expressing cells against H_2O_2-induced cytotoxicity, *via* increasing pro-survival protein BCL2 expression; however, this protective effect of NFα1/CPE was diminished in the β-arrestin KO cells. Our findings have uncovered a novel mechanism involving NFα1/CPE-5HTR1E interaction to activate β-arrestin/ERK/BCL2 signaling pathway for protecting cells against oxidative stress induced cytoyoxicity.

Disclosures: V.K. Sharma: None. **X. Yang:** None. **S. Kim:** None. **A. Mafi:** None. **D.S. Sanchez:** None. **L. Xiao:** None. **A. Inoue:** None. **W.A. Goddard:** None. **Y. Loh:** None.

Poster

P092. Metabotropic Receptors: Intracellular Signaling

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P092.03

Topic: B.03. G-Protein Coupled Receptors

Support: US National Institutes of Health (R01-DA032895, PJG)
Charles E. Kubly Mental Health Research Center

Title: Norepinephrine triggers rapid nuclear PKA activity through activation of β1-adrenergic receptors localized to the inner nuclear membrane in astrocytes

Authors: *K. C. BENTON, D. S. WHEELER, M. B. ANSARI, B. KURTOGLU, J. G. MAGLASANG, S. KHURSHEED, S. HERGENROTHER, D. LOBNER, P. J. GASSER; Biomed. Sci., Marquette Univ., Milwaukee, WI

Abstract: In the conventional model of adrenergic signaling norepinephrine activates G-protein-coupled receptors exclusively localized to plasma membranes. However, a growing body of evidence indicates norepinephrine can also initiate signaling intracellularly through activation of adrenergic receptors localized to endosomal, Golgi, and nuclear membranes. Norepinephrine is a cationic ligand, requiring carrier-mediated transport to activate intracellular-localized adrenergic receptors. We have previously demonstrated that a high-capacity transporter for norepinephrine, organic cation transporter 3 (OCT3), is localized to both plasma membranes and outer nuclear membranes in neurons and astrocytes, suggesting a potential role for nuclear adrenergic signaling in the central nervous system. We used immunocytochemistry and subcellular fractionation followed by western blot analysis to reveal that β1-adrenergic receptors are localized to inner nuclear membranes in primary cortical mouse astrocytes, and that key G_s-mediated signaling components are localized to the nuclear compartment. We identified the catecholamine transporters OCT3, OCT2, plasma membrane monoamine transporter (PMAT), dopamine transporter (DAT), and norepinephrine transporter (NET) localized to nuclear membranes in astrocytes. The present studies test the hypothesis that norepinephrine activation of nuclear adrenergic receptors is gated by catecholamine transporters. We expressed a nuclear-localized PKA activity sensor in primary astrocytes to observe real-time changes in PKA activity in the nuclear compartment. Treatment with norepinephrine induced both rapid and delayed increases in nuclear PKA activity. Pretreatment with a cocktail of inhibitors for OCTs, PMAT, NET, and DAT abolished the rapid, but not delayed, nuclear PKA response. These findings indicate that β1-adrenergic receptors are functionally coupled to key signaling components in the

nucleus of astrocytes, and that rapid nuclear PKA responses to norepinephrine require uptake by catecholamine transporters.

Disclosures: K.C. Benton: None. **D.S. Wheeler:** None. **M.B. Ansari:** None. **B. Kurtoglu:** None. **J.G. Maglasang:** None. **S. Khursheed:** None. **S. Hergenrother:** None. **D. Lobner:** None. **P.J. Gasser:** None.

Poster

P092. Metabotropic Receptors: Intracellular Signaling

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P092.04

Topic: B.03. G-Protein Coupled Receptors

Title: Effects of oxytocin analogs on neurite outgrowth and cellular signaling in human-derived SH-SY5Y cell line

Authors: *M. PIERCE[1], N. VATTEM[2], A. GORE[2], A. LESCHINSKY[2], A. AZIZ[2], M. BUTT[2], J. ABURAS[1];
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Abstract: Introduction Arginine vasopressin (AVP) and oxytocin (OT) are neuropeptides that bind to G-protein coupled receptors and influence social behaviors, and perturbations are associated with psychopathologies including autism spectrum disorder, schizophrenia, and depression. In mammals, consensus mammalian AVP and OT (Leu^8-OT) sequences are highly conserved. Six distinct OT ligand variants have been discovered in New World Monkeys, including two with in a proline in the 8th position of the peptide (Pro^8-OT; Val^3-Pro^8-OT) resulting in a polyproline helix. In addition to sequence homology between AVP and OT, structural homology between OT receptor (OTR) and vasopressin 1a receptor (V1aR) result in significant cross-reactivity between the ligands and receptors.
Methods The human-derived SH-SY5Y cell line exhibits morphological characteristics of neurons including neurites and endogenously expresses OTR, but not V1aR. Functional assays were performed using Fluo8-AM to measure ligand-induced calcium (Ca^{2+}) mobilization and FLIPR Membrane Potential (FMP) assays to assess ligand-induced hyperpolarization occurring as a consequence of stimulation of potassium (K^+) channel currents. To assess effects on neurite outgrowth, cells were treated with semi-logarithmic doses of OT analogs for 24 hours, fixed and stained with phalloidin and DAPI. The Nikon A1R confocal microscope was used to collect images to examine effects on neurite outgrowth using the Image J package FIJI with the NeuronJ plugin.
Results In SH-SY5Y cells, all three OT analogs were more potent than AVP at inducing Ca^{2+} mobilization. Specificity of the signaling at the OTR were confirmed using the V1aR antagonist SR49059 and the OTR antagonist L-371,257. Moreover, all three OT analogs displayed

equipotency at stimulating Ca^{2+}-mediated depolarization and K^{+}-mediated hyperpolarization. Finally, all OT analogs show a dose-dependent increase in neurite outgrowth.
Conclusions Together, these data suggest that OTR-mediated signaling pathways lead to neurite outgrowth in SH-SY5Y cells. Integrative studies of behavior, genetics and ligand-receptor interaction are crucial for translating signaling activation at the cellular level to effects of OT ligands on social behavior. Knowledge of how OT alters neuronal structure and function has the potential to both identify mechanisms that produce social dysfunction and to inform the development of therapeutic agents.

Disclosures: M. Pierce: None. **N. Vattem:** None. **A. Gore:** None. **A. Leschinsky:** None. **A. Aziz:** None. **M. Butt:** None. **J. Aburas:** None.

Poster

P092. Metabotropic Receptors: Intracellular Signaling

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P092.05

Topic: B.03. G-Protein Coupled Receptors

Support: NIMH Grant R01MH112729

Title: Select second generation antipsychotics are arrestin-biased agonists at D3 dopamine receptors in prefrontal cortex

Authors: *S. SCHAMILOGLU[1], E. LEWIS[3], J. ATHILINGAM[2], K. J. BENDER[2], J. L. WHISTLER[3];
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Abstract: Second generation antipsychotics (SGAs), including clozapine, quetiapine, and aripiprazole, are used to treat schizophrenia and bipolar disorder and show fewer extrapyramidal side effects than their earlier counterparts. SGAs bind with high affinity to G protein-coupled receptors (GPCRs), including D2 and D3 dopamine receptors (D2R, D3R). Their utility is thought to rely on blockade of dopamine-mediated G protein signaling. However, individual SGAs vary in their side effects, including seizures, depression, and metabolic dysfunction, suggesting that effectors other than G protein may be important for their function. SGA pharmacology is well-studied at D2R but less-characterized at D3R, which is expressed throughout brain regions implicated in psychiatric disorders, including prefrontal cortex (PFC). We have previously found that D3R agonist modulation of $Ca_V3.2$ calcium channels at the axon initial segment (AIS) requires noncanonical, G protein-independent signaling through arrestin-3. Here we demonstrate that select SGAs also engage arrestin-3 signaling at D3R, stimulating phosphorylation of ERK *in vitro* and modulating $Ca_V3.2$ at the AIS in D3R-expressing PFC

pyramidal cells. These results highlight a previously unrecognized difference in SGA signaling at D3R, that could inform the development of next-generation pharmaceuticals with improved effect/side-effect profiles.

Disclosures: S. Schamiloglu: None. **E. Lewis:** None. **J. Athilingam:** None. **K.J. Bender:** None. **J.L. Whistler:** None.

Poster

P092. Metabotropic Receptors: Intracellular Signaling

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P092.06

Topic: B.03. G-Protein Coupled Receptors

Support: Margo Cleveland Fund
John Murlin Fund

Title: G α subunits selectively modulate mu opioid receptor pharmacology as observed with a BRET assay

Authors: M. E. BARNETT, B. I. KNAPP, ***J. M. BIDLACK**;
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Abstract: The μ opioid receptor (MOR) is a G protein-coupled receptor that couples to the Gαi/o subunits. However, it is common to downplay the diversity within this class. Gαz is an often-overlooked member of this class, yet its prevalent expression in similar regions of the brain as opioid receptors suggests it may serve a distinct role in MOR signaling. Using a bioluminescence resonance energy transfer (BRET) sensor, we investigated the potential of Gαz and its regulator of G protein signaling counterpart, RGSZ, as a means to control both efficacy and potency of MOR agonists. Agonist binding to MOR resulted in an increase in BRET due to G-protein dissociation. The full agonists DAMGO, β-endorphin, endomorphin-1, methadone, etorphine, and fentanyl were more potent when MOR signaled through Gαz compared to other Gαi/o subunits. For example, the EC_{50} value of fentanyl increased 3-fold when the MOR signaled through Gαi1 compared to Gαz. Partial agonists, such as morphine, (-)pentazocine, metazocine, and ethylketocyclazocine, were more potent and more efficacious when the MOR signaled through Gαz compared to the other Gαi/o subunits. When RGSZ was present in the system, the concentration-response curves of DAMGO and morphine had a significant rightward shift. Increased activation and deactivation rates were correspondingly observed in the presence of RGSZ. Gαz has a slow rate of GTP hydroylsis in comparison to other Gαi/o proteins. To mimic the biochemical properties of Gαz, two different mutations were made in GαoA. GαoA G183S was RGS-insensitive and GαoA 41TSN43 had a slower intrinsic rate of GTP hydrolysis. Both mutants resulted in decreased rates of activation and deactivation for DAMGO and

morphine. When the MOR signaled through either mutant, the concentration-response curves of DAMGO and morphine both had a significant leftward shift. Taken together, signaling through mutant GαoA mimicked the ability of Gαz to enhance opioid potency and efficacy downstream of receptor binding. Enhanced potency and efficacy observed the MOR signaling through Gαz may be the result of slower GTP hydrolysis.

Disclosures: M.E. Barnett: None. **B.I. Knapp:** None. **J.M. Bidlack:** None.

Poster

P092. Metabotropic Receptors: Intracellular Signaling

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P092.07

Topic: B.03. G-Protein Coupled Receptors

Support: NIH Grant 5F32NS100387-02

Title: Harnessing a light-sensitive, neuronal GPCR to study ciliary localization and signaling in vivo

Authors: *A. M. WINANS, D. FRIEDMANN, E. Y. ISACOFF;
Univ. of California, Berkeley, Berkeley, CA

Abstract: Many differentiated cells in the body, including most neurons, produce a single, primary cilium. Mutations that affect primary cilia give rise to a host of diseases termed "ciliopathies" which often manifest neurodevelopmental and cognitive defects. Certain neuronal G-protein coupled receptors (GPCRs), such as serotonin and dopamine receptor subtypes, localize to the primary cilium. However, how ciliary localization modulates neuronal GPCR signaling and downstream behavior is still not well understood. Here, we compare ValopA, a ciliary non-visual opsin, and its homolog ValopB, a cell body-localized non-visual opsin, to interrogate potential effects of ciliary localization on behavior. ValopA expresses in the spinal cord of the embryonic zebrafish and inhibits spontaneous coiling when excited by blue green light. Photoinhibition of locomotion is followed by a post-excitatory phase lasting tens of minutes following cessation of the light. ValopB, when overexpressed in a ValopA knock-out animal, successfully rescues the inhibitory and excitatory phases but with markedly shorter kinetics. To probe the relationship between localization and signaling kinetics, we created a suite of ValopA/B chimeras with variable ciliary and cell body localization patterns, as well as a ValopB-Rab23 fusion protein that localizes to the cilium. Rescue experiments with these constructs show that increased ciliary localization coincides with increased photoinhibition length. Moreover, constructs with significant cell body expression will inhibit locomotion well upon repeated excitation whereas constructs with largely ciliary expression experience a decrease in inhibition efficiency and also take longer to reach full inhibition. Future experiments

will pinpoint the contribution of intrinsic signaling kinetics versus sub-cellular localization on the observed behavior.

Disclosures: A.M. Winans: None. **D. Friedmann:** None. **E.Y. Isacoff:** None.

Poster

P092. Metabotropic Receptors: Intracellular Signaling

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P092.08

Topic: B.03. G-Protein Coupled Receptors

Title: Non-visual opsin 3 functionally interacts with melanocortin receptors in hypothalamic neurons

Authors: *H. HADDAD, L. OLINSKI, C. CHUNG, E. OANCEA;
Brown Univ., Providence, RI

Abstract: Opsins are light-sensitive G-protein coupled receptors that convert external photons into intracellular signaling events. While most vertebrate opsins are well characterized, *the function and G-protein signaling of the extra-ocular opsin 3 (OPN3) in the central nervous system is unknown with untapped therapeutic potential*. Unlike other opsins mostly expressed in the retina, OPN3 is expressed in deep brain areas and peripheral tissue. In skin melanocytes, OPN3, via Gαi G-protein, inhibits the cAMP response triggered by activation of G-protein coupled receptor melanocortin 1 (MC1R), thus decreasing melanin levels in a light independent manner. Brain mapping of our OPN3-mCherry knock-in mouse model reveals high expression of OPN3 in brain areas that are associated with energy metabolism and food intake regulation, specifically the paraventricular nucleus of the hypothalamus. Neural melanocortin 4 receptor is highly expressed in this brain area and signals through cAMP to function in regulating energy balance and metabolism. More recently, MC4R was discovered to close inward rectifying potassium channel (Kir7.1) in a G-protein independent manner in response to α-melanocortin stimulating hormone (αMSH), an agonist of MC4R. This project explores the potential role of OPN3 in regulating MC4R-mediated signaling mechanisms. Through the monitoring of single-cell, real-time changes in intracellular cAMP, we found that in response to αMSH, OPN3 decreases cAMP accumulation compared to control conditions. This decrease in cAMP accumulation in OPN3 over-expressed cells was abolished after blocking Gαi activity using pertussis toxin. To examine potassium signaling, we used a thallium flux assay that measures flow of thallium through open potassium channels. By using live-cell imaging and OPN3 targeted shRNA to knock down OPN3, we discovered that knocking down OPN3 leads to a decreased thallium flux response compared to control condition when treated with αMSH. In an opposite manner, we also found that over-expressing OPN3 leads to an increased thallium flux response compared to αMSH treated control conditions. Blocking Kir7.1 channels with a

selective pore blocker (VU590) abolished the flux responses to αMSH. These findings suggest that OPN3 negatively regulates melanocortin receptor-activated cAMP signaling in a Gαi dependent manner. In terms of potassium signaling, our results indicate that OPN3 disinhibits the MC4R inhibition of Kir7.1 and leads to more channels opening upon MC4R activation. Our data reveal a novel signaling pathway with implications in energy regulation and therapies for metabolic diseases.

Disclosures: H. Haddad: None. **L. Olinski:** None. **C. Chung:** None. **E. Oancea:** None.

Poster

P092. Metabotropic Receptors: Intracellular Signaling

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P092.09

Topic: B.03. G-Protein Coupled Receptors

Support: NIH 41891

Title: Ciliary receptors in neurons: Competitive regulation and implications for excitability

Authors: *J. SHRESTHA[1], J. L. FUCHS[2];
[1]Biol. Sci., Univ. of North Texas, DENTON, TX; [2]Biol. Sci., Univ. of North Texas, Denton, TX

Abstract: Neurons commonly have a primary cilium, which is a non-motile organelle extending from the centrosome into the extracellular space. In most brain regions, cilia are highly enriched in either somatostatin type 3 receptors (SstR3) or melanin concentrating hormone type 1 receptors (MCHR1), or both. These GPCRs may affect neuronal excitability, as evidenced by heightened vulnerability to seizures in SstR3-/- mice and heightened resistance to seizures in MCHR1-/- mice. In the present study, we present evidence for competition between the two ciliary receptors. We have shown that in SstR3-/- mice, MCHR1(+) cilia are increase markedly in number and intensity. Here we also find the converse, that ciliary SstR3 was increased in MCHR1 knockouts. These striking comparisons suggest a competition between the two GPCRs. Further supporting this hypothesis is our analysis of the MCHR-/- hippocampus, in which the greatest increase in SstR3(+) cilia was in CA1, where MCHR1(+) cilia predominate in wildtypes; in contrast, little SstR3 upregulation occurred in CA3, where MCHR1(+) cilia are scarce in wildtypes. Moreover, in wildtype mice, SstR3 and MCHR1 levels were lower in cilia containing both receptor types than in cilia with only one. Receptor competition could occur at various levels, such as gene expression, access to transport systems, or space in the ciliary membrane. However, there is precedence for receptor competition at the transcriptional level: Olfactory neurons initially express multiple ciliary odorant receptors until one emerges as the sole victor, through transcriptional suppression of the alternatives. Receptor composition in neuronal primary cilia may also be governed by reciprocal competition, such that expression of

one gene entails suppression of the other(s). Accordingly, we are testing whether receptor colocalization is more common in early development, before dominance is established. Our findings have implications for understanding the functions of ciliary receptors. For example, SstR3 upregulation might contribute to seizure resistance in MCHR1-/- mice, and conversely, MCHR1 upregulation might contribute to seizure vulnerability in SstR3-/- mice. Thus, the role of any particular ciliary GPCR in normal or disease states is best understood in the context of alternative receptors. This insight will be important in designing approaches for modulating ciliary GPCRs in the treatment of epilepsy and other excitotoxic conditions.

Disclosures: J. Shrestha: None. **J.L. Fuchs:** None.

Poster

P092. Metabotropic Receptors: Intracellular Signaling

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P092.10

Topic: B.03. G-Protein Coupled Receptors

Support: UM FCVC Inaugural Fund

Title: The design of a protean agonist to RFRP-1R that improves cardiac function through functional selectivity.

Authors: *R. NICHOLS, K. PITTALA, M. LEANDER, B. MAYNARD;
Univ. of Michigan Med. Sch., Ann Arbor, MI

Abstract: The design of a protean agonist to RFRP-1R that improves cardiac function through functional selectivity.R. Nichols, K. Pittala, M. Leander, B. F. MaynardThe heart synthesizes peptides, which may play important roles in cardiovascular physiology,; yet much remains unknown about their signaling pathways. Delineating the structure-activity relationship (SAR) of a peptide is an established approach to identifying agonists and antagonists, molecular tools to dissect signaling and investigate function. In addition, peptides often act through G protein-coupled receptors (GPCRs), which may be druggable due to their defined binding pocket and ligand-receptor interactions. We discovered that the RFamide-related peptide -1 (RFRP-1; MPHSFANLPLRF-NH_2) acts through the GPCR RFRP-1R to decrease cardiac function [Nichols et al. 2010; Nichols et al. 2012]. In order to establish the RFRP-1 structure-activity relationship (SAR) for cardiac contractility, we assayed a series of single-substituted alanine and a set of N-terminal truncation analogs using isolated cardiomyocytes. From our SAR data, we identified the amidated C-terminal pentapeptide as the active core and designed a first-generation antagonist LPLAF-NH_2. We confirmed the alanyl-substituted pentapeptide competed out the effect of the active core in cardiomyocytes. We further Further, we complemented our SAR study with molecular docking software, analyzing the parent peptide, RFRP-1, active core, LPLRF-NH_2,

and antagonist, LPLAF-NH_2, to predict the binding pocket and ligand-receptor interactions involved in binding and receptor activation. A next step in our work was to design a proteolytically resistant antagonist, as peptides are susceptible to degradation by peptidases and thus limited in application. We took a two-prong approach to our design, we chose to substitute A9 with amino acid residues that would improve ligand-receptor to improve binding and also incorporated d-isomers to combat degradation and improve half-life. Next, we used molecular docking software to examine whether the analogs might interact with RFRP-1R. As we docked the series of analogs, we predicted the newly designed structures blocked RFRP-1 binding. In addition, based on their position in the binding pocket and ligand-receptor interactions, we predicted some structures may act as RFRP-1R protean agonists. Our subsequent work showed the putative protean agonist improved cardiac function in cardiomyocytes suggesting the ligand-directed RFRP-1R signaling may serve as a druggable GPCR in heart.

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Poster

P099. Vesicle Docking, Fusion, and Recycling

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Program #/Poster #: P099.01

Topic: B.05. Neurotransmitter Release

Support: NIH Grant DK027044
NanoBiology Institute, Cell Biology, Yale University

Title: Phosphorylation of Synaptotagmin by Protein Kinase C regulates vesicle fusion

Authors: *E. M. STROEVA[1], M. BERA[1], K. CHARI[2], K. WU[3], O. D. BELLO[1], J. COLEMAN[1], S. S. KRISHNAKUMAR[1], J. E. ROTHMAN[1];
[1]Cell Biol., [3]Mol. Biophysics and Biochem., [2]Yale Univ., New Haven, CT

Abstract: A family of Synaptotagmins (Syt) undergo several types of post-translational processing, which regulates neuronal growth and plasticity. Protein kinase C (PKC), which is highly expressed in the brain, has been previously shown to potentiate synaptic vesicle release via phosphorylation of the Syt1 isoform. However, the precise mechanism and functional effect of phosphorylation on Syt1 function remains unclear. In this work, we identified a new site of phosphorylation (T328/T329), located within polybasic patch of C2B domain which is functionally important for binding to PIP2 and other negative lipid headgroups. To study the effect of phosphorylation at this site, we introduced a phospho-mimetic amino acid substitution ($Syt1^{T328/329E}$). We used single-vesicle fusion assay to monitor the docking, clamping, fusion and release of a single vesicle. This fluorescence-based assay tracks individual v-SNARE containing vesicles (vSUVs) as they approach the physiologically relevant mimic of the target membrane.

vSUVs containing 24 copies of wild-type Syt1 ($Syt1^{WT}$) and 12 copies of VAMP2 remain stably docked and fuse after addition of 1 mM calcium ions. To the contrary, $Syt1^{T328/329E}$ containing vSUVs abolished clamping ability leading to spontaneous fusion. The role of phosphorylation of Syt1 by PKC was further corroborated by a γ-^{32}P radiography-based protocol for the *in vitro* phosphorylation of $Syt1^{WT}$-containing vSUVs. Phosphorylated vesicles recapitulated the loss-of-clamping behavior exhibited by the $Syt1^{T328/329E}$ -containing vesicles. Expectedly, phosphorylated Syt1 exhibited a dramatic reduction in its ability to bind negatively charged membrane in the basal (EDTA/Mg^{2+}) state and in the presence of physiologically relevant calcium concentrations. As PIP2 binding is crucial for Syt1 to oligomerize and clamp vesicle fusion, we posit that that phosphorylation of Syt1 at residue 328/329 abrogates clamping function, by disrupting Syt1 ability to oligomerize. Our findings suggest a novel mechanism of *in situ* regulation of Syt1 function by PKC.

Disclosures: E.M. Stroeva: None. **M. Bera:** None. **K. Chari:** None. **K. Wu:** None. **O.D. Bello:** None. **J. Coleman:** None. **S.S. Krishnakumar:** None. **J.E. Rothman:** None.

Poster

P099. Vesicle Docking, Fusion, and Recycling

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Title: Synaptotagmin 7 is targeted to the axonal plasma membrane through γ-secretase processing to promote synaptic vesicle docking in mouse hippocampal neurons

Authors: *J. D. VEVEA[1], G. F. KUSICK[2], K. C. COURTNEY[1], E. CHEN[2], S. WATANABE[2], E. R. CHAPMAN[1];
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Abstract: Synaptotagmin (SYT) 7 has emerged as a key regulator of presynaptic function, but its localization and precise role in the synaptic vesicle cycle remain the subject of debate. Here, we used iGluSnFR to optically interrogate glutamate release, at the single bouton level, in

SYT7KO dissociated mouse hippocampal neurons. We analyzed asynchronous release, paired pulse facilitation, and synaptic vesicle replenishment, and found that SYT7 contributes to each of these processes to different degrees. 'Zap-and-freeze' electron microscopy revealed that loss of SYT7 diminishes docking of synaptic vesicles after a stimulus and inhibits the recovery of depleted synaptic vesicles after a stimulus train. SYT7 supports these functions from the axonal plasma membrane, where its localization and stability require both γ-secretase-mediated cleavage and palmitoylation. In summary, SYT7 is a peripheral membrane protein that controls multiple modes of SV exocytosis and plasticity, in part, through enhancing activity-dependent docking of SVs.

Disclosures: J.D. Vevea: None. **G.F. Kusick:** None. **K.C. Courtney:** None. **E. Chen:** None. **S. Watanabe:** None. **E.R. Chapman:** None.

Poster

P099. Vesicle Docking, Fusion, and Recycling

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P099.03

Topic: B.05. Neurotransmitter Release

Support: NIH R01 #NS40296
NSF Grant 1122374

Title: RNA editing of the synaptic vesicle fusion clamp Complexin alters synaptic transmission and synaptic development at Drosophila glutamatergic neuromuscular junctions

Authors: *E. BRIJA[1,2], S. K. JETTI[1,2], R. W. CHO[1,2], J. T. LITTLETON[1,2,3];
[1]The Picower Inst. for Learning and Memory, [2]Dept. of Brain and Cognitive Sci., [3]Dept. of Biol., MIT, Cambridge, MA

Abstract: Chemical synaptic transmission is a critical step in neuronal communication that is extensively regulated to generate plasticity and learning. Although most studies of synaptic plasticity have focused on changes occurring within the postsynaptic compartment, presynaptic modifications to the synaptic vesicle (SV) release machinery also contribute to alterations in synaptic output. The *Drosophila* neuromuscular junction (NMJ) has emerged as an excellent model synapse to study synaptic transmission, as well as activity-dependent structural and functional plasticity. Complexin (Cpx) regulates both spontaneous and evoked neurotransmitter release through its ability to bind assembling SNARE complexes and control SNARE zippering, providing an attractive point of modulation for altering presynaptic output. The *Drosophila cpx* locus undergoes alternative splicing to produce two isoforms that differ in the C-terminal ~15 amino acids of the protein. We previously observed that activity-dependent PKA phosphorylation of the C-terminus of the Cpx7B splice variant enhanced spontaneous

neurotransmitter release and synaptic growth. Unlike Cpx7B, the more abundant Cpx7A splice variant does not undergo PKA phosphorylation, but is subject to RNA editing by ADAR (adenosine deaminase acting on RNAs). *Cpx* transcript editing produces Cpx7A protein products with varying C-terminals, including changes to an asparagine residue (N130) near the PKA phosphorylation site in Cpx7B (S126). Editing of Cpx7A generates N130G, N130S and N130D variations that alter the ability of Cpx to clamp spontaneous SV fusion and properly regulate synaptic growth. Using single cell RNA sequencing and electrophysiology, we are characterizing the differential expression and function of Cpx7A RNA edited variants across several neuronal populations. These studies should provide new insight into how differential RNA editing contributes to neuronal activity and development, as well as how changes in Cpx function can alter neurotransmitter release and synaptic growth pathways.

Disclosures: E. Brija: None. **S.K. Jetti:** None. **R.W. Cho:** None. **J.T. Littleton:** None.

Poster

P099. Vesicle Docking, Fusion, and Recycling

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P099.04

Topic: B.05. Neurotransmitter Release

Support: NINDS/NIH grant R01NS111978
Israel Science Foundation 1310/19
U.S.-Israel Binational Science Foundation grant 2019248

Title: Synapsin domains mediating alpha-synuclein function

Authors: A. STAVSKY[1], S. TAL[1], L. A. PARRA[2], J. B. KAHN[1], S. ROY[2], ***D. GITLER**[1]; [1]Dept. of Physiol. and Cell Biol., Ben-Gurion Univ. of the Negev, Beer-Sheva, Israel; [2]Depts. of Pathology and Neurosci., UCSD, La Jolla, CA

Abstract: Despite extensive effort, the physiologic role of alpha-synuclein at the synapse is contentious and unclear. We and others have reported that modest over-expression of alpha-synuclein attenuates synaptic vesicle (SV) recycling in cultured hippocampal neurons suggesting that alpha-synuclein is a physiologic attenuator of neurotransmitter release. Exploring functional interactors of alpha-synuclein, we recently found that alpha-synuclein mediated synaptic attenuation was absent in neurons lacking all three synapsin genes (synapsin triple knockouts, TKO), suggesting that alpha-synuclein/synapsin interactions played a key role in alpha-synuclein function (Atias et al., PNAS 2019, 10.1073/pnas.1903054116). Though our experiments revealed a previously unknown role of synapsin in mediating alpha-synuclein function, the identity of the synapsin isoforms involved remained unknown. Alternate splicing of mammalian synapsins generate five major isoforms - synapsin Ia, Ib, IIa, IIb and IIIa - and each synapsin isoform is

composed of a specific combination of alternative domains (called “domains A-J”). To parse out the synapsin isoform responsible for mediating alpha-synuclein function, we individually reintroduced synapsins Ia, Ib, IIa, IIb and IIIa into synapsin TKO neurons that were also over-expressing alpha-synuclein. Interestingly, while reintroduction of synapsins Ia, IIa and IIIa restored the ability of alpha-synuclein to attenuate SV-recycling, synapsins Ib and IIb did not. The Ib/IIb isoforms also interacted poorly with alpha-synuclein in immunoprecipitation assays, suggesting that the synapsin “E domain” - common to synapsins Ia/IIa/IIIa but absent in Ib/IIb - is responsible for executing synaptic alpha-synuclein functions. We propose that the synapsin E domain plays an important role in regulating the functional co-dependency between synapsins and alpha-synuclein, perhaps by helping in organizing the SV clusters near the synaptic active zone.

Disclosures: A. Stavsky: None. **S. Tal:** None. **L.A. Parra:** None. **J.B. Kahn:** None. **S. Roy:** None. **D. Gitler:** None.

Poster

P099. Vesicle Docking, Fusion, and Recycling

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P099.05

Topic: B.05. Neurotransmitter Release

Support: McDonnell Cellular and Molecular Neuroscience Grant
Klingenstein-Simons Fellowship in Neuroscience

Title: Metabolic adaptations of presynaptic function to glucose deprivation

Authors: *G. ASHRAFI[1], A. TIWARI[3], A. HASHEMIAGHDAM[2], M. LARAMIE[2]; [1]Washington Univ. Sch. of Med., Saint Louis, MO; [2]Washington Univ. Sch. of Med., St. Louis, MO; [3]Washington Univ. in St. Louis, St. Louis, MO

Abstract: The brain is the most energy-consuming organ of the body, but unlike other metabolically demanding tissues, it does not have significant energy stores. In the nervous system, neurons experience rapid energetic fluctuations particularly during intense circuit activity, fasting, or metabolic disorders. Neuronal bioenergetic plasticity is therefore essential to sustain cognitive function. Yet, the cellular pathways that enable neurons to achieve metabolic flexibility are largely unknown. A significant fraction of neuronal energy is consumed in nerve terminals during synaptic transmission. We have recently shown that nerve terminals can utilize either glucose or oxidative fuels through glycolysis or mitochondrial respiration, respectively. Neuronal access to fuels is not constant and can vary substantially. Thus, mitochondrial capacity for ATP synthesis is dynamically modulated to respond to physiological cues, such as, cellular energy demand or fuel supply. Here, we report a molecular pathway for metabolic adaptation of

neuronal mitochondria to nutrient deprivation. We performed RNA sequencing of cultured cortical neurons with differential access to glucose. We found that glucose deprivation activates a transcriptional program upregulating the expression of the mitochondrial master regulator *PGC-1α* and the mitochondrial NAD-dependent deacetylase *Sirtuin 3* (*Sirt3*), in a pathway that is dependent on the cellular energy sensor AMP kinase. Immunocytochemical analysis of cultured neurons and mouse hippocampal slices confirmed that Sirt3 localizes on axonal mitochondria of hippocampal neurons. We then examined the functional role of Sirt3 in presynaptic metabolism using quantitative optical assays. Presynaptic ATP measurements with the reporter Syn-ATP in Sirt3-deficient neurons showed that Sirt3 is required for maintaining baseline ATP levels in nerve terminals. Knockdown of Sirt3 did not affect Ca^{2+}-dependent upregulation of mitochondrial ATP production during electrical activity, further confirming its function in regulation of baseline, rather than activity-driven, mitochondrial metabolism. Sirt3 and its catalytic activity as a deacetylase are also essential for fast retrieval of synaptic vesicles after release in neurons supplied with oxidative fuels, indicating an essential role for Sirt3 in synaptic transmission. In summary, our findings suggest that the mitochondrial deacetylase Sirt3 modulates the metabolic fitness of nerve terminals and its transcriptional upregulation during energetic stress may facilitates neuronal bioenergetic plasticity.

Disclosures: G. Ashrafi: None. **A. Tiwari:** None. **A. Hashemiaghdam:** None. **M. Laramie:** None.

Poster

P099. Vesicle Docking, Fusion, and Recycling

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P099.06

Topic: B.05. Neurotransmitter Release

Support: American Heart Association [17GRNT33661156]

Title: The serotonin transporter expressed in sympathoadrenal chromaffin cells controls the kinetics and amount of catecholamine released from individual secretory vesicles

Authors: R. L. BRINDLEY, ***K. P. M. CURRIE**;
Cooper Med. Sch. of Rowan Univ., Camden, NJ

Abstract: Adrenal chromaffin cells (ACCs) comprise the neuroendocrine arm of the sympathetic nervous system and are a powerful experimental model system to investigate neurotransmitter exocytosis. ACCs synthesize and secrete catecholamines (epinephrine, norepinephrine) which play important roles in the physiological response to stress. Even though mammalian ACCs do not synthesize serotonin (5-HT), they robustly express the antidepressant-sensitive serotonin transporter (SERT) which mediates the uptake and accumulation of 5-HT by the cells (5-HT

content of wild-type adrenal glands is <0.15% of the epinephrine content). The adrenal glands from global ($SERT^{-/-}$) or sympathoadrenal system-specific SERT knockout mice ($SERT^{\Delta TH}$) displayed reduced 5-HT but no change in catecholamine content. We isolated ACCs from the mouse adrenal glands and used carbon fiber amperometry to analyze transmitter secretion from individual secretory vesicles. The charge of individual amperometric spikes (i.e. quantal size / amount of transmitter released) was significantly smaller (~35%) in $SERT^{-/-}$ and $SERT^{\Delta TH}$ cells compared to matched controls. Spike half-width (duration), and the duration of the "prespike foot" which precedes some amperometric events, were significantly shorter in knockout cells. The decrease in quantal size (spike charge) was mimicked by treating wild-type cells for >24 hours with escitalopram (an SSRI antidepressant which blocks SERT), or by depleting extracellular 5-HT from the culture medium for >24 hours (use of dialyzed serum in the culture media). In contrast, acute (minutes) or short-term (<6-8 hrs) treatment with escitalopram or 5-HT depletion had no effect. ACCs do not synthesize 5-HT because they lack the rate limiting biosynthetic enzyme tryptophan hydroxylase. However, if the product of this enzyme (5-hydroxytryptophan; 5-HTP) is provided to bypass the lack of tryptophan hydroxylase, then it can be converted into 5-HT by aromatic L-amino acid decarboxylase (AADC), which is endogenously expressed and also participates in catecholamine biosynthesis. Treating cells with 5-HTP for 1-3 hrs had no effect but 24 hr treatment fully rescued the reduced spike charge seen in SERT knockout cells. Together, our data suggest that SERT-mediated 5-HT uptake by ACCs modulates the kinetics and thus fraction of secretory vesicle contents released during a fusion event. Ongoing work will further dissect this and other potential mechanisms by which adrenal SERT regulates catecholamine secretion and hence the sympathoadrenal stress response.

Disclosures: R.L. Brindley: None. **K.P.M. Currie:** None.

Poster

P099. Vesicle Docking, Fusion, and Recycling

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P099.07

Topic: B.05. Neurotransmitter Release

Title: Genetic deletion of the membrane transporter mGAT1 in dopamine neurons selectively abolishes GABA co-release

Authors: *R. MELANI, N. X. TRITSCH;
New York Univ. Neurosci. & Physiol., New York, NY

Abstract: Midbrain dopaminergic (DA) neurons in the Substantia Nigra pars compacta (SNc) have been shown to inhibit dorsal striatum by synaptic co-release of GABA. We previously suggested that in these cells GABA release does not depend on cell-autonomous synthesis, but on axonal uptake by the membrane transporter mGAT1 (Slc6a1). However, the mechanism

responsible for this GABA co-release is still unclear. To better define the role of mGAT1, we generated conditional knocked out (cKO) mice in which Slc6a1 is ablated exclusively in DA neurons. Optogenetically stimulating DA axons in dorsal striatum, we find that dopaminergic GABA release in mGAT1cKO mice is selectively and entirely abolished. Our results show that cell-specific deletion of Slc6a1 does not impair vesicular exocytosis or cell survival, confirm that GABA co-release from DA neurons requires mGAT1 transporter function, and pave the way for future studies investigating the contribution of this transmission in modulating striatal function and behavior.

Disclosures: R. Melani: None. **N.X. Tritsch:** None.

Poster

P099. Vesicle Docking, Fusion, and Recycling

Location: Virtual Only

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Program #/Poster #: P099.08

Topic: B.05. Neurotransmitter Release

Support: National Institutes for Deafness and Communication Disorders Regulation of Synaptic Vesicle Dynamics in the Auditory System R01 DC014093
National Institutes for Deafness and Communication Disorders Development of Helper Dependent Adenoviral Vectors for Inner Ear Gene Therapy Approaches R21DC018242
Presynaptic regulation of neurotransmitter release in mammalian neuronal circuits R01 NS110742-01
University or Iowa Distinguished Scholar Award
Viral Vector Mediated Gene Therapy Approaches for Hearing and Balance Disorders

Title: Presynaptic Rac1 is a key regulator of synaptic strength at a mammalian central synapse

Authors: *M. AL-YAARI[1], C. KEINE[1], P. VALINO RAMOS[1], D. GUERRO-GIVEN[3], N. KAMASAWA[3], S. M. YOUNG, Jr.[1,2];
[1]Anat. and Cell Biology, Iowa Neurosci. Inst., [2]Dept. of Otolaryngology, Iowa Neurosci. Institute, Aging Mind Brain Initiative, Univ. of Iowa, Iowa city, IA; [3]Max Planck Florida Electron Microscopy Core, Max Plank Florida Inst. for Neurosci., Jupiter, FL

Abstract: Sound information encoding within the initial auditory processing stations requires reliable and precise synaptic transmission in response to rapid and large fluctuations in action potential (AP) firing rates. However, the number of synaptic vesicles (SVs) available for AP-evoked release is limited. Actin cytoskeleton is an important regulator of synaptic transmission. Rac1 is a core protein of the actin cytoskeleton cascades and it is implicated in several

neurodevelopmental diseases. Moreover, it plays a fundamental role in regulating synaptic transmission. Although Rac1 has been well-studied in the postsynaptic compartment, currently, the role of Rac1 in the presynaptic regulation of synaptic transmission is poorly understood. The calyx of Held is a large glutamatergic presynaptic terminal in the auditory brainstem and relies on high-fidelity transmission at frequencies of up to 1 kHz. It conveys the temporal information of sound precisely, which requires very fast and continuous synaptic transmission at high rates. Thus, it is an ideal model to determine how presynaptic Rac1 regulates synaptic transmission. We utilized a Rac1 conditional knock out (CKO) mouse in conjunction with viral vector mediate approaches to delete Rac1 in the presynaptic compartment at the matured synapse (after hearing onset) to bypass the potential roles of Rac1 in developmental stages. Our data show that Rac1 regulates synaptic transmission and plasticity, suggesting a critical role of presynaptic Rac1. Our results and their implications will be discussed.

Disclosures: M. Al-Yaari: None. **C. Keine:** None. **P. Valino Ramos:** None. **D. Guerro-Given:** None. **N. Kamasawa:** None. **S.M. Young:** None.

Poster

P099. Vesicle Docking, Fusion, and Recycling

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Program #/Poster #: P099.09

Topic: B.05. Neurotransmitter Release

Support: R01 DC014093
R21 DC018242
R01 NS110742-01
University of Iowa Distinguished Scholar Award

Title: Presynaptic $\alpha_2\delta$-3 plays a critical role in synaptic transmission at the calyx of Held

Authors: *W. MILANICK[1], T. RADULOVIC[2], S. M. YOUNG[2];
[1]Interdisiplinary Grad. Program in Neurosci., [2]Dept. of Anat. and Cell Biol., Univ. of Iowa, Iowa City, IA

Abstract: The $\alpha_2\delta$ family of proteins are critical regulators of pre- and postsynaptic functions, including voltage-gated Ca^{2+} (Ca_V) channel trafficking, Ca_V Ca^{2+} currents, synapse morphogenesis, axon regeneration, and action potential waveform. Previous research has shown that global knockout (KO) of $\alpha_2\delta$-3 can impair synapse morphogenesis and synaptic transmission and cause auditory dysfunction. However, global KO models complicate conclusions on whether functions are pre- or postsynaptic and cannot rule out developmental causes. Therefore, it is difficult to define the presynaptic roles of $\alpha_2\delta$-3 with traditional models. To study the presynaptic roles of $\alpha_2\delta$-3 we developed a novel $\alpha_2\delta$-3 conditional KO mouse model. We used the calyx of

Held/medial nucleus of the trapezoid body (MNTB) synapse as our model. The calyx is a large glutamatergic presynaptic terminal that plays critical roles in the auditory system. It is the sole driver of action potentials in the MNTB, making it a powerful model to examine the molecular mechanisms of synaptic function. Globular bushy cells give rise to the calyx and are located in the cochlear nucleus - where $\alpha_2\delta$-3 is expressed. To assess the presynaptic roles of $\alpha_2\delta$-3 we performed afferent fiber stimulation at different frequencies. We collected data from mice at postnatal 18-23, when the calyx of Held/MNTB synapse is in its mature state. Recordings were done in physiological Ca^{2+} (1.2mM), and at physiological temperature 37°C. Results will be discussed.

Disclosures: W. Milanick: None. **T. Radulovic:** None. **S.M. Young:** None.

Poster

P101. Synaptic Integration

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P101.01

Topic: B.06. Synaptic Transmission

Support: NIMH R01MH123686
NIDCD T32DC010775-10
Whitehall Foundation Grant 2018-12-9

Title: Cell type-specific nonlinear interactions of top-down and bottom-up synaptic input to posterior parietal cortex

Authors: *D. RINDNER, A. PRODDUTUR, G. LUR;
Neurobio. and Behavior, Univ. of California, Irvine, Irvine, CA

Abstract: Cognitive functions including attention, memory, and prediction rely on interactions between top-down and bottom-up information streams. Nonetheless little is known about the integration of top-down and bottom-up inputs at the cellular level, partially due to the technical difficulty of simultaneously manipulating two distinct afferent populations. We overcome this limitation using a dual-color optogenetic strategy to determine spatiotemporal features of multi-modal synaptic integration in the mouse posterior parietal cortex (PPC). We expressed Chrimson, a red light-sensitive channelrhodopsin variant, in the primary auditory cortex (A1) and ChR2, a blue light-sensitive opsin, in the anterior cingulate cortex (ACC), a frontal region commonly associated with attention and working memory. This opsin combination allowed independent control of bottom-up sensory and top-down frontal inputs to the PPC. Using whole-cell patch clamp recordings in acute brain slices we show that the PPC receives direct, monosynaptic inputs from both A1 and ACC. We found that two classes of pyramidal neurons, intrinsically bursting and regular spiking, have distinct patterns of frontal-sensory synaptic

integration. Intrinsically bursting cells displayed a robust supralinear enhancement of coincident input from A1 and ACC, an effect that diminished with increasing delays between inputs. Conversely, regular spiking cells showed linear integration of coincident input, and maximal enhancement when afferents were activated with delay. Both bursting cell coincident and regular spiking delayed enhancement were reduced by bath application of either the low-threshold calcium channel or NMDA receptor antagonists mibefradil and AP5, respectively. Notably, only bursting cell coincident enhancement was reduced when sodium channels were blocked with TTX. These results suggest that sodium channels provide a mechanism for temporally specific detection of coincident frontal-sensory inputs. Our data demonstrate that frontal-sensory synaptic interactions have cell type-specificity in the PPC and offers a novel mechanistic description of top-down and bottom-up integration.

Disclosures: D. Rindner: None. **A. Proddutur:** None. **G. Lur:** None.

Poster

P101. Synaptic Integration

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Program #/Poster #: P101.02

Topic: B.06. Synaptic Transmission

Support: NIH R21EB029740
HFSP RGY0069
ORAU Ralph E. Powe Junior Faculty Enhancement Award

Title: Synaptic cooperativity controls somatic spike precision via noise-induced stochastic gating

Authors: *S. XIAO, S. YADAV, K. JAYANT;
Purdue Univ., West Lafayette, IN

Abstract: Clustered and distributed synaptic co-operativity is a fundamental property of cortical pyramidal neurons and has been implicated in maintaining coincidence detection, feature binding and dendritic branch specific plasticity. However, the exact biophysical mechanisms by which synaptic cooperativity controls and modulates somatic output are still unclear. In basal dendrites, co-activated spines in close proximity to one another give rise to a N-methyl-D-Aspartate (NMDA) spike accompanied with both an increase in local Ca^{2+} concentration as well as sodium dependent dendritic spikes. However, how these non-linearities and distributed synapses across the vast expanse of the dendritic tree as well as multiple dendritic trees control, interact, and cooperate to modulate somatic output remains unknown. Our limited understanding of both intra- and inter-branch synaptic cooperativity stems partly from technical barriers in manipulating spine properties with high-resolution and monitoring the effects of these manipulations across different depths (as not all dendrites are at the same depth). Here, we

performed simultaneous electrophysiology, calcium imaging, and two-photon glutamate uncaging on basal dendrites of cortical pyramidal neurons in acute brain slices using a spatial light modulator (SLM) with custom built pulse compression. The SLM allows simultaneous activation up to 12 spines (~15mW per spine) with up to 24 spines within 5 ms (time for phase mask change) distributed in 3D (across 30 um) with near-diffraction-limited resolution. We found that co-activation of clustered spines on some but not all basal dendritic branches gives rise to a complex EPSP waveform, characterized with a Na^+ spikelet followed by a NMDA plateau. Compared to NMDA spikes without a Na spikelet, these complex dendritic spikes increase the temporal precision of somatic action potentials, especially when the resting potential is close to threshold. We find that temporal precision is made possible by a combination of the persistent sodium current (as it influences both the non-linear interaction and the increased input resistance) and stochastic ion-channel gating. Together, this suggests that Na^+ spikelets are a source of noise that can help bind temporally precise input patterns. More importantly, our results suggest that dendritic noise as a function of clustered inputs could serve as a way of multiplexing diverse inputs via unique somatic spike time/frequency signatures, even within the same dendritic tree. We discuss the above results through both modeling and experiments along with detailed optical characterization.

Disclosures: S. Xiao: None. **S. Yadav:** None. **K. Jayant:** None.

Poster

P101. Synaptic Integration

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P101.03

Topic: B.06. Synaptic Transmission

Support: WhiteHall2018-12-09

Title: Differences in ionic conductance and microcircuit architecture underlie distinct frontal-sensory integration in cortical neuron subclasses

Authors: *A. PRODDUTUR[1], D. J. RINDNER[1], G. LUR[2];
[1]Neurobio. and Behavior, Univ. of California Irvine, Irvine, CA; [2]Neurobio. and Behavior, Univ. of California, Irvine, Irvine, CA

Abstract: The posterior parietal cortex (PPC) is a multimodal cortical area involved in higher-order perceptual and executive processes. Critical for these functions is a convergence of bottom-up (sensory) and top-down (frontal) afferent innervation. Pyramidal neurons with complex dendritic structures perform nonlinear integration; a synaptic interaction thought to be underpinned by active and passive mechanisms. However, the precise mechanisms of how sensory and top-down synapses interact in single neurons are unknown. In the PPC, L5

pyramidal cells characterized by either regular spiking (RS) or intrinsic burst spiking (IB) perform nonlinear integration of inputs from auditory (A1) and anterior cingulate cortex (ACC). Specifically, coincident activation of A1-ACC inputs leads to supralinear integration in IB cells, whereas only delayed activation (50ms) of these inputs leads to supralinear synaptic enhancement in RS cells. Using a compartmental model, we evaluated the role of sodium (Na^+), and T-type calcium (T-Ca^{2+}) channels, and NMDA receptor kinetics in nonlinear integration of A1-ACC inputs. We found that high Na^+ conductance and faster NMDAR decay kinetics recapitulate all experimentally observed integration characteristics in IB cells.Conversely, in RS cells, higher T-Ca^{2+} conductance and slower NMDAR decay kinetics lead to supralinear integration during delayed activation. However, across the entire parameter space, our model predicted supralinear integration of coincident inputs in RS cells as well. This finding was in contrast with electrophysiology data and suggested that the mechanisms driving synaptic integration in RS cells might not be exclusively cell-autonomous. For example, dendrite targeting inhibition is known to modulate synaptic integration in pyramidal neurons. Thus, we tested the effect of disynaptic, feedforward GABAergic input on sensory-frontal integration in RS cells. We found that $GABA_A$ receptor-driven inhibition suppresses the supralinear integration in the coincident condition without affecting integration under the delayed condition. *In vitro* testing of the model's predictions revealed that RS, but not IB cells receive significant feedforward inhibition. Together, our modeling and *in vitro* data offer insights into synaptic integration in a cell-type-specific manner controlled at two levels: cell-autonomous differences in channel expression and kinetics, complemented by distinct microcircuit architecture.

Disclosures: A. Proddutur: None. **D.J. Rindner:** None. **G. Lur:** None.

Poster

P101. Synaptic Integration

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P101.04

Topic: B.06. Synaptic Transmission

Support: JSPS KAKENHI 21K06434

Title: Simulation analysis of subthreshold voltage signaling along hippocampal mossy fibers

Authors: *H. KAMIYA;
Dept. of Neurobio., Hokkaido Univ. Grad. Sch. of Med., Sapporo, Japan

Abstract: Axons are classically thought of as electrically well isolated from other parts of the neuron. Recent studies, however, revealed that subthreshold somatodendritic depolarization distributes for a substantial distance along the axons. To further explore the functional significance of subthreshold voltage propagation to the axons, the model of propagation of

excitatory presynaptic potential (EPreSP) suggested by Alle & Geiger was reconstructed and was incorporated the model of axonal sodium, potassium & leak channels by Engel & Jonas and the model of presynaptic calcium channels composed of P/Q-, N-, and R-type channels at mossy fiber boutons by Li, Bischofberger & Jonas. Preceding subthreshold EPreSPs slightly reduced the action potential-evoked presynaptic calcium currents by a decrease in the amplitude of action potentials due to the slow depolarization. EPreSPs by themselves elicited small calcium currents during subthreshold depolarization through these high-voltage activated calcium channels. Since the previous study by simultaneous pre and postsynaptic recordings demonstrated that EPreSPs enhanced action potential-evoked transmitter release from the mossy fiber terminals, it has been suggested that different mechanisms from the enhancement of action potential-evoked presynaptic calcium entry may involve enhanced transmitter release by EPreSP. Small calcium entry by subthreshold EPreSPs may enhance transmitter release from the mossy fiber terminals by high-affinity calcium sensors for enhancing transmitter release.

Disclosures: H. Kamiya: None.

Poster

P101. Synaptic Integration

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Program #/Poster #: P101.05

Topic: B.06. Synaptic Transmission

Support: National Institutes of Health [NS105804]
CURE Epilepsy
Dravet Syndrome Foundation
Brain Research Foundation

Title: Molecular and functional NMDA receptor diversity enables selective tuning of corticothalamic circuit function.

Authors: Y. CHEN[1,2], M. LADISLAV[1], S. SWANGER[1,3,4];
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Abstract: The somatosensory corticothalamic (CT) circuit processes ascending sensory signals and selects the appropriate information for our conscious brain to receive. Disruption to the balance of excitation and inhibition within CT circuitry leads to absence seizures, sleep disorders, and attention deficits. In this circuit, glutamatergic CT projections gate somatosensory information flow through the thalamus by innervating glutamatergic thalamocortical neurons in the ventral posteromedial thalamus (VPM) and GABAergic reticular thalamus neurons (nRT),

which provide robust feed-forward inhibition to the VPM. Our goal is to identify strategies for selectively modulating CT-nRT and CT-VPM synapses to correct disease-associated excitation-inhibition imbalances. N-methyl-D-aspartate (NMDA) receptors mediate glutamatergic CT synaptic transmission, but CT-nRT and CT-VPM excitatory postsynaptic currents have distinct NMDA receptor amplitudes, decay kinetics, and voltage-dependences. VPM and nRT neurons express four NMDA receptor subtypes, GluN2A-GluN2D, which have distinct functional properties. In this study, we sought to determine: 1) how GluN2 subunit diversity contributes to NMDA receptor function in nRT and VPM neurons and 2) whether NMDA receptor modulation has cell-type-specific effects on spike firing in response to corticothalamic input. Single-molecule multiplex fluorescent in situ hybridization revealed greater NMDA receptor mRNA levels in VPM neurons compared to nRT, and the relative abundance of GluN2A-GluN2D mRNAs differed between the two cell-types. Utilizing brain slice electrophysiology, we demonstrated GluN2-specific roles in regulating tonic inhibition as well as cell-type-specific NMDA receptor contributions to synaptic transmission and spike firing in response to CT input. Notably, NMDA receptor inhibition reduced VPM spike firing significantly more than nRT spike firing in response to high frequency stimulation. Furthermore, NMDA receptor inhibition reduced VPM spike firing in response to high frequency CT input significantly more than during low frequency CT input. These data suggest that NMDA receptor contributions to thalamic spike firing are cell-type-specific and frequency-dependent. Given the differential regulation of inhibitory and excitatory neuron function, we hypothesize that NMDA receptor modulation could be useful for context-dependent tuning of thalamic excitability based on which synaptic inputs are dysregulated or how excitation and inhibition are imbalanced in neurological disease.

Disclosures: Y. Chen: None. **M. Ladislav:** None. **S. Swanger:** None.

Poster

P101. Synaptic Integration

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P101.06

Topic: B.06. Synaptic Transmission

Title: Mouse cortical nerve endings release exosomes in a Ca^{2+}-dependent fashion: new insights into synaptic transmission.

Authors: G. OLIVERO[1], A. ROGGERI[1], G. VALLARINO[1], *D. DI PAOLO[2], D. MARIMPIETRI[3], M. GAGLIANI[4], M. PODESTÀ[3], K. CORTESE[4], ***A. PITTALUGA**[1]; [1]Dept. of Pharm., Univ. of Genova, Sch. of Med. and Pharma, Genova, Italy; [2]Lab. of Exptl. Therapies in Oncology, [3]Stem Cell Lab. and Cell Therapy Ctr., IRCCS Giannina Gaslini Hosp., Genova, Italy; [4]Dept. of Exptl. Medicine, DIMES, Univ. of Genova, Genova, Italy

Abstract: Whether exosomes can be actively released from presynaptic nerve terminals is matter of debate. To address the point, mouse cortical synaptosomes were incubated in basal and depolarizing (25 mM KCl enriched medium) conditions and extracellular vesicles were isolated from the synaptosomal supernatants to be characterized by dynamic light scattering, transmission electron microscopy, western blot and flow cytometry analyses. The structural and biochemical analysis unveiled that supernatants contain vesicles having the size and the shape of exosomes, that were immunopositive for the exosomal markers TSG101, flotillin-1, CD63 and CD9. The markers content increased upon the exposure of nerve terminals to the high-KCl stimulus, consistent with an active release of the exosomes from the depolarized synaptosomes. High KCl-induced depolarization elicits the Ca^{2+}-dependent exocytosis of glutamate. Interestingly, also the depolarization-evoked release of exosomes from cortical synaptosomes occurred in a Ca^{2+}-dependent fashion, since TSG101, CD63 and CD9 contents in the exosomal fraction isolated from supernatants of depolarized synaptosomes were significantly reduced when omitting external Ca^{2+}ions.. However, (±)-baclofen (10 μM), which significantly reduced the glutamate exocytosis, didn't affect the release of exosomes, suggesting that the $GABA_B$-mediated mechanism of control is inactive on this event. Our findings suggest that the exposure of synaptosomes to a depolarizing stimulus elicits a presynaptic release of exosomes that occurs in Ca^{2+}-dependent fashion. The insensitivity to the presynaptic $GABA_B$ receptors, however, leaves open the question on whether the release of exosomes could be a druggable target for new therapeutic intervention for the cure of synaptopathies.

Disclosures: G. Olivero: None. **A. Roggeri:** None. **G. Vallarino:** None. **D. Di Paolo:** None. **D. Marimpietri:** None. **M. Gagliani:** None. **M. Podestà:** None. **K. Cortese:** None. **A. Pittaluga:** None.

Poster

P101. Synaptic Integration

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P101.07

Topic: B.06. Synaptic Transmission

Support: NSF grant CRCNS-DMS-1608077

Title: Revealing the link between spiking cross-correlation patterns and the underlying subthreshold dynamics: oscillations and resonances

Authors: R. F. O. PENA, ***H. G. ROTSTEIN**;
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Abstract: A sharp peak near zero in cross-correlation functions (CCFs) indicates the presence of a putative monosynaptic connection between the pre- and post-synaptic neurons (Toyama et al.,

1981; English et al., 2017; Platkiewicz et al., 2021). However, CCFs are complex and contain significantly more information about the spiking pattern relationships and the mechanisms controlling the interactions between the participating neurons, in particular in the presence of intrinsic oscillations and resonances. Some of this information is apparent from the spiking patterns themselves, but spiking patterns are controlled by the subthreshold (membrane potential) dynamics whose effects often remain hidden in CCFs. Whether and how the subthreshold dynamic information of post-synaptic neurons can be extracted from the CCFs remains an open question. This is not a straightforward task since in vivo neuronal interactions occur in the presence of background noise, synaptic dynamics, oscillatory activity and resonances. Similar patterns may be generated by different mechanisms and this degeneracy might be difficult to resolve. We address this issue by combining biophysical modeling, numerical simulations and dynamical systems tools (phase-space analysis). We systematically focus on a wide number of representative scenarios and identify the presence of additional peaks in the CCFs, lower than the characteristic sharp peak, and link these lower peaks to the type of nonlinearities and time scales that operate at the subthreshold level and ultimately reflect the neuronal biophysical properties. Under certain conditions, the combination of these dynamic components cause a subset of trajectories in the phase-space diagrams to remain at subthreshold levels for a longer time than others before producing a spike. The presence and variability of this spike-time delay is due to a combination of noise and neuronal intrinsic dynamics. Lower peaks also emerge in the presence of other factors, such as background oscillations or ripples, but these come from a second wave of spikes. We analyze the differences between these two types of lower peaks. Our results have implications for the inference of synaptic interactions between neurons in the presence of oscillatory activity, synaptic dynamics and unidentified background patterns, and for the understanding of the biophysical and dynamics mechanisms controlling synaptic interactions between neurons in a network.

Disclosures: R.F.O. Pena: None. **H.G. Rotstein:** None.

Poster

P101. Synaptic Integration

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P101.08

Topic: B.06. Synaptic Transmission

Title: A new commissural mechanism in the medial entorhinal cortex

Authors: K. ZHENG[1], G. WANG[2], C. MIAO[1], R. MURPHY[4], Ø. SIMONSEN[1], E. NILSSEN[1], K. MIYAZAKI[6], W. N. ROSS[6], M. M. SCOTT[3], K. DEISSEROTH[7], J. F. STORM[5], M. P. WITTER[8], *J. ZHU[9];
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Charlottesville, VA; [4]Dept. of Mol. Medicine, Physiol. Section, [5]Dept. of Mol. Med., Univ. of Oslo, Oslo, Norway; [6]New York Med. Col., Valhalla, NY; [7]Bioengin & Psych, Stanford Univ. Dept. of Psychology, Stanford, CA; [8]Kavli Inst. Systems Neuroscience, Norw. Univ. Sci. & Tech., Trondheim, Norway; [9]Dept Pharmacol, Univ. VA Sch. Med., Charlottesvle, VA

Abstract: Commissures form the largest axonal tracts that wire the two brain hemispheres, yet the commissural organization and function remain largely unknown. We discovered that commissural inputs, in contrast to local parasubicular inputs, synapsed preferentially on distal dendrites of stellate cells in the mouse medial entorhinal cortex. Subsequent optogenetic experiments verified the functionally distinct anatomical inputs. Preliminary investigation showed that commissural inputs were essential for grid-like firing in behaving mice. Analysis combining octuple patch-clamp recordings, optogenetic activation, and functional Ca^{2+} and Na^{+} imaging revealed that commissural inputs interacted with local perisomatic parasubicular inputs via axosomatodendritic channels, which amplified coincident and theta rhythmic commissural synaptic events selectively in entorhinal stellate cells, but not pyramidal neurons and interneurons. Pilot neuronal simulation validated this salience selection mechanism. These data reconstruct essential neuronal constituents with their synaptic connections that encompass an excitatory commissural circuit involved in spatial coding.

Disclosures: K. Zheng: None. **G. Wang:** None. **C. Miao:** None. **R. Murphy:** None. **Ø. Simonsen:** None. **E. Nilssen:** None. **K. Miyazaki:** None. **W.N. Ross:** None. **M.M. Scott:** None. **K. Deisseroth:** None. **J.F. Storm:** None. **M.P. Witter:** None. **J. Zhu:** None.

Poster

P101. Synaptic Integration

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Program #/Poster #: P101.09

Topic: B.06. Synaptic Transmission

Support: R01NS110422
R34NS116740
Pew Charitable Trust

Title: Voltage compartmentalization by dendritic spines in vivo

Authors: *V. CORNEJO, N. OFER, R. YUSTE;
Columbia Univ., New York, NY

Abstract: Dendritic spines mediate most excitatory neurotransmission in the nervous system, so their function is likely critical for brain function. While it has been demonstrated that spines are

biochemical compartments, their electrical role is still unclear. Using a genetically-encoded voltage indicator and two-photon microscopy, we measured membrane potential in spines and dendrites from pyramidal neurons of somatosensory cortex in mice during spontaneous activity and sensory stimulation. We found different regimes for dendrite and spine voltage dynamics during supra- and sub-threshold potentials. Spine depolarizations are similar to dendritic ones when the neuron fires action potentials, but during subthreshold synaptic potentials spines often experience different voltages than parent dendrites, even activating independently. Confirming these two types of spine voltage regimes, after two-photon optogenetic activation of the parent dendrites, spine potentials are similar to dendritic potentials, but they remain compartmentalized during optogenetic activation of spine heads. Our results demonstrate that spines are voltage compartments, acting as electrical diodes, with little or no dendrite-to-spine attenuation of dendritic potentials but significant spine-to-dendrite attenuation of synaptic potentials. The regulation of the electrical behavior of spines could be important for synaptic function, plasticity and dendritic integration.

Disclosures: V. Cornejo: None. **N. Ofer:** None. **R. Yuste:** None.

Poster

P109. LTP: Kinases and Intracellular Signaling II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P109.01

Topic: B.07. Synaptic Plasticity

Support: NIH R01 grant MH 117964 to Ted Abel
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Ted Abel is supported by Roy J. Carver Charitable Trust
Laurence Ris is supported by Belgian Queen Elisabeth Medical Foundation

Title: Mice lacking the cAMP effector protein POPDC1 show enhanced hippocampal synaptic plasticity

Authors: *M. S. SHETTY[1,2], L. RIS[4], R. F. R. SCHINDLER[5], K. MIZUNO[6], L. FEDELE[5], K. P. GIESE[6], T. BRAND[5], T. ABEL[1,3];
[1]Dept. of Neurosci. and Pharmacology, Carver Col. of Med., [2]Iowa Neurosci. Inst., [3]Dept. of Neurosci. and Pharmacol., Univ. of Iowa, Iowa City, IA; [4]Dept. of Neurosci., Univ. of Mons, Mons, Belgium; [5]Natl. Heart and Lung Inst., Imperial Col. London, London, United Kingdom; [6]Dept. of Neurosci., King's Col. London, London, United Kingdom

Abstract: Activity-dependent hippocampal synaptic plasticity is a leading candidate for the cellular mechanism underlying learning and memory. Extensive research has uncovered diverse forms of synaptic plasticity and a wide array of molecular signaling mechanisms that act as positive or negative regulators. Specifically, cAMP-dependent signaling pathways have been crucially implicated in persistent forms of synaptic plasticity. In this study, we examine the role of Popeye Domain-containing Protein POPDC1 (or BVES, Blood Vessel Epicardial Substance), a cAMP effector protein expressed in brain, in modulating hippocampal synaptic plasticity. Unlike other cAMP effectors, such as PKA and EPAC, POPDC1 is membrane-bound and the sequence of the cAMP-binding cassette differs from canonical cAMP-binding domains. These properties suggest that POPDC1 may have a unique role in cAMP-mediated signaling underlying synaptic plasticity. Our results show that POPDC1 is enriched in hippocampal synaptoneurosomes. Acute hippocampal slices from *Popdc1* knockout (KO) mice show enhanced long-term potentiation (LTP) induced by a variety of stimulation paradigms, particularly in response to weak stimulation paradigms that in slices from wildtype mice induce only transient LTP. Furthermore, *Popdc1* KO mice did not display any further enhancement in forskolin-induced LTP observed following inhibition of phosphodiesterases (PDEs), suggesting a possible modulation of cAMP-PDE signaling by POPDC1. Taken together, these data reveal POPDC1 as a novel player in the regulation of hippocampal synaptic plasticity and as a potential target for cognitive enhancement strategies.

Disclosures: M.S. Shetty: None. **L. Ris:** None. **R.F.R. Schindler:** None. **K. Mizuno:** None. **L. Fedele:** None. **K.P. Giese:** None. **T. Brand:** None. **T. Abel:** None.

Poster

P109. LTP: Kinases and Intracellular Signaling II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P109.02

Topic: B.07. Synaptic Plasticity

Title: Non-canonical and canonical AKT pathways can coexist and intersect upon E-LTP expression and memory formation

Authors: *D. GOROBETS[1], A. SHEININ[2], A. PINHASOV[3], I. MICHAELEVSKI[4];
[1]Ariel Univ. Ctr., Ariel, Israel; [2]Tel Aviv Univ., Tel Aviv, Israel; [4]Integrative Brain Sci. Ctr. - Ariel, Mol. Biol., [3]Ariel Univ., Ariel, Israel

Abstract: AKT is a serine/threonine kinase involved in the broad spectrum of processes in the nervous system as neurons growth, survival and development, growth and maturation of the neuronal spines and long-term synaptic plasticity and memory. Novel results obtained in our lab emphasize that AKT activity is obligatory for expression of early phase long-term potentiation (E-LTP). Furthermore, classical downstream pathway of AKT demonstrated its redundance for

the expression of E-LTP. We hypothesized that AKT mediates E-LTP expression via previously unknown non-canonical downstream pathway. Here we report about the interplay of E-LTP expression and AKT, its non-canonical pathway, and mTORC1/GSK3β-dependent signaling pathways. We found that: a) separate or concomitant pharmacological activation of mTORC1 as well as inhibition of GSK3β fail to alter E-LTP expression; b) concomitant inhibition of GSK3β by TWS119 and AKT by AKT inhibitor VIII (A6730) rescued E-LTP expression attenuation evoked by reduction of AKT activity; c) similarly AKT inhibition on E-LTP was partially rescued by TWS119-induced inhibition of GSK3β; d) concomitant activation of mTORC1 by dorsomorphin and inhibition of AKT restored E-LTP expression as well as activation of the mTORC1 followed AKT inhibition prevents the effect of the latter; e) overexpression of the AKT1 in the CA1 region of hippocampus leading to the diminishing of the effect of its pharmacological inhibition on E-LTP; f) overexpression or knock-down of AKT1 in the hippocampus enhance short- and long-term types of both spatial and non-spatial memory. Summarizing, we suggest that AKT non-canonical and canonical pathways may serve as compensatory mechanisms for E-LTP expression, AKT isoforms may mutually compensate each other upon downregulation and maintain expression of E-LTP.

Disclosures: D. Gorobets: None. **A. Sheinin:** None. **A. Pinhasov:** None. **I. Michaelevski:** None.

Poster

P109. LTP: Kinases and Intracellular Signaling II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P109.03

Topic: B.07. Synaptic Plasticity

Support: NIH R01NS036715

Title: Pkcζ And pkcι, are not required for hippocampal synaptic plasticity

Authors: *Q. ZHU[1], M. TIAN[2], R. C. JOHNSON[4], L. J. VOLK[5], R. L. HUGANIR[3]; [1]Neurosci., Johns Hopkins Univ. Sch. of Med., Baltimore, MD; [2]Neurosci., [3]Dept. Neurosci., Johns Hopkins Univ. Dept. of Neurosci., Baltimore, MD; [4]Johns Hopkins Univ., BALTIMORE, MD; [5]Neurosci., Univ. of Texas Southwestern Med. Ctr. at Dallas, Dallas, TX

Abstract: We previously showed using constitutive and conditional knockout (cKO) techniques that the atypical PKCζ was not required for hippocampal synaptic plasticity, learning and memory. However, subsequent studies from Todd Sacktor's laboratory have suggested that our findings were due to a compensation of the loss of PKCζ by increases in the activity of a related atypical PKC, PKCι. To investigate this further we generated conditional cKOs of both atypical PKCs. These mice are constitutive KOs of PKMζ and conditional KOs of PKCι/λ using

CaMKIIpro-CRE mice. We obtained efficient KO/KD of both isoforms of PKC, with no detectable PKMζ and ~13% of PKCι/λ in the hippocampi lysates of the double cKOs. By comparing the littermates of the atypical PKCs double KOs and the PKMζ single KOs, we found that the expression and maintenance of LTP in double KOs was not affected. Combined with the previous result that the PKMζ single KO had no effect on LTP maintenance, our data indicated both atypical PKCs are not required for LTP. Moreover, we observed that LTP maintenance in the double cKO mice was still inhibited by the well-known PKCζ inhibitor zeta inhibitory peptide (ZIP) indicating PKCι/λ and PKMζ are not the relevant targets of ZIP. These results provide compelling evidence that PKCζ is not required for LTP expression and maintenance and supports the idea that the ZIP peptide is a nonspecific inhibitor.

Disclosures: Q. Zhu: None. **M. Tian:** None. **R.C. Johnson:** None. **L.J. Volk:** None. **R.L. Huganir:** None.

Poster

P109. LTP: Kinases and Intracellular Signaling II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P109.04

Topic: B.07. Synaptic Plasticity

Support: Campus Research Board Award, UIC

Title: Post-hypoxic long-term potentiation in mice lacking glutamate antiporter, system x_c-

Authors: *B. HEIT[1], A. CHU[2], J. E. RICHMOND[2], D. E. FEATHERSTONE[2], A. MCRAY[2], J. R. LARSON[2];
[1]Univ. of Illinois At Chicago, Chicago, IL; [2]Univ. of Illinois at Chicago, Chicago, IL

Abstract: Ischemic stroke remains the leading cause of adult disability in the world. Efforts to reduce stroke severity, however, have been plagued by translational failure due to gaps in our understanding of cellular and molecular mechanisms leading to brain damage after metabolic insult. Loss of blood supply to brain tissue depletes neurons of energy, which induces a sequela of events within the neuronal network. The glial-bound cystine/glutamate antiporter, system x_c-, with specific subunit xCT, supplies 60-80% of ambient extracellular glutamate in the brain**.** Using slice electrophysiology, we have previously shown that xCT-mediated glutamate release influences both the rapidity and synchrony of depolarizing events after total oxygen deprivation (anoxia). Both genetic deletion and pharmacological antagonism of system x_c- provided ischemic neuroprotection by increasing latency to anoxic depolarization (AD), attenuating AD wave amplitudes, and extending AD wave durations. The sudden onset and rapid regenerative nature of AD, however, may obscure subtle, but important, antecedent differences. Partial hypoxia, or reduced regional blood flow *in vivo*, is an ischemic condition that alters synaptic signaling, but is

not noxious enough to induce glutamate excitotoxicity or de-stabilize transmembrane ion gradients. Mild hypoxia elicits a suppression of synaptic transmission proportional to the degree of oxygen deprivation, ATP breakdown, and accumulation of extracellular adenosine. The present study employed mild hypoxia to better understand ischemia-induced alterations in neuronal responsiveness from WT and xCT KO ($xCT^{-/-}$) mice. Hippocampal slices from both genotypes were prepared, incubated, subjected to 30 minutes hypoxia (25% O_2/75% N_2), and fully re-oxygenated. Although WT and $xCT^{-/-}$ slices did not differ in hypoxia-induced synaptic suppression, mutant slices exhibited accelerated rate of recovery and post-hypoxic potentiation, which resembled LTP. Experiments using CPX, an A_1 adenosine antagonist, showed that this differential effect was not due to enhanced adenosine release in the $xCT^{-/-}$. Contrarily, further experimentation revealed that post-hypoxic potentiation in $xCT^{-/-}$ mice is driven by activation of NMDARs and enhanced calcium influx. Thus, post-hypoxic changes in mutant mice are propagated by mechanisms similar to those involved in LTP. This latter finding was remarkable as $xCT^{-/-}$ mice showed no differences in electrically-induced LTP compared to WT. Taken together, these data confirm system x_c- as a salient regulator of neuronal responses during ischemia and a therapeutic target ripe for exploration.

Disclosures: B. Heit: None. **A. Chu:** None. **J.E. Richmond:** None. **D.E. Featherstone:** None. **A. McRay:** None. **J.R. Larson:** None.

Poster

P109. LTP: Kinases and Intracellular Signaling II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P109.05

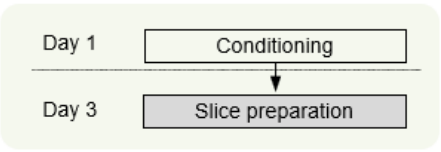
Topic: B.07. Synaptic Plasticity

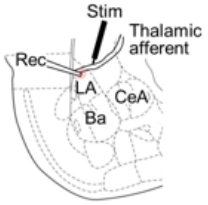
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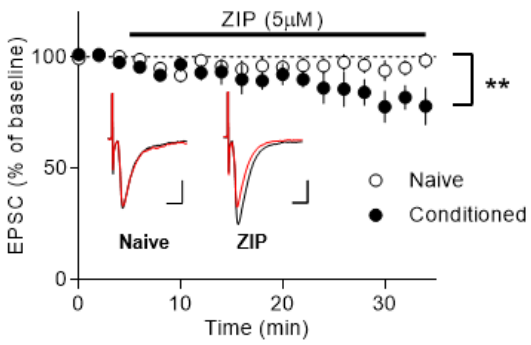
Title: Gsk-3β Activation is required for zip-induced disruption of learned fear

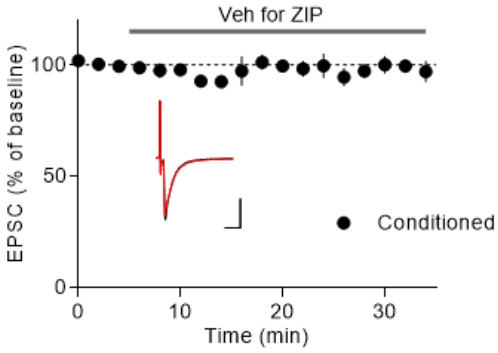
Authors: *S. SONG[1], J. KIM[2], K. PARK[3], K. PARK[3], J. LEE[4], S. PARK[4], S. LEE[1], J. KIM[1], I. HONG[5], B. SONG[6], S. CHOI[4];
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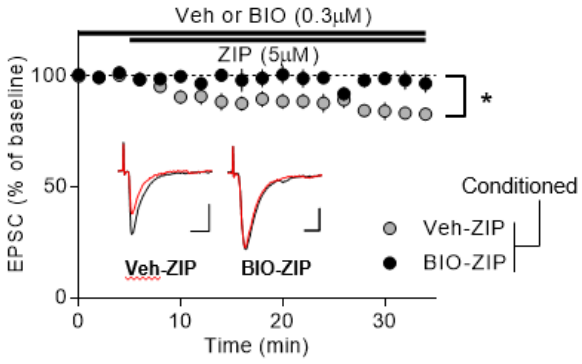
Abstract: As an peptide inhibitor for protein kinase C/Mζ (PKCζ/PKMζ), the myristoylated zeta inhibitory peptide (ZIP) is developed, and it is known to produce retrograde amnesia in various systems. However, recent findings shows ZIP induces memory loss even in the absence of PKMζ. Therefore, the mechanism of action of ZIP, remains elusive. Through a kinome-wide screen, we found that glycogen synthase kinase 3 beta (GSK-3β) was robustly activated by ZIP in vitro. While ZIP induced depotentiation (a cellular substrate of memory erasure) of fear conditioning-induced potentiation at LA synapses, it was prevented by a GSK-3β inhibitor, 6-bromoindirubin-3-acetoxime (BIO-acetoxime). Also, GSK-3β inhibition by BIO-acetoxime infusion or LA-specific GSK-3β knockdown by GSK-3β shRNA attenuated ZIP-induced disruption of learned fear. Furthermore, conditioned fear was weakened by an afterward expression of a constitutively active form of GSK-3β in the LA. Our findings suggest that GSK-3β activation leads to a disruption of memory and it is a critical step for ZIP-induced amnesic effects.

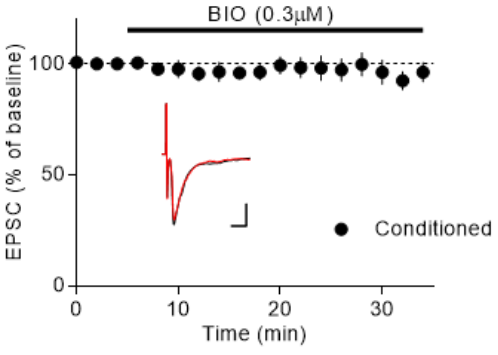


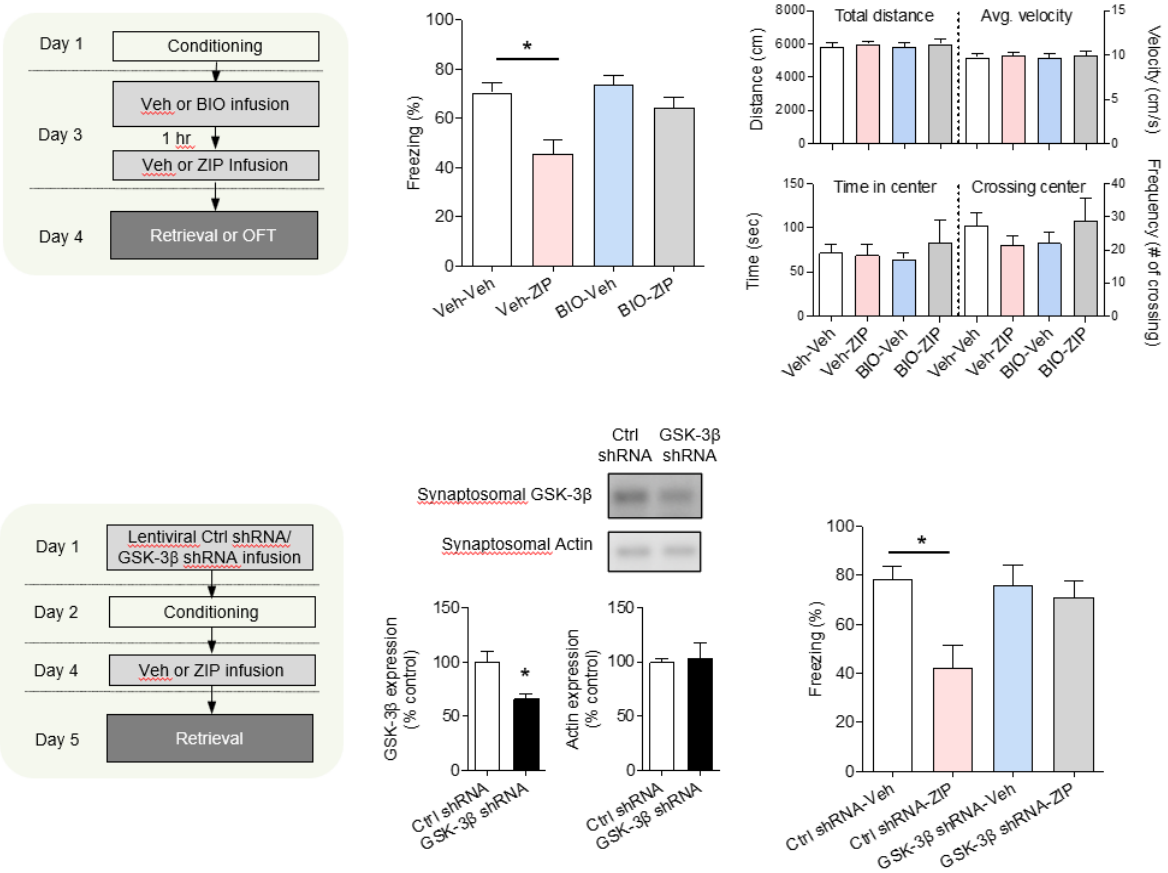












Disclosures: S. Song: None. **J. Kim:** None. **K. Park:** None. **K. Park:** None. **J. Lee:** None. **S. Park:** None. **S. Lee:** None. **J. Kim:** None. **I. Hong:** None. **B. Song:** None. **S. Choi:** None.

Poster

P109. LTP: Kinases and Intracellular Signaling II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P109.06

Topic: B.07. Synaptic Plasticity

Support: NIH NS108765 (GLH/JMB)
NIH NS108296 (GLH/JMB)
Hunter RISE grant 5R25GM060665

Title: Febrile-induced seizures during infancy promotes synaptic instability in dorsal hippocampus in adulthood

Authors: *T. RODRIGUEZ[1,2], M. L. KLOC[3], G. GUZMAN[2], S. DILAWARI[2], G. L. HOLMES[3], J. BARRY[3], P. A. SERRANO[1,2];
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Abstract: Febrile status epilepticus (FSE) in early childhood is associated with enduring cognitive impairments and temporal lobe epilepsy. However, the underlying neural mechanisms for these deficits are unknown. We used a hyperthermia-induced experimental Febrile status epilepticus (eFSE) model in infant rats and investigated the α-Amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptor, the N-methyl-D-aspartate (NMDA) receptor, and the post-synaptic density 95 (PSD95) protein levels in these rats during adulthood. AMPA receptor subunit dynamics are known to be necessary for cognition by regulating neural excitability via alteration of the c-terminal phosphorylation site for GluA1 and GluA2 subunits and their overall level of expression. The NMDA subunit NR1 plays an essential role in the calcium channel function of the receptor. The s295 phosphorylation site on PSD95 is essential for synapse stability and maturation. We performed Western blot analysis on the dorsal hippocampal tissue from Febrile status and control rats (maternal separation without seizure). In the dorsal hippocampus we show a significant decrease in GluA2, the calcium impermeable subunit important for anchoring the AMPA receptor to the synaptic membrane; a significant decrease in GluA3, the calcium permeable subunit which plays a role in synaptic strengthening; and no change in GluA1. The NR1 subunit was significantly downregulated and may contribute to a decrease in calcium influx at the post synaptic membrane. We also report a significant decrease in PSD95 and in the s295 phosphorylation site, which is important for synaptic stabilization. These results indicate that Febrile status during infancy weakens synaptic strength, destabilizes post-synaptic AMPA receptor subunits and reduces functioning of NMDA receptors in the adult dorsal hippocampus. Future work will study how these changes contribute to FSE associated memory deficits as a consequence of putative cellular hyperexcitability in different hippocampal subfields and changes to entorhinal-hippocampal synaptic efficacy.

Disclosures: T. Rodriguez: None. **M.L. Kloc:** None. **G. Guzman:** None. **S. Dilawari:** None. **G.L. Holmes:** None. **J. Barry:** None. **P.A. Serrano:** None.

Poster

P109. LTP: Kinases and Intracellular Signaling II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P109.07

Topic: B.07. Synaptic Plasticity

Support: FRM Equipe
INSERM

AMU

Title: Synaptic and intrinsic potentiation in oriens lacunosum moleculare interneurons induced by theta frequency stimulations

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Abstract: Functional plasticity in cortical neurons generally includes two complementary sides, i.e. synaptic modifications and plasticity of intrinsic neuronal excitability. In inhibitory interneurons, synaptic plasticity has been mostly investigated and except for a few rare cases, intrinsic plasticity in GABAergic interneurons is largely overlooked. Oriens lacunosum moleculare (OLM) interneurons display anti Hebbian long term synaptic potentiation (LTP) conferred by calcium permeable AMPA receptors (CP AMPAR) that preferentially open under hyperpolarization of the postsynaptic membrane. So far, LTP in OLM cells has been induced with induction protocols that are not physiologically realistic. We report here induction of both synaptic and intrinsic potentiation in OLM interneurons following stimulation of afferent glutamatergic inputs in the theta frequency range (5 Hz). LTP is induced by synaptic activation of CP AMPAR whereas long term potentiation of intrinsic excitability (LTP IE) results from the group I metabotropic glutamate receptor (mGluR1) dependent down regulation of Kv7 and hyperpolarization activated cyclic nucleotide gated (HCN) channels. We also investigate the signal transduction pathway involved in these down regulations, more specifically the role of phosphatidylinositol 4,5 bisphosphate (PiP2), phospholipase C (PLC) and protein kinase C (PKC). We conclude that theta frequency stimulations induce intrinsic plasticity and synaptic potentiation in OLM interneurons through two separated pathways.

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Poster

P109. LTP: Kinases and Intracellular Signaling II

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Topic: B.07. Synaptic Plasticity

Support: NIH R01 Grant 5R56MH095980
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NSF NN2 Grant 2014862

Title: Permissive environment of perisynaptic astroglia in hippocampal dentate gyrus during LTP

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Abstract: Perisynaptic astroglial processes (PAPs) play a major role in modulating synapse formation and synaptic transmission. Both *in vivo* and *in vitro* experiments have demonstrated the motile nature of these processes, which can affect the degree that PAPs influence synapses. Long-term potentiation (LTP), a widely studied cellular mechanism of learning and memory, is associated with increased informational content at hippocampal dentate gyrus synapses. Given the intertwined nature of astrocyte-neuron interactions, it is important to examine the role that PAPs play in LTP. While previous work has focused primarily on area CA1 of the hippocampus, we explored PAP structure during control stimulation and with plasticity following the induction of LTP in the dentate gyrus. LTP was induced in adult rats *in vivo* via delta burst stimulation administered through electrodes implanted in the medial perforant pathway. The contralateral hemisphere of each animal served as the control, and received only baseline stimulation. Serial section transmission electron microscopy was used to capture high resolution images of the dentate gyrus neuropil. Three-dimensional reconstructions were then generated from the electron micrographs and used to compare PAP structural changes in the LTP versus control conditions. The results demonstrated the highly interdigitating nature of PAPs. Approximately 80% of the synapses for both conditions exhibited PAP apposition at the axon-spine interface (ASI). This is compared to around 60% of synapses with PAP ASI apposition for area CA1. All synapses had at least some glia present at one or more of the following locations: ASI, presynaptic axon only, postsynaptic dendritic spine only, or pre- and postsynaptic structures but not at the ASI. Those synapses with PAP ASI apposition had a wider range of postsynaptic density areas compared to the synapses with no PAPs at the ASI. Interestingly, there was no significant difference in the percent of the ASI surrounded by PAPs 2 hours after LTP induction versus control. These preliminary results suggest the increased information content for synapses occurs in close proximity to astrocytic processes. Moreover, additional variables beyond the degree of ASI apposition, such as subcellular resource distribution or surface-to-volume ratio, may be the important determinants of the effects that PAPs exert at dentate gyrus synapses.

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Poster

P148. Epilepsy: Animal Models

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Topic: B.10. Epilepsy

Support: NIH CA096236
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Title: Adgrb1 (Bai1) deletion leads to social deficits, increased seizure susceptibility, and altered brain development

Authors: *F. SHIU[1], J. C. WONG[1], R. H. PURCELL[2], T. LALA[3], S. OWINO[4], D. ZHU[5], E. G. VAN MEIR[6], R. A. HALL[7], A. ESCAYG[1];
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Abstract: The adhesion G protein-coupled receptor BAI1/ADGRB1 plays an important role in suppressing angiogenesis, mediating phagocytosis, and acting as a tumor suppressor. Previous research demonstrated in vitro and in vivo knockdown of BAI1 leads to immature dendrite formation while overexpression leads to dendrite retraction. BAI1 also functionally interacts with autism-relevant proteins such as Neuroligin 1 and IRSp53. However, little is known about the relationship between altered BAI1 function and clinically relevant neurological phenotypes. Therefore, we evaluated the effect of reduced Bai1 expression on behavior, seizure susceptibility, and brain morphology in Adgrb1 mutant mice. We compared adult homozygous (*Adgrb1*$^{-/-}$), heterozygous (*Adgrb1*$^{+/-}$), and wild-type (WT) littermates using a battery of tests to assess social behavior, anxiety, repetitive behavior, locomotor function, and seizure susceptibility. We found that *Adgrb1*$^{-/-}$ mice showed deficits in cognition and social behavior, and increased vulnerability to induced seizures. *Adgrb1*$^{-/-}$ mice also weighed significantly less than littermates beginning at P15 for males and P9 for females, but body weights were comparable between same-sex littermates by P37. We also compared brain weights between Adgrb1 mutants and WT littermates. While no significant differences were observed in P1 mice, average brain weights were lower in *Adgrb1*$^{-/-}$ mice compared to *Adgrb1*$^{+/-}$ and WT littermates at 3 weeks and 2-3 months after birth. Reduced neuron density was observed in 2-3 months old *Adgrb1*$^{-/-}$ mice, while levels of astrogliosis and microgliosis were comparable to WT littermates. Finally, higher levels of apoptosis were observed in the hippocampus and cortex of *Adgrb1*$^{-/-}$ mice at P1, but not at later timepoints (3 weeks and 2-3 months). These results suggest that Bai1 dysfunction might be associated with a broader range of clinically relevant neurological phenotypes than previously reported.

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Poster

P148. Epilepsy: Animal Models

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Program #/Poster #: P148.02

Topic: B.10. Epilepsy

Support: Senior Research Fellowship (ICMR Grant 45/01/2019-PHA/BMS)
IBRO International Travel Grant

Title: Setting a new platform for antiseizure drug testing: a mouse model with impaired mitochondrial oxidative phosphorylation recapitulates the human drug-resistant epilepsy.

Authors: *S. KUMAR[1], A. A. LAGUNIN[2], R. K. GOEL[1];
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Abstract: Epilepsy is characterized by recurrent disabling seizures. Every third patient with epilepsy remains resistant to drug therapy with devastating consequences on the quality and expectancy of the patient's life. The mechanisms governing drug-resistant epilepsy are diverse and imperfectly known therefore, recreating drug-resistant epilepsy in laboratory animals continues to be a challenge in epilepsy research. We propose to select a clinically relevant drug-resistance mechanism, using system biology and network pharmacology approach, to produce a new animal model with high face, construct, and translational validity. Several databases have been searched to gather information on 42-licensed antiseizure drugs (Drugs@FDA Database), 175-molecular drug targets (DrugBank and DrugCentral), and 601-epilepsy-gene associations (DisGeNET, and Disease-Connect). Collected information has been transformed into two datasets (Venn analysis) i.e. Disease-drug targets (n=58) and Disease non-drug targets (n=543). The most suitable mechanism of drug-resistance i.e. Mitochondrial oxidative phosphorylation has been extracted from 543 disease non-drug targets (using DAVID web-service and DIAMOnD algorithm) and manually verified from clinical reports. For in-vivo modeling, a commercially available specific inhibitor of mitochondrial complex-I was injected into the adult mice via an intraperitoneal route (2.5mg/kg rotenone). Rotenone pre-treated mice were daily exposed to electric shock (20V, 15 mA, 6Hz) twice a day for ~15 days or injected with chemoconvulsant (35mg/kg pentylenetetrazole, i.p) for ~35 days. Drug resistance epilepsy was characterized by licensed antiseizure drugs applied as described by International League Against Epilepsy. According to our observations, the rotenone kindling model shows significant resistance ($P<0.05$) to Lamotrigine (15mg/kg), Levetiracetam (40mg/kg), Carbamazepine (40mg/kg), Zonisamide (100mg/kg), Gabapentin (224mg/kg), Pregabalin (30mg/kg), Phenytoin (35mg/kg), Topiramate (300mg/kg), Valproate (200mg/kg), and two drug combinations (lamotrigine + aforementioned drugs), with less than 10% mortality. Pre-clinical drug testing through the rotenone-kindling model may uncover more effective drug therapies for the management of drug-resistant epilepsy because the rotenone-kindling model shows resistance to third-generation antiseizure drugs as well as their two-drug combinations.

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Poster

P148. Epilepsy: Animal Models

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Topic: B.10. Epilepsy

Support: Interagency agreement (AOD21005-001 00000) between the NIH and the USAMRICD

Title: Comparison of soman-induced epileptogenesis and neuropathology in juvenile and adult male rats treated with midazolam

Authors: D. A. NGUYEN[1], B. MARRERO-ROSADO[1], C. R. SCHULTZ[1], M. F. STONE[1], M. DE ARAUJO FURTADO[2], *L. A. LUMLEY[1];
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Abstract: Organophosphorus nerve agents (OPNAs) are acetylcholinesterase inhibitors that lead to pharmacoresistant *status epilepticus* when treatment with benzodiazepines is delayed. In adult male rodents, exposure to the OPNA soman leads to extensive loss of neurons, epileptogenesis and functional impairment when seizure is unmitigated. While the majority of research on OPNA exposure has evaluated its short-term effects in adult male rodents, much less is known about the detrimental responses in younger animals. Our laboratory previously reported that young rats (postnatal day [PND] 7 and 14) are highly susceptible to the lethal effects of OPNAs, while juvenile (PND42) rats are less susceptible compared to adults. This current study evaluated age differences in the development of epileptogenesis, neurodegeneration and neuroinflammatory responses following soman exposure. **Methods.** Juvenile and adult male and female rats implanted with telemetry transmitters for electroencephalographic recordings were exposed subcutaneously (SC) to an equitoxic 1.2 x median lethal dose of soman (PND42: female, 159.6 μg/kg; male 224.4 μg/kg; PND70: female, 88.8 μg/kg; male: 118.1 μg/kg) and treated intramuscularly (IM) with atropine sulfate (2 mg/kg) and HI-6 dimethanesulfonate salt (DMS; 118.5 mg/kg) 1 min after exposure, and with midazolam (3 mg/kg; IM) 40 min after seizure onset. Rats were euthanized at 3, 14 or 30 days after exposure for evaluation of neuronal degeneration and microglial activation. **Results.** Delayed midazolam treatment did not prevent epileptogenesis, neurodegeneration or neuroinflammation following soman exposure. Fewer juvenile rats exposed to soman tended to develop spontaneous recurrent seizure compared to adult rats. By 2 weeks after soman exposure, more extensive neuronal loss and greater microglial activation were observed in adult rats compared to juvenile rats in the thalamus, amygdala and piriform cortex. **Conclusions**. This study further demonstrates that delayed treatment with

midazolam is not fully protective against soman-induced epileptogenesis and neuropathology, exemplifying the need for adjunct anti-seizure treatment. Also, juvenile rats had better outcomes compared to adult rats treated with delayed midazolam, despite receiving a greater dose of soman. The views expressed are solely those of the authors and do not necessarily represent the official views of the CCRP, NIAID, NIH, HHS, USAMRICD or DoD

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Poster

P148. Epilepsy: Animal Models

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Fondation pour la Recherche Médicale, grant FDT202012010523
Fondation Assitance Publique-Hôpitaux de Paris (EPIRES, Marie Laure PLV Merchandising)

Title: Focal cortical inhibition of voltage-dependent potassium channels recapitulates the human tonic-dystonic seizures in anti-LGI1 encephalitis.

Authors: *P. BAUDIN[1,2], S. WHITMARSH[1,2], L. COUSYN[1,2,3], D. ROUSSEL[1,2], S. LECAS[1,2], K. LEHONGRE[1,2], S. CHARPIER[1,2], S. MAHON[1,2], V. NAVARRO[1,2,3];
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Abstract: Autoimmune encephalitis associated with auto-antibodies directed against the Leucin rich, glioma inactivated 1 protein (anti-LGI1 AIE) is responsible for pathognomonic tonic-dystonic seizures (TDS). Several anti-LGI1 antibodies-related changes in neuronal activity and excitability, notably associated with dysfunctions in voltage-dependent potassium channels and AMPA receptors, have been previously reported. However, their relation to seizures remains unknown. We developed a new *in vivo* experimental model to test the hypothesis that a focal cortical inhibition of voltage-dependent potassium channels could recapitulate the human TDS. Comparing electro-clinical features of TDS in the model with those recorded in a cohort of anti-LGI1 AIE patients revealed striking similarities between human and rodent seizures, including a similar frequency of recurrence and motor semiology, and their systematic precession by a large-amplitude slow EEG wave. By combining multi-site extracellular and intracellular recordings of cortical pyramidal neurons in sedated rats, we demonstrated that a sequence of changes in

neuronal excitability and synaptic activity concurs to initiate the massive membrane depolarization, and associated high-frequency firing, underlying the paroxysmal EEG activity. This new *in vivo* animal model could serve future investigations for further delineation of the ictogenesis mechanisms in this frequent auto-immune encephalitis.

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Poster

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Semmes Foundation, Inc. (JH)

Title: Enhanced GABA-mediated calcium activity regulates aberrant neurogenesis in a mouse model of epilepsy.

Authors: *Z. LYBRAND[1], S. GOSWAMI[2], J. ZHU[3], V. JARZABEK[2], N. MERLOCK[2], M. AKTAR[4], C. SMITH[2], L. ZHANG[3], P. VARMA[2], K. CHO[5], S. GE[6], J. HSIEH[2];
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Abstract: Aberrant neurogenesis is the generation of pro-epileptic adult-born granule cells (abGC) in the mouse hippocampus that is characterized by abnormalities abGC migration, dendritic development, intrinsic excitability, synaptic connectivity, and is associated with spontaneous recurrent seizures (SRS). In this study, we identified enhanced intracellular calcium events associated with early stages of aberrant neurogenesis in a pilocarpine mouse model of epilepsy. Using chemogenetic tools to prevent this increased calcium activity selectively in abGC at different stages of maturation, we identify a critical window that promotes the aberrant maturation of abGCs. Suppressing this calcium activity during this critical window of aberrant neurogenesis prevents the abnormal development of abGCs and reduces SRS. Using mono-

synaptic pseudotyped rabies tracing, we further demonstrated that chemogenetic modulation of calcium activity in aberrant abGCs decreased recurrent CA3 back-projections and restored proper entorhinal connections typically lost in pilocarpine models of epilepsy. Furthermore, we demonstrate that this enhance calcium activity that promotes aberrant neurogenesis is mediated by early GABA A receptor activity. These results illustrate the pathological potential of adult-born granule cells in the hippocampus. Despite their limited number, abGCs have an unlimited capacity to disrupt the hippocampus circuitry.

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Poster

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Topic: B.10. Epilepsy

Support: NIH Grant R15NS088776

Title: Memory disruption and alterations of the PI3K/AKT/mTOR signaling pathway following single kainate induced seizure

Authors: *G. SULLENS, K. BLANDIN, D. NARVAIZ, P. D. WOMBLE, D. SANTANA COELHO, J. N. LUGO, JR;
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Abstract: Individuals with epilepsy report memory deficits and disturbances as a major quality of life concern. Most studies have focused on the long-term effects of kainate (KA) induced status epilepticus (continuous seizures) on learning and memory. We examined the acute effects of a single KA induced seizure on learning and memory following a single seizure episode. Male and female C57BL/6J mice underwent Delay Fear Conditioning one or six hours after cessation of seizures following 10mg/kg i.p injection of kainic acid. Mice were tested for contextual and cued memory either 24 hours or one week after seizure induction. KA induced seizures resulted in a contextual fear memory deficit 24 hours later for both one- and six-hour training cohorts. Mice that received KA seizures had a deficit in contextual fear memory when trained one hour later ($p < .05$), but there was no effect of KA nor training time for cued fear memory 24 hours after seizure induction. One week after seizure induction, only mice that seized one hour prior to training showed a cued fear memory deficit compared to saline controls trained at the same time and both KA and saline controls trained six hours after recovery, $p < .05$. Levels of AKT, pAKT, S6, and pS6 were measured by western blotting as indicators of alterations in the mTOR

signaling pathway in the hippocampus. Twenty-four hours after seizure induction there was an overall decrease in total AKT for KA mice compared to controls, $p < 0.01$. This decrease was not present one week later. No alterations in total S6 were observed at either time point. KA increased levels of pS6/S6 and pAKT/AKT 24 hours after seizure induction compared to controls, $p < 0.01$, but not one week later. We also measured changes in the levels of microglia and astrocytes in the hippocampus. We found Iba1 was significantly increased after seizures compared to controls 24 hours and one week after seizure induction, $p < .05$. This study adds to the growing literature which indicates time dependent learning and memory deficits after a single seizure. Additionally, this study indicates there are transient alterations in the mTOR signaling pathway and persistent inflammatory responses within the hippocampus which may play a role in the memory disturbances observed.

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Poster

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Topic: B.10. Epilepsy

Support: FAPEG
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CAPES

Title: Responses to Obstructive Sleep Apnea and Chemoreflex Stimulation in Rats with Epilepsy Induced by Pilocarpine

Authors: C. QUINTINO[1], P. P. GHAZALE[2], P. P. BRAGA[1], K. P. GOMES[1], C. E. G. OLIVEIRA[1], D. H. MATTA[1], G. H. SCHOORLEMMER[2], T. S. MOREIRA[3], E. P. MENDES[1], C. H. CASTRO[1], D. B. COLUGNATI[1], ***A. P. PANSANI**[1];
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Abstract: Obstructive sleep apnea (OSA) and chemoreflex alterations are common in patients with epilepsy and may contribute to sudden unexpected death in epilepsy (SUDEP). The aim of our study was to characterize in rats with chronic epilepsy 1) the latency to awake from obstructive apneas made in sleep, 2) changes in heart rate and respiratory effort during obstructive apnea in sleep, and 3) peripheral chemoreflex sensitivity. Epilepsy was induced with pilocarpine (350 mg/kg, i.p.) in 6 male Wistar rats. Six control rats received a saline injection. Sixty days after the first spontaneous seizure, we implanted a tracheal balloon to induce

obstructive apnea, EEG and EMG electrodes to detect sleep phases, ECG electrodes to measure heart rate, and a thoracic balloon to record respiratory effort (thoracic pressure variation). For 8 hours during the light period, we induced apneas (up to 1 apnea/min) during sleep. Each apnea ended as soon as the rat awoke (triggered by increased muscle tone). Subsequently, cannulas were implanted in the femoral artery and vein. The next day, the peripheral chemoreflex was triggered by i.v. injection of 40 µg/kg potassium cyanide (KCN). The apnea duration showed interaction between group and sleep phase in the median quartile. Both control and epileptic rats woke up after about 6.4 s when apneas were made during NREM. However, when apneas were made during REM, the latency to awake was lower in rats with epilepsy than in controls (median: 7.3 vs. 8.2 s, linear regression model and t-test, $p < 0.01$). To analyze the effect of epilepsy on respiratory effort and heart rate during apnea, we selected apneas with a duration of 10 s, and compared the mean curves of response against time by functional ANOVA. Respiratory effort increased, and heart rate fell during apneas made in REM and NREM sleep, but the mean curves in epileptic rats were not different from those in controls (functional ANOVA and Cuevas Heteroscedastic - CS test, $p > 0.05$). KCN injection induced transient bradycardia, respiratory effort, and hypertension in both groups. In epileptic subjects, but not in controls, the pressor response was followed a transient hypotension (functional ANOVA and CS test, $p<0.05$). In conclusion, our study demonstrated that rats with epilepsy had a reduced latency to awake from apnea in REM, but not in NREM, which could be possible related with alteration on cholinergic system. The changed arterial pressure response after chemoreflex stimulation in rats with epilepsy suggests changes in sympathetic control. Further studies should be performed to understand if and how these alterations affect SUDEP risk.

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Poster

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Title: Impact of raptor or rictor deletion on hippocampal dentate granule cell morphology during epileptogenesis

Authors: *C. M. GODALE[1], N. MAYER[2], E. PARKINS[1], K. KRAUS[1], C. GROSS[3], S. C. DANZER[3];

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Abstract: The mechanistic target of rapamycin (mTOR) signaling pathway is hyperactivated during epileptogenesis. The central regulators of this pathway are mTOR complexes, mTORC1 and mTORC2, which are controlled by the regulatory-associated protein of mTOR (Raptor) and the rapamycin-insensitive companion of mTOR (Rictor), respectively. It is known that hippocampal dentate granule cells are a target of mTOR hyperactivation in temporal lobe epilepsy (TLE). However, the exact roles played by mTORC1 and mTORC2 in regulating pathologies found in TLE are unknown. Granule cell pathology has been hypothesized to mediate the development of TLE; therefore, understanding how mTOR regulates granule cell dysmorphogenesis may have therapeutic benefits. We developed a viral cre-recombinase driven strategy to produce sparse deletions of Raptor or Rictor from hippocampal dentate granule cells in mice during epileptogenesis. Sparse deletions were titrated to produce knockout (KO) cells in each mouse without impacting epileptogenesis. One week after pilocarpine-induced status epilepticus (SE), Raptor$^{fl/fl}$, tdTomato$^{+/-}$ or Rictor$^{fl/fl}$, tdTomato$^{+/-}$ mice received hippocampal injections of AAV9.CamKII.HI.eGFP and AAV9.CamKII.Cre, producing a mixture of eGFP+ wildtype and tdTomato+ Raptor or Rictor knockout cells in each mouse. Animals were video monitored for seizures during week seven, and brains were collected at the end of the eighth week for histology. Soma area measurements revealed Raptor KO cells had consistently smaller soma areas than adjacent wildtype cells, while Rictor KO cells were statistically identical to wildtype cells. Studies are ongoing to reveal the roles of mTORC1 and mTORC2 signaling during epileptogenesis in granule cell TLE pathologies.

Disclosures: C.M. Godale: None. **N. Mayer:** None. **E. Parkins:** None. **K. Kraus:** None. **C. Gross:** None. **S.C. Danzer:** None.

Poster

P148. Epilepsy: Animal Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P148.09

Topic: B.10. Epilepsy

Support: NIH Grant NS35439, NS108296

Title: Cell-type specific expression and function of NRSF in the developing dentate gyrus: lessons from epilepsy

Authors: *Y. CHEN, N. KAMEI, J. DAGLIAN, K. CHEN, A. L. PHAM, T. Z. BARAM; Univ. of California Irvine, Irvine, CA

Abstract: Background: Neuron-restrictive silencing factor (NRSF) is a transcriptional repressor of many activity-dependent neuronal genes that carry NRSF-response elements (NRSE). NRSF is upregulated by seizures and insults, which promotes epileptogenesis by regulating a group of genes that critically influence hippocampal neuronal function. We have reported that experimental febrile status epilepticus (eFSE) leads to epileptogenesis and disruption of spatial memory network development yet blocking the activity of hippocampal NRSF after FSE ameliorates spatial memory defects and selectively rescues dendritic structure of dentate gyrus (DG) granule cells. The granule cell layer contains cells at different developmental stages and cells of different ages contribute differently to the formation of an aberrant neuronal circuit. Notably, the expression and function of NRSF varies by cell type and age and some granule cells may not express NRSF after eFSE. To determine the contribution of NRSF to eFSE-induced DG network disruption at the required resolution in time and space, we investigate the cell-type specific expression of NRSF in the developing DG and the developmental stage of granule cells expressing NRSF after eFSE. **Methods:** eFSE were generated as described, by increasing core and brain temperatures of P10-11 rats to >38.5°C for 30 min, which generates limbic FSE-like seizures. NRSF expression was detected via single cell resolution in situ hybridization, validated by NRSF ICC, then coupled with ICC of several cellular markers. The age of NRSF-expressing cells was determined using Edu birth-dating. **Results:** Within 48-72h, eFSE upregulated the expression of NRSF in selective subpopulations of DG cells. These were neurons, apparent from NeuN expression and absence of the microglial marker Iba1. NRSF-expressing cells in the GC layer co-expressed Prox1 indicating that they are granule cells. In the subgranular zone (SGZ) of eFSE rats, NRSF cells co-expressed GFAP, suggesting their immaturity. Studies of overt NRSF expression in newborn, infant, or mature GCs are ongoing.

Disclosures: Y. Chen: None. **N. Kamei:** None. **J. Daglian:** None. **K. Chen:** None. **A.L. Pham:** None. **T.Z. Baram:** None.

Poster

P151. Antiepileptic Therapies III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P151.01

Topic: B.10. Epilepsy

Support: NINDS R01 NS34509

Title: Genetic compensation at the AIS: Scn8a, Kcna1 and Kcnq2.

Authors: *S. HILL[1], P. JAFAR-NEJAD[2], F. RIGO[3], M. H. MEISLER[4];
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Abstract: The neuronal voltage-gated sodium channel $Na_v1.6$ is concentrated at the axon initial segment (AIS), where it plays a key role in initiation of action potentials. Gain-of-function mutations in *SCN8A* are a cause of developmental and epileptic encephalopathy (DEE). Administration of an antisense oligonucleotide (ASO) that reduces expression of *Scn8a* results in delayed seizure onset in a mouse model of *Scn8a* DEE, and this ASO also rescues survival of Dravet mice with mutation of *Scn1a* (*Lenk et al, Ann. Neurol. 87:339-346, 2020*). We asked whether reduction of *Scn8a* expression could ameliorate the seizure phenotypes of mice with mutations in two genes encoding potassium channels that are also localized at the AIS: *Kcnq2* and *Kcna1*. Mice with homozygous mutations of *Kcna1* (*Smart et al Neuron 1998*), *Kcnq2* (*Soh et al, J. Neurosci. 34:5311-5321, 2014*) and *Lgi1* (*Chambrol et al, Brain 133:2749-2762*) were treated on postnatal day 2 by intracerebroventricular injection of the ASO that reduces the level of *Scn8a* transcript by 50%. Surviving mice were given repeated doses of the ASO at four-week intervals. The number of convulsive seizures and length of survival were monitored. In both the *Kcna1* and *Kcnq2* mutant mice, treatment with the *Scn8a* ASO significantly increased the length of survival, demonstrating functional interaction between ion channels at the AIS. We also examined the effect of *Scn8a* reduction on mice with mutation of the synaptic protein LGI1, which resulted in an increase in survival from 3 weeks to 4 weeks. The data demonstrate amelioration of genetic epilepsies by modulation of AIS-localized ion channel expression. An *SCN8A* ASO may have therapeutic relevance for a range of genetic epilepsies.

Disclosures: S. Hill: None. **P. Jafar-Nejad:** A. Employment/Salary (full or part-time):; Ionis Pharmaceuticals. **F. Rigo:** A. Employment/Salary (full or part-time):; Ionis Pharmaceuticals. **M.H. Meisler:** None.

Poster

P151. Antiepileptic Therapies III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P151.02

Topic: B.10. Epilepsy

Support: Programa E022-INP 085/2010
CONACYT-86784

Title: Kcc2 expression in dentate gyrus of rat fed ketogenic diet with seizures induced by amygdala kindling

Authors: *T. E. JUÁREZ ZEPEDA[1], C. RUBIO[2], V. CUSTODIO[2], L. E. BAENHA ALVAREZ[1], L. HERNÁNDEZ LÓPEZ[2], K. JERÓNIMO CRUZ[1], J. G. MARTÍNEZ-GALINDO[2], M. TAPIA RODRÍGUEZ[3], E. COBALLASE URRUTIA[1], L. CARMONA APARICIO[1], A. VANOYE CARLO[1], C. PAZ TRES[2], L. GRANADOS ROJAS[1];

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Abstract: Ketogenic diet (KD), is a high fat, low carbohydrate and adequate protein regimen, which has been used as a non-pharmacological treatment of refractory epilepsy. However, their mechanism of action is unknown. In patients with refractory epilepsy and in rat epilepsy models, KCC2 expression cation-chloride cotransporter is down-regulated. KCC2 transports chloride out of neurons, contributing thus the intracellular concentration of this ion, which determines the strength and polarity of the gamma-aminobutyric acid (GABA)-mediated neurotransmission. Alterations in homeostasis of KCC2 could explain the hyperexcitability observed in epilepsy. Therefore, the aim of this study was to analyze the effect of the KD on cotransporter KCC2 expression in the three layers of the dentate gyrus, in rats with seizures induced by amygdala kindling. Groups of intact animals and with kindling fed on normal or ketogenic diet were evaluated. 50 μm thick cuts of the brain were obtained and processed with immunodetection techniques for the KCC2 cotransporter. Subsequently, the optical density of the immunohistochemical staining of the KCC2 cotransporter was obtained to evaluate its expression. The results indicated that animals fed a ketogenic diet increase the concentration of glucose and decrease the concentration of β-hydroxybutarate. Kindling animals fed a ketogenic diet reduced their body weight. The optical density of the KCC2 cotransporter in the three layers of the dentate gyrus was reduced in kindling animals. The ketogenic diet reversed the reduction of the KCC2 cotransporter produced by kindling in all three layers of the dentate gyrus. Also, ketogenic diet per se increased the expression of the KCC2 cotransporter. It is likely that the mechanism of action of the ketogenic diet to control seizures is through the modulation of the expression of the KCC2 cotransporter.

Disclosures: T.E. Juárez Zepeda: None. **C. Rubio:** None. **V. Custodio:** None. **L.E. Baenha Alvarez:** None. **L. Hernández López:** None. **K. Jerónimo Cruz:** None. **J.G. Martínez-Galindo:** None. **M. Tapia Rodríguez:** None. **E. Coballase Urrutia:** None. **L. Carmona Aparicio:** None. **A. Vanoye Carlo:** None. **C. Paz Tres:** None. **L. Granados Rojas:** None.

Poster

P151. Antiepileptic Therapies III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P151.03

Topic: B.10. Epilepsy

Support: NINDS R21NS114795-01

Title: Cannabidiol administration significantly improves social behavior and increases seizure resistance in an Scn8a mouse model

Authors: *J. C. WONG, L. SHAPIRO, A. ESCAYG;
Emory Univ., Atlanta, GA

Abstract: An increasing number of *de novo SCN8A* mutations have been identified in patients with catastrophic, treatment-resistant childhood epilepsy. *De novo SCN8A* mutations have also been found in patients with autism, intellectual disability and developmental delay but with less severe epilepsy. Since *SCN8A*-associated epilepsy mutations have only recently been identified, it is unclear which treatments are the most efficacious for these patients. Furthermore, attempts at seizure control are not always effective at improving behavioral outcomes and can sometimes exacerbate behavioral abnormalities. Recent clinical trials demonstrate that cannabidiol (CBD) significantly reduces seizure frequency in patients with Dravet syndrome and Lennox-Gastuat syndrome. It is hypothesized that the antiepileptic effects of CBD may be mediated, in part, by reducing resurgent sodium currents. In HEK293T cells, CBD was shown to reduce resurgent current in both wild-type (WT) $Na_v1.6$ and mutant $Na_v1.6$ channels, suggesting that CBD may be beneficial in *SCN8A*-associated disease.

Using CRISPR/Cas9, we knocked in the human *SCN8A* R1620L mutation into the corresponding position in the mouse *Scn8a* gene. We previously showed that heterozygous mutants (RL/+) exhibit increased seizure susceptibility and a range of behavioral abnormalities, including hyperactivity and social behavior deficits. To evaluate whether CBD could protect against induced seizures, RL/+ mutants and WT littermates were administered 200-360 mg/kg CBD and seizures were induced using the 6 Hz paradigm and pentylenetetrazole (PTZ). We found that 320 mg/kg CBD was the minimum dose required to observe a statistically significant increase in protection against 6 Hz seizures in the RL/+ mutants (N = 10-12/dose). CBD (200 mg/kg) was able to increase resistance to PTZ-induced seizures in WT littermates; however, a higher dose of CBD (360 mg/kg) was required to significantly increase resistance in the RL/+ mutants (N = 9/group).

To determine whether CBD could also improve behavior, RL/+ mutants and WT mice were administered 10-100 mg/kg CBD two hours prior to behavioral testing. The mice were subjected to a battery of behavioral assessments to evaluate locomotor activity, motor coordination, and social behavior. CBD was able to reduce locomotor activity and restore normal social behavior in the RL/+ mutants at doses of 100 mg/kg and 10 mg/kg, respectively (N = 12-13/group). Our results demonstrate that CBD can significantly increase seizure resistance and improve social behavior in an *Scn8a* mouse model. These findings suggest that CBD may be therapeutic in patients with *SCN8A*-associated disease.

Disclosures: J.C. Wong: None. **L. Shapiro:** None. **A. Escayg:** None.

Poster

P151. Antiepileptic Therapies III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P151.04

Topic: B.10. Epilepsy

Title: Novel synthetic cannabidiol enantiomer modulates delta and beta wavelength EEG activity

Authors: *A. CONTRERAS[1], W. A. MAIO[2], D. J. HINES[1], R. M. HINES[1];
[1]Psychological and Brain Sci., Univ. of Nevada, Las Vegas, Las Vegas, NV; [2]New Mexico State Univ., Las Cruces, NM

Abstract: Novel synthetic cannabidiol enantiomer modulates δ and β wavelength EEG activity April Contreras[1], William A. Maio[2], Dustin J. Hines[1], and Rochelle M. Hines[11]Interdisciplinary Neuroscience PhD Program, Department of Psychological and Brain Sciences,University of Nevada, Las Vegas[2]Department of Chemistry, New Mexico State University
Cannabidiol (CBD) is the major non-psychotropic cannabinoid (CB) of *Cannabis* with therapeutic potential as an anticonvulsant and anxiolytic. Compared to other constituents of *Cannabis*, CBD has a low affinity for CB1 and CB2 receptors. Novel synthetic CBD derivatives have been designed to improve affinity for CB receptors and receptor activation, however little is known about the behavioral and electroencephalographic (EEG) effects of novel CBD congeners. In the present study, we examine the effects of caraway derived CBD congeners in mice to further our understanding of these compounds, with the larger goal of developing novel therapeutics for childhood epilepsy syndromes and anxiety disorders. A series of (+)-*enantiomer*-CBD congeners ((+)-*ent*-CBD) with elongated alkyl chains were synthesized using an innovative 4-step method using conversion of (+)-carvone into isopiperitenol. (+)-*ent*-CBD congeners with elongated alkyl chains were administered to mice via oral gavage following one hour of EEG baseline recording. Three of the (+)-*ent*-CBD congeners elicited a significant change in the beta (β) frequency of the EEG compared to baseline. Of these, the congener with the longest alkyl chain ((+)-*ent*-CBD-**8**) had particularly potent effects on both the β and delta (δ) frequency bands. This suggests that variations in alkyl chain length of synthetic congeners may subtly modulate central nervous system activity. Our findings contribute to the understanding of novel synthetic CBD derivatives on neuronal signaling, with major implications for the development of novel anticonvulsant and anxiolytic therapies which target CB1 and CB2 receptors with differential specificity.
Keywords Cannabidiol (CBD), cannabinoid (CB), CB1, CB2, synthetic derivative, enantiomer, congener, modulation, EEG

Disclosures: A. Contreras: None. **W.A. Maio:** None. **D.J. Hines:** None. **R.M. Hines:** None.

Poster

P151. Antiepileptic Therapies III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P151.05

Topic: B.10. Epilepsy

Title: Comparative potencies of cannabidiol (CBD), cannabigerol (CBG) and cannabichromene oil (CBC oil) in the mouse maximal electroshock test

Authors: *A. DHIR[1], S. FERBER[2], M. MASCAL[3], N. SHEVCHENKO[3], M. A. ROGAWSKI[1]; [1]Dept. of Neurology, Sch. of Med., Univ. of California, Davis, Sacramento, CA; [2]Flawless Creations, Los Angeles, CA; [3]Dept. of Chem., Univ. of California, Davis, CA

Abstract: Cannabidiol (CBD), a biologically active constituent of the cannabis (hemp) plant, has been approved for the treatment of seizures associated with Lennox-Gastaut syndrome, Dravet syndrome, and tuberous sclerosis complex. CBD protects against tonic hindlimb extension (THLE) in the mouse maximal electroshock (MES) test, a standard model used in the identification of antiseizure agents. Although CBD has been extensively studied, many other plant cannabinoids have not been assessed for potential utility in epilepsy therapy. The current study determined the effectiveness and potencies of cannabigerol (CBG) and cannabichromene (CBC) in the mouse MES test. Samples of CBD, both from decarboxylation of cannabidiolic acid (CBDA) and a commercial source, were tested in parallel as a comparator. Samples of CBD and CBG were prepared by decarboxylating their respective active precursors CBDA and cannabigerolic acid (CBGA; 95+% pure) by heating at 102°C for 60 min. The precursor compounds were isolated from hemp. For comparison, the antiseizure activity of pure, commercial, hemp-derived CBD and chemically synthesized CBG in crystalline form was also determined in the MES test. We also investigated whether CBDA and CBGA have antiseizure potential at the highest dose tested (150 mg/kg) in this model. Male CF1 mice were used in the current study. Sixty min after IP injection of the cannabinoids, the animals were subjected to a 0.2-s, 60-Hz, 50-mA electrical stimulus delivered with corneal electrodes (5 mm diameter stainless steel balls) wetted with 0.9% saline. Animals failing to show THLE were scored as protected. CBD obtained from CBDA isolates protected animals from THLE at doses between 25-150 mg/kg with an ED50 value of 62.7 ± 5.6 mg/kg. CBG obtained from CBGA was also protective in a similar range of doses 65-200 mg/kg with an ED50 value of 99.03 ± 7.01 mg/kg. A commercial sample of crystalline, hemp-derived CBD and crystalline, synthetic CBG had similar efficacies and potencies. The ED50 value of commercial CBD was 83.7 ± 5.7 mg/kg. Pure crystalline CBG protected 62.5%, 75% and 62.5% of animals at 100 mg/kg, 150 mg/kg and 200 mg/kg. In contrast, CBC oil was less potent than CBD and CBG. At doses of 50, 100, 150 and 250 mg/kg, the fraction of animals protected was 11.1%, 12.5%, 12.5% and 25%, respectively. CBDA had no antiseizure activity at the highest dose tested whereas CBGA protected 25% of animals tested. The rank order of potencies of the cannabinoids was CBD > CBG > CBC. CBG is sufficiently effective and potent that it may have clinical utility.

Disclosures: A. Dhir: A. Employment/Salary (full or part-time):; University of California, Davis. **S. Ferber:** None. **M. Mascal:** A. Employment/Salary (full or part-time):; University of California, Davis. **N. Shevchenko:** A. Employment/Salary (full or part-time):; University of

California, Davis. **M.A. Rogawski:** A. Employment/Salary (full or part-time):; University of California, Davis.

Poster

P151. Antiepileptic Therapies III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P151.06

Topic: B.10. Epilepsy

Support: NRF-2020R1A2C2011808
NRF- 2021R1A4A1028713
NRF-2019M3C1B8090805
S-2021-0693-000

Title: Adverse Effects of Transcranial Focused Ultrasound in Treating Epilepsy

Authors: *M. KOO[1], T. CHOI[2], J. JOO[3], Y.-M. SHON[3,4,5], J. PARK[1,2];
[1]Biomed. Engin. - Intelligent Precision Healthcare Convergence, [2]Dept. of Intelligent Precision Healthcare Convergence, Sungkyunkwan Univ., Suwon, Korea, Republic of; [3]Samsung Med. Ctr., [4]Biomed. Engin. Res. Center, Samsung Med. Center, Sch. of Med., [5]Samsung Advanced Inst. for Hlth. Sci. & Technol., Sungkyunkwan Univ., Seoul, Korea, Republic of

Abstract: Transcranial focused ultrasound stimulation (tFUS) has demonstrated its effectiveness in alleviating the pathological hyperexcitability of small animal epilepsy models. However, no studies have reported the functional adverse effects of tFUS in treating epilepsy. With chemical and electrical stimuli, paradoxical responses of post-inhibitory rebound (PIR) which triggers excitatory responses following a brief activation of inhibitory GABAergic neurons were previously demonstrated. Thus, tFUS for treating epilepsy could also adversely trigger proconvulsive responses. This study aims to explore tFUS stimulus sequences which could generate adverse effects of amplifying the epileptiform activities. The experiment was conducted via pentylenetetrazol-injected acute epilepsy rat model (Male Sprague-Dawley rat, 8wk, n=26) where tFUS was applied for six times at the thalamic region under urethane anesthesia. While the acoustic pressure was fixed to 0.25 MPa, the burst length (10s, 20s, 40s) and interval (50s, 40s, 40s) of the acoustic transmissions were varied. Brain electrical activities were evaluated via electroencephalograph. Immunohistochemistry analysis was performed with c-Fos and GAD65 antibodies to figure out the excitatory and inhibitory cell responses, respectively. The ratio of the number of epileptic discharges after tFUS to the baseline decreased from 2.56 (SEM=0.35) to 1.20 (SEM=0.26) as the stimulation length was elongated from 10s to 40s. After the brief suppressive responses identified as the reduced number of peak ratio during the 10s stimulation, the significant increased ratio (p=0.0001) compared to the non-stimulated group (2.31; SEM=0.47) was presented in the post-stimulus period. The c-Fos-positive cell ratio analysis at

the dentate gyrus (DG) granular layer showed higher proportions of c-Fos labeled cells in 10s stimuli group (80.72%; SEM=2.00%) than the non-stimulated group (71.73%; SEM=5.81%). However, the expression ratio was dramatically reduced in the 40s stimuli group (24.22%; SEM=14.134%). In the GAD65-positive cell density analysis at the DG, 10s stimuli group (6.41 cells/mm^2; SEM=1.06) showed higher expression than the non-stimulated group (2.33 cells/mm^2; SEM=0.49), while 40s stimuli (13.58 cells/mm^2; SEM=1.58) showed the highest value. Although an anti-seizure effect was remarkably provoked by the longer period of ultrasonic stimuli, the shorter transmission (10s) of tFUS may trigger excitatory responses following brief inhibitory responses. Thus, adverse effects caused by PIR excitatory responses need to be considered when tFUS sequence is scheduled to provide for neuro-therapeutics.

Disclosures: M. Koo: None. **T. Choi:** None. **J. Joo:** None. **Y. Shon:** None. **J. Park:** None.

Poster

P151. Antiepileptic Therapies III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P151.07

Topic: B.10. Epilepsy

Support: NIH Grant NS109668
NIH Grant NS113955

Title: Novel naphthalenyl substituted aminothiazoles prevent acute neural excitotoxic injury in a rat model of temporal lobe epilepsy

Authors: *T. KYLLO[1], V. SINGH[2], H. SHIM[2], L. SINGH[2], H. M. NGUYEN[2], Y.-J. CHEN[2], E. TERRY[1], M. TEBBE[1], H. WULFF[2], *J. D. ERICKSON[1];
[1]Neurosci. Ctr., LSUHealth, New Orleans, LA; [2]Pharmacol., Univ. of California Davis, Davis, CA

Abstract: Excessive glutamate release from synapses during seizures leads to neural excitotoxic injury in vulnerable forebrain neurons and subsequent epileptic disease in the kainic acid animal model of temporal lobe epilepsy. Here, we have further characterized neuron-specific, calcium and neural activity regulated, high-affinity methylaminoisobutryic acid (MeAIB) / glutamine transport activity in mature hippocampal neuronal cultures that is inhibited by **riluzole**, a benzothiazole anti-convulsant and known neuroprotective agent against glutamate-induced excitotoxicity. We have sequentially screened two libraries of riluzole derivatives to identify compounds that inhibit spontaneous neural activity-regulated, high affinity MeAIB transport potently like riluzole and have synthesized new chemical thiazole derivatives of riluzole that are more selective for the high-affinity MeAIB transport system over neuronal sodium channel blockade than riluzole *in vitro*. One of the most potent transport inhibitors in our *in vitro* screens,

SKA-378, is orally active, brain penetrant and prevents kainic acid-induced hippocampal neural injury in all hippocampal pyramidal neuron CA subfields when given before or after kainic acid-induced status epilepticus when injury is evaluated after 3d. SKA-378 also protected neurons in other established interconnected limbic regions including the subiculum and the entorhinal and pyriform cortex, and in mid-line amygdalar and select thalamic neurons. These results indicate that our novel riluzole aminothiazole derivatives may be useful agents to explore mechanisms involved in the development, prevention, and management of chronic temporal lobe epilepsy.

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Poster

P158. Astrocytes: Disease Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P158.01

Topic: B.11. Glial Mechanisms

Support: 3-R01-DA048815-02

Title: Investigating the degree of chronic reactive astrocyte heterogeneity during Toxoplasma gondii infection

Authors: *Z. FIGUEROA[1], W. AGNEW-SVOBODA[1], E. VIZCARRA[2], M. RICCOMAGNO[3], T. A. FIACCO[3], E. WILSON[4];
[1]Neurosci. Grad. Program, [2]Biomed. Sci. Grad. Program, [3]Neurosci., [4]Biomed. Sci., Univ. of California, Riverside, Riverside, CA

Abstract: Neuroinflammation is common to neurodegenerative diseases and brain injuries, resulting in immune responses characterized by changes in astrocytes. Astrocytes ordinarily provide critical support for neurons but become reactive in response to brain disease, injury, or infection. Reactive astrocytes (RAs) are defined by their increased proliferation, enlarged cell bodies and processes, and change in function. A longstanding and unresolved issue is whether RAs contribute to or help alleviate disease progression, but overall, RAs have been discovered to be an important contributor to several neurological diseases. *Toxoplasma gondii* infects a third of the world's population and is asymptomatic except during times of immunocompromise when infection leads to severe neurological damage. *Toxoplasma* is a highly successful neurotropic parasite that causes persistent subclinical neuroinflammation due to cyst formation in neurons lasting for the lifetime of the host, making this a key resource for our studies. During chronic *Toxoplasma* infection, astrocytes demonstrate a neuroprotective function by inhibiting parasite replication via STAT1-mediated mechanisms, while simultaneously demonstrating detrimental

effects by downregulating GLT-1, suggesting heterogeneity exists among RAs. Utilizing flow cytometry, we have characterized surface integrin subsets found during chronic *Toxoplasma* infection. Experiments demonstrate that during infection potential chronic reactive astrocyte subsets are present and may be contributing directly to known functions present during infection. However, variability exists within these heterogeneous astrocytic naïve and infected subsets. To further determine if astrocytes define a chronic inflammatory state, single cell RNA sequencing experiments on astrocytes at various stages of infection were conducted to determine if similar populations are expressed during acute compared to chronic infection. In addition, to investigate RA function we have created a transgenic reporter mouse, $Lcn2^{CreERT2}$; $Rosa26^{lsl\text{-}tdTomato}$, that will allow for isolation of RA populations during infection for the first time, providing a novel tool for future use.

Disclosures: Z. Figueroa: None. **W. Agnew-Svoboda:** None. **E. Vizcarra:** None. **M. Riccomagno:** None. **T.A. Fiacco:** None. **E. Wilson:** None.

Poster

P158. Astrocytes: Disease Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P158.02

Topic: B.11. Glial Mechanisms

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ALF Gothenburg 724421
ALF Gothenburg
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Hjärnfonden

Title: Reactive Astrocytes Prevent Maladaptive Plasticity after Ischemic Stroke

Authors: M. ASWENDT[1], U. WILHELMSSOM[2], A. STOKOWSKA[2], Y. DE PABLO[2], M. HOEHN[1], M. PEKNA[2], ***M. PEKNY**[2];
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Abstract: Restoration of functional connectivity is a major contributor to functional recovery after stroke. We investigated the role of reactive astrocytes in functional connectivity and recovery after photothrombotic stroke in mice with attenuated reactive gliosis ($GFAP^{-/-}Vim^{-/-}$). Infarct volume and longitudinal functional connectivity changes were determined by *in vivo* T2-weighted magnetic resonance imaging (MRI) and resting-state functional MRI. Sensorimotor function was assessed with behavioral tests, and glial and neural plasticity responses were

quantified in the peri-infarct region. Four weeks after stroke, *GFAP*$^{-/-}$*Vim*$^{-/-}$ mice showed impaired recovery of sensorimotor function and aberrant restoration of global neuronal connectivity. These mice also exhibited maladaptive plasticity responses, shown by higher number of lost and newly formed functional connections between primary and secondary targets of cortical stroke regions and increased peri-infarct expression of the axonal plasticity marker Gap43. We conclude that reactive astrocytes are required for optimal recovery-promoting plasticity responses after ischemic stroke.

Disclosures: M. Aswendt: None. **U. Wilhelmssom:** None. **A. Stokowska:** None. **Y. de Pablo:** None. **M. Hoehn:** None. **M. Pekna:** None. **M. Pekny:** None.

Poster

P158. Astrocytes: Disease Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P158.03

Topic: B.11. Glial Mechanisms

Support: NIH Grant NS057499

Title: The role of Orai1 in astrocyte-mediated neuroinflammation

Authors: *M. NOVAKOVIC, M. PRAKRIYA;
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Abstract: Rationale: Neuroinflammation mediated by astrocytes and other glia are implicated in a growing list of neurological pathologies from depression and stroke to dementia. In these disease states, astrocytes become reactive, releasing cytokines and inflammatory factors which affect neuronal survival, synaptic transmission, and cognitive functions. However, the cellular checkpoints and signaling that control the transformation of astrocytes from their resting state to the reactive state and how these checkpoints contribute to neuronal dysfunction are poorly resolved. This study is designed to understand how astrocytic calcium signaling through store-operated Orai1 channels mediates reactive astrogliosis and neuroinflammation. Methods: All cellular experiments were performed on astrocytes cultured from neonatal C57BL/6 mice. Orai1 knockout (KO) and wild-type (WT) astrocytes were obtained using the cre/loxp system: WT (Orai1 fl/fl) and Orai1 KO (Orai1 fl/fl; nestin-cre+) astrocytes from littermates were used for the in vitro experiments. Behavioral studies were performed on both male and female astrocyte-specific Orai1 KO mice obtained using the inducible cre line Ald1h1-Cre. Tamoxifen-injected cre-negative mice served as littermate controls for the Orai1 KO mice. Results: Orai1 mediates the production of multiple pro-inflammatory cytokines through regulation of transcription factors, NFAT and NFκB. The global effect of astrocytic Orai1 signaling was assessed in a behavioral model where neuroinflammation triggered by lipopolysaccharide (LPS) injection

induces depressive-like behaviors in mice. In this paradigm, astrocyte-specific Orai1 knockout mice are partially protected from the neuroinflammation-induced depression, and differences between male and female behavioral patterns were observed. Together, these studies implicate Orai1 signaling in astrocytes as a vital component of the neuroinflammatory response, and provide insight into the role of astrocytes in neuroinflammatory disease.

Disclosures: M. Novakovic: None. **M. Prakriya:** None.

Poster

P158. Astrocytes: Disease Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P158.04

Topic: B.11. Glial Mechanisms

Support: MEXT Grant-in-Aids for Scientific Research 17H2216
MEXT Grant-in-Aids for Scientific Research 18K19494

Title: Deletion of Bergmann glial glutamate transporters causes progressive motor deficits and astrogliosis without Purkinje cell degeneration

Authors: *H. BI[1], Y. HIRAOKA[1], Y. OKUBO[3], V. T. NGUYEN-MINH[2], T. WANG[2], I. SUGIHARA[2], K. SAKIMURA[4], K. TANAKA[1];
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Abstract: Glutamate transporters play critical roles in the maintenance of glutamatergic homeostasis. In the cerebellum, the glia glutamate transporters, GLAST and GLT1 are co-localized in the processes of Bergmann glia (BG), specialized astrocytes that enwrap excitatory synapses on Purkinje cells (PC). Various studies have suggested loss of GLAST and/or GLT1 is involved in the pathogenesis of neurological disorders. However, the contributions of GLAST and GLT1 to the pathophysiology of neurological disorders are poorly understood. To investigate the role of GLAST/GLT1 in cerebellar dysfunction, we generate conditional double knockouts (cDKOs) to ablate GLAST and GLT1 selectively in BG. We crossed mice expressing the Cre-recombinase under the BG-specific Septin 4-promoter (Sept4-iCre) with floxed-GLAST and floxed-GLT1 mice. GLAST/GLT1 expression was completely lost in BG at around 6 weeks in cDKOs. We investigated the motor behavior of cDKOs using rotarod and an elevated balance beam test. cDKO mice showed motor discoordination starting from 6-week-old. In addition, cDKOs exhibited episodes of motor attacks including ataxia, tremor and dystonia when exposed to physical or emotional stress. Motor deficits in male and female cDKOs are progressive. Patch clamp recordings of EPSCs of PCs in acute slice preparations revealed that the decay time

constants of CF-EPSCs and PF-EPSCs are significantly prolonged in cDKOs. Histological analysis revealed normal development of the cerebellum in cDKOs. Immunohistochemistry showed that deletion of GLAST/GLT1 did not affect the density and morphology of PC. In contrast, the density of BG in PC layer is increased in 9-month-old cDKOs. Furthermore, the unique cellular architecture of BG was disrupted in cDKOs, such as the retraction of BG fine processes, abnormal translocation of BG into the molecular layer and transformation into stellate-shaped cells by 9-month-old. We also found intracellular Ca^{2+} concentration of BG is increased in cDKO mice compared to controls after PF stimulation by Ca^{2+}imaging. These results suggest loss of glia glutamate transporters in BG causes motor deficits with alternation in morphology, number and Ca^{2+} homeostasis of BG without PC loss. Our finding may provide novel insights into the mechanism of how motor diseases arise without neurodegeneration.

Disclosures: H. Bi: None. **Y. Hiraoka:** None. **Y. Okubo:** None. **V.T. Nguyen-Minh:** None. **T. Wang:** None. **I. Sugihara:** None. **K. Sakimura:** None. **K. Tanaka:** None.

Poster

P158. Astrocytes: Disease Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P158.05

Topic: B.11. Glial Mechanisms

Support: NIH Grant R01 ES024064

Title: Differential dysregulation of glutamate transporter expression contributes to relative susceptibility to methylmercury of wild type, and Nrf2-knockout derived spinal astrocytes.

Authors: *D. WIWATRATANA[1], W. D. ATCHISON[2];
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Abstract: Dysregulation of redox and glutamate homeostasis is involved in methylmercury (MeHg)- induced neuronal and astrocytic degeneration. MeHg disrupts not only the expression of Nrf2, a master regulator of the antioxidant pathway, but also glutamate transporter expression and its concentration. To characterize the role of Nrf2 in MeHg- induced spinal astrocyte degeneration, spinal cord astrocytes (SCAs) viability, and antioxidant genes from Nrf2 knockout derived SCAs (Nrf2-KO SCA) were compared to the wild-type derived SCA (WT-SCAs). The results showed that Nrf2-KO SCAs were more susceptible to MeHg compared to WT-SCAs. A one-hour exposure to 5 µM MeHg significantly reduced the viability of Nrf2-KO SCAs by 60% compared to WT-SCAs. The viability of Nrf2-KO SCAs further decreased and was considerably lower than the WT-SCA with prolonged exposure to MeHg. While antioxidant genes such as *Gclc*, *GPx1*, and *GPx4* involved in glutathione synthesis were significantly reduced in both WT-

and Nrf2-KO SCAs during MeHg exposure, the cystine/glutamate antiporter (or system Xc-) gene (*Slc7a11*) increased in WT-SCA (5-fold induction) and appeared to reduce in Nrf2-KO SCAs. The excitatory amino acid transporter 2 (EAAT2) gene, *Slc1a2* was significantly reduced following MeHg exposure for 18h in both WT- and Nrf2-KO SCAs compared to their same genotype treated with the vehicle. The EAAT1 gene, *Slc1a3* was significantly reduced only in Nrf2-KO derived SCAs, but only slightly affected in the WT SCAs. There was a differential expression of *Slc1a3* and *Slca1a2* in Nrf2 KO SCAs in which the *Slc1a3* expression was significantly higher than WT-SCAs (4-fold higher), and *Slc1a2* expression was about 2-fold higher than WT SCAs. The perturbation of Nrf2 in MeHg-induced SCA toxicity involved the reduction of GSH synthesis genes. Differential susceptibility between Nrf2-KO SCA and WT-SCA through the differential glutamate transporter expressions suggests a genetic background also plays a role in differential susceptibility to MeHg.

Disclosures: D. Wiwatratana: None. **W.D. Atchison:** None.

Poster

P158. Astrocytes: Disease Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P158.06

Topic: B.11. Glial Mechanisms

Support: NIH Grant NS111270

Title: Glial-specific knockdown of a group of evolutionarily conserved genes rescues hypoxia-induced lethality in Drosophila melanogaster

Authors: *D. ZHOU[1], J. XUE[1], C. MARSH[1], T. STOBDAN[1], G. G. HADDAD[1,2,3]; [1]Pediatrics, [2]Neurosciences, UCSD, La Jolla, CA; [3]The Rady Children's Hospital, San Diego, San Diego, CA

Abstract: Hypoxia, lack of oxygen supply to the tissues/cells, is one of the critical pathological elements in many human diseases, including ischemic stroke, myocardial infarction and solid tumors. Understanding the mechanisms regulating hypoxia tolerance or susceptibility is essential for developing effective strategies for medical therapy. In this regard, we have generated *Drosophila melanogaster* populations that tolerate severe hypoxia through laboratory-directed evolution, sequenced and analyzed their genomes. In parallel, we took advantage of the natural evolution of humans in high-altitude regions and sequenced/analyzed the whole genomes of Ethiopian and Andean highlanders. Through a comparative genomic approach, we obtained a group of evolutionarily conserved genes (28 human/23 *Drosophila* genes) that are potentially involved in regulating hypoxia tolerance in both human highlanders and the hypoxia-tolerant flies. Indeed, we confirmed so far that ubiquitous knocking down of the *grn/GATA3*, *inv/EN1*,

Mkk4/MAP2K4, Pxt/DUOX1/DUOX2, pyd/TJP1, RapGAP1/RAP1GAP2, Shal/KCN and *shep/RBMS3* dramatically enhanced hypoxia tolerance *in vivo* in *Drosophila*. In addition, we found that glial-specific knocking down of *Egfr, grn, pyd* and *Shal* rescued hypoxia-induced lethality. Some of them (i.e., *bnl, croc,* and *Mkk4* and *shep*) mediate Notch activation-conferred survival in hypoxic environment, and the others (i.e., *Egfr, grn, pyd* and *Shal*) regulate hypoxia tolerance through Notch-independent mechanisms. We believe that these evolutionarily conserved mechanisms are novel targets that have a strong potential to be translated into therapeutic strategies to treat hypoxia-related diseases.

Disclosures: D. Zhou: None. **J. Xue:** None. **C. Marsh:** None. **T. Stobdan:** None. **G.G. Haddad:** None.

Poster

P158. Astrocytes: Disease Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P158.07

Topic: B.11. Glial Mechanisms

Support: Leon Levy Foundation - Fellowship in Neuroscience
T32 MH019524

Title: Macrophage-derived amphiregulin modulates astrocyte network reactivity

Authors: *M. L. COOPER[1], M. SELLÉS[1], S. A. LIDDELOW[2], M. V. CHAO[1];
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Abstract: Early in neurodegenerative disease, astrocytes can expand their gap junctional networks to redistribute resources and maintain neuronal function. How is this beneficial form of astrocyte reactivity induced? To answer this, we looked at similar mechanisms in other organs and applied them to the nervous system. In the heart, cardiac macrophages induce gap-junctional network expansion in myocytes during arrhythmia. There, macrophage-derived amphiregulin (AREG) signals upon epidermal growth factor receptor (EGFR) to induce phosphorylation of connexin 43 (cx43), recruiting it to the cell membrane to form additional gap junctions and rescue the tissue. Here, we test this mechanism in the resident macrophages of the brain (microglia) and the resident cx43 network mediators (astrocytes). Like cardiac macrophages, cultured primary microglia (n = 2 isolations) release AREG in response to stress (LPS, 1ug/ml; $p < 0.001$ vs baseline). Cultured primary astrocytes (mRNA n = 3 isolations; protein n = 5 isolations) respond to AREG (100ng/mL) like cardiac myocytes: they phosphorylate cx43, with a 3-fold increase 9 minutes after AREG ($p < 0.001$). Phosphorylation returns to baseline within 90 minutes. Three minutes after AREG administration, cx43 mRNA increases 2-fold; protein levels

increase 2-fold by 11 minutes (mRNA $p = 0.007$; protein $p \leq 0.01$). Although astrocytes constitutively express little dimerized EGFR, AREG administration increases EGFR dimerization 2-fold within a minute ($p = 0.006$) and 3-fold in 7 minutes ($p < 0.001$). To measure the change in functional gap-junction network size induced by AREG, astrocytes were cut-loaded with biotin (244Da, traverses gap junctions) and dextran (10kDa, marks initially loaded astrocytes). We found a 3-fold increase in astrocyte connectivity in as little as 10 min after AREG ($p < 0.001$). Intriguingly, higher levels of AREG (1000 ng/mL) have the opposite effect – cx43 mRNA is reduced 2-fold in 3 minutes ($p = 0.02$), and cx43 protein declines 2-fold within 30 minutes ($p = 0.01$; $n = 3$ isolations). This is likely due to differential dimeric conformations of the EGFR extracellular region. Weakened dimerization through lower concentrations of partial agonists such as AREG elicits more sustained EGFR signaling, inducing cellular expansion and process outgrowth pathways. In contrast, strong dimerization through high concentrations of partial agonists or full agonists, such as EGF or TGFα, activates rapid proliferation and inflammation pathways. This differential response to EGFR activation may dictate whether astrocytes enlarge or contract their network to respond to a stressor through beneficial or harmful forms of reactivity.

Disclosures: M.L. Cooper: None. **M. Sellés:** None. **S.A. Liddelow:** None. **M.V. Chao:** None.

Poster

P158. Astrocytes: Disease Mechanisms I

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Program #/Poster #: P158.08

Topic: B.11. Glial Mechanisms

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Title: Hiv-1 induces region-specific astrocyte dysfunction in the hiv^+ human brains

Authors: *A. DONNER[1], L. CHEN[2], M. ASH[3], J. SCHNEIDER[3], L. AL-HARTHI[4], X.-T. HU[2];
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Abstract: Combined antiretroviral therapy (cART) has improved life expectancy for people living with HIV/AIDS (PLWH); but that is association with HIV-1 associated neurocognitive

disorders (HAND). The mechanism underlying HAND is unknown; but it is likely related to HIV-induced interactive neuron/astrocyte dysfunction in the brain regions that regulate neurocognition. HIV induces neurotoxicity, causing dysregulation, injury, and in severe cases, death of neurons in such brain regions, which could be worsened by astrocyte dysfunction. Among astrocytes, HIV not only dysregulates cytokines/chemokines, but also disturbs K^+ buffering (mediated by inwardly rectifying $K_{ir}4.1$ channels), glutamate uptake (mediated by excitatory amino acid transporters, GluT-1), and cell-to-cell communication (mediated by gap-junction protein connexin, Cx43); all could promote/induce neurotoxicity. However, previous studies focusing on assessing HIV-induced astrocyte dysfunction mainly use animal or cell culture models. Very little is known if, how and to what extent HIV actually alters astrocyte function in human brains, for instance, by disrupting $K_{ir}4.1$ channel, GluT-1, and Cx43 function; and that has not been studied. Here we assessed HIV-induced changes in the protein/mRNA levels of $K_{ir}4.1$ channels, GluT-1s and Cx43 in the prefrontal cortex (PFC), caudate nucleus (CN; two key regulators of neurocognition), and cerebellum (a non-regulator of cognition) from post-mortem HIV^+ human brains (at age of 44~97; HIV^+ n=6 vs. HIV^- n=4 HIV^+). We found that the protein levels of $K_{ir}4.1$ channels and GluT-1s were significantly decreased ($p<0.01$ and $p<0.05$, respectively); while $K_{ir}4.1$ channel mRNA levels were significantly increased ($p<0.05$), along with a trend of increase in GluT-1 and decrease in Cx43 mRNA (likely compensatory responses to the reduced protein levels), in HIV^+ human PFC compared to HIV^- controls. We also found that, differing from that seen in PFC, the protein levels of $K_{ir}4.1$ channels and GluT-1 ($p<0.05$) were significantly increased in HIV^+ human CN compared to HIV^- controls; and there was no significant change in their mRNA levels. In contrast, there was no significant change in the expression of such proteins/mRNAs in HIV^+ human cerebellum. Collectively, our novel findings demonstrate that HIV induces astrocyte dysfunction in the human brain by altering the expression/function of $K_{ir}4.1$ channels, GluT-1s, and Cx43. Importantly, such astrocyte dysfunction is region-specific, occurring specifically in the key brain regions that regulate neurocognition. Further studies are needed to determine if such dysfunction contribute to the mechanism underlying HAND.

Disclosures: A. Donner: None. **L. Chen:** None. **M. Ash:** None. **J. Schneider:** None. **L. Al-Harthi:** None. **X. Hu:** None.

Poster

P158. Astrocytes: Disease Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P158.09

Topic: B.11. Glial Mechanisms

Support: R01 AA012404
U01 AA020926

Title: Spatial transcriptomics and single nuclei RNA sequencing identify spatially resolved overlapping prefrontal cortex cell-type specific mechanisms between alcohol dependence and immune activation mouse models.

Authors: *N. A. SALEM[1], O. PONOMAREVA[1], L. MANZANO[2], G. R. TIWARI[1], A. J. ROBERTS[3], M. ROBERTO[4], R. MAYFIELD[1];
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Abstract: Alcohol use disorders (AUDs) affected 8.6% of men and 1.7% of women globally and attributed to 5.3% of all global deaths in 2016. We aim to identify novel molecular mechanisms implicated in the escalation of alcohol consumption, which will provide the potential for therapeutics development to prevent or reverse this escalation. We utilized chronic intermittent exposure treatment paradigm (CIE), an alcohol dependence model that produces escalated drinking in mice, and neurobiological and behavioral adaptations in mice that mimic those found in humans with AUD. We have previously shown that immune genes in brain glial cells are important regulators of alcohol drinking behavior. TLR7, key player in neuroimmune activation, is differentially expressed in mice following CIE exposure. Chronic treatment with R848, a TLR7 activator, increases alcohol consumption in mice. We performed single nuclei RNA sequencing (snRNA-seq) on micro-punches obtained from pre-frontal cortex (PFC) of CIE and control mice, to determine cell-type-specific alcohol-induced gene expression changes. Analyses pipelines resulted in the identification of 24 clusters, the expression levels of canonical markers of each cell type were used to determine the clusters' cellular identity. We identified differentially expressed (DE) genes and their enriched pathways in each cluster. Astrocytic and microglial DE genes were enriched in inflammation-related pathways (Il-2, CXCR4, IL-17, and chemokine signaling). To understand the spatial context of overlapping mechanisms between CIE and immune activation model, we generated 10x Genomics Visium Spatial transcriptomics data from coronal PFC sections (10 µm) from C57BL/6J mice treated by a single injection of the R848 (50 µg, i.p.) or saline. We utilized an 'anchor'-based integration workflow introduced in Seurat on snRNA-seq data obtained from PFC micro-punches (reference) and the spatial RNA sequencing data (query), this pipeline outputs, for each spatial capture spot, a prediction score for each of the snRNA-seq derived cell types. Our analysis showed increased microglial prediction scores in layers 2/3 and astrocytic prediction scores in layers 5/6, indicating location-specific cell-type specific response to R848 treatment.Additionally, we identified 19 DE genes in R848 brain slices overlapping with CIE astrocytes and microglia DE genes. Gene ontology analyses show association of those genes to response to stress/stimuli. Integration of the two datasets aims to identify spatially resolved transcriptomic changes induced by immune activation in the PFC mediating the escalation of alcohol consumption.

Disclosures: N.A. Salem: None. **O. Ponomareva:** None. **L. Manzano:** None. **G.R. Tiwari:** None. **A.J. Roberts:** None. **M. Roberto:** None. **R. Mayfield:** None.

Poster

P158. Astrocytes: Disease Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P158.10

Topic: B.11. Glial Mechanisms

Support: PAPIIT UNAM No. IN221820
APEC No. APEC-INB-20-21
Presupuesto Interno de la Facultad de Medicina- UNAM
Instituto de Fisiología Celular- Bioterio

Title: Palmitic acid reduces cell viability and alters VEGF-A and VEGFR2 expression in Müller glial cells.

Authors: *A. E. MEDINA ARELLANO[1], L. D. OCHOA DE LA PAZ[2];
[1]Biochem., Univ. Nacional Autónoma De México, Mexico city, Mexico; [2]BIOCHEMISTRY, UNIVERSIDAD NACIONAL AUTÓNOMA DE MÉXICO, Mexico city, Mayotte

Abstract: Lipids perform a diversity of functions in biological processes. Recent studies have established a relationship between dyslipidemia and the progress of diabetic retinopathy (DR), the most common microvascular disease associated with diabetes. In this pathology, palmitic acid (PA), a saturated fatty acid, increase levels in plasma of DR patients. In the retina, Müller glial cells (MGC) regulate a plethora of homeostatic functions, such as potassium ion buffering, neurotransmitter recycling, and angiogenic factors regulation. This study aimed to investigate the role of PA in MGC viability, morphology, and expression of angiogenic molecules such VEGF. We also explore the effect of PA on $GABA_A$ receptors. MGC primary cultures from CD1 mice were exposed to 250 μM PA and vehicle (0.008% DMSO) during 24, 48, 72, or 96 hours. Cell viability was measured by trypan blue and CCK-8 assays. Cell morphology was evaluated by immunofluorescence (IF) against CRALBP, and VEGF protein expression was confirmed by IF. VEGFR2 and HIF-1α transcripts were also determined by RT-PCR. We find that PA reduces cell viability in MGC cultures after 72 hours of exposition (92.07 ± 1.05 vs 48.52 ± 4.55, $p<0.001$). The vehicle was unable to mimic these effects (92.07 ± 1.05 vs 91.33 ± 3, ns). PA also induced morphological changes in this retinal cells, altering cellular and nuclear area (1440μm^2 ± 104.5 vs 860.0 ± 102.1, $p<0.01$ and 168.6 ± 5.11 vs 97.8 ± 8.72, $p<0.001$, respectively). By immunofluorescence assays, we detect a significant increase of VEGF-A in MGC cultures exposed to PA (13321 ± 2519 vs 43590 ± 4979 a.u., $p<0.001$). Transcriptional expression of molecules related to VEGF signaling was also studied. VEGFR2 mRNA expression decreased in MGC cultures during PA treatments (0.3210 ± 0.0050 vs 0.2787 ± 0.0071, $p<0.05$), but HIF-1α not change during this exposition (0.4611 ± 0.0208 vs 0.4520 ± 0.0152, ns). Interestingly, PA downregulate $GABA_AR$ alfa 1 subunit mRNA (0.2326 ± 0.1137 vs 0.093 ± 0.0154, $p<0.01$), but not beta2 (0.2046 ± 0.0126 vs 0.1699 ± 0.0606, ns). These results together suggest that MGC plays a pivotal role in the VEGF synthesis, possibly through HIF-1α activation and modulating VEGFR2 expression, in response to increment in the PA levels, an event associated with DR pathology. Also, the AP ability to regulate $GABA_AR$ subunit expression suggests a homeostatic

disruption related to the synaptic process during DR. This observation makes imperative to explore the mechanism involved in this process.

Disclosures: A.E. Medina Arellano: None. **L.D. Ochoa de la Paz:** None.

Poster

P158. Astrocytes: Disease Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P158.11

Topic: B.11. Glial Mechanisms

Support: Conacyt, Investigación Básica #183867
Conacyt, Beca Posgrado #288719

Title: L-kynurenine prevents oxidative damage induced by pro-oxidants in rat brain homogenates and primary astrocytes culture: an independent effect of kynurenic acid

Authors: *T. BLANCO-AYALA[1,2], P. UGALDE-MUÑIZ[4], R. LUGO-HUITRÓN[5], D. RAMÍREZ-ORTEGA[3], B. PINEDA[3], D. F. GONZÁLEZ-ESQUIVEL[3], J. PEDRAZA-CHAVERRÍ[6], L. A. RAMOS-CHÁVEZ[7], G. I. VÁZQUEZ-CERVANTES[3], A. JIMÉNEZ-ANGUIANO[1], V. PÉREZ DE LA CRUZ[3];
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Abstract: L-kynurenine (L-KYN) is an endogenous metabolite from the tryptophan ctabolism which can be metabolized further metabolized to 3-hydroxykynurenine, kynurenic acid (KYNA) or anthranilic acid. L-KYN has been used as a neuroprotective strategy in some experimental models; however, its protective effects have been attributed to KYNA production. Recently, L-KYN has also shown to be susceptible toward oxidation, suggesting that can exert redox effects. The aim of this study was to describe the scavenger and antioxidant properties of L-KYN both and evaluate whether this effect is independent to KYNA production. First, we investigated whether L-KYN itself can act as a scavenger for superoxide anion ($O_2^{\bullet -}$), hydrogen peroxide (H_2O_2), hydroxyl radical (●OH) and peroxynitrite ($ONOO^-$). Our data show that L-KYN can scavenge OH● and $ONOO^-$, but not $O_2^{\bullet -}$ or H_2O_2 in chemical combinatory assays. L-KYN prevents the DNA and protein degradation induced by ●OH in a concentration dependent-manner. Therefore, ROS production was evaluated in brain homogenates exposed to $FeSO_4$ (5 µM), 3-NP (5 mM) and $ONOO^-$ (25 µM) in presence or absence of L-KYN (0 - 5 µM). ROS production increased in a time dependent-manner with all the pro-oxidants used, whereas the co-

incubation with L-KYN decreased ROS levels. Lipid peroxidation (LP) increased and MTT reduction decreased with the oxidants, and these effects were abolished in presence of L-KYN. Furthermore, in order to clarify if the KYN effect involves KYNA scavenger activity, two different strategies were employed: 1) primary astrocytes cultures obtained from kynurenine aminotransferase II - main enzyme involved in KYNA production - knock-out mice (KAT-II KO), exposed to different pro-oxidants and 2) using a polyclonal KYNA antibody that recognize KYNA, which is produced by the of L-KYN - ROS interaction in a synthetic system of combinatory chemistry. ROS production was increased in astrocytes exposed to 3-NP and $ONOO^-$ and was attenuated in presence of L-KYN. The peroxynitrite scavenger effect of L-KYN in combinatory chemical assays was maintained even when the KYNA antibody was used in combination with L-KYN. Additionally we evaluated Nrf2 levels in brain tissues from rats treated with a daily injection of L-KYN (75 mg/kg, i.p., 5 days) to further characterize L-KYN antioxidant profile. Altogether, these findings strongly suggest that L-KYN can be considered as a potential endogenous antioxidant, regardless of its ability to produce KYNA.

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Poster

P203. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P203.01

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: The Behavioral and Pathological Effects of Tamiflu in an Alzheimer's Disease Mouse Model

Authors: *C. FASTENAU, L. SOLANO, J. L. WICKLINE, S. SMITH, K. ODFALK, K. F. BIENIEK, S. C. HOPP;
UT Hlth. Sci. Ctr. San Antonio, San Antonio, TX

Abstract: Many patients with viral symptoms are treated with the widely-used antiviral agent Tamiflu (oseltamivir phosphate), which is highly efficacious against influenza. However, observational studies have reported acute and delayed adverse neuropsychiatric side effects in patients with age-related neurodegenerative diseases such as Alzheimer’s Disease (AD). Here, we test the hypothesis that an interaction between Tamiflu and AD pathology may contribute to abnormal behavioral side effects. Specifically, we hypothesize that Tamiflu’s mechanism as a neuraminidase inhibitor blocks the removal of sialic acid residues and leads to an increase in sialylation levels in extracellular proteins, including the pathognomonic feature of AD amyloid-

beta (Aβ) plaques. We have observed enhanced α2–6 sialic acid bonds surrounding Aβ plaques in the 5XFAD mouse model of AD and in human AD cases which we hypothesized could increase following Tamiflu treatment leading to exacerbation of behavioral deficits. To test this hypothesis, we investigated the effects of Tamiflu (or vehicle) treatment on aged 5XFAD mice and wild-type (WT) littermates. After initial treatment (daily for five days), the mice were tested with the Open Field (OF) Behavior for anxiety-like behaviors. Following drug administration, a second OF test was performed followed by a two-day Novel Object Recognition (NOR) test. Analyses of behavioral data demonstrated no overall drug effect in this cohort; however, 5XFAD mice exhibited greater velocity in a 2-Way ANOVA compared to WT mice for OF (5.249 cm/s vs 4.403 cm/s, p=0.01) and NOR (4.361 cm/s vs 3.1 cm/s, p=0.002) possibly indicative of hyperactivity. Following behavioral testing, the mice were euthanized and their brains were fixed in 10% neutral buffered formalin (24 hours) and processed to generate paraffin-embedded tissue blocks. Preliminary lectin staining of aged 5XFAD and WT brains suggest that plaque-associated microglia are sialylated at the α2–6 and α2–3 bonds while there is minimal α2–6 and α2–3 sialic acid labeling present in WT mice. Taken together, these observations suggest that disease associated microglia are heavily sialylated in AD. Qualitative analysis of the experimental 5XFAD mice suggests different structures are sialylated at the α2–6 and α2–3 bonds between treatment groups. The α2–6 specific lectin appears to be staining plaque associated microglia while the α2–3 specific lectin is staining a variety of extracellular features. Further analysis is required to define the sialic acid interactions within treatment groups. Thus, this study aims to untangle the complex role sialylation may play in disease pathogenesis and aging.

Disclosures: C. Fastenau: None. **L. Solano:** None. **J.L. Wickline:** None. **S. Smith:** None. **K. Odfalk:** None. **K.F. Bieniek:** None. **S.C. Hopp:** None.

Poster

P203. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P203.02

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant K99 AG066960

Title: Membralin deficiency in astrocytes disrupted ER homeostasis and induced reactive status of astrocytes

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Abstract: The endoplasmic reticulum (ER) is the primary organelle for synthesizing membrane proteins, secretory proteins, and lipids. Disturbance of ER homeostasis in peripheral immune

cells induces inflammatory responses. Glial cells, including astrocytes and microglia, are the immune cells in the brains, becoming reactive/gliosis during the progression of Alzheimer's disease (AD) and many other neurodegenerative disorders. However, whether glial protein homeostasis mediates the reactive status of glial cells is yet to be investigated. Previously, we identified an ER-associated degradation protein, membralin, as an important mediator in neurodegeneration. Loss of membralin impaired the turnover of nicastrin protein, which increased γ-secretase complex formation and activity. Moreover, deletion of membralin in astrocytes decreased excitatory amino acid transporter 2 (EAAT2) expression and induced excitotoxicity. Using adeno-associated viral (AAV) vectors to elevate membralin levels in *SOD1* ^{G93A} ALS mouse model can significantly extend the lifespan of the animals. Here, we show that membralin is critical in mediating the reactive status of astrocytes. Astroglial membralin knockout animals developed strong neuroinflammation phenotypes through pathological analysis and transcriptomic analysis. Loss of membralin in astrocytes disrupted ER homeostasis and nuclear envelop and induced the production of pro-inflammatory cytokines. In contrast, the elevation of membralin in astrocytes can alleviate the induction of C3+ astrocytes. Further, elevating membralin levels in *5XFAD* mouse model can significantly reduce the AD pathologies. Therefore, modulation of atroglial membralin levels can be a promising target to regulate neuroinflammation for AD therapy.

Disclosures: L. Jiang: None. **X. Li:** None. **S. Myers:** None. **T. Huang:** None. **R.J. Kaufman:** None.

Poster

P203. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P203.03

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Loss of circulating memory CD8 T cells that induce AD-like pathology in mice correspond to decreased CSF Abeta42 and increased CSF total Tau and pTau181 in human patients

Authors: *C. WHEELER[1,2,3], D. VAN DAM[4,6], Y. VERMEIREN[4,6], H. DE REU[5], P. DE DEYN[4,6], V. YAMAMOTO[7], K. TRUJILLO[1];
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Abstract: Objective: We assessed the relationship between Amyloid Precursor Protein (APP)-peptide-specific, KLRG1^{+} CD8 T cell levels in blood and CSF Aβ1-42, total Tau, and pTau181

in Alzheimer's (AD), mild cognitively impaired (MCI), and normal aging patients.
Rationale: We previously engineered mice to accumulate age-related, antigen-specific memory CD8 T cells as in aging humans. These mice spontaneously develop all major hallmarks of AD with aging. Analogous T cells reactive to an APP epitope were found in aging humans, accumulated in AD brain, and decreased in AD blood where their levels tracked the disease with high accuracy. The relationship of these cells to established AD biomarkers could suggest the mechanism(s) by which they impact pathology and define the limits of their ability to track the AD continuum.
Methods: Antigen-specific CD8 T cells in blood were quantified by flow cytometry after staining with anti-CD8, anti-KLRG1 and APP peptide-HLA-A2 multimers in confirmed late-onset AD patient, MCI with or without CSF biomarkers of AD, and normal aging cohorts (n = 50). Aβ1-42, total Tau, and pTau181 were quantified in CSF, and cognitive performance assessed by MMSE.
Results: Percentage of APP-specific memory CD8 T cells was significantly decreased with dementia (0.68 + 0.29% for MMSE <25; 1.63 + 0.32% for MMSE >24), reaching minimal levels in AD. Although this precluded meaningful correlations of APP-specific T cells within AD alone, decreases in the larger parental ($KLRG1^{+}$) memory CD8 T cell population correlated with decreased CSF Aβ1-42 in AD (r = 0.511; *P* = 0.003; n = 31). Decreasing APP-specific CD8 T cells in blood also correlated with lower CSF Aβ1-42 in normal aging patients (r = 0.518; *P* = 0.028; n = 18). In addition, patients with the highest third of APP-specific CD8 T cells in blood had significantly elevated total Tau and pTau181 in CSF (482$\pm$39 vs. 370$\pm$47 total Tau, *P* = 0.011; 70$\pm$4 vs. 60$\pm$6 pTau181, *P* = 0.027).
Conclusion: Our results reveal that age-related memory CD8 T cells in blood decrease with dementia, and in proportion to established CSF biomarkers for AD, especially Aβ1-42. Given that decreased CSF Aβ1-42 is among the earliest biomarkers for AD, this supports the notion that antigen-specific memory CD8 T cells are associated with the earliest detectable pathologic changes in AD, and as such may represent novel candidates to predict and track the disease.

Disclosures: C. Wheeler: A. Employment/Salary (full or part-time):; T-Neuro Pharma, Inc., StemVax Therapeutics. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Stock ownership, T-Neuro Pharma, Inc., Patent author with revenue-sharing agreement (2 relevant). **D. Van Dam:** None. **Y. Vermeiren:** None. **H. De Reu:** None. **P. De Deyn:** None. **V. Yamamoto:** None. **K. Trujillo:** A. Employment/Salary (full or part-time):; T-Neuro Pharma, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Stock ownership, T-Neuro Pharma, Inc..

Poster

P203. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P203.04

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: N-as-triggered spms are direct regulators of microglia in a model of alzheimer’s disease

Authors: B. CHOI[1], K. PARK[2], J.-S. BAE[2], *H. JIN[1];
[1]Col. of Vet. Medicine, Kyungpook Natl. Univ., Daegu, Korea, Republic of; [2]Sch. of Medicine, Kyungpook Natl. Univ., Daegu, Korea, Republic of

Abstract: Sphingosine kinase1 (SphK1) is an acetyl-CoA dependent acetyltransferase which acts on cyclooxygenase2 (COX2) in neurons in a model of Alzheimer’s disease (AD). However, the mechanism underlying this activity was unexplored. Here we show that N-acetyl sphingosine (N-AS) is first generated by acetyl-CoA and sphingosine through SphK1. N-AS then acetylates serine 565 (S565) of COX2, and the N-AS-acetylated COX2 induces the production of specialized pro-resolving mediators (SPMs). In a mouse model of AD, microglia show a reduction in N-AS generation, leading to decreased acetyl-S565 COX2 and SPM production. Treatment with N-AS increases acetylated COX2 and N-AS-triggered SPMs in microglia of AD mice, leading to resolution of neuroinflammation, an increase in microglial phagocytosis, and improved memory. Taken together, these results identify a role of N-AS in the dysfunction of microglia in AD.

Disclosures: B. Choi: None. **K. Park:** None. **J. Bae:** None. **H. Jin:** None.

Poster

P203. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P203.05

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH/NIA grant AG056114

Title: Reprogramming Transcriptional Activation State in Plaque-associated Microglia in 5xFAD Mice with Increased Gene-dosage of Wildtype TREM2 but not TREM2-R47H Variant

Authors: *C. LEE[1], **A. J. DE LA ROCHA**[1], X. GU[1], A. DAGGETT[1], K. INOUYE[1], P. LANGFELDER[1], N. WANG[1], X. W. YANG[1,2];
[1]Semel Inst. for Neurosci. and Human Behavior, [2]Brain Res. Inst., UCLA, Los Angeles, CA

Abstract: In late onset Alzheimer’s disease (AD), the rare variant of Triggering Receptor Expressed on Myeloid Cells 2 (TREM2), TREM2-R47H, is associated with about two-fold higher disease risk. TREM2 expression is highly enriched in the microglia, the resident innate immune cells in the brain. In AD brains, TREM2 is proposed to mediate the formation of a microglial neuroprotective barrier surrounding amyloid-β plaques, as well as the activation of a

disease-associated transcriptomic signature. Recently, our lab showed that elevated TREM2 gene-dosage in BAC transgenic mice is beneficial in amyloid AD mouse models resulting in reduced plaque loads, reprogramming of microglial transcriptomic activation state, altered plaque-associated microglial morphology, and rescued behavioral deficits. However, 5xFAD mice with a BAC transgene overexpressing the human TREM2-R47H variant, unlike those overexpressing wild-type human TREM2, showed a significant reduction in the beneficial effects of TREM2 gene-dosage increase. Additionally, cortical RNA-sequencing studies showed that the microglial transcriptomic signature of TREM2-mediated microglial reprogramming is abolished in the 5xFAD/TREM2-R47H mice. We next applied RNAscope, a multiplexing *in situ* hybridization method, to examine the transcriptional activation states of plaque-associated microglia. Our results reveal that elevated wild-type TREM2 gene-dosage induces an alternative transcriptional activation state in plaque-associated microglia, where the expression of a subset of TREM2 dosage-dependent (TD) genes decreased while another subset of TD genes increased. Importantly, this TREM2 gene-dosage dependent activation state for plaque-associated microglia was absent in 5xFAD/TREM2-R47H mice. Together, our study demonstrates that TREM2 gene-dosage confers an alternative activation state in plaque-associated microglia, and this i*n vivo* function of human TREM2 in the BAC model is abolished with TREM2-R47H, the AD-risk variant.

Disclosures: C. Lee: None. **A.J. De La Rocha:** None. **X. Gu:** None. **A. Daggett:** None. **K. Inouye:** None. **P. Langfelder:** None. **N. Wang:** None. **X.W. Yang:** None.

Poster

P203. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P203.06

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: P01 DA028555
R01 NS36126
P01 NS31492
P01 MH64570
P01 NS43985
P30 MH062261
R01 AG043540

Title: Cd4+ effector t cells accelerate alzheimer's disease in mice

Authors: *J. MACHHI[1], P. YEAPURI[2], Y. LU[2], R. MOSLEY[3], H. E. GENDELMAN[4]; [2]Pharmacol. and Exptl. Neurosci., [3]Dept Pharmacol. and Exp Neurosci., [4]Prof & Chair Dept Pharmacol, [1]Univ. of Nebraska Med. Ctr., Omaha, NE

Abstract: Background: Alzheimer's disease (AD) is a progressive neurodegenerative disorder characterized by cognitive impairments linked to pathological deposition of misfolded self-protein amyloid beta (Aβ) which in kind facilitates tau aggregation and neurodegeneration. Neuroinflammation is accepted as key disease driver caused by innate microglia activation. Recently, adaptive immune alterations have been uncovered beginning early and extending throughout the disease. How these occurs and whether they can be harnessed to affect disease progress is unclear. We propose that self-antigens would induct autoreactive effector T cells (Teff) serving to drive pro-inflammatory and neurodestructive immunity. Here, we investigated the role of effector immunity could affect cellular level disease pathobiology in an AD animal model. **Methods:** In this report, we developed and characterized amyloid beta (Aβ) reactive type 1 T helper (Th1) and type 17 Th (Th17) cloned lines and studied their role in AD pathogenesis. The cellular phenotype and antigen-specify of Aβ-specific Th1 and Th17 clones were confirmed using flow cytometry, immunoblot staining and Aβ T cell epitope loaded haplotype-matched major histocompatibility complex II IA^b (MHCII-IA^b-KLVFFAEDVGSNKGA) tetramer binding. Aβ-Th1 and Aβ-Th17 clones were adoptively transferred into the APP/PS1 double transgenic mice expressing a chimeric mouse/human amyloid precursor protein and a mutant human presenilin 1 and mice were assessed for memory impairments. Finally, mice were sacrificed and blood, spleen, lymph nodes and brain were harvested for immunological, biochemical, and histological analysis. **Results:** The propagated Aβ-Th1 and Aβ-Th17 cells were confirmed stable and long-lived. Treatment of APP/PS1 mice with Aβ reactive effector T cells (Teffs) accelerated memory impairment and systemic inflammation, increased amyloid burden, elevated microglia activation, and exacerbated neuroinflammation. Both Th1 and Th17 Aβ reactive Teffs progressed AD pathology by downregulating anti-inflammatory and immunosuppressive regulatory T cells (Treg) as recorded in the periphery and within the central nervous system. **Conclusions:** These results underscore an important pathological role for CD4+ Teffs in AD progression. We posit that aberrant disease-associated effector immune responses can be controlled by the induction of Aβ reactive Tregs.

Disclosures: J. Machhi: None. **P. Yeapuri:** None. **Y. Lu:** None. **R. Mosley:** None. **H.E. Gendelman:** None.

Poster

P203. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P203.07

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: RF1AG069378
R01AG048993
P20GM103442

U54GM128729
P20GM113123

Title: Influence of intestinal bacteria on colitis-induced exacerbation of Alzheimer's disease

Authors: *L. JOHNSON[1], B. SAHU[2], M. SOHRABI[2], C. K. COMBS[2];
[2]Biomed. Sci., [1]Univ. of North Dakota, Grand Forks, ND

Abstract: Alzheimer's disease (AD) is characterized by progressive cognitive decline and is a leading cause of death in the United States. Neuroinflammation has been implicated in the progression of AD and several recent studies suggest that peripheral immune dysfunction is also an aspect of disease. Continuing evidence indicates that intestinal dysbiosis is an attribute of AD and inflammatory bowel disease (IBD) has been shown to aggravate cognitive impairment. Previously, we demonstrated that an IBD-like condition exacerbates, and a probiotic intervention attenuates, AD-related changes in the brains of AppNL-G-F mice. In this study, we investigated whether dietary intervention with a probiotic, VSL#3, could attenuate IBD-potentiated changes in the brains of these mice. Age matched 6-10-month-old male C57BL/6 wild type (WT) and AppNL-G-F mice were randomly divided into 4 groups: Vehicle control, oral VSL#3, dextran sulfate sodium (2% DSS dissolved in drinking water), and DSS given with VSL#3. Mice were sacrificed at the end of the experiment and colons and brains were collected for biochemical and immunohistochemical analyses. Colons were used to quantify lipocalin levels via ELISA as a measure of inflammatory change. Fixed brain sections were used for performing immunohistochemistry for amyloid beta (Aβ). Aβ ELISAs and cytokine arrays were performed on hippocampal and cortical lysates, respectively. As anticipated, VSL#3 treatment attenuated the DSS-induced colitis disease activity index and weight loss in WT and AppNL-G-F mice. Although VSL#3 significantly attenuated the DSS-mediated increase in WT colonic lipocalin levels, it was less protective in the AppNL-G-F DSS treated group. In spite of apparent intestinal benefit, VSL#3 treatment did not attenuate DSS-dependent increases in plaque load or Aβ levels in the AppNL-G-F brains. Corresponding with this lack of efficacy, VSL#3 treatment did not alter many of the DSS-induced increases in brain cytokines in AppNL-G-F mice and even potentiated levels of some including IFN-γ and IL-6. Altogether, our data indicates that dietary probiotic intervention with VSL#3, while sufficient to provide some protection against the colitis-like condition, may exacerbate communication of IBD-associated inflammation to the brain.

Disclosures: L. Johnson: None. **B. Sahu:** None. **M. Sohrabi:** None. **C.K. Combs:** None.

Poster

P203. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P203.08

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: CIHR grant no 331652

Title: Conditional genetic deletion of CSF1 receptor in microglia ameliorates the physiopathology of Alzheimer's disease

Authors: *V. PONS[1], S. RIVEST[2];
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Abstract: Background: Alzheimer's disease (AD) is a progressive neurodegenerative disorder and the most common form of dementia in the world. Microglia are the innate immune cells of CNS, their proliferation, activation and survival in pathologic and healthy brain have previously been shown to be highly dependent on CSF1R. **Methods:** Here we investigate the impact of such receptor on AD etiology and microglia. We deleted CSF1R using Cre/Lox system, the knockout (KO) is restricted to microglia in the APP/PS1 mouse model. We induced the knockout at 3-month-old, before plaque formation and evaluated both 6 and 8-month-old groups of mice. **Results:** Our findings demonstrated that CSF1R KO did not impair microglial survival and proliferation at 6 and 8 months of age in APP cKO compared to their littermate-control groups $APP_{Swe/PS1}$. We have also shown that cognitive decline is delayed in CSF1R-deleted mice. Ameliorations of AD etiology is associated with a decrease in plaque volume in cortex and hippocampus area. A compensating system seems to take place following the knockout, since TREM2/β-Catenin and IL-34 expression are significantly increased. Such a compensatory mechanism may promote microglial survival and phagocytosis of Aβ in the brain. **Conclusions:** Our results provide new insights on the role of CSF1R in microglia and how it interacts with the TREM2/β-Catenin and IL-34 system to clear Aβ and ameliorates the physiopathology of AD.

Disclosures: V. Pons: None. **S. Rivest:** None.

Poster

P203. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P203.09

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant U01 AG052460
NIH Grant R21 NS10167

Title: Behavioural and anti-inflammatory outcomes of EP2 antagonist treatment in a two-hit mouse model of Alzheimer's disease

Authors: *A. BANIK, R. AMARADHI, M. SAU, R. J. DINGLEDINE, T. GANESH;
Pharmacol. and Chem. Biol., Emory Univ., Atlanta, GA

Abstract: Alzheimer's disease (AD) is one of the leading causes of dementia. Approximately 50 million people are living with AD and related forms of dementia globally while adequate disease modifying treatment for AD is still not available. Neuroinflammation is widely evident in AD brains and thought to contribute and exacerbate the AD-pathology and ensuing cognitive and behavioral deficits in patients. Prostaglandin-E_2 (PGE_2) receptor EP2, which is activated downstream of cyclooxygenase-2 (COX-2) signaling, emerged as a neuroinflammatory target in several neurodegenerative diseases including AD, and treatment with a selective small molecule antagonist of EP2 mitigates the neuroinflammation in acute brain injury models. Therefore, we investigated the effect of a selective EP2 antagonist on behavioral deficits in 5xFAD transgenic mice subjected to mild, systemic inflammation by lipopolysaccharide (LPS). We have exposed a cohort of 5xFAD mice in B6SJL background with once-a-week intraperitoneal injection of LPS (1 mg/kg), and treated with either vehicle or EP2 antagonist TG11-77. HCl (100 mg/kg/day) for 12 weeks in drinking water. Complete blood count (CBC) analysis was performed to demonstrate that LPS induced an anemia of inflammation, and induction of inflammatory gene expression (mRNA) in these mice brains is being measured. Further, we aim to determine the effect of EP2 antagonist treatment on these inflammatory markers and behavioral parameters such as working memory and spatial memory (Y-maze and Morris Water Maze). The outcome of this study will indicate whether EP2 antagonism is a prudent therapeutic strategy in reducing brain inflammation and behavioral deficits in this two-hit model of AD (transgenic line with LPS).

Disclosures: A. Banik: None. **R. Amaradhi:** None. **M. Sau:** None. **R.J. Dingledine:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); R.D. is a founder of, and has equity in Pyrefin Inc, which has licensed technology from Emory University in which R.D. is an inventor. **T. Ganesh:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); T.G. is a founder of, and has equity in Pyrefin Inc, which has licensed technology from Emory University in which T.G. is an inventor.

Poster

P203. Alzheimer's Disease: Neuroinflammation and Immune Actions: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P203.10

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: P50AA26117
T32AA007565

Title: The Effects of Chronic Voluntary Alcohol Consumption in a Mouse Model of Alzheimer's Disease

Authors: *S. C. GIRONDA[1], S. M. DAY[2], S. L. MACAULEY[3], J. L. WEINER[2];
[1]Neurosci., [2]Integrative Physiol. and Pharmacol., [3]Gerontology and Geriatric Med., Wake Forest Sch. of Med., Winston Salem, NC

Abstract: The majority of Alzheimer's disease (AD) cases are sporadic with unknown causes. Many dietary factors, including alcohol intake, have been reported to increase the risk to develop AD. The effect of alcohol on cognitive function and AD pathogenesis remains elusive. Nearly 6% of Americans, or 15 million people are currently diagnosed with alcohol use disorder (AUD). Seeing the concordance between AUD and AD, we sought to identify the effects of chronic voluntary alcohol consumption on affective behavior, cognitive performance, metabolism, amyloid plaque deposition, and immune activation in a mouse model of dementia. 17 male APP/PS1 transgenic mice and 17 male wild-type control mice at four months of age were singly housed and randomly assigned to one of two conditions. The first provided access to 20% alcohol and water (12 hrs/4 days week during the dark phase) and the second provided access to only water in their home cages. Prior to alcohol exposure, mice were run through a battery of behavioral tests to assess baseline affective state and cognitive performance. At 4 months of age, mice were provided access to alcohol for 10 weeks. Throughout the drinking exposure, food consumption and weights were monitored and recorded. There were no genotype effects on alcohol intake or preference however alcohol intake selectively decreased food consumption in APP/PS1 mice. During the last five weeks of drinking, the baseline behavioral battery was repeated on non-alcohol-drinking days. While there were no significant differences between the groups prior to alcohol exposure, alcohol consumption elicited changes in several measures of affective state including the open field test, marble burying, and nest building in the alcohol-exposed APP/PS1 mice. We did not find any differences in performance on cognitive tests such as novel object recognition. In addition, alcohol significantly increased small amyloid plaque deposition. These findings are consistent with research in the clinical literature suggesting that alterations in affective state are a precursor to changes in cognitive performance and influenced by chronic alcohol consumption in AD. This model may prove useful to elucidate the neural mechanisms through which alcohol consumption exacerbates behavioral and neurobiological AD phenotypes.

Disclosures: S.C. Gironda: None. **S.M. Day:** None. **S.L. Macauley:** None. **J.L. Weiner:** None.

Poster

P223. Tau: Animal and Cellular Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P223.01

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIA Grant R01AG062840
Joseph H. Stahlberg Foundation Grant

Title: The effects of a high-affinity transferrin receptor antibody-Erythropoietin fusion protein in a mouse model of tauopathy

Authors: *J. YANG[1], W. OU[2], J. SIMANAUSKAITE[4], M. CHOI[5], J. SUN[2], M. MELVILLE[1], D. CASTELLANOS[1], S. CHILUKURI[6], A. KOKA[7], D. H. CRIBBS[8], R. K. SUMBRIA[3]; [1]Keck Grad. Inst., Claremont, CA; [2]Dept. of Biomed. and Pharmaceut. Sci., [3]Biopharmaceutical Sci., Chapman Univ. Rinker Campus, Irvine, CA; [4]Dept. of Neurosci., Pomona Col., Pomona, CA; [5]Keck Sci. Dept., Claremont McKenna Col., Claremont, CA; [6]La Salle High Sch., Pasadena, CA; [7]Oak Park High Sch., Oak Park, CA; [8]Neurol., Univ. of California Irvine Dept. of Neurol., Irvine, CA

Abstract: Alzheimer’s disease (AD) is a chronic neurodegenerative disease with limited treatments. Erythropoietin(EPO), a neurotrophin, is a potential therapeutic for AD but has limited blood-brain barrier (BBB)permeability. A fusion protein of EPO and the transferrin receptor monoclonal antibody (cTfRMAb) actsas a molecular trojan horse by shuttling the EPO into the brain via the transvascular route. We previouslyshowed that this BBB-penetrating cTfRMAb-EPO is protective in a mouse model of amyloidosis, but theprotective effects of this BBB-penetrating EPO on tauopathy are unknown (Yang et al, PMID: 32579360,2020). We engineered an aglycosylated cTfRMAb-EPO fusion protein, designated TfRMAb-N292G-EPO, by removing the Fc N-linked glycosylation site at position 292, to reduce TfRMAb Fc-effectorfunction related adverse effects. This study aimed to investigate the effects of cTfRMAb-N292G-EPO inthe PS19 mouse model of tauopathy. Male and female hemizygous Tg(Prnp-MAPT*P301S)PS19Vle(PS19) 6-month-old mice were injected intraperitoneally with saline (n=9) or cTfRMAb-N292G-EPO (10mg/kg, n=10), two to three days a week for 8 weeks. Age-matched wildtype littermates were injectedintraperitoneally with saline (n=12). Locomotion, hyperactivity, and anxiety were assessed by the open-field test. Sagittal mouse brain sections were analyzed by blinded readers for phospho-tau (ser202,thr205) and microgliosis (iba1) by immunofluorescence in the cerebral cortex, hippocampus, amygdala,and entorhinal cortex. The cTfRMAb-N292G-EPO significantly reduced the phospho-tau-positive area inall regions analyzed ($p<0.01$) and microglia-positive area in the entorhinal cortex and amygdala ($p<0.05$).This trend was seen in both sexes. The locomotion hyperactivity and reduced anxiety observed in thePS19 mice were significantly ($p<0.05$) depreciated by the cTfRMAb-N292G-EPO compared to thesaline-treated PS19 mice. Overall, this study demonstrated the therapeutic effects of the BBB-penetratingerythropoietin in a PS19 mouse model of tauopathy.

Disclosures: J. Yang: None. **W. Ou:** None. **J. Simanauskaite:** None. **M. Choi:** None. **J. Sun:** None. **M. Melville:** None. **D. Castellanos:** None. **S. Chilukuri:** None. **A. Koka:** None. **D.H. Cribbs:** None. **R.K. Sumbria:** None.

Poster

P223. Tau: Animal and Cellular Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P223.02

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH/NIA R56AG068063
NIH R01 AG068063-01A1

Title: The relationship between sleep disruption and tau pathology in the progression and treatment of Alzheimer's Disease

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Abstract: Sleep is an essential physiological behavior that supports brain health and cognitive function. In Alzheimer’s disease (AD), a devastating neurodegenerative disorder, patients experience accelerated sleep loss which can be correlated with AD onset and contribute to AD progression. Tau is an axonal microtubule stabilizing protein that forms aggregates in AD and contributes to cognitive decline, synapse loss and neuronal death. Tau mislocalization and aggregation at synapses may impair sleep and restorative sleep-dependent homeostatic plasticity. I hypothesize that sleep disruption occurs early in AD progression and subsequently drives further tau mislocalization and aggregation, and cognitive decline. Preliminary data using P301S (PS19) transgenic mice shows that differences in sleep behavior arise as early as 3 months in females and 6 months in males. Accumulation of tau becomes apparent between 6-9months. These results highlight sex differences in the onset of sleep disruption and support sleep disruption as an early-stage symptom. Endocannabinoids provide an intriguing avenue for therapeutic intervention because of their role in promoting sleep and anti-inflammatory signaling. Preliminary work shows that sleep disruption in PS19 mice can be acutely reversed by increasing the endocannabinoid anandamide. Therefore, the objectives of this work are to test the link between sleep loss and tau aggregation through correlation and causation and to identify the therapeutic window targeting endocannabinoid signaling during sleep in advance of tau pathology and cognitive decline. These studies will provide a deeper understanding of the behavioral and molecular changes that occur during abnormal sleep in AD and highlight endocannabinoids as a suitable signaling pathway for enhancing the restorative benefits of sleep.

Disclosures: S. Martin: None. **M. Ye:** None. **J. Lord:** None. **L. Sharek:** None. **G.H. Diering:** None.

Poster

P223. Tau: Animal and Cellular Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P223.03

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Else-Kroener-Fresenius Stiftung
Koeln Fortune
Studienstiftung des Deutschen Volkes

Title: Investigating isoform-specific functions of Tau under healthy and pathological conditions using Tau-deficient hiPSC derived cortical neurons

Authors: *S. BACHMANN, M. BELL, M. AL KABBANI, J. KLIMEK, H. ZEMPEL;
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Abstract: One hallmark of many neurodegenerative diseases, such as Alzheimer's disease (AD), is the formation of neurofibrillary tangles by hyperphosphorylated Tau in the brain, resulting in neuronal death and cognitive decline. In the adult human brain, six Tau isoforms are expressed, originating from alternative splicing of exons 2, 3, and 10 of the *MAPT* gene. The isoforms differ in the number of N-terminal inserts (0, 1, or 2N) and the C-terminal repeat number (3 or 4R). Normally, 3R and 4R isoforms are expressed in equal ratios throughout the brain however, mutations in the *MAPT* gene affecting splicing can alter the Tau isoform ratio and ultimately cause neurodegenerative diseases. Recent results from murine primary neurons and human neuroblastoma cells highlight that the six human-specific isoforms are differentially localized within neurons and influence microtubule dynamics in an isoform-specific manner. To further investigate the Tau isoforms in a human neuronal context, we generated three Tau KO iPSC cell lines derived from Ngn2-WTC11 cells using CRISPR/Cas9. Basic characterization of the Tau KO cell lines showed no difference in the morphology or pluripotency of iPSCs, the capability of Ngn2-induced neuronal differentiation or neuronal morphology compared to WT iPSCs and induced neurons. In addition, live-cell calcium imaging revealed no difference between WT and Tau KO neurons in spontaneous activity. Initial results from re-expressed Tau isoforms in Tau KO human neurons support the differential localization of the isoforms observed previously in rodent neurons. Further experiments will investigate the isoform-specific role of Tau regarding microtubule dynamics, axonal branching, and synapse formation under basal and pathological conditions. Expected results will provide a major understanding of basic biological functions of Tau and help to identify potential therapeutic targets for the treatment of AD and related neurodegenerative diseases.

Disclosures: S. Bachmann: None. **M. Bell:** None. **M. Al Kabbani:** None. **J. Klimek:** None. **H. Zempel:** None.

Poster

P223. Tau: Animal and Cellular Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P223.04

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: R01MH107507

Title: Expression of pathological tau in hippocampal interneurons

Authors: *E. LEGGETT, A. R. MCQUISTON;
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Abstract: Alzheimer's Disease (AD), a progressive disease of cognitive decline, is the sixth leading cause of death in the United States. No medications exist to stop disease progression. One pathological hallmark at early stages of AD is the accumulation of hyperphosphorylated and oligomeric forms of microtubule-associated tau protein (tau) in the entorhinal cortex and one of its downstream targets, the CA1 region of the hippocampus. Mouse models expressing mutant forms of human tau, which cause pathological tau variant formation in familial frontotemporal lobe dementia (FTLD) patients, display dementia-like symptoms in mice. We used a similar strategy to express hybrid FTLD human tau mutants (hTau-P301L or hTau-P301L-S320F) (Koller et al 2019, Strang et al 2018) in the medial entorhinal cortex (MEC) or hippocampal CA1 of genetically-modified mouse lines via intracranial injection of Cre-recombinase-dependent adeno-associated viruses. Expression of hTau-P301L in layer 3 pyramidal cells of the MEC of pOXR1-Cre mice resulted in the trans-synaptic spread of hTau-P301L into both pyramidal cells and select inhibitory interneurons in hippocampal CA1. One subclass of interneuron in hippocampal CA1 vulnerable to trans-synaptic spread was the calretinin-positive interneuron-selective interneurons. Calretinin and calbindin were co-expressed with hTau-P301L, but parvalbumin, another calcium-binding protein, was not. To further examine pathological tau expression in calretinin interneurons, a Cre-recombinase-dependent AAV carrying the coding sequence of hTau-P301L-S320F was injected into ventral hippocampal CA1 of Calb2-Cre mice. The presence of hyperphosphorylated or misfolded tau was examined using the antibodies AT8, PHF1, and MC1. In separate experiments using the same tissue, thioflavin-S staining was used to detect the presence of neurofibrillary tangles. Images were analyzed for the presence of tau forms at different timepoints following injection to examine the progression of pathological tau variant formation in the hippocampus. These results identified the vulnerability of select subtypes of hippocampal interneurons to the trans-synaptic spread of tau. Using these data, we can target neuronal subtypes of hippocampal CA1 that are vulnerable to the trans-synaptic spread of tau for future functional experiments.

Disclosures: E. Leggett: None. **A.R. McQuiston:** None.

Poster

P223. Tau: Animal and Cellular Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P223.05

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH grant R01AG063743

Title: Rev-erb-alpha Regulates tau accumulation via modulating microglia phagocytosis in a sex-dependent manner

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Abstract: REV-ERBα is a nuclear receptor and circadian clock component that has been implicated in the regulation of microglial activation. Disrupted REV-ERBα (*Nr1d1*) expression abrogates endogenous circadian machinery in microglia and increases their phagocytic activity. Microglia exhibit sex-dependent responses to disease-associate stimuli, but is incompletely understood how this is regulated. Here, we show that microglial REV-ERBα influences Tau pathology in a sex-dependent manner and investigate sex-specific transcriptome changes of REV-ERBα deficient microglia. Microglial-specific REV-ERBα deletion in female Tau P301L-expressing mice resulted in less accumulation of phosphorylated Tau, but slightly increase tau in male mice. We verify that the absence of REV-ERBα in microglia accelerates the clearance of injected FITC-conjugated Tau monomer protein in the hippocampus, particularly in female mice. These data implicate that understanding the sex-dependent effects of REV-ERBα on tauopathy will inform future therapeutic efforts targeting REV-ERBs, and shed light on the mechanisms underlying the increased risk of AD in women.

Disclosures: J. Lee: None. **J.M. Dimitry:** None. **E.S. Musiek:** None.

Poster

P223. Tau: Animal and Cellular Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P223.06

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: The role of ovarian hormones in mediating the neuroprotective effects of irisin in tauopathy model mice

Authors: *S. J. TERRILL, K. A. BRETLAND, L. LIN, G. LEONE, S. M. FLEMING, C. M. DENGLER-CRISH;
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Abstract: Tauopathies, including Alzheimer's disease (AD), progressive supranuclear palsy, corticobasal syndrome, and chronic traumatic encephalopathy, are progressive neurodegenerative disorders that are pathologically defined by neurofibrillary tangle formation, a result of pathological aggregation of misfolded and abnormally phosphorylated tau protein. These progressive and fatal brain disorders gradually destroy an individual's memory, ability to learn, reason, and communicate. Despite substantial scientific effort over the last few decades, there are currently no established therapies that modify disease progression of tauopathies. Recently, the novel exercise-induced hormone irisin has emerged as a potential therapeutic agent for the treatment of neurodegenerative diseases. New evidence suggests this hormone can protect the brain against neuronal degeneration, yet the biological mechanisms mediating the neuroprotective effects of irisin remain poorly understood. Recently, we have demonstrated that peripherally administered recombinant human irisin reduces hippocampal tau hyperphosphorylation and neuroinflammation in female but not male htau mice, a transgenic mouse model that develops tauopathy similar to AD and frontotemporal dementia. Here we directly assessed whether endogenous female sex hormones are necessary for the expression of neuroprotective factors following irisin treatment in female htau mice. Following bilateral ovariectomy (OVX) or sham surgical procedure, female htau and control mice received weekly injections of recombinant irisin (i.p.). Following 4 weeks of injections, brain tissue was processed for changes in key proteins associated with inflammation and pathology after vehicle or irisin treatment. Our results demonstrate that ovarian hormones may mediate irisin efficacy in reducing early accumulation of hyperphosphorylated tau in the hippocampus of female htau mice. However, our results also suggest that irisin's ability to reduce neuroinflammation (TNFα) does not require intact ovarian cycling. Collectively, these preliminary findings suggest that female sex hormones may facilitate the neuroprotective effect of irisin in female htau mice. Ongoing analyses will further elucidate neurobiological mechanisms through which irisin and estrogens modulate brain pathology in htau mice.

Disclosures: S.J. Terrill: None. **K.A. Bretland:** None. **L. Lin:** None. **G. Leone:** None. **S.M. Fleming:** None. **C.M. Dengler-Crish:** None.

Poster

P223. Tau: Animal and Cellular Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P223.07

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Angelo State Faculty Research Enhancement Program (FREP)

Title: The effects of high-fat diet administration on cognitive and non-cognitive behaviors in a tau mouse model of Alzheimer's disease

Authors: *S. L. P. LIPPI[1], B. A. COLLINS[1], A. E. HOWARD[1], M. N. RODRIGUEZ[1], G. L. PARRISH[1], J. A. OLIVAREZ[2], A. PORRAS[1];
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Abstract: Alzheimer's disease (AD) is a neurodegenerative condition that leads to deficits in memory, alterations in behavior, and changes in activities of daily living. While poor dietary choices, in the form of high-fat diet (HFD) administration, have been shown to impair spatial memory and exacerbate AD pathology in APP mice, few studies have assessed the effects of HFD on mice modeling solely neurofibrillary tangle (NFT) pathology. We examined the effects of HFD on both cognitive and noncognitive behaviors in rTg4510 (P301L) mice. Preliminary analysis reveals that HFD does lead to significant weight gain ($p < 0.05$) and while genotype effects were strong in the Morris Water Maze (MWM) spatial memory task, with tau mice performing worse than wildtype (WT) mice ($p < 0.01$), HFD had no effect on spatial memory measures. Tau mice spent significantly less time in the open arms of the elevated zero maze (EZM) than WT mice ($p < 0.05$) and took longer to enter into the open arms ($p < 0.05$). No differences in distance (cm) or time spent in the center of the open field test were seen. While HFD did not have any effects on anxiety or spatial memory, it did have an effect on nesting behavior (non-cognitive behavior). Tau mice built significantly poorer nests than WT mice ($p < 0.001$) and HFD significantly affected nest building; however, it was dependent on genotype. In WT mice, HFD impaired nest building ($p = 0.002$) while in tau mice HFD improved nesting score ($p < 0.001$) compared to the control diet (CD) counterparts. Results from other studies show that HFD impairs spatial memory while the current preliminary data suggest otherwise. Upcoming analyses on brain tissue examining inflammatory markers, phosphorylated tau, and neurotrophins (BDNF) aim to explore the effects of HFD on NFT pathology and brain health in this tau mouse model.

Disclosures: S.L.P. Lippi: None. **B.A. Collins:** None. **A.E. Howard:** None. **M.N. Rodriguez:** None. **G.L. Parrish:** None. **J.A. Olivarez:** None. **A. Porras:** None.

Poster

P236. Therapeutic Strategies: Preclinical Animal Models and Non-Pharmacological Therapeutic Approaches I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P236.01

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: AG014449
AG017617
AG043375

Title: Stereological analysis of cholinergic and GABAergic neuron morphology in the basal forebrain of young and aged Ts65Dn mice following maternal choline supplementation (MCS)

Authors: *M. K. GAUTIER[1,3,4], C. M. KELLEY[8], S. LEE[2,5], E. J. MUFSON[8,9], S. D. GINSBERG[1,4,6,7];
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Abstract: Down syndrome (DS) is caused by the triplication of chromosome 21. By early middle-age, individuals with DS develop Alzheimer's disease (AD) neuropathology including amyloid-β plaques, neurofibrillary tangles, and loss of cholinergic neurons in the basal forebrain. The trisomic Ts65Dn mouse recapitulates key aspects of DS/AD pathology, including deficits in hippocampal dependent learning and memory, dysregulation of the endosomal-lysosomal (EL) system, and degeneration of basal forebrain cholinergic neurons (BFCNs). We previously showed maternal choline supplementation (MCS), a well-tolerated treatment modality, was able to attenuate overexpression of EL regulatory genes and rescue early endosomal defects within BFCNs of 10-12 month old (MO) Ts65Dn mice. To explore whether amelioration of endosomal pathology within the septohippocampal circuit affects neuronal atrophy, we performed morphometric analyses of vulnerable neuronal populations in the medial septum and vertical limb of the diagonal band (MS/VDB) of the basal forebrain. Utilizing novel, custom-designed stereology tools in ImageJ (COANSI), choline acetyltransferase (ChAT)-immunoreactive BFCNs were compared in Ts65Dn mice and normal disomic (2N) littermates at two timepoints: 3-4 MO (pre-BFCN degeneration) and 10-12 MO (post-BFCN degeneration). Quantitative analysis of fluorescent z-stacks revealed 10-12 MO Ts65Dn mice receiving perinatal MCS had significantly increased numbers of ChAT-positive BFCNs in the MS/VDB compared to age-matched trisomic mice from dams on a normal choline diet. Significant increases in neuronal density and area of the MS/VDB occupied by neuropil were also observed. MCS treated 3-4 MO Ts65Dn and 2N offspring also had increases in BFCN number and regional area, suggesting early life choline delivery has organizational effects on neurodevelopment of BFCNs independent of genotype. To assess the efficacy of MCS treatment on non-cholinergic populations, we investigated GABAergic neurons within the MS/VDB of 10-12 MO mice. Preliminary data suggests aged Ts65Dn mice experience a loss of parvalbumin-positive GABAergic neurons compared to 2N littermates, and that MCS rescues this population from degeneration. Taken together, these data suggest perinatal MCS has long-lasting neuroprotective benefits on diverse neuronal populations within the septohippocampal circuit and possibly throughout the CNS. Behavioral improvements in MCS treated Ts65Dn mice may reflect an ability to shield vulnerable basal forebrain neurons from progressive neurodegenerative processes.

Disclosures: M.K. Gautier: None. **C.M. Kelley:** None. **S. Lee:** None. **E.J. Mufson:** None. **S.D. Ginsberg:** None.

Poster

P236. Therapeutic Strategies: Preclinical Animal Models and Non-Pharmacological Therapeutic Approaches I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P236.02

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: The Effect of Melatonin on spatial learning and Aβ load in an Alzheimer's Disease Mouse Model

Authors: *A. G. RILEY-DIPAOLO[1], E. MILIOTOU[2], **A. J. STAVNEZER**[3];
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Abstract: The neurohormone melatonin is released fromform the pineal gland and is an endogenous antioxidant and free radical scavenger that regulates sleep/wake cycle and circadian rhythm. Decline of nocturnal melatonin secretion with severe sleep disturbance has been consistently reported in Alzheimer's Disease (AD), a neurodegenerative disease characterized by cognitive deficits and neuronal loss. However, exogenous melatonin supplementation has been repeatedly found tocan reduce Aβ neurotoxicity and formation while improving cognitive performance and sleep quality. In this study, the long-term influence of melatonin on behavioral and neuropathological changes were evaluated in the 5xFAD mouse model, when treatment was started in adolescence. The 5xFAD mouse is a transgenic model of AD that mimics the accumulation of senile plaques, neuronal apoptosis and memory deficits. After ~204 weeks of orally administered melatonin (10 mg/kg), open field and Morris water maze behaviour were was assessed in 54 mice, followed by quantification in hippocampal tissue. Overall, transgenic mice showed decreased anxiety, impaired learning, and a significant increase in plaque load, replicating previous results with this strain. Melatonin marginally improved learning in transgenic mice by decreasing distance travelled in the Morris water maze and slightly decreasing concentration in the hippocampus of 5xFAD mice compared with untreated 5xFAD. These results support the hypothesis that long-term melatonin supplementation may be beneficial at early stages of the disease process and should be explored more fully. Abnormal accumulation of neurotoxic forms of Aβ occur in brain regions involved in the regulation of sleep in AD patients and the disturbed regulation of sleep that occurs early in the disease may contribute to cognitive decline by decreasing Aβ clearance. Early melatonin interventions may be one of the most promising strategies in the development of methods to retard or prevent Aβ and memory deficits during this stage of the disease. In sharp contrast to conventional antioxidants, melatonin

crosses the blood-brain barrier, is relatively devoid of toxicity, and constitutes a straight-forward potential therapeutic candidate in AD treatment, perhaps due to improving sleep quality and clearing Aβ plaques.

Disclosures: A.G. Riley-DiPaolo: None. **E. Miliotou:** None. **A.J. Stavnezer:** None.

Poster

P236. Therapeutic Strategies: Preclinical Animal Models and Non-Pharmacological Therapeutic Approaches I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P236.03

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant 1R01AG060754-01A1

Title: The effects of electrical stimulation of the cholinergic forebrain on visuospatial working memory and neurotrophic and beta-amyloidogenic markers in a murine model

Authors: *J. KUMRO[1], A. TRIPATHI[3], J. SWORD[1], A. PILLAI[4], A. I. VAZDARJANOVA[5], A. V. TERRY, JR[2], D. T. BLAKE[6];
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Abstract: The cholinergic forebrain or nucleus basalis of Meynert (NBM) provides acetylcholine to the entire cortical mantle and degenerates sooner than other brain regions during Alzheimer's disease (AD). The current frontline AD therapy of cholinesterase inhibitors improves cognition for about half of a year until becoming ineffective. Our previous research demonstrated that intermittent, instead of continuous, electrical stimulation of the NBM improves working memory in a young adult primate model. Here we implanted bilateral electrodes into the cholinergic forebrain of wild-type and 5xFAD mouse models at 4 months old. Mice received daily NBM intermittent stimulation for 1 hour at 100uA until the age of 9 months. Both stimulated mouse groups outperformed unstimulated groups during learning of the Morris Water Maze task. However, only 5xFAD treated mice significantly outperformed their control group during the probe test. In both groups, treatment increased expression of nerve growth factor's receptor TrkA while reducing expression of the amyloidogenic enzyme beta-site APP cleaving enzyme 1 (BACE1), as well as displaying reduction in Aβ(1-42) concentration. To determine the necessity of cholinergic receptor subtypes and their roles in eliciting these expression changes, we then bilaterally implanted NBM electrodes into 4 month-old mice and unilaterally stimulated one hemisphere for 2 weeks while simultaneously antagonizing different cholinergic receptor subtypes. Comparing the hemispheric differences of expression of

neurotrophic and beta-amyloidogenic markers between receptor subtype groups identifies which subtypes elicit the observed treatment response.

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Poster

P236. Therapeutic Strategies: Preclinical Animal Models and Non-Pharmacological Therapeutic Approaches I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P236.04

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Evaluating routes of administration for AAV-vectored immunotherapy for Alzheimer's Disease

Authors: *K. J. SPILLER[1], M. HORIUCHI[2], B. DAWE[1], C. J. HINDERER[2], A. BOTTELBERGS[3], R. V. ARAS[1], L. KELLAM[2], E. SCHAEFFER[4], J. M. WILSON[2], E. A. RAMSBURG[5];
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Abstract: Background: Although the pathogenesis of Alzheimer's Disease (AD) is not completely understood, most researchers agree that the symptoms of AD result from the accumulation of aggregated tau and amyloid beta, both intra- and extracellularly. The 3D6 monoclonal antibody (mAb) has previously been shown to clear amyloid plaques when repeatedly delivered passively in mouse models of AD and a humanized version of the antibody (bapineuzimab) progressed to Ph3 clinical trials at which time development was halted because of lack of efficacy in the tolerable dose range. We reasoned that vectored delivery might enable efficient clearance of amyloid with only a single injection, and might even mitigate vascular toxicities, depending on the route of administration. **Methodology & Results:** In partnership with UPenn, we tested AAV-vectored antibody therapeutics for AD. After delivery of AAV vector-encoded 3D6 (1e11 genome copies, single injection), we show target engagement in the brains of three different mouse models of Alzheimer's disease by measuring the Aβ plaque-binding. Further, using both histological readouts in APP-KI mice and binding measurements in rat interstitial fluid, we show that there is limited spread of this secreted transgene product after neuronal transduction in the parenchyma. Finally, we used aged amyloid transgenic mice (PDAPP) that overexpress human amyloid precursor protein (APP) to compare efficacy of AAV vector-encoded 3D6 (1e11 genome copies, single injection) versus passive delivery of 3D6 (3

mg/kg/week). Animals receiving AAV had equivalent amyloid clearance when compared to those receiving 3D6 passively. Unfortunately, vascular toxicities occurred after both delivery methods. **Conclusion:** Our collaborative efforts demonstrate that a one-time AAV injection is an effective vehicle for antibody delivery in neurodegenerative disorders. In these rodent models, intramuscular delivery of a vectored antibody allowed for widespread target engagement in the brain. Further, CNS-directed gene therapy can concentrate therapeutic molecules in the brain to ameliorate pathologies associated with neurodegenerative disorders.

Disclosures: K.J. Spiller: A. Employment/Salary (full or part-time):; Janssen R&D. **M. Horiuchi:** None. **B. Dawe:** A. Employment/Salary (full or part-time):; Janssen R&D. **C.J. Hinderer:** None. **A. Bottelbergs:** A. Employment/Salary (full or part-time):; Janssen R&D. **R.V. Aras:** A. Employment/Salary (full or part-time):; Janssen R&D. **L. Kellam:** None. **E. Schaeffer:** A. Employment/Salary (full or part-time):; Janssen R&D (at the time of the study). **J.M. Wilson:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Sponsored research agreements with Ultragenyx, Biogen, Janssen, Precision Biosciences, Moderna, Inc., Scout Bio, Passage Bio, Amicus Therapeutics, and Surmount Bio.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Paid advisor to and holds equity in Scout Bio and Passage Bio; he holds equity in Surmount Bio. **E.A. Ramsburg:** A. Employment/Salary (full or part-time):; Janssen R&D (at the time of the study).

Poster

P236. Therapeutic Strategies: Preclinical Animal Models and Non-Pharmacological Therapeutic Approaches I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P236.05

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH T32 AG 066596
NIH 5R01AG048218-06

Title: In vivo testing of a CRISPR based gene therapy for Alzheimer's disease

Authors: *B. AULSTON[1], J. SUN[2], K. BRANES[1], N. CHECKA[1], S. ROY[1];
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Abstract: To date there is no therapeutic agent that can slow down the relentless course of Alzheimer’s disease (AD). Surprisingly, very few therapeutic strategies target amyloid precursor

protein (APP) - a gene that plays an unequivocal role in AD pathogenesis. Key evidence includes APP-duplications that lead to familial disease and an APP “Icelandic” mutation is protective for sporadic AD. Our lab has developed a novel CRISPR-based APP-editing approach that favorably modulates APP-cleavage - attenuating pathologic ß-cleavage, while upregulating neuroprotective a-cleavage. Importantly, the vast majority of the APP molecule - including the elaborate N-terminus and membrane-spanning domain - remains intact in this setting, ensuring that physiological processes continue unabated. Our strategy uses gene editing to effectively delete the YENPTY endocytic domain at the extreme C-terminus of APP, a pentapeptide motif facilitating ß-cleavage and Aß production. In this submission, we present in vivo evidence that our APP-editing strategy is both safe and efficacious. First, in order to analyze our strategy in “an ideal setting” we created two novel mouse lines in which the C-terminus of APP was genomically edited using APP CRISPRs. To generate mice, APP gRNA-Cas9 ribonucleoproteins were injected into APP knockin ($APP^{NL/G/F}$) and Wt, C57 zygotes. Two types of indels were identified at the APP C-terminus editing site in P0 mice: a thymine insertion (InsT) and 5 bp deletion (Del5) both of which shift the reading frame of APP and disrupt the Y188 motif. APP KI and C57 “ΔC-term” term mice were then bred to homozygosity for experiments. Homozygous APP KI ΔC-term showed attenuated amyloid pathology and neuroinflammation compared to APP KI controls and no gross histological defects were observed in C57 ΔC-term mice. In a second set of experiments we introduced APP CRISPRs into $APP^{NL/G/F}$ mice using AAV vectors. Control (scrambled sequence) and APP targeting gRNA constructs were packaged into AAV-PHP.eB vectors and viruses were retro-orbitally injected into 6week-old APP KI, Cas9 expressing mice. Mice were maintained for 2.5 weeks post-injection then sacrificed for tissues analysis. IHC and WB analysis of brain tissue revealed robust editing of the APP C-terminus in APP CRISPR treated mice which was associated with a dramatic reduction of amyloid plaques. In total, these data demonstrate the in vivo feasibility of APP C-terminus CRISPR editing as a treatment strategy for AD.

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Poster

P236. Therapeutic Strategies: Preclinical Animal Models and Non-Pharmacological Therapeutic Approaches I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P236.06

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Can Lineage negative stem cells derived from Umbilical Cord Blood of female Yoga practitioner reverse Amyloid beta induced memory loss in mice?

Authors: *P. NADHOLTA[1], P. SAHA[1], V. SURI[1], A. SINGH[2], K. SONI[1], S. RAIKAR[2], M. LELE[2], S. JINDAL[2], P. VERMA[3], K. MAITY[2], A. ANAND[1];
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Abstract: Can Lineage negative stem cells derived from Umbilical Cord Blood of female Yoga practitioner reverse Amyloid beta induced memory loss in mice?
Authors: Pooja Nadholta*, Pradip K Saha, Vanita Suri, Amit Singh, Krishan K Soni, Shravani V Raikar, Mugdha R Lele, Sheetal Jindal, Prashant Verma, Kalyan Maity, Akshay Anand
Conflict of Interest: None.
Abstract: Amnesia is a major clinical problem among elderly. Alzheimer disease is one of Neuro-degenerative disease which is associated with memory loss during old age and no reliable treatment is available despite of many clinical trials. In this study we aimed to determine the efficacy of Lin-ve stem cells (SC) derived from Umbilical cord blood (UCB) of Yoga practitioner mothers during pregnancy in reversal of Amyloid beta (Aβ) induced memory loss and simultaneously the pathways involved with this reversal. The efficacy of the UCB has already been studied in enhancing memory in such memory loss models. We want to see whether the UCB from Yoga practitioner mothers has more stemness and effects in memory reversal or not. This study included both human population (Pregnant women aged b/w 18 to 35 , n=13) and animal model (Swiss albino,6-8 weeks old, 6 groups) of brain injury, which was created via administration of Aβ in hippocampal region of mice brain. Participants were given Yoga intervention from 16th-18th week onward till delivery (4 months). UCB was collected, Lin+ve and Lin-ve population were separated and stemness of Lin-ve SC was assessed through flow cytometery for markers such as CD34 and CD117. Aβ was injected in hippocampus stereotactically to create memory loss and after 21 days of Aβ administration, Lin-ve SCs were injected through same path at the same location. Neurobehavioral tests (Morris Water Maze & Passive avoidance), Immunohistochemistry (IHC), and qPCR techniques were used in this study to examine memory loss, memory reversal and then various molecules involved in the pathway. Neuro-behavioral analysis after 10 days of transplantation
showed improved memory. Yoga can be adopted as a lifestyle during pregnancy for improved outcomes and Lin-ve SCs derived from these practitioners can be considered a good source of memory reversal in amnesia and dementia patients however, more studies are warranted in the arena.

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Poster

P236. Therapeutic Strategies: Preclinical Animal Models and Non-Pharmacological Therapeutic Approaches I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P236.07

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Reduction of beta-amyloid pathology and improvement of cognitive functions in an Alzheimer's disease mouse model by targeting acid sphingomyelinase in blood with a novel anti-ASM monoclonal antibody

Authors: M. PARK[1], **S.-B. HONG**[2], B. CHOI[3], K. PARK[1], M. HONG[2], B. SHIN[2], J.-I. CHO[2], W. KIM[4], J. LEE[4], M. SO[4], H. JIN[3], *J.-S. BAE[1];
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Abstract: Elevated activities of acid sphingomyelinase (ASM) in the blood of Alzheimer's disease (AD) patients and normal aged persons are implicated in the AD pathology, and age-related blood-brain barrier dysfunction and subsequent neurodegeneration. Here we sought to determine whether secreted ASM in blood is pivotal in AD pathogenesis and can be a therapeutic target by inhibiting ASM activity with a novel anti-ASM monoclonal antibody in a mouse model of AD. A high affinity monoclonal anti-ASM scFv (clone#: 9104), cross-reactive to both human and mouse ASM was identified by screening a synthetic human scFv phage library. A chimeric anti-ASM antibody (9104-mIgG1; human Fab with mouse IgG1 Fc) was generated for animal studies. Although the anti-ASM antibody did not inhibit purified recombinant ASM, it inhibited ASM activity present in the plasma membrane and thus reduced the levels of ceramide, a catabolized product of sphingomyelin, in the plasma membrane. As previously demonstrated, ASM activity was significantly elevated in the plasma of AD model mice (APP/PS1) compared to wildtype littermates (wildtype: 0.145±0.04 nmol/mg/h; APP/PS1: 0.251±0.05 nmol/mg/h). Upon repeated intraperitoneal injection of the anti-ASM antibody (50 mg/kg, Q2W for 8 weeks), ASM activity was reduced by 64% in the plasma (APP/PS1-9104-mIgG1: 0.090±0.10 nmol/mg/h). On the contrary, ASM activities in the brain (cortex and hippocampus) were not affected possibly due to blood-brain-barrier that limits penetration of the antibody. Cognitive functions were significantly improved as demonstrated by Morris' water maze and fear conditioning tests. Amyloid plaques and soluble amyloid beta peptides (Aβ40 and Aβ42) were significantly reduced in both cortex and hippocampus. GFAP (astrocyte marker) expression was also significantly reduced in both cortex and hippocampus, while phospho-Tau and Iba-1 (microglia marker) expression was lower but not to the level of significance. The presented results indicate that elevated ASM in blood is a potential therapeutic target for AD and a monoclonal anti-ASM antibody would be an applicable therapy that indirectly reduces amyloid-beta pathology.

Disclosures: M. Park: None. **S. Hong:** None. **B. Choi:** None. **K. Park:** None. **M. Hong:** None. **B. Shin:** None. **J. Cho:** None. **W. Kim:** None. **J. Lee:** None. **M. So:** None. **H. Jin:** None. **J. Bae:** None.

Poster

P236. Therapeutic Strategies: Preclinical Animal Models and Non-Pharmacological Therapeutic Approaches I

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Program #/Poster #: P236.08

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIA Grant AG057895-01A1
Bass Connections Grant, Duke University
R01AG066184

Title: Exercise-type specific increases in memory and decreased anhedonia in CVN-AD mice, a model of familial Alzheimer's disease, accompanied by increased global cerebral vascular perfusion

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Abstract: Alzheimer's disease (AD) is a complex, age-dependent condition influenced by genetic and environmental factors, and there is increasing evidence to support the role of physical exercise in slowing AD progression. This study aimed to examine the relationship between AD behavioral impairments, types of chronic exercise and cerebral blood flow (CBF). Female, 52 wk old, CVN-AD mice (APPSwDI/mNos2-/- which have mutated APP and a lowered expression of NOS2) were exercised, from 28 to 52 weeks of age (WOA) on one of two exercise regimens: Voluntary (VOL) - wheel running 6-8 hrs/day, 5 days/wk or voluntary plus enforced (ENF) - wheel running plus 45 min treadmill training 2 days/wk. Sedentary (SED) CVN-AD mice were controls. Daily wheel running distance was greater in ENF mice than VOL mice and body wts of all mice decreased with increased cardiovascular effort. Because symptoms of AD include both memory loss and anhedonia, mice were trained after 8 and 24 wks of exercise on a novel object recognition task and a sucrose preference test. At both timepoints, SED mice showed no memory for the familiar object and no consistent preference for sweetened water. While VOL mice also showed no memory for the familiar object after 8 wks of running, they did show novel object preference after 24 wks. Like the SED mice they did not show consistent sucrose preference. In contrast, ENF mice showed novel object preference at both

time points and consistent sucrose preference after 24 wks of training, demonstrating that the combined exercise regimen was best at improving memory and reducing anhedonia. In vivo MRI used a 7.1T Bruker imaging system with 20 cm bore and 440 mT/m gradient set, with a 72 mm volume excitation coil, and a 4 channels surface receive coil. We used pseudo continuous Arterial Spin Labeling for estimation of CBF. To quantify CBF, T1 maps were acquired at the same resolution using an inversion recovery sequence, and spin labeling efficiency was measured in the brain feeding arteries using an FcFLASH protocol in one single slice, for each mouse. In vivo perfusion imaging of whole brain of AD-mice revealed CBF was the most variable in SED mice, followed by ENF mice. ANOVA revealed that perfusion values differed with exercise ($F(2,24)=3.7$, $p=0.04$). Post hoc tests revealed that CBF values were higher in VOL relative to SED mice ($p=0.04$), suggesting that CBF increases with chronic voluntary exercise, but added treadmill training prevents this increase, perhaps due to chronic stress from enforced exercise. Interestingly, higher CBF did not correlate with more exercise, improved memory, or lower anhedonia. Regional blood flow analysis in targeted brain regions may better explain these data.

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Poster

P236. Therapeutic Strategies: Preclinical Animal Models and Non-Pharmacological Therapeutic Approaches I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P236.09

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH 800105

Title: Novel bi-level gene regulation underlies Tip60 HAT mediated neuroprotection in Alzheimer’s disease

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Abstract: Epigenetic mechanisms, such as histone acetylation modifications, play a crucial role in Alzheimer’s disease (AD) pathogenesis. Accordingly, our lab has shown that downregulation of Tip60 histone acetyltransferase (HAT) in the human AD hippocampus and the *Drosophila* AD

brain results in reduced chromatin acetylation and concomitant epigenetic repression of critical synaptic genes. Remarkably, these phenotypes are prevented by Tip60 overexpression in the *Drosophila* AD brain, supporting a neuroprotective role for Tip60 in AD. Although Tip60's role in chromatin dynamics has been well established, alternative Tip60 cellular mechanisms remain unexplored. Here, we show a novel RNA binding function for Tip60 HAT *in vivo* in the *Drosophila* brain. Using genome-wide sequencing of the RNA molecules bound with Tip60, Tip60's RNA binding function was found to be highly specific, selective and reproducible with its RNA targets being enriched for critical neuronal processes and AD. Remarkably, 77% of Tip60's RNA targets overlap with its gene targets, suggesting Tip60 orchestrates a bi-level gene regulation at the chromatin and the RNA levels that is unprecedented for any other HAT. Further, Tip60's RNA binding function in *Drosophila* is conserved in human hippocampal tissue and appeared compromised in AD patients, demonstrating mammalian conservation of Tip60-RNA interactions while further underscoring human AD relavance. Preliminary data suggests a role for Tip60 in alternative splicing and loss of Tip60 in AD could negatively affect splicing of its RNA synaptic gene targets, therefore contributing towards AD pathogenesis. We are first to uncover an RNA binding function for Tip60 HAT and propose a novel bi-level gene regulation that may underly Tip60's neuroprotection in AD-linked neurodegeneration.

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Poster

P259. Alpha-Synuclein: Transmission

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Program #/Poster #: P259.01

Topic: C.03. Parkinson's Disease

Support: Aufzien Family Center for the Prevention and Treatment of Parkinson's Disease

Title: Intranasal administration of CSF-derived extracellular vesicles from patients with Parkinson's Disease induce Parkinson-like symptoms and pathology

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Abstract: Parkinson's disease (PD) is characterized by the gradual appearance of intraneuronal lewy aggregates (Lewy bodies), which are primarily composed of misfolded α-synuclein (α-syn), resulting in cytotoxicity and neural death. Recent *in-vitro* and *in-vivo* studies support the idea of transcellular spread of misfolded α-syn in a prion-like transmission via extracellular vesicles (EVs) secretion and induce pathological aggregates in healthy neurons. EVs derived from brain

lysates and cerebrospinal fluid (CSF) of PD patients have been shown to propagate α-syn aggregation in healthy cells. Braak et al. hypothesized the olfactory bulb as a primary propagation site for the initiation of the disease. To examine whether pathological α-syn can actively spread from the nasal cavity to the brain via EVs and initiate pathological aggregate in the brain, we isolated CSF-derived EVs from PD patients or from non-synucleinopathies patients (non-PD). Healthy 2-months old C57/Bl mice were given EVs isolated from patient's CSF (PD or non-PD) by internasal administration. 3 months after treatment mice were subjected to behavior testing and examined for PD-related brain pathology. Intranasal delivery of CSF-derived EVs from PD patients to healthy mice induced PD-like symptoms and pathology; PD-treated mice had impairments in motor behavior, hyposmia and elevated anxiety compared to non-PD treated and healthy untreated mice. Striatums of PD-EVs treated mice revealed increased phosphorylated ser129 α-syn aggregation, decreased tyrosine hydroxylase expression, astrogliosis and impaired autophagy compared to non-PD treated and healthy treated mice. Together, these data suggest that EVs from PD patients are sufficient for propagating α-syn aggregation *in-vivo* and initiating PD-like symptoms and pathology in healthy mice when delivered by intranasal routes.

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Poster

P259. Alpha-Synuclein: Transmission

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Program #/Poster #: P259.02

Topic: C.03. Parkinson’s Disease

Title: Mao-b inhibition facilitates α-synuclein secretion via an abc transporter-mediated pathway and delays its aggregation

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Abstract: α-Synuclein (α-syn) is secreted under physiological conditions in mouse primary neurons and brain interstitial fluids, showing that extracellular α-syn may play a key role in cell-to-cell transmission of its pathology in Parkinson’s disease (PD). However, the mechanisms that regulate extracellular α-syn secretion are not fully elucidated. In this study, we investigated effects of monoamine oxidase-B (MAO-B) inhibition on α-syn secretion in cultured cells (n=3~6 per group) and its aggregation in recombinant adeno-associated virus (rAAV)-based rat models (male Sprague-Dawley rat, n=3~8 per group). In SH-SY5Y cell line stably expressing wild-type α-syn, both inhibition of MAO-B activity by selegiline or rasagiline and siRNA-mediated

knockdown of MAO-B facilitated α-syn secretion. Conversely, TVP-1022, the 1,000 times less active S-isomer of rasagiline, did not alter α-syn secretion. This MAO-B inhibition-induced increase in α-syn secretion was not blocked by brefeldin A, which inhibits endoplasmic reticulum/Golgi classical transport, while it was blocked by probenecid and glyburide, which inhibit ATP-binding cassette (ABC) transporter function. MAO-B inhibition preferentially facilitated the secretion of detergent-insoluble α-syn protein and reduced its intracellular accumulation under chloroquine-induced lysosomal dysfunction. Also, in mouse primary cortical neurons, α-syn secretion was facilitated by MAO-B inhibition and blocked by ABC transporter inhibition. Moreover, in a rat model injected rAAV-A53T α-syn into the substantia nigra, administration of selegiline delayed the striatal formation of Ser129-phosphorylated α-syn aggregates and loss of nigrostriatal dopaminergic neurons. Selegiline delayed α-syn aggregation and dopaminergic neuronal loss in a cell-to-cell transmission rat model generated by injecting rAAV-wild-type α-syn and externally inoculating α-syn fibrils into the striatum. These findings suggest that MAO-B inhibition modulates the intracellular clearance of detergent-insoluble α-syn via the ABC transporter-mediated non-classical secretion pathway in vitro, and temporarily delays the formation and transmission of α-syn aggregates in rat models. We propose a new function of MAO-B inhibition that modulates α-syn secretion and aggregation in PD treatment.

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Poster

P259. Alpha-Synuclein: Transmission

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Program #/Poster #: P259.03

Topic: C.03. Parkinson's Disease

Support: NS108686
NS112540

Title: Loss of tau expression attenuates neurodegeneration associated alpha synuclein pathology in vivo

Authors: *S. C. VERMILYEA[1], A. CHRISTENSEN[1], B. SINGH[1], R. KARIM[1], J. MEINTS[1], M. K. LEE[1,2];
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Abstract: Parkinson's Disease (PD) is a progressive neurodegenerative disease characterized by the loss of several neuronal populations, including dopaminergic neurons of the substantia nigra pars compacta. Another hallmark of the disease is the identification of intracellular proteinaceous inclusions called Lewy bodies (LBs) and Lewy neurites (LNs). The small presynaptic protein α-

synuclein (αS) is the major component of LBs and LNs in PD and highly related LB Dementia (LBD). Neuronal dysfunction and degeneration linked to α-synuclein (αS) pathology is thought to be responsible for the neurodegeneration in PD and LBD. However, studies indicate bidirectional pathological relationships between αS pathology and microtubule associated protein tau abnormalities. For example, we showed that A53T mutant human αS (HuαS) can cause tau dependent post-synaptic and cognitive deficits. Therefore, we examined whether tau is involved in the onset and progression of overt α-synucleinopathy. We induced α-synucleinopathy by intramuscular (IM) injections of HuαS preformed fibrils (PFF) in A53T mutant transgenic mice (TgA53T), in $mTau^{-/-}$ or wildtype background. IM inoculation of TgA53T mice leads to motor dysfunction onset by ~70 days post inoculation (dpi) and end stage paralysis by ~100 dpi. Therefore, motor behavior at 70 dpi and disease progression were evaluated as well as biochemical and histological analysis at 40dpi, 70dpi, and end stage.Significantly, $TgA53T/mTau^{-/-}$ mice exhibit reduced motor deficits at 70 dpi measured by rotarod performance as well as a significant delay of disease onset, progression, and time to reach end stage criteria. Analysis of neuropathology shows that end stage TgA53T mice and $TgA53T/mTau^{-/-}$ mice show comparable pathology. However, at 70 dpi, $TgA53T/mTau^{-/-}$ mice had modest yet significant reductions of α-synucleinopathy, as well as inflammatory markers Iba1 and GFAP. In addition, tau removal delayed ventral horn motor neuron degeneration at 70 dpi. Interestingly, no differences were observed at 40 dpi. Similarly, *in vitro* application of PFF to primary hippocampal neurons demonstrated no change of PFF uptake and processing, nor pS129αS aggregation as a function of tau expression. However, PFF-induced neurotoxic indicators including morphology and postsynaptic density deficits in wildtype neurons were prevented with tau removal. We conclude that while tau expression does not impact onset and progression of αS aggregation, loss of tau expression protects neurons from downstream toxic effects of αS aggregation. Therefore, tau reduction and the pathways activated may represent novel therapeutic targets for α-synucleinopathy.

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Poster

P259. Alpha-Synuclein: Transmission

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Topic: C.03. Parkinson's Disease

Support: NS012093
NS086074

Title: Differential metabolism of α-Syn pre-formed fibril (PFFs) in neuronal and non-neuronal cells

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Abstract: Parkinson's disease (PD) and other α-synucleinopathies are characterized by the accumulation of α-synuclein (αS) aggregates. Studies indicate that αS pathology can spread via the cell-to-cell transmission of αS pathology. To better understand how various brain cells contribute to the spreading of αS pathology, we examined the metabolism of αS pre-formed fibril (PFFs) by neurons and glial cells (microglia, astrocytes, and oligodendrocytes). Our results show that microglia and astrocytes rapidly metabolize αS PFF where the half live of αS PFF in these glial cells are ~5 hours. In neurons, while the full-length αS rapidly disappears following αS PFF uptake, truncated αS accumulates with the half-life of over 48 hours. Epitope mapping and fractionation studies indicate that in αS PFF is truncated at C-terminal region and remains insoluble/aggregated. The differential processing of αS can be recapitulated in cell lines as differentiated CLU neuronal cell lines show stable accumulation of truncated αS while undifferentiated cells rapidly metabolize αS. Immunolocalization and subcellular fractionation studies show that αS PFF is initially localized to endosomes followed by lysosomes. Inhibition of lysosomal function leads to stabilization of full-length and truncated αS in all cell types. Our data show that lysosomal degradation system is involved in the metabolism of extracellular αS aggregates taken up by various cell types. Further, our studies indicate that glial cells protect neurons αS aggregates by rapid clearance of αS aggregates while αS aggregates taken up by neurons are metabolized to aggregation prone C-terminally truncated αS.

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Poster

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Indiana Clinical and Translational Sciences Institute

Title: Brain region-specific characteristics of alpha-synuclein propagation in synucleinopathy models

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Abstract: In Parkinson's disease (PD) and other synucleinopathy disorders, aggregated forms of the presynaptic protein alpha-synuclein (aSyn) can spread throughout the brain via anatomically connected neuronal pathways. Data collected over the past 10 years have revealed that the propagation mechanism involves the internalization of pathogenic aSyn aggregates by neurons via an endocytic mechanism, after which the internalized protein stimulates the conversion of endogenous cytosolic aSyn to the fibrillar form via a seeding mechanism. Despite these insights, factors that determine brain region-specific differences in propagation efficiency and propagation-related toxicity are poorly understood. To address this knowledge gap, we characterized aSyn preformed fibrils (PFFs) in terms of their brain region-specific spreading efficiencies and molecular perturbations in cellular and *in vivo* propagation models. We observed that neurons prepared from the hippocampal and cortical regions (but not the midbrain region) of embryonic rat brain showed evidence of aSyn inclusions. This selectivity was found to reflect differences in levels of aSyn expression and PFF-mediated proteasomal dysfunction, but not the efficiency of PFF uptake, in different types of neurons. Current efforts are focused on characterizing brain region-specific proteomic and lipidomic perturbations in rats injected with aSyn PFFs in the frontal cortex, amygdala, and substantia nigra. Data from an initial proteomics study revealed that elongation factor-2 was selectively down-regulated in the amygdala of PFF-treated animals. Moreover, a pilot lipidomics analysis revealed an apparent increase in lipids in the PC/SM, PE, DAG, and ceramide families in the amygdala and/or cortex (but not the SN) of PFF-injected rats. Collectively, these findings suggest that seeded aSyn aggregation occurs more readily in certain types of neurons and elicits brain region-specific proteomic and lipidomic changes. Ultimately, the results of these studies will deepen our understanding of the molecular underpinnings of the spread of pathology in synucleinopathy disorders, laying the groundwork for developing well-tailored intervention strategies in the brains of patients.

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Poster

P259. Alpha-Synuclein: Transmission

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Topic: C.03. Parkinson's Disease

Support: Spanish Ministry of Science and Innovation, Ref PID2020-120308RB-I00

Title: Prion-like spread of endogenous alpha-synuclein in the non-human primate brain

Authors: *A. FAJARDO-SERRANO[1], J. CHOCARRO[1], A. J. RICO[1], E. RODA[1], G. ARIZNABARRENA[1], E. LORENZO-RAMOS[1], S. ARRIETA[1], A. PEJENAUTE[1], A. HONRUBIA[1], A. VAZQUEZ[2], A. I. RODRIGUEZ-PEREZ[3], J. L. LABANDEIRA-GARCÍA[3], J. L. LANCIEGO[1];
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Abstract: Lewy bodies (LBs) are intracellular aggregates of misfolded alpha-synuclein observed in dopaminergic neurons of the substantia nigra pars compacta (SNpc) in Parkinson's disease (PD), representing the main pathological hallmark of this neurodegenerative disorder. Available experimental evidence supported that misfolded alpha-synuclein can spread in a prion-like fashion trans-synaptically, i.e. between interconnected neurons. In this regard, it has been postulated that synucleinopathy propagates from the enteric system to the dorsal motor nucleus of the vagus, then spreading rostrally until reaching the SNpc and finally the cerebral cortex (the so-called "Braak hypothesis"). To what extent this spread drives the clinical course of PD still is a matter of debate, in particular when evaluating both the functional and clinical relevance for this alpha-synuclein propagation. Surprisingly, first evidence for this spread came from PD patients transplanted with embryonic dopaminergic cells, where postmortem evaluation showed LB pathology in grafted cells. Although at the experimental level there is conclusive evidence for a prion-like dissemination of preformed alpha-synuclein fibrils through interconnected brain areas, interactions between exogenous fibrils and endogenous alpha-synuclein remained unclear. In other words, evidence showing transneuronal spread of endogenous alpha-synuclein is still lacking. Here we took advantage of a viral vector coding for human tyrosinase for driving a time-dependent neuromelanin (NMel) accumulation in dopaminergic cells of *Macaca fascicularis*. Furthermore, accumulation of NMel induced synucleinopathy (Lewy and Marinesco bodies) in SNpc neurons. Most importantly, LB-pathology was observed in pyramidal neurons located in cerebral cortical regions receiving dopaminergic afferents, such as the dorsal frontal gyrus and the precentral gyrus. In other words, here we provide the first evidence showing spread of endogenous alpha-synuclein in the macaque brain, thus providing a straightforward demonstration of the transneuronal propagation of endogenous alpha-synuclein.

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Poster

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JPB Foundation

Title: Neurexin 1alpha Mediates Export of Pathologic alpha-Synuclein

Authors: *X. YIN[1], X. MAO[1], S. LIU[2], S. S. KARUPPAGOUNDER[1], H. WANG[1], J. SONG[2], V. L. DAWSON[1], T. M. DAWSON[1];
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Abstract: Cell-to-cell transmission and accumulation of misfolded pathologic α-synuclein (α-syn) underlies Parkinson's disease (PD). What drives the export of pathologic α-syn out of cells is unknown. Here we show that Neurexin 1α (Nrxn1α) binding to α-syn preformed fibrils (α-syn PFF) initiates the exosome-associated exocytosis of pathologic α-syn. Disruption of Nrxn1α in cultured neurons blocks α-syn PFF propagation, which leads to accelerated pathologic α-syn accumulation, enhanced phospho-Ser129 α-syn phosphorylation and neurotoxicity. In vivo, the lack of Nrxn1α prevents the spread of α-syn PFF from the striatum to neighboring brain regions, resulting in a reduction in α-syn pathology throughout the brain. The reduction in the spread of pathologic α-syn leads to less DA neuron loss and neurophysiological and behavioral deficits. These results support an essential role for Nrxn1α in the pathogenesis of PD through facilitation of pathologic α-syn exosome-based release and identifies new opportunities to develop effective approaches to halt the progression of PD.

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Poster

P307. Mechanisms of Neurodegeneration

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P307.01

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

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GIST Research Institute (GRI) grant
NRF-2019R1A2C2087565

Title: Effects of cereblon on stress-activated chaperones and redox proteins.

Authors: U. AKBER[1], S. BONG[2], Z.-Y. PARK[2], *C.-S. PARK[1];
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Abstract: Effects of cereblon on stress-activated chaperones and redox proteins.
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***Correspondence:** Chul-Seung Park (cspark@gist.ac.kr)
Conflict of interests: The authors declare no conflict of interest.**Acknowledgments:** This work was supported by Mid-career Research Grant (NRF2019R1A2C2087565) and Original Technology Research Program for Brain Science (Grant NRF2018M3C7A1056293) of the National Research Foundation of Korea, funded by the Korean government, Ministry of Science and ICT.
AbstractChronic ultra-mild stress (CUMS) has been reported to induce an anhedonic-like state in mice that resembles some of the symptoms of human depression. Many individuals with depression have prevalent pathology of α-synuclein (αsyn) and tau in the brain whereas molecular chaperones protect neurons from the deleterious effects of αsyn and tau misfolding. In the present study, utilizing CUMS animal model of depression, we report the role of cereblon (CRBN, the substrate receptor of the CUL4A/B-Roc1-RBX1 E3-ubiquitin ligase) in regulating the activities of chaperones DNAJA1 (DJ2) and DNAJB1 (DJ1) which in turn modulate the pathological effects of tau and αsyn, respectively. Pathological phosphorylation of tau and αsyn was significantly low in the neuronal tissues of *Crbn*$^{-/-}$ mice model of CUMS. Moreover, *Crbn*$^{-/-}$ mice showed resilient behavioral responsiveness as compared to *Crbn*$^{+/+}$ mice. LC-MS/MS analysis showed an enriched metabolic and antioxidant status in the brains of *Crbn*$^{-/-}$ subjects as compared to *Crbn*$^{+/+}$ ones. Certain antioxidant proteins were elevated in the neuronal tissues of *Crbn*$^{-/-}$ mice; for instance, quinone oxidoreductase, superoxide dismutase (Sod1), glutathione S-transferase (Gstm1), peroxiredoxin-2 and biliverdin reductase A. Similarly, certain isoforms of molecular chaperones and co-chaperones were higher in the brains of *Crbn*$^{-/-}$ mice; for example, Hspa8 heat shock cognate 71, Hspd1 isoform 1 of 60 kDa heat shock protein, DJ1, and DJ2. The present study highlights the roles of CRBN in regulating the typical core behaviors, reshaping metabolic status and the fate of aggregation-prone proteins in the brains of mice exposed to CUMS. A better understanding of the molecular mechanism of depression and neurodegeneration will improve the development of better and novel treatments.

Disclosures: U. Akber: None. **S. Bong:** None. **Z. Park:** None. **C. Park:** None.

Poster

P307. Mechanisms of Neurodegeneration

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Title: Hippocampal peroxisomal proliferator-activated receptor β/δ deficiency induces cognitive alterations

Authors: T. ESPINOSA-JIMÉNEZ[1,2,3], M. CARRASCO[1,2,3,4], *J. CASTAÑEDA-CABRAL[5], M. E. UREÑA-GUERRERO[5], C. BEAS-ZÁRATE[5], M. VÁZQUEZ-CARRERA[1,6,7], C. AULADELL[2,3,8,9], E. VERDAGUER[2,3,8], J. FOLCH[2,4], A. CAMINS[1,2,3], M. ETTCHETO[1,2,3]; [1]Dept. of Pharmacology, Toxicology and Therapeut. Chemistry, Fac. of Pharm. and Food Science, Univ. of Barcelona, Barcelona, Spain; [2]Biomed. Res. Networking Ctr. in Neurodegenerative Dis., Madrid, Spain; [3]Inst. of Neuroscience, Univ. of Barcelona, Barcelona, Spain; [4]Dept. of Biochem. and Biotechnology, Fac. of Med. and Life Science, Univ. Rovira i Virgili, Reus, Spain; [5]Univ. de Guadalajara, Zapopan, Mexico; [6]Inst. of Biomedicine of the Univ. of Barcelona (IBUB), Univ. of Barcelona, Barcelona, Spain; [7]Spanish Biomed. Res. Ctr. in Diabetes and Associated Metabolic Dis. (CIBERDEM), Inst. de Salud Carlos III, Madrid, Spain; [8]Dept. of Cell. Biology, Physiol. and Immunology, Fac. of Biology, Univ. of Barcelona, Barcelona, Spain; [9]Inst. of Nanoscience and Nanotechnology (IN2UB), Univ. of Barcelona, Barcelona, Spain

Abstract: PPARβ/δ, the most PPAR abundant isoform in the central nervous system, is expressed in its major cell types, and is involved in the modulation of microglial homeostasis and energetic metabolism. Several studies demonstrated that people suffering from type 2 diabetes mellitus (T2DM) develop cognitive decline turning insulin resistance one of the best predictors of this disturbance. Although numerous investigations have studied the role of PPARβ/δ in metabolism, its role in neuronal and cognitive function has been underexplored. Therefore, the aim of the study is to determine the role of PPARβ/δ in the neuropathological pathways involved in the development of cognitive decline and as to whether a risk factor involved in cognitive loss such as obesity modulates neuropathological markers. In the present work, 6-month-old male PPARβ/δ-null (PPARβ/$\delta^{-/-}$) and wild type (WT) littermates with the same genetic background (C57BL/6X129/SV) were used. After the weaning (at 21 days old), animals were fed either with conventional chow (CT) or with a palmitic acid-enriched diet containing 45% of fat mainly from hydrogenated coconut oil (HFD). Thus, four groups were defined: WT CT, WT HFD, PPARβ/$\delta^{-/-}$ CT and PPARβ/$\delta^{-/-}$ HFD. Since it has been described that behavior alterations could be observed

among hybrids/strains, open field test was performed to compare C57BL/6X129/SV and C57BL/6 mice to assess anxiety levels. Our results confirmed that C57BL/6X129/SV showed significantly increased levels of anxiety compared to C57BL/6. Therefore, in order to evaluate cognitive decline, behavioral tests were dismissed, and dendritic spine quantification and other biochemical biomarkers were performed. In this line, PPARβ/$\delta^{-/-}$ mice exhibited a decrease in dendritic spine density and synaptic markers, such as PSD95 and BDNF, suggesting an alteration in cognitive function and synaptic plasticity. Likewise, our study demonstrated that the lack of PPARβ/δ receptor enhances the gliosis in the hippocampus, contributing to astrocyte and microglial activation and also, induced an increase in neuroinflammatory biomarkers such as TLR4 and PTP1B. Additionally, alterations in the hippocampal insulin receptor pathway were found. A fact to note is that HFD intake did not enhance the effects observed in the PPARβ/$\delta^{-/-}$ mice at the hippocampal level the studied mechanism. Taken together, these findings suggest that the loss of PPARβ/$\delta^{-/-}$ affects neuronal and synaptic structure, contributing to cognitive dysfunction and, they also present this receptor as a possible new target for the treatment of cognitive decline.

Disclosures: T. Espinosa-Jiménez: None. **M. Carrasco:** None. **J. Castañeda-Cabral:** None. **M.E. Ureña-Guerrero:** None. **C. Beas-Zárate:** None. **M. Vázquez-Carrera:** None. **C. Auladell:** None. **E. Verdaguer:** None. **J. Folch:** None. **A. Camins:** None. **M. Ettcheto:** None.

Poster

P307. Mechanisms of Neurodegeneration

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Program #/Poster #: P307.03

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: Global MS Research Booster Award
National MS Society Fellowship

Title: Demyelination and interferon signaling in axons cause retrograde upregulation of neuronal ISGylation pathway genes

Authors: *B. D. S. CLARKSON, C. L. HOWE;
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Abstract: Cognitive impairment in MS is associated with diffuse gray matter injury, which remains poorly understood. Given that axons span both white and gray matter regions, it's possible that signals within these axons contribute to the spread of diffuse injury in the brain. We show that retrograde IFNg signaling in axons (in vitro human and murine) and demyelination (murine in vivo) cause transcriptional / translational changes in neuronal cell bodies. Chief among the candidate signaling pathways identified is ISGylation, a process whereby a multitude

of cellular proteins are modified by the attachment of one or more ISG15 molecules. Using custom adeno-associated viral vectors we tested how altering ISGylation in neurons affects neuronal synaptic function and how it affects neuronal responsiveness to specific inflammatory factors in cell cultures. We also measured ISGylation in MS brain tissues to see if neuronal ISGylation correlates with gray matter injury in MS. We report early evidence that increased neuron ISGylation alters the composition of neuron-derived extracellular vesicles. Microglia treated with extracellular vesicles from neurons overexpressing ISGylation pathway genes exhibited morphology consistent with increased phagocytic activity and increased mRNA expression of inflammatory cytokines including: CCL2, IL1b, TNFa, iNOS, CXCL10, CCL5, and IL6. At this point, it is still unclear how these changes are related to gray matter pathology. However, it is possible that microglial activation by these vesicles is involved with driving synapse loss as activated microglia are known to be capable of stripping neuronal synapses.

Disclosures: B.D.S. Clarkson: None. **C.L. Howe:** None.

Poster

P307. Mechanisms of Neurodegeneration

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Program #/Poster #: P307.04

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: Pittsburgh Foundation IREG
NIH Grant R03AG063295

Title: Phagocytic glia drive prion-like spreading of mutant huntingtin aggregates in Drosophila brains

Authors: *M. PANNING PEARCE, K. M. DONNELLY, A. ZAYA, G. DAVIS, O. DELORENZO, W. THU, G. PISANO;
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Abstract: A key pathological and diagnostic feature of most neurodegenerative diseases is appearance of insoluble aggregates as a result of protein misfolding in the central nervous system (CNS). Accumulation of toxic protein aggregates in the brain is partially offset by a number of clearance mechanisms, such as the ubiquitin-proteasome pathway, autophagy, and engulfment by phagocytic glial cells; however, these mechanisms fail or become overwhelmed over time. A growing body of evidence supports the hypothesis that pathogenic aggregates associated with many neurodegenerative diseases behave similarly to infectious prions--they spread from cell-to-cell and nucleate the aggregation of natively-folded versions of the same protein, events which are thought to propagate aggregate-induced neurotoxicity along synaptic tracts the brain. We have recently reported that mutant huntingtin (mHTT) aggregates associated with the inherited

neurodegenerative disorder Huntington's disease spread in a "prion-like" manner between synaptically-connected neurons and glial cells in the *Drosophila* CNS. mHTT aggregates formed in presynaptic olfactory receptor neuron (ORN) axons directly nucleate the aggregation of soluble wild-type HTT (wtHTT) proteins expressed in the cytoplasm of post-synaptic partner projection neurons (PNs) or nearby glial cells. ORN-to-PN and ORN-to-glia transfer of mHTT aggregates is enhanced when ORNs are genetically silenced, slowed when caspase-dependent apoptosis in ORNs is blocked, and remarkably, require expression of the conserved glial scavenger receptor Draper/MEGF10. Further, mHTT aggregate transmission between synaptically-connected ORNs and PNs requires a transient visit to the glial cytoplasm, indicating that phagocytic glia can directly mediate aggregate transfer between neurons in vivo. Preliminary data from our lab indicate that entry of phagocytosed neuronal mHTT aggregates into the glial cytoplasm involves certain Rab GTPases with proposed roles in intracellular vesicle membrane fusion, suggesting that inefficient or stalled phagosome maturation and/or phago-lysosome fusion events can generate a reservoir of seeding-competent mHTT aggregate species in the glial cytoplasm. In support of this, expression of mHTT in ORNs accelerates age-related decline in phagocytic capacity of nearby glial cells. Together, our findings add to a growing understanding of phagocytic glia as double-edged players in neurodegeneration—these cells can clear neurotoxic aggregates, but age- and/or disease-related defects in this process can paradoxically promote aggregate propagation through in the brain.

Disclosures: M. Panning Pearce: None. **K.M. Donnelly:** None. **A. Zaya:** None. **G. Davis:** None. **O. DeLorenzo:** None. **W. Thu:** None. **G. Pisano:** None.

Poster

P307. Mechanisms of Neurodegeneration

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Program #/Poster #: P307.05

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: Robert J. and Nancy D. Carney Institute for Brain Science Innovation Award
NIAAA R01AA024434
IMSD: 5R25GM083270-11 (MPI)

Title: Alcohol-induced alternative splicing in drosophila memory circuits

Authors: *T. M. BROWN[1], E. PETRUCCELLI[3], S. J. GRATZ[2], A. G. WATERMAN[1], K. M. NUÑEZ[2], K. M. O'CONNOR-GILES[1], K. R. KAUN[2];
[1]Neurosci., [2]Brown Univ., PROVIDENCE, RI; [3]Biol. Sci., Southern Illinois University, Edwardsville, Edwardsville, IL

Abstract: Repeated alcohol experiences produce long-lasting memories for sensory cues associated with intoxication. These memories can problematically trigger relapse in individuals recovering from alcohol use disorder (AUD). The molecular mechanisms by which ethanol changes memories to become long-lasting and inflexible remain unclear. We recently demonstrated that formation of these memories results in expression of alternative transcript isoforms in memory-encoding neurons in *Drosophila melanogaster*. *Drosophila* rely on mushroom body (MB) neurons to make associative memories, including memories of ethanol-associated sensory cues. Decreasing expression of genes that play a role in splicing in adult MB neurons reduces formation of these memories, demonstrating the necessity of RNA processing in ethanol memory formation. Moreover, decreasing expression of alternatively spliced targets like *Dop2R* and *Stat92E* in adult MB neurons reduces ethanol memory formation. This suggests that the splicing changes in these transcripts has functional implications for maladaptive behaviors like addiction. To test this, we engineered mutant *Drosophila* lines with forced expression of the naïve or ethanol memory-associated isoform of *Dop2R* and *Stat92E*. We assessed mutant preference for an odor previously associated with ethanol following Pavlovian conditioning in a novel three-choice assay. We hypothesized that naïve or ethanol memory associated isoforms would reduce or enhance ethanol cue preference, respectively. Our results suggest that single target-specific splicing within memory-encoding neurons has functional consequences. These findings highlight the functional role of dynamic splicing in the nervous system and demonstrate a role for alternative splicing in the development of AUD.

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Poster

P307. Mechanisms of Neurodegeneration

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Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: Intramural Research Grant for Neurological and Psychiatric Disorders of NCNP (30-5; 1-5)
Grant-in-Aid for Scientific Research (B) from Japan Society for the Promotion of Science

Title: Selective phosphorylation of serine 345 on p47-phox serves as a priming signal of ROS-mediated axonal degeneration

Authors: *T. ARAKI, S. WAKATSUKI;
Dept PNS Res., Natl. Inst. Neurosci, NCNP, Tokyo, Japan

Abstract: Axonal degeneration is observed as a major component in many neurodegenerative diseases. We previously reported that the E3 ubiquitin ligase ZNRF1 promotes axonal degeneration by degrading AKT. We showed that, in response to reactive oxygen species (ROS) generated by NADPH oxidases, the EGFR-dependent phosphorylation of ZNRF1 at the 103rd tyrosine residue (pY103) induces its ubiquitin ligase activity in transected neurites. Here, we show that a NADPH oxidase subunit p47-phox/neutrophil cytosolic factor 1 regulates ZNRF1 activity. Interestingly, transection-induced phosphorylation of p47-phox at the 345th serine residue by p38 MAPK serves as an initiating signal to activate ZNRF1. The phosphorylated p47 (pS345) or a phospho-mimetic mutant p47-phox binds directly to ZNRF1 whereas a phosphorylation-resistant mutant p47-phox cannot bind to ZNRF1 and its overexpression in neurites significantly suppresses pY103 of ZNRF1, AKT ubiquitination, and degeneration after transection, suggesting that pS345 might enhance the EGFR-mediated phosphorylation-dependent activation of ZNRF1. Collectively, our findings suggest that pS345 might represent an important checkpoint to initiate the ZNRF1-mediated neurite degeneration and could serve as a novel target for neurite protection.

Disclosures: T. Araki: None. **S. Wakatsuki:** None.

Poster

P307. Mechanisms of Neurodegeneration

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Program #/Poster #: P307.07

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant NS069844

Title: Metabolic stress in neurons delays injury-induced Wallerian degeneration in Drosophila motoneuron axons

Authors: *T. WALLER, M. MALIK, C. A. COLLINS;
Molecular, Cellular, and Developmental Biol., Univ. of Michigan, Ann Arbor, MI

Abstract: The long length of axons creates distinct metabolic and cellular challenges to synaptic terminals and axons that are physically distant from their cell bodies. Axonal function requires support from glial cells as well as physical transport of proteins and organelles from the cell body. When transport is acutely interrupted by injury to the axon, the distal axon undergoes Wallerian degeneration, a cell autonomous breakdown process triggered by the activation of the NADase enzyme Sarm1 (Sterile Alpha And TIR Motif Containing 1). The regulation and function of Sarm1 is closely linked with the synthesis and breakdown of NAD+, an essential co-enzyme and metabolite. It is thought that activated Sarm1 promotes axonal degeneration through metabolic catastrophe. We were therefore surprised to find that multiple genetic manipulations

that are expected to impair energy homeostasis in neurons counterintuitively lead to a delay in Wallerian degeneration of injured axons. These manipulations include knockdown of a key glycolytic enzyme pyruvate kinase (PyK), and ATP synthase subunit G (ATPsynG) which is essential for ATP generation by oxidative phosphorylation. Mutations that impair mitochondrial localization in axons (for example in the mitochondrial rho GTPase miro), and, strikingly, ectopic expression of a constitutively active NADase enzyme (via a self-dimerizable TIR domain from Sarm1) leads to a delay in the degeneration of injured axons compared to control axons. We hypothesize that energy impairments in axons trigger a stress response pathway that alters the kinetics of axonal degeneration, and note that activation of nuclear markers of JNK signaling is a shared feature of the manipulations that impair Wallerian degeneration. Overexpression of Dicer 2 (Dcr2) is required to see the delay in injury-induced degeneration following NADase activation, which may indicate a role for Dcr2 in regulating this process. We are currently implementing ATP reporters in live Drosophila axons to probe the relationship between axonal energetics and the Wallerian degeneration pathway.

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Poster

P307. Mechanisms of Neurodegeneration

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Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: R01NS069726
R01NS094539
13GRNT17020004
NCRG-IRG 16IRG27780023

Title: Metabolomic and transcriptional profiling reveals bioenergetic stress and neuroinflammation in vivo after neuronal deletional of NAMPT

Authors: *S. LUNDT, N. ZHANG, Z. ZHANG, L. ZHANG, S. DING;
Univ. of Missouri, Columbia, MO

Abstract: Nicotinamide phosphoribosyltransferase (NAMPT) is the rate-limiting enzyme in the NAD^+ salvage pathway. Our previous study demonstrated that deletion of NAMPT gene in projection neurons using Thy1-$NAMPT^{-/-}$ conditional knockout (cKO) mice causes neuronal degeneration, muscle atrophy, neuromuscular junction abnormalities, paralysis and eventually death. Here we conducted a combined metabolomic and transcriptional profiling study *in vivo* in an attempt to further investigate the mechanism of neuronal degeneration at metabolite and mRNA levels after NAMPT deletion. Here using steady-state metabolomics, we demonstrate

that deletion of NAMPT causes a significant decrease of NAD^+ metabolome and bioenergetics, a buildup of metabolic intermediates upstream of glyceraldehyde 3-phosphate dehydrogenase (GAPDH) in glycolysis, and an increase of oxidative stress. RNA-seq shows that NAMPT deletion leads to the increase of mRNA levels of enzymes in NAD metabolism, in particular PARP family of NAD^+ consumption enzymes, as well as glycolytic genes Glut1, Hk2 and PFBFK3 before GAPDH. GO, KEGG and GSEA analyses show the activations of apoptosis, inflammation and immune responsive pathways and the inhibition of neuronal/synaptic function in the cKO mice. The current study suggests that increased oxidative stress, apoptosis and neuroinflammation contribute to neurodegeneration and mouse death as a direct consequence of bioenergetic stress after NAMPT deletion.

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Poster

P307. Mechanisms of Neurodegeneration

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Program #/Poster #: P307.09

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: Giving Back Fund
Stand By Eli Foundation

Title: Identification of a novel disease mechanism and a potential treatment for newly discovered neurodevelopmental disease “NEDAMSS”

Authors: *S. SINHA RAY[1], D. DUTTA[3], C. N. DENNYS[5], F. ROUSSEL[1], S. POWERS[2], X. ZHANG[1], P. BISWAS[1], J. CAROLE[1], N. WEIN[1], H. J. BELLEN[6], S. LIKHITE[1], P. C. MARCOGLIESE[4], K. C. MEYER[7];
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Abstract: NEDAMSS is a recently discovered neurodevelopmental disorder with growth regression, abnormal movements, loss of speech and seizures. The disease is caused by heterozygous mutations in the Interferon Regulatory Factor 2 Binding Protein-Like (IRF2BPL) gene. IRF2BPL is a ubiquitously expressed intron-less transcriptional regulator. The function of the protein is currently unknown, but the observed disease phenotype in patients indicates a role in development and maintenance of the nervous system. To study the disease mechanism, we used patient skin-derived fibroblasts with nonsense variants in the gene and utilizing an *in vitro*

modelling system we directly reprogrammed them into induced neuronal progenitor cells (iNPCs). The iNPCs were then differentiated into astrocytes, the most numerous cell type within the central nervous system. Additionally, we also used a direct conversion method to generate neurons directly from the skin fibroblasts to observe any morphological defects. Using these methods and performing molecular and functional analysis on the astrocytes, we identified a novel disease mechanism not previously described. While IRF2BPL is localized in the nucleus in healthy individuals, we found that several patient cell lines displayed mis-localization of the full-length protein in the cytoplasm both *in vitro* and *in vivo* within a fly model, caused by the dimerization with truncated mutant proteins. Moreover, the patient astrocytes also showed caused neuronal death in co-culture assays and abnormal mitochondrial ATP-linked respiration levels. Based on these observations, we treated our cell lines with CuATSM, a small molecule, neuroprotective drug currently in clinical trials for ALS. We found that CuATSM successfully rescued neuronal survival and reduced ATP-linked respiration to normal levels in NEDAMSS astrocytes. In summary, we identified a novel disease mechanism involved in NEDAMSS disease and found a promising treatment that might be able to slow or halt the progression of the disease.

Disclosures: S. Sinha Ray: None. **D. Dutta:** None. **C.N. Dennys:** None. **F. Roussel:** None. **X. Zhang:** None. **P. Biswas:** None. **J. Carole:** None. **N. Wein:** None. **H.J. Bellen:** None. **S. Likhite:** None. **P.C. Marcogliese:** None. **K.C. Meyer:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Alcyone Therapeutics, Kiadis, Amicus Therapeutics, Inc.

Poster

P307. Mechanisms of Neurodegeneration

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Program #/Poster #: P307.10

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH (NINDS): ZIA NS003106

Title: The bacterial microbiome controls neurodegenerative death in Drosophila

Authors: *A. K. SHUKLA, K. R. JOHNSON, E. GINIGER;
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Abstract: There has been a growing recognition that the microbiome plays a pivotal role in aging and neurodegenerative disease (ND), but the complexity of the interaction between microbe and host has made it difficult to dissect. To better understand how host-microbe interactions affect NDs, we used a *Drosophila melanogaster* model of neurodegeneration, activation of the endogenous tau kinase, Cdk5, by overexpression of its activating subunit, Dp35.

Under conventional, microbe-rich conditions, overexpressing Dp35 reduces lifespan by ~40%. To our surprise, however, we found that lifespan is ***completely*** rescued in flies overexpressing Dp35 if they are raised without bacteria (axenic hereafter) (Median survival 39.5±7.7 days in conventional and 69.5±3.5 days in axenic). To uncover the underlying molecular basis for the pathological interaction between the microbiome, aging, and ND we performed gene expression profiling on axenic and conventionally raised flies at various ages. Gene ontology (GO) analyses of these data demonstrate that the microbiome-driven effect on lifespan in the ND condition is associated with profound changes in chromatin structure and genome integrity, mediated specifically via altered regulation of histone genes and transposons. Given the profound effect of the microbiome on lifespan in the ND condition, we next investigated how the microbial population changes with host age. In male flies, the total bacterial load does not change much over the lifespan, but 16S ribosomal rDNA sequencing revealed that the diversity of the microbiome undergoes drastic simplification over the course of the fly's lifespan. Remarkably, the drop in bacterial species diversity is greatly accelerated in OE Dp35. Moreover, the species that remain at older ages are altered by overexpression of Dp35 in the host. We are currently working to understand why bacterial diversity changes with age, why it depends on Cdk5/Dp35, and whether the particular species that remain are significant. We are also investigating how chromatin modification is associated with the microbiome-dependent neurodegenerative death of the organism. Overall, this work highlights the importance of host-microbe interactions as a fundamental regulatory system in modulating neurodegenerative death.

Disclosures: A.K. Shukla: None. **K.R. Johnson:** None. **E. Giniger:** None.

Poster

P314. Neuroinflammation: Neurodegeneration I

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Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: Grant-in-Aid for Scientific Research C 15K10993, 19K09442

Title: Neurological deficit appeared 3 weeks post heat exposure by cerebellar dysfunction in the rodent model of heatstroke.

Authors: K. MIYAMOTO[1], H. OHTAKI[3], *A. YOSHIKAWA[4], H. YAMAGA[1], M. NAKAMURA[1], K. SUZUKI[1], K. HONDA[2], K. DOHI[1];
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Abstract: Introduction: Brain is extremely sensitive to hyperthermia. Disorientation, confusion, delirium and seizures are often seen in human heatstroke. Such consciousness disturbance improves once the core body temperature falls below the critical level in acute phase. However, there are cases that neurological sequelae remain after heatstroke that interrupts the patients getting the back to social life. Among them, cerebellar structures are vulnerable to heat, therefore cerebellar ataxia is often appeared. Interestingly, there are several interesting reports cerebellar ataxia appearing not immediately after heatstroke but after several weeks have passed. Thought, the pathomechanism of this delayed cerebellar ataxia after heatstroke is not well understood. Therefore, we focused on the delayed cerebellar ataxia after heatstroke using the rodent heatstroke model.**Methods:** Male C57BL6J mice were subjected to heat exposure (HE), 41℃, RH>95% for 60 min as a heatstroke group (HS). The animals which were not exposed to heat were prepared as a control (Ct). Behavioral test (Rota-rod) was performed 1, 3, 5, 7 and 9 weeks post heat exposure and running time was compared between Ct and HS group. Then, another two group (Ct, HS) were prepared and brains were removed 1, 3 and 9 weeks post heat exposure. Kluver-Barrera (KB) staining was performed to evaluate the demyelination at cerebellum and corpus callosum. Immunostaining of Calbindin was performed to semi-quantify of the Purkinje cells. Moreover, we performed immunostaining of Synaptophysin and Postsynaptic density-95 (PSD-95) to evaluate the pre and post synapses around Purkinje cells post heat exposure. **Results:** Motor coordination disorder in HS group significantly appeared 3 weeks post heat exposure and gradually improved to some extent over time. Demyelination was detected 1, 3 weeks and the number of Purkinje cell significantly decreased 1,3 and 9 weeks post heat exposure. The expression of Synaptophysine and PSD-95 around Purkinje cells were weakest in 3 weeks post heat exposure and improved over the time.**Conclusion:** Delayed cerebellar ataxia in heatstroke appeared from 3 week and recovered to some extent over time. Our results suggested that permanent Purkinje cells loss, transient demyelination and synaptic impairment at cerebellum might correlated with delayed cerebellar ataxia of heatstroke.

Disclosures: K. Miyamoto: None. **H. Ohtaki:** None. **A. Yoshikawa:** None. **H. Yamaga:** None. **M. Nakamura:** None. **K. Suzuki:** None. **K. Honda:** None. **K. Dohi:** None.

Poster

P314. Neuroinflammation: Neurodegeneration I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P314.02

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NARSAD Young Investigator Grant #26804 to JW

Title: Prenatal activation of the maternal immune system alters prefrontal cortex neuroanatomy in adult offspring

Authors: *V. R. RIESGO[1], E. P. SELLINGER[2], J. M. JURASKA[2], J. WILLING[1];
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Abstract: In light of the COVID-19 pandemic, it is vitally important to further our understanding of how the maternal immune system can affect brain development in offspring. During the prenatal period, the developing brain is especially sensitive to exogenous and endogenous factors that can disrupt vital developmental processes, including the optimization of regional neuron number. Maternal infection has been shown to affect certain cells of the prefrontal cortex (PFC) and PFC-mediated behaviors. However, little is known about the long-term effects of maternal infection on neuron number and volume of the medial prefrontal cortex (mPFC), a region critical for executive functions and behavior inhibition. The present study assesses the effects of prenatal lipopolysaccharide (LPS), which activates the maternal immune system, on mPFC anatomy in young adult offspring. 100 µg/kg LPS was administered intraperitoneally to pregnant rats on embryonic days 15 and 16, and brain tissue was collected from offspring on postnatal day (P) P60. Maternal LPS treatment reduced the number of successful pregnancies and reduced litter size compared to controls. Additionally, stereological analysis of young adult male and female offspring showed that LPS reduced mPFC neuron number, mPFC volume and white matter volume under the PFC (n=10-11 males and females per group). These results indicate that maternal infection can permanently alter the structure of this region and may further implicate a role for maternal infection in psychiatric illnesses characterized by PFC dysfunction.

Disclosures: V.R. Riesgo: None. **E.P. Sellinger:** None. **J.M. Juraska:** None. **J. Willing:** None.

Poster

P314. Neuroinflammation: Neurodegeneration I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P314.03

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Title: Generation of a Glia-Midbrain Neuron Co-Culture System Derived From Human Pluripotent Stem Cells

Authors: N. LEBLANC[1], J. WANG[1], J. CHAN[1], A. C. EAVES[1,2], S. A. LOUIS[1], *E. KNOCK[1];
[1]STEMCELL Technologies Inc., Vancouver, BC, Canada; [2]Terry Fox Laboratory, BC Cancer, Vancouver, BC, Canada

Abstract: Astrogliosis is a hallmark of Alzheimer’s and Parkinson’s diseases, and astrocytes and microglia play key roles in both neurodevelopment and neuroinflammation. To allow

investigation of neuron-glia interactions in the context of disease modeling, we developed STEMdiff™ Midbrain Neuron and STEMdiff™ Astrocyte Differentiation and Maturation Kits to efficiently generate functional hPSC-derived midbrain neurons and astrocytes. hPSC-derived neural progenitor cells (NPCs) generated using the STEMdiff™ Neural Induction kit monolayer protocol were either plated into STEMdiff™ Astrocyte Differentiation Medium or STEMdiff™ Midbrain Neuron Differentiation Medium. Astrocytes were passaged every week using the recommended protocol. On the third passage, the medium was replaced with STEMdiff™ Astrocyte Maturation Medium and similarly maintained for an additional two weeks. Midbrain neurons were passaged after one week into STEMdiff™ Midbrain Maturation Medium and cultured for an additional two weeks. Cell identity was confirmed using immunocytochemistry for S100β, GFAP, and DCX (astrocytes), or BIIITUB and TH (midbrain neurons). We then co-cultured the astrocytes and neurons for one week. Our data show that using STEMdiff™ Astrocyte Differentiation and Maturation Kits can result in a highly pure population of astrocytes (S100β: 72.0% ± 20.4%; GFAP: 33.8% ± 30.0%; DCX: 0.8% ± 1.5%; n = 6). Our STEMdiff™ Midbrain Neuron Differentiation and Maturation Kits produce a subpopulation of BIIITUB+ neurons expressing tyrosine hydroxylase (TH), the enzyme implicated in dopamine biosynthesis (BIIITUB: 93.46% ± 6.93; TH: 16.57% ± 10.67; n = 13). Functional data using MEA show electrical activity up to 6 months in culture (up to 0.85 ± 0.33 Hz mean firing rate). Interestingly, after one week of co-culture, the number of BIIITUB-positive and TH-positive neurons was higher in the astrocyte co-culture group than in the neuron-only control (BIIITUB: 2.76 ± 1.50-fold higher, P = 0.0003; TH: 2.63 ± 0.99-fold higher, P=0.01; n = 6, unpaired t-test), which indicates co-culture with astrocytes could lead to better survival and maturation of neurons in vitro. These data show that STEMdiff™ Midbrain Neuron Differentiation and Maturation Kits can generate functional hPSC-derived dopaminergic-midbrain neurons, which, used alongside STEMdiff™ Astrocyte Differentiation and Maturation Kits, create co-culture models suited for in vitro studies of neurodegeneration.

Disclosures: **N. LeBlanc:** A. Employment/Salary (full or part-time):; STEMCELL Technologies. **J. Wang:** A. Employment/Salary (full or part-time):; STEMCELL Technologies. **J. Chan:** A. Employment/Salary (full or part-time):; STEMCELL Technologies. **A.C. Eaves:** A. Employment/Salary (full or part-time):; STEMCELL technologies. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Owner of STEMCELL Technologies. **S.A. Louis:** A. Employment/Salary (full or part-time):; STEMCELL Technologies. **E. Knock:** A. Employment/Salary (full or part-time):; STEMCELL Technologies.

Poster

P314. Neuroinflammation: Neurodegeneration I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P314.04

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: MITACs
WinSanTor Inc

Title: Muscarinic acetylcholine type 1 receptor antagonists augment mitochondrial function through AMPK in human neuroblastoma cells

Authors: *F. NAZNIN, P. FERNYHOUGH;
Univ. of Manitoba, Winnipeg, MB, Canada

Abstract: Introduction Impairments in mitochondrial physiology play a vital role in the progression of multiple degenerative conditions in diabetes, such as diabetic peripheral neuropathy, and activation of muscarinic acetylcholine type 1 receptor (M1R) has been linked with mitochondrial dysfunction. In rodent models of diabetes, inhibition of M1R in sensory neurons induced AMP-activated protein kinase (AMPK) activity and nerve regeneration. We *hypothesized* that specific (muscarinic toxin 7; MT7) and selective (pirenzepine; PZ) antagonists of M1R, would enhance AMPK activity and contribute to augmentation of mitochondrial function in human neuroblastoma SH-SY5Y cells. This comprehensive study aimed at evaluating mitochondrial parameters including oxygen consumption rate (OCR), mitochondrial membrane potential (MMP) and expression of component proteins of the mitochondrial complexes (OXPHOS).
Methods SH-SY5Y cells expressing M1R were treated with different doses of M1R antagonists (PZ and MT7) at different time points in the presence or absence of inhibitors or siRNAs of AMPK. Cellular bioenergetic status was assessed using a Seahorse Biosciences XF-24 Analyzer. The intracellular MMP was evaluated by using a fluorescent dye (JC-1) and the fluorescence intensity was measured by microplate reader. Expression analysis of AMPK and OXPHOS proteins was done by Western blot. Data are reported as the mean ± SEM, and one-way ANOVA with Tukey's or Dunnett's multiple comparison post-hoc tests were applied. A value of $p<0.05$ was considered statistically significant.
Results Both PZ and MT7 treatment in SH-SY5Y cell culture significantly increased AMPK phosphorylation in a dose-dependent manner at different time points. Mitochondrial protein expression (OXPHOS) and OCR (including parameters for maximal respiration and spare respiratory capacity), ATP production. MMP were also augmented in PZ and MT7 treated cells. Exposure to PZ for 6-h increased the MMP with earlier time points without effect. Compound C or AMPK (α1 and α2)-specific siRNA suppressed the PZ or MT7 induced elevation of mitochondrial function and MMP.
Conclusion. PZ and MT7 treatment in SH-SY5Y cells improved mitochondrial function through a M1R-modulated pathway involving activation of AMPK and may represent a potential therapeutic target to regulate mitochondrial function and nerve regeneration in diabetic neuropathy.

Disclosures: F. Naznin: None. **P. Fernyhough:** None.

Poster

P314. Neuroinflammation: Neurodegeneration I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P314.05

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH grant AA011605

Title: Chronic alcohol exposure following TLR3 agonist changes neuroimmune genes expression in the insular cortex and nucleus accumbens of female rat brains

Authors: *W. LIU[1], K. J. VAN VOORHIES[1], J. LIU[1], J. BESHEER[2];
[1]Univ. of North Carolina at Chapel Hill, Chapel Hill, NC; [2]Psychiatry; Bowles Ctr. for Alcohol Studies, Univ. of North Carolina - Chapel Hill, Chapel Hill, NC

Abstract: Chronic alcohol consumption can result in permanent damage to brain, and promote changes in the expression of neuroimmune genes. Toll-like receptors (TLRs) are key components of neuroimmune activation. Our previous study showed that treatment with poly(I:C), a TLR3 agonist, escalated alcohol self-administration. The goal of the present study was to examine the effects of chronic alcohol treatment on neuroimmune signaling triggered by TLR3. Female Long Evans rats were randomly assigned to two groups. Alcohol group was administered ethanol (5 g/kg, i.g.) once daily for 14 days, control group with water. General locomotor activity was assessed following the 13th day of alcohol exposure in the open field test. Chronic alcohol exposure decreased both the percentage of center time and total activity in the open field relative to the water group. Rats were allowed to withdraw for 24 hours after last alcohol exposure. Both control and alcohol groups were subdivided into two groups matched on body weight. One group was injected with poly(I:C) (3 mg/kg, i.p.), and another with saline. All rats were sacrificed 24 hours after poly(I:C). To investigate the effect of poly(I:C) following chronic alcohol exposure on neuroimmune gene expression, TLR3, TLR7, IRF3, IRF7, IL1β, IL6, TNFα, CD11b and GLT-1 were examined in the anterior insular cortex (IC) and nucleus accumbens core (AcbC) using RT-qPCR. Our studies found that poly(I:C) increased TLR3 and TLR7 gene expression in both IC and AcbC. Alcohol-poly(I:C) significantly potentiated alcohol-induced increase in TLR3 and TLR7 in the IC. Chronic alcohol exposure and poly(I:C) upregulated TNFα gene expression, and chronic alcohol exposure potentiated TLR3 activation-induced increase in both IC and AcbC. Poly(I:C) significantly increased IRF7 gene expression in the IC and AcbC, chronic alcohol exposure potentiated IRF7 expression in the IC, but not in AcbC. IL1β, IL6 and IRF3 gene expression were not changed in the IC or AcbC. CD11b, a myeloid cell integrin receptor which are expressed by both macrophages and microglia, were increased by chronic alcohol and poly(I:C) in the IC and AcbC. TLR3 activation enhanced alcohol-induced CD11b gene expression in the IC, but not in AcbC. Upregulation of the astroglial glutamate transporter GLT-1 was observed in the IC following chronic alcohol exposure, and the increase in GLT-1 gene expression was blunted by TLR3 activation in the IC,

and not changed in the AcbC. These findings indicate that TLR3 activation can potentiate chronic alcohol-induced changes in neuroimmune gene expression, which varies in the IC and AcbC, two regions known to be involved in modulating alcohol drinking.

Disclosures: W. Liu: None. **K.J. Van Voorhies:** None. **J. Liu:** None. **J. Besheer:** None.

Poster

P314. Neuroinflammation: Neurodegeneration I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P314.06

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: Johns Hopkins Lyme Disease Treatment Center, Anonymous Donation

Title: Hippocampal gray matter changes are associated with post-treatment Lyme disease

Authors: *D. BHATTACHARYA[1], A. W. REBMAN[2], C. B. NOVAK[2], E. A. MIHM[2], J. N. AUCOTT[2], C. L. MARVEL[1];
[1]Dept. of Neurol., [2]Dept. of Med., Johns Hopkins Univ. Sch. of Med., Baltimore, MD

Abstract: Approximately 10% of patients who are treated for Lyme Disease (LD) continue to have persistent symptoms, known as Post-Treatment Lyme Disease (PTLD). Patients with PTLD often report lingering cognitive difficulties, such as poor memory and “brain fog”. Few neuroimaging studies have been conducted with PTLD patients, however, to understand the underlying mechanisms of these cognitive symptoms. The hippocampus is a brain region that has been associated with learning and memory and is a major component of cognition. Therefore, this study used voxel-based morphometry (VBM) to assess whether gray matter (GM) changes in the hippocampus were associated with cognitive performance in PTLD. Twelve human participants’ structural brain images were obtained via magnetic resonance imaging, including 7 men and 5 women, with an average age of approximately 45 years, and education level of approximately 16 years. Their mean duration of illness was 2.6 years. They were administered standard neuropsychiatric tests related to verbal recall, working memory, and processing speed. An exploratory VBM analysis of GM volume in the hippocampus was conducted using hippocampal masks to delineate CA1, CA2, CA3, dentate gyrus, entorhinal cortex, and subiculum sub-regions. GM within these sub-regions were compared to cognitive performance. Results revealed negative associations between hippocampal gray matter and delayed verbal memory recall, digit span, and processing speed. These associations were strongest in the subiculum. These results suggest that hippocampal GM increases were associated with lower cognitive function. Further research is needed to understand these underlying mechanisms, and how interventions can aid the treatment of cognitive difficulties in PTLD.

Disclosures: D. Bhattacharya: None. **A.W. Rebman:** None. **C.B. Novak:** None. **E.A. Mihm:** None. **J.N. Aucott:** None. **C.L. Marvel:** None.

Poster

P314. Neuroinflammation: Neurodegeneration I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P314.07

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Title: Oxidative stressors induced glycan modifications of extracellular vesicles derived from HT22 murine hippocampal cells

Authors: *T. SATO[1], A. SHIMODA[2], N. MIYANISHI[3], T. NEDACHI[2];
[2]Grad. Sch. of Life Sci., [3]Grad. Sch. of Food and Nutritional Sci., [1]Toyo Univ., Gunma, Japan

Abstract: It is widely accepted that oxidative stress has been implicated in the progression of neurodegenerative diseases. We have previously revealed that the protein/RNA contents of extracellular vesicles (EVs) released from HT22 murine hippocampal cells was significantly altered by oxidative stress. On the other hands, although glycan modification of EVs is crucial for their internalization into the target cells, the impact of oxidative stress on glycan modification of EVs has not been well understood. In this study, we established a new method for analyze surface glycosylation of EVs released from HT22 cells and investigated the impact of oxidative stress on the glycosylation. HT22 cells were cultured with or without 250 μM H_2O_2 for 48 hours and the released EVs were purified by using the ultracentrifugation methods. The purity of EVs was then routinely confirmed by western blotting, transmission electron microscopes, and nano particle analysis. In order to analyze surface glycans of EVs, we used the permethylation method to replace all reactive glycan hydrogens into methyl groups, and the permethylated glycans were analyzed by using MALDI/TOFMS analysis. This method was suitable for robust analysis of small amounts of glycan samples, such as purified EVs. The results indicated that both the N-linked glycan and O-linked glycans in EVs contained sialic acids, and the proportion of sialylated glycan in EVs was significantly increased by oxidative stress. To elucidate physiological roles of this glycan modification of EVs, we measured the uptake of control EVs or EVs rich in sialic acids, into HT22 cells. In brief, HT22 cells were treated with PKH26-labeled EVs for 24 hours, and the uptake of EVs into HT22 cells was monitored by fluorescence microscopy. Control EVs were effectively incorporated into HT22 cells, whereas little amounts of EVs rich in sialic acids were uptaken by the cells. Overall, these suggested that the glycosylation of EVs released from HT22 cells were tightly engaged to oxidative states of the cells, and the modification of EVs possibly regulated their incorporation into the target cells. Given that the release and incorporation of EVs in central nervous systems were important for

the progression of several neurodegenerative diseases, the glycan modification of EVs may be an attractive therapeutic target as wells as an useful biomarker for neurodegenerative diseases.

Disclosures: T. Sato: None. **A. Shimoda:** None. **N. Miyanishi:** None. **T. Nedachi:** None.

Poster

P314. Neuroinflammation: Neurodegeneration I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P314.08

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: DGE-1745038

Title: Regulation of neutrophil infiltration into the CNS by the astrocyte circadian clock

Authors: *J. LAWRENCE[1], A. OBGASLASE[1], A. ARCHAMBAULT[1], G. F. WU[2], E. S. MUSIEK[3];
[1]Washington Univ. in St. Louis, St. Louis, MO; [2]Neurol., Washington Univ. in St. Louis, Saint Louis, MO; [3]Neurol., Washington Univ. In St. Louis, Saint Louis, MO

Abstract: Inflammation is a critical component for the return to homeostasis post insult or injury; however, inflammation in the central nervous system (CNS) can lead to irreparable damage. The balance between pro- and anti-inflammation in the CNS rapidly shifts towards a neurotoxic proinflammatory state during neurodegenerative diseases such as Alzheimer's Disease (AD) or Multiple Sclerosis (MS). These dramatic inflammatory responses are also coupled with extensive blood brain barrier (BBB) leakage and breakdown. This BBB breakdown enables the entry of leukocytes, including neutrophils, which mediate early inflammatory responses and modulate pathogenesis of both AD and MS. We propose a unique mechanism by which circadian regulation of CXCL5 in astrocytes allows for tight temporal and spatial control over these mechanisms. CXCL5 is a potent chemokine that both induces BBB breakdown and neutrophil chemotaxis - two immune responses critical during neurodegeneration. Our preliminary data shows clear circadian rhythms in *Cxcl5* expression in astrocytes, and that deletion of the circadian transcription factor *Bmal1* in astrocytes causes a striking increase in CXCL5 expression, an effect that is exacerbated in an AD mouse model. We will use both an astrocyte specific *Bmal1* knock-out (KO) and CXCL5 overexpression to address the role of astrocytic CXCL5 in different models of neuroinflammation. Ultimately, we hope to better understand if circadian regulation of astrocytic CXCL5 levels mediates daily rhythms in neutrophil recruitment into the brain, and how disrupting this influences CNS immune responses and BBB integrity in mouse models of AD and multiple sclerosis.

Disclosures: J. Lawrence: None. **A. Obgaslase:** None. **A. Archambault:** None. **G.F. Wu:** None. **E.S. Musiek:** None.

Poster

P314. Neuroinflammation: Neurodegeneration I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P314.09

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: MH087332
MH104131
MH105330
DA052209

Title: Characterizing the Role of IFNβ in Neuronal Injury Induced by HIV-1

Authors: *H. SINGH, J. KOURY, R. MAUNG, M. KAUL;
Univ. of California, Riverside, Riverside, CA

Abstract: The HIV-1 virus can enter the central nervous system (CNS) early in infection and continues to cause HIV-associated neurocognitive disorders (HAND). This is of global concern as 38 million people worldwide are diagnosed with HIV, and an estimated 50% of people living with HIV develop HAND. The pathogenic mechanism of HAND and HIV-associated dementia (HAD), the most severe neurological complication of HIV/AIDS, remains incompletely understood, and there is no specific treatment available. One model that shares key features of neuropathology, differential gene expression, and impaired learning and memory with people living with HIV and HAND/HAD and encephalitis (HIVE) is a transgenic (tg) mouse expressing the HIV envelope glycoprotein gp120 (HIVgp120) in the CNS under the control of the glial fibrillary acidic protein (GFAP) promoter in astrocytes. Interferons (IFNs) play a major role as the first line of host defense against HIV in the brain. Our microarray study revealed that brains of HIVgp120tg mice share a significant fraction of differentially regulated genes with HIV and HIVHIVE patients with neurocognitive impairment, including transient expression of genes induced by interferon-β (IFNβ). We have demonstrated that intranasal treatment with IFNβ prevents in vivo neuronal damage triggered by HIVgp120, and our in vitro experiments showed that neuroprotection depended on IFNAR1 and CCL4. However, in the absence of protective amounts of IFNβ, IFNAR1 (the receptor for type I IFNs) can contribute to neuronal injury in the presence of HIVgp120. To characterize the role of endogenous IFNβ, we cross-bred HIVgp120tg with IFNβ knockout (IFNβKO). The brains of these mice were analyzed for hallmarks of HIV-induced neuropathology and gene expression. While the IFNβKO did not overall worsen the neuropathology, it was critical for the upregulation of IRF7, -9, and RIG-I, but not IRF3, in

HIVgp120tg brains. Thus, endogenous IFNβ contributes to the innate immune response of the brain but is insufficient to prevent neuronal injury in the long-term.

Disclosures: H. Singh: None. **J. Koury:** None. **R. Maung:** None. **M. Kaul:** None.

Poster

P323. Mechanisms of Spreading Depolarizations and Transmitter Release

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P323.01

Topic: C.08. Ischemia

Support: NIH Grant NS106901-01
GM109089

Title: Nimodipine inhibition of spreading depolarization: metabolic dependency in brain slices

Authors: *M. BENNETT[1], J. WEISEND[1], A. CARLSON[2], C. SHUTTLEWORTH[1];
[1]Neurosci., [2]Neurosurg., Univ. of New Mexico, Albuquerque, NM

Abstract: Introduction: Spreading depolarizations (SD)s have recently been identified as key contributors to progression of acute brain injuries, but specific mechanisms of neuronal injury after SD remain incompletely explained. Prior work showed that L-type calcium channel antagonists (including nimodipine) can reduce the initiation and propagation of SD, due to block of neuronal Zn^{2+} accumulation. However that prior work could not address neuronal injury as, under control conditions with adequate substrate supply, neurons recovered fully after each SD. The goal of the present study was to explore L-type channel involvement in conditions that model the vulnerable ischemic penumbra. **Methods:** SD was induced in the hippocampal CA1 region of murine brain slices, and SD progression and swelling monitored with intrinsic optical imaging, extracellular recordings of DC potential shifts and field excitatory postsynaptic potentials (fEPSPs). Responses in control conditions were compared with metabolically compromised conditions, using flow restriction as previously described (Exp Neurol 305 (2018) 121-128). **Results:** As previously reported, reduction in metabolic substrate availability conferred vulnerability to neuronal damage after SD. Thus, in control conditions, SD caused a brief period of synaptic silencing followed by full recovery, whereas metabolically compromised slices did not recover after a single SD. In vulnerable slices, SD led to persistent inhibition of fEPSPs and a marked decrease in IOS signal that is attributed to structural disruption. Nimodipine pre-exposure (10μM) prevented deleterious effects of SD in compromised slices. fEPSPs recovered, and IOS decreases were prevented (27.5% decrease vs 10.97% increase at 10 min post-SD, $P<0.02$, n=6-8). Nimodipine also reduced the second phase of SD-induced DC shift to near control levels ($P=0.0049$; n=6-8) as well as reduced overall duration of DC shift ($P=0.0464$; n=6-8). Like nimodipine, zinc chelation via TPEN (50μM) also resulted in slice

recoverability following SD in vulnerable slices. Conclusions: These results suggest that L-type channels can be involved in damaging effects of SD in metabolically compromised tissue. Zn^{2+} is released at high levels during SD, and prevention of neuronal Zn^{2+} and Ca^{2+} overload following SD, while avoiding hypotensive or other deleterious effects, could provide a useful additional approach to limit damaging effects of SD in stroke and other injuries. Funding: NS106901, GM109089

Disclosures: M. Bennett: None. **J. Weisend:** None. **A. Carlson:** None. **C. Shuttleworth:** None.

Poster

P323. Mechanisms of Spreading Depolarizations and Transmitter Release

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P323.02

Topic: C.08. Ischemia

Support: NIH Grant NS106901
NIH Grant T32HL007736
NIH Grant GM109089

Title: Basal Excitability and CaMKII mediate SD induced Potentiation

Authors: *J. WEISEND[1], A. CARLSON[2], C. W. SHUTTLEWORTH[1];
[1]Neurosciences, [2]Neurosurg., Univ. of New Mexico, Albuquerque, NM

Abstract: Spreading depolarizations (SDs) are transient recurring waves of extended depolarization associated with stroke. SD can generate long lasting synaptic potentiation, but mechanisms and pathophysiological relevance are unestablished. We examined mechanisms of synaptic potentiation in conditions modeling either vulnerable tissue contributing to infarct expansion or healthy peri-infarct tissue. Focal microinjection of KCl initiated SDs in the hippocampal CA1 subregion of murine brain slices. Intrinsic optical signals (iOS) were recorded to monitor SD propagation and tissue swelling. Field excitatory postsynaptic potentials (fEPSPs) were fully recoverable after SD in control recording conditions, but persistent synaptic depression and iOS depression (both signaling tissue injury) was seen in recording conditions where metabolic substrate supply was reduced by flow restriction (Exp Neuro 305 (2018) 121-128). In control conditions, a single SD produced the well-described transient abolition of fEPSP (~10 min), and was followed by recovery to a potentiated level (125.4% ± 17.5 at 30 min post-SD, n=18). Studies of repetitive SDs showed that synaptic potentiation was saturated after a single SD. Tissue swelling, a potential confound, was resolved ~12 min after SD, and neither peak swelling nor recovery rates correlated with the degree of sustained potentiation. Adenosine accumulation after SD delays fEPSPs recovery and may mask the degree of potentiation.

However adenosine accumulation typically resolves in ~10min, and pre-exposure with the adenosine A1 receptor antagonist DPCPX (300nM), did not unmask additional potentiation (n=6). Potentiation was unaffected by an inhibitor of protein kinase C activity (K252a, 200nM), but was significantly reduced by an inhibitor of Ca^{2+}/calmodulin-dependent protein kinase II (Arcyriaflavin A, 250nM, n=6, $p<0.05$). In metabolically compromised tissues SD is followed by persistent suppression of fEPSPs. This is partly due to persistent adenosine accumulation, as DPCPX recovered fEPSP amplitude towards baseline values, but did not unmask persistent potentiation. These data suggest a single SD saturates long-term plasticity and that this could involve CaMKII-dependent signaling, as described previously for experimentally-evoked LTP in this preparation. SD induced synaptic strengthening is evident in control conditions that model peri-infarct tissue destined to survive following stroke, but absent in conditions that model expansion of an infarct core. These findings provide a basis for future studies of whether SD-induced plasticity may contribute to adaptation and recovery around stroke lesions.

Disclosures: J. Weisend: None. **A. Carlson:** None. **C.W. Shuttleworth:** None.

Poster

P323. Mechanisms of Spreading Depolarizations and Transmitter Release

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P323.03

Topic: C.08. Ischemia

Support: R01 NS106901
COBRE P206GM109089

Title: Identification of neuronal injury and survival pathways after spreading depolarization using transcriptome sequencing

Authors: *M. DELL'ORCO[1], J. WEISEND[1], N. PERRONE-BIZZOZERO[1], R. A. MORTON[1], A. CARLSON[2], C. W. SHUTTLEWORTH[1];
[1]Neurosciences, [2]Neurosurg., Univ. of New Mexico Sch. of Med., Albuquerque, NM

Abstract: Spreading depolarization (SD) is a slowly propagating wave of profound depolarization that sweeps through cortical tissue. SD is not harmful to healthy brain tissue, but can cause irrecoverable injury to metabolically compromised tissue, exacerbating acute brain injuries. SDs occur in stroke brain, usually originating near ischemic foci and propagating throughout peri-infarct and surrounding tissue leading to tissue loss. Previous studies have shown changes in expression levels of genes known to regulate synaptic plasticity and neurogenesis following SD. This study aimed to perform an extensive, unbiased analysis to identify a more complete range of biological pathways modified by SD using RNAseq. SDs were induced repetitively (4 SDs in 2 hours every 30 min) in healthy, female mice and confirmed with

Intrinsic Optical Signal imaging. SD induction was with either focal application of KCl (NaCl for sham controls, n=6) in C57Bl/6 mice or with optogenetic stimulation (2 mW for 20 sec) in Thy1-ChR2-YFP mice (n=6). Two hours after onset of the initial SD, total cortical RNA was extracted and subjected to Illumina paired-end RNAseq to identify differentially expressed (DE) genes (fold change >1.25, p value <0.05). We identified 122 significantly upregulated genes after KCl-induced SDs and 169 after optogenetic induction; of these 63 genes (27.6%) where in common. Consistent with previous studies, top DE genes include the neurotrophic factor BDNF and VGF, as well as intermediate early genes ARC, FOS, JUN and EGR. Among other DE genes with significantly increased levels include HOMER1, COX2, ADRB1, NR4A1 nuclear receptor, COX2, DUSP gene family, GABRB3 GABA receptor, and KCNJ2 potassium channel. Through pathway analysis we found significant increases in the expression of genes associated with axogenesis, branching of axons, neuritogenesis, dendritic growth, and regeneration of neurites. Upstream analysis also showed significant activation of MAPKs signaling. Interestingly, we also found a significant decrease in the expression in genes associated with cell death, apoptosis and neuronal degeneration. These results indicate that SD in healthy tissues causes relatively rapid upregulation of pathways that could potentially be involved in plasticity or circuit modification. Additionally, downregulation of pathways involved in neuronal injury could contribute to previous reports of ischemic preconditioning by SD. These results also identify a range of novel targets that could be used to test whether clusters of SD may enhance plasticity or recovery in surviving peri-infarct tissue, in addition to the well-established role of SD in expansion of the infarct core.

Disclosures: M. Dell'Orco: None. **J. Weisend:** None. **N. Perrone-Bizzozero:** None. **R.A. Morton:** None. **A. Carlson:** None. **C.W. Shuttleworth:** None.

Poster

P323. Mechanisms of Spreading Depolarizations and Transmitter Release

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P323.04

Topic: C.08. Ischemia

Support: Heart and Stroke Foundation of Canada Operating Grant
New Frontiers in Research Fund – Exploration Award
NSERC Operating Grant

Title: A hemolysis assay to detect release of a molecular activator of spreading depolarization

Authors: C. LOWRY[1], B. M. BENNETT[1], R. D. ANDREW[2];
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Abstract: The channel that opens to drive spreading depolarization (SD) may not be a conventional channel under normal physiological conditions, one reason why it has eluded identification for +30 years. The Na^+/K^+-ATPase (the pump) is a transporter that normally shuttles cations across the neuronal plasma membrane. But when brain tissue loses blood flow, we hypothesize that a chemical activator is released which binds to the pump, converting it into an open channel that then drives SD. To test this concept of a released SD *activator* (SD*a*), we collected samples (containing putatively released SD*a*) from live brain slices undergoing SD evoked by simulated ischemia (O_2/glucose deprivation, OGD). After slices were removed, the artificial cerebrospinal fluid (aCSF) that bathed the slices was superfused over a healthy slice. SD was evoked in ~64% of these naïve slices, and this was not a result of elevated K^+ released by the slices. While this is a crude bioassay for SD*a* release, a higher throughput assay would be useful to help isolate and identify the SD*a*. Palytoxin (PLTX) is a marine poison. Its potency converting the Na^+/K^+-ATPase into an open channel can be measured using an hemolysis assay (Habermann et al. 1981). That is, pump molecules on the red blood cell (RBC) are converted to an open channel by PLTX binding. The RBCs swell and burst, releasing hemoglobin which is then colorimetrically assayed. If our SD*a* acts like PLTX, then SD*a* samples should induce RBC hemolysis. First, we replicated the Habermann RBC bioassay, generating a concentration-response curve for log [PLTX] vs % hemolysis ranging from [PLTX] = 0.01 to 1 nM. Hemolysis by 0.03 nM PLTX was increasingly inhibited by 100 to 500 μM ouabain which competes with PLTX binding at the Na^+/K^+-ATPase. Next, we tested the hemolytic activity of our SD*a* fractions. Hemolysis significantly increased in tubes containing aCSF which had bathed slices that generated SD (`post-SD` aCSF) as compared to pre-SD aCSF (n=4; $p<0.0001$).The latter evoked half the hemolysis of post-SD aCSF. This was repeated and replicated using slices in aCSF where SD was induced with brief hyperthermia (44°C). These data indicate a PLTX-like hemolytic effect by a compound released from those brain slices undergoing SD. Interestingly, the hemolytic efficacy of the putative SD*a* required a small priming amount of PLTX (.01 nM) which itself evoked only minimal hemolysis. These data support our previous electrophysiological work implicating the release of a molecule from metabolically stressed gray matter that promotes SD by converting the Na^+/K^+-ATPase from transporter to open channel in a PLTX-like fashion.

Disclosures: C. Lowry: None. **B.M. Bennett:** None. **R.D. Andrew:** None.

Poster

P323. Mechanisms of Spreading Depolarizations and Transmitter Release

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Program #/Poster #: P323.05

Topic: C.08. Ischemia

Support: NIMH Grant 2R01MH086638-11

Title: Tissue-scale computer modeling of spreading depolarization, ischemia and cytotoxic edema

Authors: *C. KELLEY[1], A. J. H. NEWTON[2], M. L. HINES[3], R. A. MCDOUGAL[4], W. W. LYTTON[5];
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Abstract: Spreading depolarization (SD) is characterized by a breakdown in the homeostatic maintenance of intracellular and extracellular ion concentrations, leading to a wave of depolarization spreading slowly through a neuronal population at 2-7 mm/min with a period of neuronal silence lasting minutes following in its wake. SD at the cellular level produces the spreading depression seen in extracellular field potentials. Using the NEURON simulator and its reaction-diffusion module, we developed tissue-scale models 0.125 - 1.0 mm^3 in size that include neurons with dynamic volumes in realistic densities, extracellular diffusion of ions and O_2, and glial buffering of extracellular K^+. We modeled SD for *in vitro* (O_2 diffuses into tissue from bath solution) and *in vivo* (O_2 replenished throughout tissue) environments, normoxic (0.04 mM O_2, extracellular volume fraction (α) 0.2, extracellular tortuosity (λ) 1.6) and ischemic (0.01 mM O_2, α= 0.07, λ= 1.8) conditions, and in presence of cytotoxic edema (cell swelling). SD was initiated by elevating extracellular K^+ (20-40 mM; 200 μm diameter) in a central core. Realistic neuronal surface:volume ratio (S:V) greater than 1 μm^{-1} was essential for producing SD with observed speeds, and SD wave speed increased roughly linearly with S:V. In the ischemic *in vitro* condition (neuronal volume fraction 0.24), SD could not be initiated with S:V= 0.02 μm^{-1}. The SD wave traveled at 1.2 mm/min with S:V= 1.0 μm^{-1}, 2.4 mm/min with S:V= 2.0 μm^{-1}, 4.8 mm/min with S:V= 3.0 μm^{-1} which is typical of cortex, and 6.0 mm/min with S:V= 4.0 μm^{-1}. SD wave speed is also dependent on tissue oxygenation, traveling at 2 mm/min in the normoxic state and 4.8 mm/min in the anoxic state when SA:V= 3 μm^{-1}. In the *in vivo* environment, the thresholds of required core size and elevated extracellular K^+ were higher in the normoxic condition than in ischemic conditions. Furthermore, when there was an ischemic core with cytotoxic edema but no initially elevated extracellular K^+, SD initiated spontaneously within 4 seconds then traveled at a speed of 1.8 mm/min. Our model demonstrated how imbalances in extracellular K^+ concentration, alterations to the extracellular space from edema, and impaired tissue oxygenation during ischemia produce SD. Using this approach to study SD, we can bridge the gap between empirical findings from SD elicited in experimental settings and SD associated with neurological conditions like ischemic stroke, traumatic brain injury, and migraine aura.

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Poster

P323. Mechanisms of Spreading Depolarizations and Transmitter Release

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Program #/Poster #: P323.06

Topic: C.08. Ischemia

Support: NIH Grant R01NS121426
NSF GRFP 2035701

Title: Probing the mechanism of rapid guanosine release during ischemia

Authors: *M. E. WEESE, A. E. ROSS;
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Abstract: Guanosine is a purinergic signaling molecule implicated in a range of neuroprotective functions, including ischemia. While adenosine's role in neuroprotection has been investigated, guanosine's role remains largely unknown. Guanosine is released rapidly following injury to reduce damage and prevent glutamate excitotoxicity. Similarly, guanosine minimizes reperfusion injury and greatly decreases the amount of infarct tissue following ischemic attack. Our lab recently discovered spontaneous transient guanosine release in the CA1 of the hippocampus, which increases significantly in concentration during ischemia. Despite this work, the mechanism of rapid release and the extent to which release is impacted during ischemia remains unknown. Here, we investigate guanosine's rapid release mechanism in both normoxic and ischemic conditions *ex vivo* using fast-scan cyclic voltammetry (FSCV). Mechanistic information was determined by evaluating the impact of Ca^{2+} chelation, voltage-gated Na^{+} channel blockage, inhibition of GTP hydrolysis, and adenosine A_1R antagonism on transient guanosine release. On average, guanosine transients are 222 ± 59 nM per event, last approximately 0.93 ± 0.05s, and occur approximately every 37 ± 7.3 s. Calcium chelation significantly decreased the frequency of guanosine events during normoxia and the events that remained were longer in duration (n = 16, $p < 0.0001$). These results indicate vesicular release drives the primary release mechanism. Preliminary work from the competitive inhibition of extracellular GTP hydrolases suggests both direct guanosine and GTP release. Understanding the mechanism of rapid guanosine release under both healthy and ischemic conditions will provide insight into guanosine's neuroprotective properties and open pathways for understanding its role during ischemia.

Disclosures: M.E. Weese: None. **A.E. Ross:** None.

Poster

P323. Mechanisms of Spreading Depolarizations and Transmitter Release

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Topic: C.08. Ischemia

Support: University of Cincinnati
NIH Grant R01NS121426

Title: Spontaneous release of hippocampal guanosine during normoxia and ischemia

Authors: *M. T. CRYAN, A. E. ROSS;
Dept. of Chem., Univ. of Cincinnati, Cincinnati, OH

Abstract: Guanosine is an important neuromodulator that is believed to protect the central nervous system during chemical injuries like stroke and ischemia. Previous reports have supported slow release of guanosine in response to chemical injury over the course of hours to days using liquid chromatography and microdialysis; however, it is not currently known if guanosine signals on a more rapid timescale in immediate response to these kinds of injuries. In this study, we used fast-scan cyclic voltammetry (FSCV) to electrochemically detect and characterize subsecond release of guanosine in the hippocampal CA1 region of the *ex vivo* rat brain. Guanosine transient release was quantitated under normal conditions and during a period of global ischemia. Transient event concentration and duration were significantly increased during the ischemic period. Guanosine was also shown to be released in a more burst-like, clustered fashion during ischemia compared to normoxic conditions. These changes in release behavior during ischemic stress gave rise to an overall increase in total guanosine concentration in response to tissue injury. POM1, an NTPDase inhibitor which blocks the breakdown of purine triphosphates to their ribonucleoside form, was used to support the observed transients as purinergic in nature and explore direct release of guanosine. The number of transient events was reduced during administration of POM1 compared to the predrug control. Total guanosine concentration was also lowered, implying that most transient guanosine is a result of breakdown from GTP. We report the first measurements of rapid guanosine release during normal conditions and during episodes of ischemic stress characterized with FSCV.

Disclosures: M.T. Cryan: None. **A.E. Ross:** None.

Poster

P333. Ischemia and Motor Control

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P333.01

Topic: C.08. Ischemia

Title: Refinement of cortico-rubro tract activity during long-term skilled motor learning and post-stroke rehabilitation

Authors: *H. GHUMAN, F. APARICIO, S. BARATI, K. GANGULY;
Univ. of California, San Francisco, San Francisco, CA

Abstract: Motor impairments after a stroke has been linked to the structural and functional integrity of the pathways that connect motor cortex and spinal cord. Besides the pyramidal tract (PT) in humans, alternate motor fibers (AMFs) have been suggested to play a key role in modulating recovery after stroke. In rodents, AMFs consist of the cortico-rubro-spinal and cortico-reticulo-spinal tracts. Lesioning of PT in rodents have shown to induce axonal sprouting of the cortico-rubro tract (CRT) neurons, but the precise role of CRT in skilled motor learning and execution remains poorly understood. Using retrograde labeling and head-mounted miniscope imaging, we first investigated the timing of when the CRT in primary motor cortex (M1) is recruited during learning a skilled reach-to-grasp task in free-behaving mice. We trained the mice for 10-12 weeks (250 trials/day, 5 days/week) to examine the stability of the CRT during long term motor learning and execution. We then induced a stroke in the secondary motor cortex (M2) of skilled mice using photothrombosis and investigated the interaction between CRT activity in M1 and functional recovery during post-stroke rehabilitation. In vivo calcium imaging of CRT neurons during motor task revealed a sequential activation of task-modulated neurons that remained task-engaged during long term learning. Improvements in task performance during early learning paralleled with the recruitment and activation of task-modulated CRT neurons. During the early (< 2 weeks) post-stroke rehabilitation, the motor performance of the skilled mice was severely impaired due to variable reach trajectories and unsuccessful grasps. The activity of the M1 CRT neurons during this period was significantly reduced and showed high inter-trial variability. During 8 weeks of post-stroke rehabilitation, task-modulated M1 CRT neurons again emerged that correlated to the functional recovery of the forelimb. These findings suggest that CRT plays a key role in modulating long term motor performance and recovery after a stroke.

Disclosures: H. Ghuman: None. **F. Aparicio:** None. **S. Barati:** None. **K. Ganguly:** None.

Poster

P333. Ischemia and Motor Control

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P333.02

Topic: C.09.Stroke

Title: Change over time feature of recruitment of motor unit of the paretic muscle of hemispheric stroke during the recovery phase

Authors: *M. YOSHIDA[1,2], T. ITO[1], H. FUNAKOSHI[1], T. MASUDA[3], T. KOKUBUN[4];
[1]Grad. school of Saitama Prefectural Univ., Koshigaya, Japan; [2]Rehabil. Amakusa Hosp.,

Koshigaya, Japan; [3]Fukushima Univ., Fukushima, Japan; [4]Physical Therapy, Saitama Prefectural Univ., Koshigaya, Japan

Abstract: The paralysis after the stroke induces the abnormality of the motor unit (MU) recruitment pattern. Generally, normal MU is recruited according to the size principle and regulates a firing rate. However, chronic hemiparetic stroke recruits small MU selectively, and it has been reported that it regulates a firing rate. From these reports, we thought that we might clarify the recruitment process of MU of the paralysis if we tracked a change of MU after the stroke. This pilot study aims to investigate the features of the motor unit recruitment of the paretic muscle in the recovery period of hemispheric stroke. One stroke patient participated in this study. This study was approved by the ethics review committee. We measured the muscle activity of the biceps brachii using surface electromyography (Delsys. Inc) with six-time points at the recovery period after stroke. We also measured elbow flexure torque using a handheld dynamometer (HHD) simultaneously. The raw EMG data were decomposed into motor unit action potential (MUAP). Then mean firing rate (MFR) and peak amplitude of MUAP were calculated by decomposed MUAP. Additionally, coefficients and intercepts of the regression line were calculated by MFR and MUAP's peak amplitude. The coefficient of the linear regression line was adopted as the outcome to evaluate a range of the recruited MU size, and the intercept of the linear regression line was adopted as the outcome to evaluate the change of the mean firing rate of MU recruited early. The elbow joint maximum flexure torque was calculated by maximum torque and the forearm length. The standard deviation of the root means square (RMS) of the biceps brachii was calculated by EMG data before the decomposition. The change over time of each time point was evaluated by the changing of three elements; MU data (regression, coefficient, and intercept), elbow joint maximum flexure torque, and the standard deviation of RMS of the biceps brachii. The coefficient and intercept of the regression line, elbow joint flexure torque, and the standard deviation of RMS the biceps brachii showed the tendencies to increase over time. The time-dependent increase of the regression coefficient showed a restrictive MU recruitment pattern. In addition, the time-dependent change of the intercept of the regression line showed that the early recruited MU firing rate increased. This pilot study clarified the MU properties of change over time in the paretic muscle of hemispheric stroke. The features to recruit a small MU over time and to increase firing rates were observed. The MU recruitment pattern in the paralysis muscle of the patient with convalescence stroke changes over time and small MU is selectively recruited.

Disclosures: M. Yoshida: None. **T. Ito:** None. **H. Funakoshi:** None. **T. Masuda:** None. **T. Kokubun:** None.

Poster

P333. Ischemia and Motor Control

Location: Virtual Only

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Program #/Poster #: P333.03

Topic: C.09.Stroke

Support: NIH Grant R01NS064136C

Title: Investigation of acute hyperglycemia on immune responses and stroke outcome after ischemic stroke

Authors: *H. CHEN[1], T. C. CHIANG[2], A. KIM[1], T. BLISS[3], M. Y. CHENG[2], G. K. STEINBERG[4];
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Abstract: Background: Hyperglycemia occurs in over 40% of ischemic stroke patients, and exacerbates hemorrhagic transformation (HT) and worsens stroke outcomes. Previous rodent studies reported deleterious effects of hyperglycemia on stroke outcome during acute stroke (within hours to days), however, the impact of hyperglycemia during the subacute stroke period (days to weeks) remains unclear. Moreover, the mechanism of hyperglycemia worsening stroke outcome remains elusive. In this study, we investigated the effects of acute hyperglycemia on stroke outcome and its effect on the peripheral and central immune responses, in both the acute and subacute phase of ischemic stroke.Method: C57BL6 male mice were subjected to ischemic stroke by intraluminal middle cerebral artery occlusion (MCAO) for 30 min followed by reperfusion. Glucose was injected intraperitoneally 10 min before MCAO to induce hyperglycemia (HG). One cohort was sacrificed at 24hr (acute stroke phase), and brain infarct, hemorrhagic transformation and brain edema were quantified. Another cohort was allowed to survive for two weeks post-stroke (subacute stroke phase). Neurological deficits were assessed by neurological scores on post-stroke days (PD) 1, 3 and 7. Horizontal rotating beam tests were performed to evaluate motor-sensory functions on pre-stroke for baseline and PD 4, 7, 11 and 14. Peripheral and brain immune cells were analyzed by flow cytometry.Result: In the acute phase (24hr), hyperglycemia did not alter brain infarct volume but significantly induced brain edema and HT, indicating that hyperglycemia-enhanced brain edema and HT are independent of the brain infarct. In the subacute phase, hyperglycemia stroke mice exhibited greater body weight loss ($p<0.0001$) and significant increase in mortality: 100% of hyperglycemic mice died by PD 8 compared to 25% mortality for euglycemic mice ($p=0.0008$). Post-mortem results show that 100% of hyperglycemia mice had macro-visible HT. Hyperglycemia stroke mice also had worsened neurological deficit at PD 1, 3, and 7 ($p<0.001$), and significantly impaired performance on the rotating beam test compared to the euglycemia group. Our pilot data suggests that hyperglycemia increased myeloid cell infiltration into ischemic brains, including neutrophils and monocytic cells.Conclusion: Hyperglycemia adversely impacts stroke outcomes, with higher mortality and worsened functional deficit. Modulation of the post-stroke immune response may contribute to these detrimental effects of hyperglycemia. Understanding the mechanisms by which hyperglycemia exacerbates stroke injury will be important to improve clinical outcomes.

Disclosures: H. Chen: None. **T.C. Chiang:** None. **A. Kim:** None. **T. Bliss:** None. **M.Y. Cheng:** None. **G.K. Steinberg:** None.

Poster

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Topic: C.09.Stroke

Support: NIH Grant T32 HD007418

Title: Altered motor drive to the lower limbs in individuals with bilateral cerebral palsy during a step-up task: a pilot study

Authors: *V. GOYAL[1,2], T. SUKAL-MOULTON[2,3];
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Abstract: Individuals with bilateral cerebral palsy (CP) often experience a loss of selective voluntary motor control (SVMC) in the lower limbs, such as abnormal coupling between the hip adductors and sagittal plane extensors. Reduced SVMC ability is postulated to be a peripheral biomarker for damage to corticospinal tracts and upregulation of corticobulbospinal tracts in humans. Individuals with bilateral CP spend the majority of the inclined gait cycle in the stance phase, yet investigation into the loss of SVMC during this phase has been limited. The stance requirements of stair climbing include a high degree of coordination between the hip abductors and sagittal plane extensors, making this activity of daily living an ideal task to investigate abnormal extensor coupling. Progressive weighting of the limbs is a technique that has previously been used to test the limits of SVMC ability in cerebral palsy. Thus, we combined a step-up task with load modulation to investigate abnormal extensor coupling in individuals with bilateral CP.
We included 5 participants with typical development (one male aged 18 years, four females aged 6, 11, 14, and 19 years) and 1 participant with bilateral CP (male aged 15 years) in our pilot study. Participants were outfitted with reflective markers for motion tracking and instructed to step-up repeatedly onto a 4-inch platform mounted on force plates. Body weight was modulated from -20% to +15% in 5% increments using a weighted vest and the Zero-G. Data analysis focused on two stance phases: the push-off phase of stance, where the trailing leg is pushing the body up onto the platform, and the pull-up phase of stance, where the leading leg is pulling the body up onto the platform. To quantify a stereotypical adductor/extensor coupling, peak support moments (the sum of hip, knee, and ankle sagittal plane moments) and hip abduction moments at the time of peak support moments were identified for each stance phase. All joint moments were normalized to body weight and the zero-load condition for comparison.
Participants with typical development showed increases in peak support moment and concurrent hip abduction moment with increased load in both stance phases. However, the participant with bilateral CP showed an increase in peak support moment and a decrease in concurrent hip abduction moment with increased load in the push-off phase of stance. These early results

indicate that abnormal extensor coupling, possibly from use of extrapyramidal motor tracts, may challenge strategies during dynamic activities of daily living. We suggest that the experimental paradigm is useful to explore the limits of SVMC in a practical and functional context.

Disclosures: V. Goyal: None. **T. Sukal-Moulton:** None.

Poster

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Topic: C.09.Stroke

Support: NIH

Title: Within-subject comparison of the neural activity patterns in premotor and somatosensory cortex pre- and post-ischemic injury to primary motor cortex in the rat.

Authors: *F. BARBAN[1,2], H. M. HUDSON[3], R. J. NUDO[3,4], M. CHIAPPALONE[1,2], D. J. GUGGENMOS[3];
[1]Italian Inst. of Technol., Genova, Italy; [2]Dept. of Informatics, Bioengineering, Robotics and Systems Engin., Univ. of Genoa, Genova, Italy; [3]Dept. of Rehabil. Med., [4]Landon Ctr. on Aging, Univ. of Kansas Med. Ctr., Kansas City, KS

Abstract: Acquired brain injuries, such as stroke, are a leading contributor of long-term disability worldwide. When an injury occurs within primary motor cortex (M1) there is a loss of descending output information to the spinal cord, a disruption in the integration of somatosensory and motor planning inputs (from primary somatosensory cortex (S1) and premotor cortex (PM), respectively) and ultimately global changes in functional connectivity and activity between spared, interconnected regions throughout the brain. While the loss of integrative and descending information contributes to the severity of observed motor deficits, some motor recovery can and does occur in spite of the permanent loss of M1 inputs and outputs. It is likely that the neuroplastic changes and their evolution that occur in the previously connected, spared brain regions are critical for the recovery process. While these changes have been demonstrated through anatomical and motor representational analyses, it is unclear what changes in neural activity occur following an ischemic injury to M1 and how this activity contributes to motor impairment and recovery. To further elucidate this effect, we designed a within-subject experiment in which Long-Evans rats were trained to perform a skilled reaching task. Following task acquisition, each rat was chronically implanted with fixed, 32-channel intracortical microwires arrays in PM and S1. Two, 1-hour long behavioral sessions were taken while full-bandwidth neural activity was recorded from all electrode sites and synchronized to video feeds box sensors. Following these baseline sessions, an ischemic injury was induced in

M1 unilateral to the cortical implants through endothelin-1 injections. Two additional behavioral sessions were acquired at Day 4 and Day 7 post-injury. Neural activity was processed using custom tools to analyze both Local Field Potential and multi-unit activity within and between S1 and PM and synchronized with the reaching behavior. Preliminary results in the week immediately following ischemic injury suggest there is a global reduction in LFP power and variance a cross all bands. A disruption in the connectivity structure, estimated using power envelope correlation and phase coherence, was also revealed between the arrays implanted in PM and S1 after the lesion. These results offer direct evidence of the disruption in firing patterns within and between PM and S1 following M1 injury. Ultimately, this study will help us better understand the neural mechanisms underlying stroke recovery and possibly provide spatiotemporal stimulation targets for electroceutical rehabilitation therapies.

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Poster

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Topic: C.09.Stroke

Support: NIH K99HD098300
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NIH R37CA225608
NIH R01DC014358

Title: Bihemispheric motor cortex connectivity to the jaw and tongue muscles after unilateral stroke in young and aged rats

Authors: M. J. CULLINS, N. P. CONNOR;
Dept. of Surgery, Univ. of Wisconsin, Madison, WI

Abstract: Introduction: Oral motor deficits, including dysphagia, commonly occur after stroke, yet the mechanisms of corticobulbar plasticity after stroke are not well understood. Due to the bihemispheric control of swallowing, it has been suggested that plasticity of the contralesional (intact) cortex drives recovery of swallowing. Age may be an important factor in that stroke most commonly occurs in aged individuals, age is an independent predictor of dysphagia after stroke, and neuroplasticity may be reduced with age. We sought to determine the impact of unilateral stroke and age on oromotor cortical plasticity by examining the volume of the sensorimotor cortex that activates the jaw and tongue muscles in both hemispheres. **Methods**: Using the middle cerebral artery occlusion rat stroke model, intracortical microstimulation (ICMS) was

used to map regions of sensorimotor cortex that activate tongue and jaw muscles in both hemispheres. Larger cortical maps suggest greater functional connectivity. Young adult (7 mo) and aged (30 mo) F344xBN rats underwent either a stroke via left middle cerebral artery occlusion or a control sham surgery, followed by ICMS mapping 8 weeks later. N = 11 aged stroke, 8 aged sham, 13 young stroke, 13 young sham. **Results**: The volume of sensorimotor cortex that activates the tongue in the ipsilesional (damaged) hemisphere was reduced with stroke in the aged group (aged sham = 4.73 mm^3, aged stroke = 0.85 mm^3, $p < 0.001$), but failed to reach significance in the young group (young sham = 4.36 mm^3, young stroke = 3.04 mm^3, $p = 0.124$). Previous studies found that swallowing bolus area was reduced with stroke. In this study, within the aged stroke group, bolus area was correlated with the volume of cortex activating the tongue in the ipsilesional hemisphere (Pearson's $r = 0.74$, $p = 0.008$). Stroke in the aged group was also associated with an increased jaw representation in the contralesional hemisphere ($p = 0.009$). Both age groups exhibited increased stimulation thresholds in the ipsilateral hemisphere (young $p = 0.007$; aged $p = 0.007$). No significant differences in motor map size with age were found within the sham group. **Conclusion**: Unilateral MCA stroke is associated with a significant loss of functional connectivity between the sensorimotor cortex and the tongue muscles, which is greater with age. This reduced cortical input is likely to represent reduced volitional control of the lingual musculature. An increase in the jaw representation in the intact hemisphere is in agreement with studies that have suggested a role for contralesional cortical plasticity in the recovery of oral motor function after stroke.

Disclosures: M.J. Cullins: None. **N.P. Connor:** None.

Poster

P333. Ischemia and Motor Control

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P333.07

Topic: C.09.Stroke

Title: Comparison of stroke induction procedures on olfactory discrimination and performance in mice

Authors: T. O. COTHREN, J. T. HOLSTEN, K. B. PRESCOTT, T. C. PETERSON, *D. A. MACQUEEN, III;
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Abstract: Stroke is a leading cause of disability in the United States. A stroke can cause both physical and cognitive deficits. Levels of impairment on physical and cognitive functioning depend on the brain structures affected by the stroke. The current study compared the behavioral effects produced by two different stroke induction procedures on an automated Odor Discrimination Task (ODT). Twelve C57BL/6J mice were trained to stability on the ODT, which

consists of five odors assigned as target stimuli and one odor assigned as a non-target. Across a series of trials, mice are required to lick in response to target-odors to receive a sucrose reward, and to inhibit licking in response to the non-target odor. Measures derived from the ODT allow for the evaluation of both sensorimotor and cognitive effects. Following training, subjects were counterbalanced into groups based upon performance. Each subject received either a sham operation (n = 4), a photothrombotic stroke (P stroke; n = 4), or a distal middle cerebral artery occlusion (dMCAO; n = 4). The dMCAO models the most common type of ischemic stroke observed in humans. The photothrombotic stroke surgery utilized administration of a light-sensitive dye (Rose Bengal) and a high intensity light stereotactically placed above the primary motor cortex to create microcoagulation. Measures derived from the ODT include sensitivity index, responsivity index, trials completed, short samples (trials terminated prematurely), lick density, hit rate, and correct rejection rate. A 2-way ANOVA of time by condition was used to analyze data for each response parameter. There was a significant time X condition interaction for trials completed (p = 0.008), hit rate (p = 0.007), and lick density during the initial odor exposure period ($p < 0.001$). Only P stroke led to significantly greater lick density (during the odor release period) compared with baseline; observable on post-surgery day 1 and points up to 2-weeks following surgery. All groups exhibited significant decreases from baseline in trials completed at various points within the post-surgery period. No group evidenced significant differences from baseline with regard to hit rate. Though photothrombotic stroke procedures induced a transitory impact on lick density, performance was similar across conditions by the conclusion of the study (28 days). The lick monitoring procedures of the ODT can be used to detect differential motoric effects of stroke induction procedures.

Disclosures: T.O. Cothren: None. **J.T. Holsten:** None. **K.B. Prescott:** None. **T.C. Peterson:** None. **D.A. MacQueen:** None.

Poster

P333. Ischemia and Motor Control

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P333.08

Topic: C.09.Stroke

Title: Large Vessel Stroke as a Presenting Neurological Manifestation after Covid Vaccination in a Young Adult

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Abstract: Adenovirus-based COVID 19 vaccines from AstraZeneca and Johnson & Johnson have been associated with a syndrome of thrombosis, mainly involving the venous system, as well as thrombocytopenia akin to heparin-induced thrombocytopenia. Most of the cases were in women. Arterial thrombosis associated with these vaccines is relatively rare; however, a few cases have been reported with the AstraZeneca vaccine but not Johnson and Johnson. Here we report a case of a 24-year-old African American with a history of PTSD was brought to the ED, a week after his COVID vaccination, on account of mental status alteration, dysarthria with a right facial droop. Besides testing positive for cannabinoids in his urine, his laboratory results including platelets were within normal limits. CT angiogram showed an infarct in the distribution of the left middle cerebral artery with a relatively small infarct area with a large penumbra. The lesion was confirmed with brain MRI . . Neurosurgery was consulted; however, he did not meet the criteria for thrombectomy. and also did not receive TPA as he did not meet the criteria giving the extent of his lesion. An echocardiogram revealed no intramural thrombosis and no arrhythmia on telemetry. A thrombophilia workup did not reveal any predisposition to clot formation. Interestingly, his symptoms completely resolved within two days of admission and he was successfully discharged to follow up as an out-patient. Given the rarity of a stroke at his age and his apparent lack of risk factors, it is quite likely that his stroke was due to increased coagulation due to the vaccine. The fact that he did not have thrombocytopenia and HIT-like features or a thrombogenic risk presents the possibility of alternative mechanisms to thrombosis due to the vaccine. His quick recovery could be attributed to his youth and the possible reversibility of the pathogenic process. However, more data is required for a clearer picture of the incidence, distribution, and pathogenesis of stroke in young people after JnJ vaccination so that appropriate recommendations and interventions can be put in place for optimal management.

Disclosures: K. Affram: None. **L. Ayari:** None. **M. John:** None. **B. Kirmani:** None.

Poster

P375. Pain Processing: Itch: Models and Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P375.01

Topic: D.02. Somatosensation

Title: Neuroprotectin D1 analogue modulates spinal cord synaptic transmission in a mouse model of chronic itch

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[2]Pain Res. Division, Anesthesiol., [1]Duke Univ. Med. Ctr., Durham, NC

Abstract: Patients with cutaneous T cell lymphomas (CTCL) frequently have severe chronic itching. Our lab established an animal model of CTCL in nude mice, showing robust lymphoma growth and chronic itch on the back of mice (Han et al., Neuron, 2018). Specialized pro-

resolving mediators (SPMs), such as resolvins, protectins, and maresins, have demonstrated potent anti-inflammation, pro-resolution, and antinociception actions in different animal models of inflammation (Xu et al., Nat Med, 2010). Recently, we reported that a stable analogue of neuroprotection D1 (NPD1), 3-oxa-PD1n-3 DPA (Oxa), produced potent inhibition in itching behaviors (scratching) in CTCL mice following intrathecal injection (Nesman et al., 2021). However, the mechanism by which Oxa suppresses chronic itch is unknown. Here, we show that Oxa can rapidly modulate excitatory and inhibitory neurotransmission in the cervical spinal dorsal horn (SDH) neurons. We used whole-cell patch clamp recordings from SDH neurons of naïve and CTCL mice. Baseline frequency of spontaneous excitatory postsynaptic current (sEPSC) was significantly higher in CTCL mice compared with that of naïve mice ($P = 0.0075$). Perfusion of spinal cord slices with Oxa (10 ng/ml) rapidly (< 3 min) decreased the sEPSC frequency in SDH neurons from CTCL mice but had no effects in SDH neurons of naïve mice. There were no significant effects of Oxa on the amplitude of sEPSC in both groups of mice. Next, we recorded spontaneous inhibitory postsynaptic current (sIPSC). The baseline frequency and amplitude of sIPSC were not different between CTCL mice and naïve mice. Notably, perfusion of Oxa (10 ng/ml) increased the frequency of sIPSC in the CTCL mice. These results indicate that a stable analogue of NPD1, 3-oxa-PD1 can potently inhibit the release of excitatory neurotransmitters and enhance the release of inhibitory neurotransmitters from presynaptic nerve terminals in the spinal cord in chronic itch conditions. Our results provide a new mechanism by which SPM relieves chronic itch.
References Han et al., Neuron. 2018;99(3):449-463.e6Xu et al., Nat Med. 2010;16(5):592-7Nesman et al., Org Biomol Chem. 2021;19(12):2744-2752.

Disclosures: K. Furutani: None. **O. Chen:** None. **R. Ji:** F. Consulting Fees (e.g., advisory boards); Boston Scientific.

Poster

P375. Pain Processing: Itch: Models and Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P375.02

Topic: D.02. Somatosensation

Support: F31 NS113407

Title: Molecular determinants of central sensitization in chronic itch - computational modeling studies

Authors: *R. D. GRAHAM[1], H. LEE[2], B. DUAN[2], S. F. LEMPKA[1];
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Abstract: Chronic itch is a debilitating condition that can be symptomatic of many clinical disorders (e.g., chronic kidney disease, depression). Recent work has found that spinal interneurons that express urocortin 3 (Ucn3+) are sensitized in chronic itch conditions. Furthermore, Ucn3+ neurons receive inhibitory synaptic inputs from interneurons that express neuropeptide Y (NPY+), and this inhibition is reduced in chronic itch conditions. However, we do not understand the molecular constituents governing the development of chronic itch phenotypes. Uncovering these mechanisms may assist in developing novel treatments for chronic itch. To study chronic itch pathogenesis, we developed multi-compartment models of Ucn3+ and NPY+ spinal interneurons. We represented both model types using a ball-and-stick morphology described in previous studies of spinal interneurons. We simulated the active membrane properties of each neuron using Hodgkin-Huxley-style dynamics. Experimental data showed that the expression levels of several ion channels ($Na_v1.6$, SK, $Ca_v2.3$, BK) are significantly affected in a calcipotriol-treated mouse model of atopic dermatitis. We used a Markov Chain Monte Carlo method to generated and test thousands of combinations of ion channel conductances, which produced hundreds of biophysically plausible Ucn3+ and NPY+ neuron models in control and chronic itch conditions. We validated these model populations on their ability to reproduce experimentally measured excitability metrics (e.g., rheobase, spike interval). The model populations reproduced many of the distributions of experimentally measured excitability metrics. To determine which ion channels contribute to chronic itch pathogenesis, we systematically increased or decreased the conductance of each channel type in each cell model to mimic the changes in channel expression observed experimentally. We found that increasing $Na_v1.6$ conductance in model Ucn3+ neurons produced a modest increase in Ucn3+ excitability, whereas decreasing $Na_v1.6$ conductance in model NPY+ neurons produced a considerable decrease in excitability typical of chronic itch conditions. Furthermore, decreasing $Na_v1.6$ conductance in NPY+ neurons increased the likelihood that Ucn3+ neurons would generated action potentials in response to afferent inputs. Therefore, decreased $Na_v1.6$ expression in NPY+ neurons may be a key mechanism of chronic itch pathogenesis. Preliminary data suggest that electrical stimulation of Abeta axons in the dorsal roots may decrease the output of Ucn3+ neurons. Going forward, we will further explore the use of electrical stimulation therapies in treating chronic itch.

Disclosures: R.D. Graham: None. **H. Lee:** None. **B. Duan:** None. **S.F. Lempka:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Medtronic, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Presidio Medical, Inc., Hologram Consultants, LLC. F. Consulting Fees (e.g., advisory boards); Abbott Neuromodulation.

Poster

P375. Pain Processing: Itch: Models and Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P375.03

Topic: D.02. Somatosensation

Support: NIH Grant R01NS105680
Muscular Dystrophy Association
University of Arizona (Technology and Research Incentive Fund)

Title: Tmem184b is necessary for IL-31 induced itch

Authors: E. G. LARSEN[1], T. S. CHO[1], M. L. MCBRIDE[1], J. FENG[2], B. MANIVANNAN[1], C. MADURA[3], N. E. KLEIN[1], ***E. B. WRIGHT**[1], H. D. GARCIA-VERDUGO[1], C. JARVIS[1], R. KHANNA[3], H. HU[2], T. M. LARGENT-MILNES[3], M. R. C. BHATTACHARYA[1];
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Abstract: Nociceptive and pruriceptive neurons in the dorsal root ganglia (DRG) convey sensations of pain and itch to the spinal cord, respectively. One subtype of mature DRG neurons, comprising about 5% of neurons in the ganglia, is responsible for sensing mediators of acute itch and atopic dermatitis, including the cytokine IL-31. How itch-sensitive (pruriceptive) neurons are specified is unclear. Here we show that *Tmem184b*, a gene with roles in axon degeneration and nerve terminal maintenance, is required for the expression of a large cohort of itch receptors, including those for IL-31, Leukotriene C4, and Histamine. Male and female mice lacking *Tmem184b* show reduced responses to IL-31, but maintain normal responses to pain and mechanical force, indicating a specific behavioral defect in pruriception. Calcium imaging experiments indicate that a reduction in IL-31 induced calcium entry is a likely contributor to this phenotype. We identify an early failure of proper Wnt-dependent transcriptional signatures and signaling components in Tmem184b mutant mice that may explain the improper DRG neuronal subtype specification. Accordingly, lentiviral re-expression of Tmem184b in mutant embryonic neurons restores Wnt signatures. Together, these data demonstrate that Tmem184b promotes adult somatosensation through developmental Wnt signaling and promotion of proper pruriceptive gene expression. Our data illuminate a new key regulatory step in the processes controlling the establishment of diversity in the somatosensory system

Disclosures: E.G. Larsen: None. **T.S. Cho:** None. **M.L. McBride:** None. **J. Feng:** None. **B. Manivannan:** None. **C. Madura:** None. **N.E. Klein:** None. **E.B. Wright:** None. **H.D. Garcia-Verdugo:** None. **C. Jarvis:** None. **R. Khanna:** None. **H. Hu:** None. **T.M. Largent-Milnes:** None. **M.R.C. Bhattacharya:** None.

Poster

P375. Pain Processing: Itch: Models and Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P375.04

Topic: D.02. Somatosensation

Support: NIH R01AR074062
Stanley Glaser Foundation Research Award UM SJG 2019-18

Title: Npy2r^{+} neurons play distinct roles in itch and pain in the amygdala

Authors: *D. PAVLENKO[1], K. SANDERS[1], R. VERPILE[1], A. MARKAN[2], H. ISHIDA[1], T. AKIYAMA[1];
[1]Dermatol., [2]Univ. of Miami, Miami, FL

Abstract: Title: NPY2R^{+} neurons play distinct roles in itch and pain in the amygdala The amygdala plays an important role in the neural pathways of itch and pain. Recent studies have demonstrated that stimulating itch-responsive neurons in the amygdala increases itch- and anxiety-related behaviors. In contrast, stimulation of the $GABA_A$ receptor in the amygdala results in a reduction in itch-related behavior. Neuropeptide Y receptor type 2 (NPY2R) has been found primarily in the amygdala and hypothalamus. It has been categorized as a presynaptic GCPR whose activation leads to a downstream inhibition of glutamate release. In the present study, we investigated the involvement of NPY2R-expressing amygdala neurons in itch and pain.To manipulate the activity of NPY2R-expressing neurons, we prepared NPY2R-cre mice and injected a cre-dependent AAV into the amygdala of these mice. The cre-dependent AAV carried tdTomato and chrimsonR, allowing us to activate NPY2R-expressing neurons with light and map the projection patterns of NPY2R-expressing amygdala neurons in the brain. After surgery, the intradermal injections were performed in light on and light off conditions. When the light is turned on, the NPY2R neurons are activated, and when mice were injected with 5-HT (serotonin) and histamine, the itch-related behavior was suppressed. Finally, there was no difference in mouse response in mechanical or thermal pain test. Based on these findings, we concluded that NPY2R-expressing amygdala neurons are involved in the regulation of acute itch, but not acute pain.

Disclosures: D. Pavlenko: None. **K. Sanders:** None. **R. Verpile:** None. **A. Markan:** None. **H. Ishida:** None. **T. Akiyama:** None.

Poster

P375. Pain Processing: Itch: Models and Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P375.05

Topic: D.02. Somatosensation

Title: Anoctamin 1/TMEM16A in pruritoceptors is essential for Mas-related G protein receptor-dependent itch

Authors: *H. KIM, H. KIM, S. PAK, S. LIM, U. OH;
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Abstract: Itch is an unpleasant sensation that evokes a desire to scratch. Pathologicconditions such as allergy or atopic dermatitis produce severe itching sensation. Mas-related G protein receptors (Mrgprs) are receptors for many native pruritogens. However, signaling pathways downstream to these receptors in dorsal root ganglion (DRG) neurons are not yet understood. We found that Anoctamin 1 (ANO1/TMEM16A), a Ca2+-activated chloride channel is a primary transduction channel mediating Mrgprs-dependentitch signals. The majority of MrgprA3 positive DRG neurons expressedANO1. Genetic ablation of Ano1 in DRG neurons (AdvCREAno1fl/fl) displayed a significant reduction in scratching behaviors in response to agonists for Mrgprs as well as in Mrgprs-dependent chronic itch models. In-vivo Ca2+ imaging revealed that chloroquine, a potent Mrgpr receptor agonist, excited DRG neurons via ANO1. Chloroquine, SLIGRL, and bilirubin activate ANO1 via MrgprA3, MrgprC11, and MrgprA1, respectively. More importantly, the overexpression of Ano1 in DRG neurons of Ano1-deficient mice rescued the impaired itching. These results demonstrate that ANO1 mediates the Mrgprs-dependent itch signaling in pruriceptors and provides clues to treating pathologic itch syndromes.

Disclosures: H. Kim: None. **H. Kim:** None. **S. Pak:** None. **S. Lim:** None. **U. Oh:** None.

Poster

P375. Pain Processing: Itch: Models and Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P375.06

Topic: D.02. Somatosensation

Support: NIH Grant 5R01NS109170

Title: Molecular determinants of central sensitization in chronic itch - electrophysiological studies

Authors: *H. LEE[1], R. D. GRAHAM[2,3], S. F. LEMPKA[2,3], B. DUAN[1];
[1]Dept. of Molecular, Cell. and Developmental Biol., [2]Dept. of Biomed. Engin., [3]Biointerfaces Inst., Univ. of Michigan, Ann Arbor, MI

Abstract: Chronic itch, a debilitating symptom of skin inflammation, is associated with sensitization of the somatosensory nervous system. Recent studies have identified the selective neural circuits that transmit acute itch; however, the mechanisms by which itch transforms into a pathological chronic state remain largely unknown. We have previously shown that Aβ

mechanoreceptors, together with spinal Urocortin 3-positive (Ucn3+) excitatory interneurons and neuropeptide Y-positive (NPY+) inhibitory interneurons, form a microcircuit that transmit and gate mechanical itch. Sensitization of this spinal circuit contributes to the development of chronic itch. Here, using whole-cell path clamp recordings, we observed increased intrinsic excitability and reduced inhibitory synaptic transmission in Ucn3+ neurons under a chronic itch condition. In contrast to Ucn3+ neurons, intrinsic excitability of NPY+ neurons were largely reduced under a chronic itch condition. To explore the molecular mechanisms underlying sensitization of this circuit, we then used single-cell qRT-PCR to examine the mRNA expression levels of voltage-gated sodium, potassium, and calcium channels in spinal Ucn3+ and NPY+ neurons under a chronic itch condition. We found that the expression levels of Nav1.6 and Cav2.3 channels were increased in Ucn3+ neurons, while the expression level of SK3 channel was decreased. By contrast, we observed that the expression levels of Nav1.6 and BK channels were decreased in NPY+ neurons. Furthermore, our ongoing computational work reveals that decreasing Nav1.6 conductance in NPY+ neurons plays a major role in opening the itch gate in this microcircuit under a chronic itch condition. However, changing SK, BK, or Cav2.3 conductance had negligible effects on sensitization of this circuit. Therefore, our results suggest that changes in Nav1.6 expression are a key component of chronic itch sensitization. The findings presented here may open a new avenue for developing pharmaceutical strategies to treat chronic itch.

Disclosures: H. Lee: None. **R.D. Graham:** None. **S.F. Lempka:** None. **B. Duan:** None.

Poster

P375. Pain Processing: Itch: Models and Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P375.07

Topic: D.02. Somatosensation

Support: NIH Grant NS111643

Title: Loss of NPY-Y1 inhibition worsens pruritis during aging

Authors: *D. ACTON, M. D. GOULDING;
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Abstract: The sensation of itch is generated either by mechanical stimuli, such as an insect crawling across the skin, or by chemical stimuli, such as histamine released from activated mast cells. Both forms of stimulus generate the urge to scratch, which has the effect of removing harmful irritants from the skin. In healthy animals they do this by activating discrete neural pathways in the skin and spinal cord; however, dysregulated crosstalk between these pathways gives rise to chronic itch, characterized by spontaneous scratching and elevated sensitivity to

mechanical stimuli (alloknesis). Chronic itch is a common debilitating condition that becomes more prevalent during aging, and for which treatments are lacking. We recently demonstrated that neuropeptide Y (NPY) acts at the inhibitory NPY receptor type 1 (Y1) to establish thresholds for touch discrimination and mechanical itch responses in healthy mice (Bourane et al. 2014; Acton et al., 2019). Ablation of $Y1^+/Lbx1^+$ dorsal horn neurons prevents mechanical itch responses but spares chemical itch sensitivity. Conversely, pharmacological blockade of Y1 or conditional knockout of *Npy1r* in the dorsal horn strongly potentiates mechanical but not chemical itch. Here, we report that NPY-Y1 signaling strongly suppresses itch responses in models of both acute and chronic pruritis in young adult (8-week-old) mice. Using the Y1 antagonist BIBP 3226, which potentiates itch by disinhibiting $Y1^+/Lbx1^+$ dorsal horn neurons, we show that NPY derived from spinal INs suppresses alloknesis induced by intradermal histamine, as well as in dry skin and allergic contact dermatitis models of chronic itch. Aged (18-month-old) mice display elevated baseline levels of spontaneous and evoked scratching, as well as increased scratching in models of acute alloknesis and chronic itch. Remarkably, the protective effects of endogenous NPY are lost in aged mice, likely accounting for these elevated scratching responses. By contrast, aged mice remain sensitive to the selective Y1 agonist [Leu^{31}, Pro^{34}]-NPY, suggesting reduced availability of NPY, rather than loss of Y1 signaling. Consistent with this, expression of NPY but not Y1 is reduced in aged mice. Current efforts are directed at identifying the cellular processes that drive the attenuation of NPY signaling and increase itch sensitivity during aging.

Disclosures: D. Acton: None. **M.D. Goulding:** None.

Poster

P375. Pain Processing: Itch: Models and Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P375.08

Topic: D.02. Somatosensation

Title: Role of dopamine and adenosine receptor ligands on spinal reflexes in an inducible iron-deficient model of Restless Legs Syndrome

Authors: S. WOODS[1], M. SCHAUB[1], M. DIAZ-RIOS[2], V. CASADÓ ANGUERA[3], V. CASADO[3], ***S. CLEMENS**[1];
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Abstract: Restless Legs Syndrome (RLS) is a common sensorimotor disorder that heavily affects sleep. Dopaminergics that target the dopamine (DA) D3 receptor (D3R) subtype are one treatment option for RLS. Brain iron deficiency (BID) is tightly associated with RLS in the clinic, and animal models have shown a correlation between BID and alterations in DA D1

receptor (D1R) and adenosine (ADE) neurotransmission in the striatum. However, the effects of BID in the spinal cord remain unclear. Therefore, we here probed the effects of an iron-deficient (ID) diet on spinal reflex latencies and their modulation by select DA and ADE receptor ligands. Male and female mice (C57BL/6) were separated upon weaning into control diet cohorts and cohorts on an ID diet (Envigo Teklad TD.99397, 2-6 ppm Fe). A Hargreaves system was used to measure thermal pain reflex withdrawal latencies under control conditions (no drugs), and after i.p. injections of DA (D3R agonist: pramipexole, PPX, D1R antagonist: SCH 39166, both at 0.5 mg/kg) and ADE receptor ligands (caffeine and N6-cyclopentyl-adenosine, CPA, at 50 and 1 mg/kg, respectively). Following behavioral testing, spinal cords were harvested and probed for expression of DA and ADE receptors, and proximity ligation assays (PLAs) were performed to assess A1R-D1R receptor heteromer expression.
We found that reflex latencies decreased after 3 weeks compared to controls, suggesting increased spinal cord excitability in male and female ID cohorts. Treatment with PPX restored reflex latencies of ID animals to those of the control cohorts, but it also induced rearing activities in the ID animals. Treatment with SCH 39166 increased reflex latencies in male but not female ID animals, while treatment with CPA increased reflex latencies in both male and female ID animals, without inducing rearing behaviors. Tissue analyses revealed an increase in D1R expression and a significant decrease in A1R-D1R heteromers.
These data indicate that diet-induced iron-deficiency leads to an increase in spinal cord excitability and that DA and ADE receptor ligands can restore normal reflex latencies. We suggest that ID affects DA and ADE neuromodulation by way of A1R-D1R and possibly D1R-D3R heteromers.

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Poster

P410. Somatosensory Cortical Processing

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P410.01

Topic: D.04. Somatosensation – Touch

Support: DARPA Grant

Title: Enhanced tactile sensitivity using intracortical microstimulation

Authors: *L. E. OSBORN[1], B. P. CHRISTIE[1], D. P. MCMULLEN[2], A. S. PAWAR[3], M. C. THOMPSON[1], M. A. ANAYA[3], S. J. BENSMAIA[4], P. A. CELNIK[3], M. S. FIFER[1], F. V. TENORE[1];
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Abstract: Enhanced tactile sensitivity using intracortical microstimulationLuke Osborn, Breanne Christie, David McMullen, Ambarish Pawar, Margaret Thompson, Manuel Anaya, Sliman Bensmaia, Pablo Celnik, Matthew Fifer, Francesco TenoreIntroducing low levels of noise to a weak vibrotactile signal can increase the detectability of the weak signal (Enders et al 2013). Known as stochastic resonance, this phenomenon appears to improve humans' tactile detection ability by delivering a noninformative subthreshold vibrotactile signal in addition to the stimulus of interest. Researchers showed that even remote vibrotactile noise (i.e., vibration on the wrist) has potential to enhance tactile sensitivity at the fingertips (Enders et al). Currently, it is unknown if other tactile stimuli modalities can also enhance the detectability of vibrotactile stimuli. In this study, we explored the role of cortical stimulation in improving tactile sensitivity in a human participant using a brain-computer interface. Microelectrode arrays were implanted in sensorimotor regions of the brain in a human participant with a sensory incomplete spinal cord injury (C5/C6 ASIA Impairment Scale Grade B). Intracortical microstimulation (ICMS) of primary somatosensory cortices elicited tactile sensations in the participant's hands, who also retained native tactile sensation in those same regions.In six different sites across both left and right hands, we delivered haptic stimuli using a 300 Hz vibrating motor. We measured the haptic detection thresholds while delivering subthreshold ICMS (approximately -1.5 dB) as well as with suprathreshold ICMS (>1.5 dB). The ICMS was delivered through electrodes that could activate neural populations that elicited percepts overlapping with the placement of the vibrating motor. Specifically, the sites tested on the right hand were thumb fingertip, index fingertip, base of index finger, base of middle finger, ring fingertip and on the left hand the thumb fingertip. Averaging across all sites, we found that subthreshold ICMS decreased the haptic threshold by 26% below baseline (-1.58 dB) with standard error 7.18% (0.42 dB). The suprathreshold ICMS condition increased the haptic threshold by 24% over baseline (0.50 dB) with standard error 14.40% (0.50 dB). These results show that ICMS to targeted regions of the somatosensory cortex is able to enhance native haptic sensitivity, as measured by detection thresholds, of a human participant. These findings have implications for rehabilitating, restoring, and improving native tactile sensitivity and perceptions in individuals living with spinal cord injury, deficits from stroke, or diabetic neuropathy.

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Poster

P410. Somatosensory Cortical Processing

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P410.02

Topic: D.04. Somatosensation – Touch

Support: APL internal grant

Title: Manipulating the perception of artificial touch using non-informative vision

Authors: *B. CHRISTIE[1], L. OSBORN[1], D. MCMULLEN[2], A. PAWAR[3], F. TENORE[1], M. S. FIFER[1];
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Abstract: The brain continuously receives information from multiple sensory pathways. These inputs are combined to form a cohesive picture of our surroundings, allowing us to perform adaptive behaviors. To interact with objects in our environment, the brain must integrate visual and tactile signals. Visual feedback can actually improve tactile acuity [1], but we still do not fully understand the neural mechanisms behind visuotactile integration. Visual and tactile signals are thought to be integrated in frontal, parietal, and subcortical regions of the brain [2]. In this study, we stimulated the primary somatosensory cortex (S1), bypassing subcortical regions such as the thalamus, to assess if vision can still affect touch when touch is delivered directly through S1.

We delivered intracortical microstimulation (ICMS) to a human subject with penetrating microelectrode arrays implanted in S1 to deliver artificial tactile stimuli to the hand [3] while the subject received non-informative visual feedback. The term “non-informative” refers to when an individual receives visual feedback of a body part that is being stimulated, but has no vision of the tactile stimulus itself. Non-informative vision has been previously shown to enhance tactile acuity [1]. We tested seven electrodes that each evoked percepts in at least two spatially discrete regions of the hand. The participant was asked to focus his gaze away from the hand or on one of the two discrete regions, and then to report where he felt tactile stimuli on a diagram of the hand. Each electrode and visual condition pairing was repeated up to nine times. We compared the reporting frequency in each region of the hand across visual conditions.

We found that the perceived locations of tactile percepts elicited by ICMS were not affected by non-informative vision. Across visual conditions, reporting frequency neither increased in attended regions nor decreased in unattended regions of the hand. Future experiments could include running control tests with vibration or electrocutaneous stimulation.

In summary, our results suggest that subcortical brain regions, such as the thalamus, may be required for integrating vision and touch, which aligns with prior literature [2]. This study also has important implications for the field of brain-computer interfaces (BCIs): some training may be required for BCI users to learn to integrate visual inputs with tactile percepts evoked by ICMS. Future experiments could evaluate if a training paradigm is necessary to learn this association, or if it could be passively learned over time with long-term use outside of the laboratory.

[1] Kennett et al. 2001, [2] Gentile et al. 2010, [3] Fifer et al. 2020

Disclosures: B. Christie: None. **L. Osborn:** None. **D. McMullen:** None. **A. Pawar:** None. **F. Tenore:** None. **M.S. Fifer:** None.

Poster

P410. Somatosensory Cortical Processing

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P410.03

Topic: D.04. Somatosensation – Touch

Support: DARPA Contract HR001120C0120 ("NEO")

Title: Virtualized Touch of Holographic Objects in Augmented Reality via Intracortical Microstimulation of Human S1

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Abstract: Augmented reality (AR) technologies enable immersive experiences with simulated virtual objects. Currently, haptic feedback in AR can be provided via wearable gloves or exoskeletons. Haptic devices may be able to relay touch information with extremely high fidelity but have substantial limitations: (1) haptic devices often occlude the skin, so they may only replace touch sensation rather than augment natural touch, (2) haptic devices are often bulky, encumbering natural dexterous finger movements. Direct electrical stimulation of the peripheral nerves and/or central nervous system may be a promising strategy for unobtrusively providing haptic feedback to individuals with sensory loss and/or commercial users as neural interfaces become more widespread. The present study explores the potential for direct neural stimulation of primary somatosensory cortex (S1) to provide haptic feedback in AR. We worked with a human subject who was implanted bilaterally with microelectrode arrays in S1. The participant retained sufficient proximal arm and hand functionality to enable interactions with holographic objects. Virtual objects (cubes) were holographically projected into the workspace via a Microsoft Hololens 2. Intracortical microstimulation (ICMS) was delivered in one of two modes: (1) a fixed-duration, fixed frequency pulse delivered to finger-specific site(s) upon that tracked finger's entry or exit from the virtual object, (2) fixed-frequency stimulation to finger-specific site(s) while that finger was within the virtual object. In initial exploratory sessions, the subject was able to physically scan the workspace and reliably hover within invisible virtual objects in either ICMS delivery mode. In subsequent sessions, the participant was able to differentiate virtual objects by their assigned haptic magnitudes (i.e., ICMS pulse magnitude). In a culminating session, the subject was able to interact with several virtual objects whose compliance was simulated via object scale change and stimulation intensity that scaled linearly with finger movement distance. In this session, the subject was reliably able to select the least

compliant virtual object (of two), and sort a set of four virtual objects by their simulated compliance.This study provides a compelling baseline experience for augmenting or bypassing touch feedback in AR scenarios via direct neural stimulation, worthy of further exploration with noninvasive and/or implanted stimulation devices. For individuals with sensory loss, neurally-enabled haptics may increase the accessibility of virtual experiences as mixed reality technologies become more widely used.

Disclosures: M.S. Fifer: None. **L.E. Osborn:** None. **D.P. McMullen:** None. **B.P. Christie:** None. **A.S. Pawar:** None. **S.J. Bensmaia:** None. **P.A. Celnik:** None. **F.V. Tenore:** None.

Poster

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Title: Distinct neocortical mechanisms underlie human SI responses to median nerve and laser evoked peripheral activation examined with the Human Neocortical Neurosolver modeling tool

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Abstract: Non-invasive, clinically-relevant tools like magneto- and/or electro-encephalography (M/EEG) have long been used to measure electromagnetic fields associated with non-nociceptive and nociceptive processing in the human primary somatosensory cortex (SI). The physiological mechanisms that produce such macroscale signals, however, have yet to be fully articulated. We examined the neocortical circuit mechanisms contributing to previously reported innocuous median nerve (MN) and noxious laser evoked (LE) responses from human SI using the Human Neocortical Neurosolver (HNN) modeling software tool specifically designed for biophysically principled interpretation of M/EEG signals (Neymotin et al., 2020; https://hnn.brown.edu). We analyzed two datasets consisting of MEG and EEG recordings during MN and laser stimulation, respectively. Oriented current dipole inverse solution analysis revealed robust features of the respective SI MN and LE responses that were conserved across recordings from prior literature. The timing and orientation of peaks in the SI responses showed that the positive orientation of the first peak in the MN response at ~20 ms corresponds to outward deep-to-superficial electrical current flow, while the first LE response deflection occurs later at ~170 ms and is oriented in the opposite direction. Simulating the circuit-level mechanisms accounting for

these and later peaks with HNN's detailed laminar cortical column model revealed that the MN response can be simulated with a sequence of layer-specific exogenous excitatory feedforward and feedback synaptic drives. This sequence was similar to that previously reported for SI tactile evoked responses (Jones et al., 2007; Neymotin et al., 2020), with the important addition of an early excitatory feedback input to superficial layers at ~30 ms that helped generate the prominent first negative deflection in the MN response. To accurately account for the LE response, a burst of repetitive gamma-frequency (~40 Hz) supragranular feedback drives followed by a later feedforward drive was required. These novel model-derived predictions are consistent with prior reports of evoked gamma oscillations from LE responses in both humans and animals (Heid et al., 2020; Hu and Iannetti, 2019; Yue et al., 2020) and make several multiscale predictions about the laminar circuit mechanisms underlying temporal and spectral features in SI MN and LE responses. These findings can guide future studies that test and expand our predictions with invasive recordings or other imaging modalities and may ultimately help with the development of treatments for somatosensory disorders including chronic pain.

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Poster

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Fondation Bertarelli

Title: The cortical representation of touch reflects extensive subcortical integration of signals from mechanoreceptor subtypes

Authors: *A. J. EMANUEL[1], B. P. LEHNERT[1], S. PANZERI[2], C. D. HARVEY[1], D. D. GINTY[1];
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Abstract: How are tactile signals represented and processed within the nervous system? Current models to explain how signals emanating from cutaneous mechanoreceptors generate representations of touch are based on comparisons of the tactile responses of mechanoreceptor subtypes and neurons in somatosensory cortex. We used mouse genetic manipulations to causally investigate the contributions of peripheral mechanoreceptor subtypes to cortical responses to touch. Cortical neurons exhibited remarkably homogeneous and transient responses to indentation applied to the glabrous skin of the paws and these responses resembled those of Aβ rapidly adapting low-threshold mechanoreceptors (RA-LTMRs). Concurrent disruption of signals from both Aβ RA-LTMRs and Aβ slowly adapting (SA-) LTMRs eliminated cortical responses to light indentation forces. However, disruption of signals from either subtype alone caused opposite shifts in cortical sensitivity but otherwise largely unaltered tactile responses, indicating that both subtypes contribute to normal cortical responses. Selective optogenetic activation of single action potentials in Aβ RA-LTMRs or Aβ SA-LTMRs drove low-latency responses in the majority of mechanically sensitive cortical neurons. Likewise, most somatosensory thalamic neurons were also driven by optogenetic activation of Aβ RA-LTMRs or Aβ SA-LTMRs. These findings support a new model in which signals from physiologically distinct mechanoreceptor subtypes are extensively integrated and transformed within the subcortical somatosensory system to generate cortical representations of touch.

Disclosures: A.J. Emanuel: None. **B.P. Lehnert:** None. **S. Panzeri:** None. **C.D. Harvey:** None. **D.D. Ginty:** None.

Poster

P410. Somatosensory Cortical Processing

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Topic: D.04. Somatosensation – Touch

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Title: Expectation shapes neuronal response along the somatosensory pathway

Authors: G. ENGLISH[1], N. GHASEMI NEJAD[1], M. F. YANIK[2], W. VON DER BEHRENS[1]; [1]ETH Zurich, Zurich, Switzerland; [2]ETH, Zuerich, Switzerland

Abstract: Predictive coding theories hint at the existence of mechanisms that allow agents to more optimally navigate complex environments. However, experimental evidence elucidating the details of the precise neural circuitry underlying predictive coding remains limited. To address this gap, we recorded extracellular neural activity at different stations of the somatosensory hierarchy while delivering novel sensory stimulation protocols aimed at isolating the effect of precision-weighted expectations. We find that the magnitude of the neural response

across the cortical layers of both the primary and secondary somatosensory cortices scales with the error size of an interruption to a precision-weighted expectation. When the sensory background supports precise expectations about forthcoming stimuli, the local field potentials evoked by unexpected stimuli in the primary somatosensory cortex signal also the specific type of error that was encountered. Additionally, unexpected stimuli induced increased power in the beta and gamma-frequency bands in the supragranular and granular layers of the primary somatosensory cortex to subsequent stimuli, potentially representing a prediction update process that is active for up to 700 ms after the original prediction error. After the application of the NMDA antagonist Ketamine, the variation in neural response between unexpected stimuli with different precision-weighted expectations is reduced, in particular in the secondary somatosensory cortex, indicating a role for NMDA receptors in the learning of complex statistical regularities unfolding over time. Overall, our results point towards the specific cortical instances and the potential mechanisms that may be critical for predictive coding.

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Poster

P410. Somatosensory Cortical Processing

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Program #/Poster #: P410.07

Topic: D.04. Somatosensation – Touch

Title: Microstimulation and optogenetics: A combined stimulation strategy

Authors: *H. BAEZ[1], M. DUHAIN[2], M. GOMEZ-RAMIREZ[3];
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Abstract: The ability to selectively control neural activity is key to determine the role of brain areas in perception and action. Intracortical Microstimulation (ICMS) has been a leading method for upregulating neural activity in vivo. Although ICMS has led to major discoveries, the widespread effects of the electrical current evoked by ICMS limit selectively control neural ensembles in areas lacking topographical structure. Further, ICMS only provides upregulation of neural activity, and it is indiscriminate about the stimulated cell type(s). In contrast, optogenetics provides spatially-localized and rapid control of cell-type specific (i.e., excitatory and/or inhibitory) neural circuits via light. However, behavioral effects of optogenetics in large-brain animals (e.g., monkeys) have been mixed, likely because light fails to activate a sufficient portion of the neural circuit mediating the behavior. Increasing light intensity can lead to tissue damage, and less specificity of the stimulated ensemble. To overcome these limitations, we propose a novel method that combines ICMS and optogenetics to provide refined stimulation of non-topographically aligned circuits. As proof of principle, we developed a biologically-

principled model to determine the effects of ICMS and/or optogenetics on different cell populations in area 1 of somatosensory cortex. As expected, we observed that neighboring neurons to the stimulation site were activated more strongly by ICMS or optical light. We further found that ICMS activated distant neurons with axons oriented towards the stimulated site. This effect was obviated with optogenetics by targeting opsin expression to the soma. Importantly, optimization techniques showed that ICMS+optical stimulation was the most effective method for selectively activating non-topographically aligned cell ensembles. Combined ICMS and optogenetics requires less stimulation power, providing a more targeted and less-damaging stimulation strategy as compared to each method alone. These data indicate that combined ICMS+optogenetics is a feasible and efficient approach for studying the role neural circuits in perception and action.

Disclosures: H. Baez: None. **M. Duhain:** None. **M. Gomez-Ramirez:** None.

Poster

P417. Stimulus Features, Responses, Coding: Non-Human Primates

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Program #/Poster #: P417.01

Topic: D.04. Somatosensation – Touch

Support: NIH F31 NS096952
R01 NS095162
R35 NS122333

Title: Neural computations in the cuneate nucleus of primates

Authors: *C. M. GREENSPON[1], A. K. SURESH[1], Q. HE[2], J. M. ROSENOW[3], L. E. MILLER[4], S. J. BENSMAIA[2];
[2]Dept. of Organismal Biol. and Anat., [1]Univ. of Chicago, Chicago, IL; [3]Neurosurg., [4]Dept. of Physiol., Northwestern Univ., Chicago, IL

Abstract: In primates, the coding of tactile information has been studied extensively in the peripheral nerves and primary somatosensory cortex (S1). These studies reveal that the responses of individual S1 neurons reflect convergent input from multiple classes of nerve fibers, and that this input is subjected to computations. Indeed, cortical neurons exhibit spatiotemporal response properties that confer to them an idiosyncratic selectivity for specific spatiotemporal features in their inputs. The classic textbook view is that these response properties reflect computations that are effected in cortex, implying that sensory signals are not meaningfully processed in the two intervening structures - the cuneate nucleus (CN) and the thalamus. However, CN responses have not been investigated using stimuli whose representation in the nerve had been quantitatively characterized, which precludes a quantitative analysis of how tactile signals are

transformed in CN. To fill this gap, we recorded the responses evoked in individual CN neurons to a battery of tactile stimuli that have been extensively used to characterize the response properties of tactile nerve fibers and cortical neurons, including skin indentations, vibrations, random dot patterns, and scanned edges. We then compared CN responses to their upstream and downstream counterparts to assess the degree to which tactile signals are transformed in CN. We found that the responses of CN neurons were more similar to their S1 counterparts than they were to their inputs: CN neurons receive input from multiple sub-modalities, have spatially complex receptive fields, and exhibit selectivity for geometric features. Thus, the CN is not a simple relay station and instead plays a key role in the processing of tactile information.

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Poster

P417. Stimulus Features, Responses, Coding: Non-Human Primates

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

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Topic: D.04. Somatosensation – Touch

Support: R01 NS 095162

Title: Movement gating of tactile signals in the cuneate nucleus of primates

Authors: *Q. HE[1], C. VERSTEEG[3], A. K. SURESH[1], L. E. MILLER[4], S. J. BENSMAIA[2]; [2]Dept. of Organismal Biol. and Anat., [1]Univ. of Chicago, Chicago, IL; [3]Biomed. Engin., [4]Dept. of Physiol., Northwestern Univ., Chicago, IL

Abstract: A key function of sensory systems is to filter sensory input to enhance behaviorally relevant signals and suppress irrelevant ones. An instance of this filtering is the documented movement gating of cutaneous signals: During movement, the cutaneous signals triggered by the resulting skin compression and stretch are suppressed, resulting in decreased cutaneous sensitivity, measured psychophysically. The cuneate nucleus (CN) receives projections from sensorimotor cortex, which have been shown to modulate the transmission of field potentials reflecting afferent input. However, the role of this top down modulation in movement gating has not been demonstrated at the single-cell level. To fill this gap, we had rhesus macaques perform a reach-to-grasp task while we monitored neural activity in CN using chronically implanted electrode arrays. At the same time, we measured hand kinematics using a camera-based motion tracking system, and the grasp forces exerted on the manipulandum using a multi-axis sensor. Throughout the task, we delivered tactile stimuli placed over the receptive fields (RFs) of CN neurons. From the resulting behavioral and neuronal data, we assessed the degree to which CN responses to an unchanging stimulus were modulated during different task epochs. We found that

the responses of almost all cutaneous neurons with RFs on the proximal limb were suppressed during movement. This was not the case for neurons with cutaneous RFs on the hand, for proprioceptive neurons, or when movements were imposed on the arm rather than actively generated. We conclude that the movement gating of cutaneous signals is already present in the CN and is triggered by volitional movement signals, likely stemming from the cortex.

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Poster

P417. Stimulus Features, Responses, Coding: Non-Human Primates

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Topic: D.04. Somatosensation – Touch

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ANR-11-LABX-0042
ANR-11-IDEX-0007

Title: Diversity of functional connectivity patterns between the fundus of the intraparietal sulcus and the primary somatosensory area S1 in awake macaque monkeys

Authors: *W.-A. SHENG[1], S. CLAVAGNIER[1], M. FROESEL[1], W. VANDUFFEL[2,3], S. BEN HAMED[1];
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Abstract: The functions of a cortical area are largely determined by its extrinsic connections with other brain regions. Cortical networks can be mapped anatomically with tracer injections. More recently, the correlation of BOLD fMRI signals in a resting brain has been used to reveal functional associations between regions.
The intraparietal sulcus (IPS) is involved in multisensory integration, and the ventral intraparietal area (VIP) in the IPS fundus responds to visual, vestibular, tactile and auditory signals, with tactile receptive fields over-representing the face. However, anatomical projections from areas 1, 2 and 3a (jointly defining primary somatosensory area S1) to VIP are mainly from the hand and the face/hand boundary representations. Mapping functional connections between fundal IPS and S1 reveals a complex organization of face and hand connectivity fields between VIP and S1, shedding a new light on VIP functions.
To identify sparse or local connections between VIP and S1, we proceeded as follows. Awake resting state fMRI data from 10 monkeys, both hemispheres was co-registered in a common space (voxels of 1mm3). Posterior-anterior IPS to lateral-medial S1 connectivity patterns were

identified using single-voxel seeds running along the posterior-anterior axis of IPS fundus. At the group level, these patterns reveal hand-face borders in area 3a, 3b and 2 matching the IPS tip, as described in the literature. This implies that the awake resting state connectivity measurement is a potential alternative when localizers are unavailable. Based on these borders, we observed different IPS-S1 face and hand connectivity fields: according to the IPS seed positions, the face pattern has one anterior and one posterior peak, while the hand pattern is dominated by the anterior peak.
At a finer grain, face connectivity fields among individuals are diverse. There are slightly more double-peak face than single-peak face patterns in area 3a and 3b, while the former is the dominant pattern in area 2. The single-peak patterns can be further divided into three subcategories based on their peak IPS seed position and the majority has its peak seed position in the anterior IPS.
Overall, this study indicates that fundal IPS somatosensory activities arise from heterogeneous connectivity patterns with S1, predicting distinct overarching behavioral functions.

Disclosures: W. Sheng: None. **S. Clavagnier:** None. **M. Froesel:** None. **W. Vanduffel:** None. **S. Ben Hamed:** None.

Poster

P417. Stimulus Features, Responses, Coding: Non-Human Primates

Location: Virtual Only

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Program #/Poster #: P417.04

Topic: D.04. Somatosensation – Touch

Title: Analysis of the firing rate of layer V sensory neurons as an indicator for cryoanesthesia and thermosensation

Authors: *C. Y. LEE[1], S. R. NASON[2], A. J. BULLARD[2], C. A. CHESTEK[2], K. P. PIPE[1]; [1]Mechanical Engin., Univ. of Michigan, Ann Arbor, Ann Arbor, MI; [2]Biomed. Engin., Univ. of Michigan, Ann Arbor, MI

Abstract: Neural mechanisms for cold sensation and cold-induced anesthesia are important for developing non-opioid local analgesics and understanding of neuropathic diseases. Yet, coding of these mechanisms in the sensory cortex is not widely investigated. Prior studies have reported human subject scoring to imprecise cooling methods (e.g. ice, cold air) and lack quantitative data on CNS behavior to broad ranges of cold temperatures (*T*). The present work aims to identify the dynamic neural response to cutaneous thermal and mechanical stimuli using non-human primate models (rhesus, male, n=2). Animals were implanted with Utah arrays (Blackrock Microsystems) in the hand area of the primary sensory cortex, containing neurons receptive to both thermal and mechanical stimuli. The cooling device used is a custom feedback-controlled thermoelectric probe (±0.001 °C, ≤5 °C/sec, dia. 0.25 in) that allows simultaneous or independent blunt poking

and cooling of a target area. The following measurements were performed with IACUC approval over the course of 6 months: (1) baseline neural response to the environment (20°C) while the animal is in a seated and restrained position (baseline data acquired prior to all trials, with position and climate maintained), (2) response to constant *T* (-5, 0, 5, 10, 35 °C), (3) response to innocuous jabs on skin cooled to a constant *T*, with stimuli spaced irregularly over a trial period of 60-90 secs to gauge the effects of cooling on poking sensation while reducing chances of adaptation or allodynia, (4) immediately following the conclusion of (3), raising *T* to 35 °C (monkey normal skin *T*) with continued jabs, to test for adaptation vs. numbing from cold, and (5) response to rapid changes in *T* while maintaining contact. Preliminary findings show a steady decline in firing rate from a series of pokes during extended cooling, with a slight increase in firing rate once the probe *T* was raised to 35 °C. Surges in firing rate were noted at the initial contact and removal of the cold probe, and the firing rate at initial contact was consistently less than the first poking stimulus, except when *T* = 35 °C. Overall, firing rate in individual neurons as a function of *T*, contact sensation (pressure from cold tip), and time is a potential indicator of cryoanesthesia, but the magnitude of direct responses is minimal; there is a need to amplify responses to only cold and poking, and reduce other activity. Future work will include measuring responses to dynamic *T*, establishing a model that predicts neural response to cold and mechanical stimuli, and engaging human subject testing to pair human sensation scores to monkey neural data.

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Poster

P417. Stimulus Features, Responses, Coding: Non-Human Primates

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Topic: D.04. Somatosensation – Touch

Support: NS34086
NS18787

Title: Multi-finger representations in primary somatosensory cortex: Revising our understanding of the spatio-temporal integration properties of area 3b

Authors: *M. GOMEZ-RAMIREZ[1], S. S. HSIAO[2], N. TRZCINSKI[3];
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Abstract: The prevailing model of somatosensory processing indicates that area 3b neurons have receptive fields (RFs) that are confined to a pad within an individual finger. Based on this RF

property and its downstream projections, area 3b is thought to primarily serve as a cortical relay site encoding tactile features comprising an object. As such, object sensing and manipulation (i.e., haptics) is believed to emerge in area 2 and beyond, which have cells with complex spatio-temporal RFs that integrate inputs across multiple fingers. However, our knowledge of 3b cells' RF size is primarily derived from anesthetized studies in non-human primates. The findings from these studies are limited because anesthesia can significantly impact the strength of neural activation, and undermine cells' responses to non-preferred digit stimulation. Similarly, the vast majority of area 3b studies in awake-behaving animals are limited because researchers failed to systematically stimulate each finger. In these awake-behaving experiments (as well as anesthetized studies), researchers typically localize the finger with the maximum response using unreliable hand held probes, and then use motorized devices to systematically stimulate the preferred finger only (i.e., the finger evoking the greatest response). Thus, the field fails to have a detailed understanding of area 3b's classical and non-classical spatio-temporal RF properties across the fingers in the hand. Here, we systematically mapped the RFs of area 3b cells by presenting bars of different orientations to digits 2, 3, 4, and 5 in awake-behaving non-human primates. Contrary to the leading tenet, we found that the vast majority of cells in area 3b have classical RFs that extend to multiple digits. We further observed that multidigit RFs evolved as a function of time, indicating that they are driven by feedback cortical mechanisms. We also found that differences in orientation preference across fingers ranged between 22.5 and 67.5 degrees, an ideal range for encoding objects with curved shapes. Taken together, these data show that area 3b plays a more integral role in generating tactile object representations than previously thought.

Disclosures: M. Gomez-Ramirez: None. **S.S. Hsiao:** None. **N. Trzcinski:** None.

Poster

P417. Stimulus Features, Responses, Coding: Non-Human Primates

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Title: Self-awareness from multisensory integration: neuronal evidence of mirror self-recognition in the Macaque SII

Authors: *R. BRETAS[1], M. TAOKA[1], S. HIHARA[1], A. CLEEREMANS[2], A. IRIKI[1];
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Japan; [2]Consciousness, Cognition, and Computation Group (CO3), ULB Neurosci. Institute, Univ. Libre de Bruxelles, Brussels, Belgium

Abstract: Multisensory integration in the parietal cortex has been proposed as one of the mechanisms supporting bodily self-awareness. The responses of multimodal neurons in the secondary somatosensory cortex (SII) of macaque to both somatosensory and visual stimuli within the animal's peripersonal space, with receptive fields sometimes as large as the entire body, seem to account for more than a pure somatosensory function. Therefore we tested in an exploratory manner whether these responses are maintained when the stimulus is seen through a mirror, indicating a possible role in merging sensory stimuli in a self-body schema, a foundation for self-awareness.

For the experiment one female Japanese macaque was habituated to a mirror until developing the ability of mirror self-recognition. The animal was then stimulated by touch and/or movement within her peripersonal space by the experimenter, objects, or by self movement/touch while sitting in front of a full-body mirror. During the task, single neuron activity was observed bilaterally from the SII proper and parietal ventral area, with visual receptive fields being detected in 96 neurons. Of these visually responsive neurons, 31% were found to respond to stimuli seen through the mirror but out of direct view from the animal, 6% to stimulation to the mirror directly (e.g. experimenter touching the animal image in the mirror), 7% responded to the monkey touching her own body, and 5% to touching another body or object. Neurons were further grouped into self (39%) or non-self (11%) related. Other neurons showed nonspecific responses. All responses were qualitatively accessed and cross checked by two experimenters, with 12 neurons being recorded alongside a video of the task.

Our findings provide neuronal evidence of mirror self-recognition in macaques, even as an acquired skill when the animal did not show innate mirror self-recognition, corroborating behavioral indicators such as the mark test. Single cell activity in the SII, besides responding to both visual and somatosensory stimuli, could also consistently discriminate between the self and non-self. As one of the first areas in which whole-body multisensory information is integrated, the SII could be situated as a rudimentary but fundamental precursor to self-awareness. Taken together, these results place the SII in an ideal position to process a body-in-the-world map, incorporating sensory information of the external world and the self as a whole, indivisible body.

Disclosures: R. Bretas: None. **M. Taoka:** None. **S. Hihara:** None. **A. Cleeremans:** None. **A. Iriki:** None.

Poster

P417. Stimulus Features, Responses, Coding: Non-Human Primates

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P417.07

Topic: D.04. Somatosensation – Touch

Support: NIH Grant NS18787
NSF Grant 1150209

Title: Temporal Coding Along the Somatosensory Neuroaxis

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Abstract: The tactile perception of vibration and texture has been shown to rely on precise and repeatable temporal spiking patterns in both the somatosensory nerves and somatosensory cortex. While the prevalence and precision of temporal information has been shown to differ in the nerves and in the brain, these have not been systematically compared. Furthermore, spike timing remains to be characterized at intermediate stages of processing along the medial lemniscal pathway, namely the cuneate nucleus and thalamus, and in higher order cortical fields, including secondary somatosensory cortex (S2) and parietal ventral area (PV). In the present study, we analyzed neuronal responses to skin vibrations and textures at each at each stage of processing along the neuraxis of rhesus macaques, from nerve through S2/PV. First, we systematically characterized the precision of temporal spiking patterns using a variety of previously developed metrics - including vector strength and harmonic ratio - and quantified the informativeness of spike timing, by comparing the performance of decoders that take timing into account to that of decoders that do not. Second, we developed neuronal models - informed by known features of somatosensory neurons and their interconnections - to evaluate the conditions under which precise spike timing can propagate through multiple synapses, given realistic levels of convergence and synaptic noise. These models replicated the reduction in the prevalence of temporal coding at successive stages of processing. Third, we assessed the degree to which stimulus information carried in temporal spiking sequences at one stage is reflected in the firing rates at the following stage. Indeed, we have shown that cortical firing rates are far more informative than are their peripheral counterparts, implying a conversion from temporal to rate codes. We sought to characterize this transformation along each stage. These analyses informed models - again inspired by known properties of neurons along the somatosensory neuraxis - that demonstrate physiologically plausible mechanisms for the observed temporal to rate transformation. Together these analytical and modeling efforts shed light on how information is carried in temporal spiking patterns and how this information is transformed as it ascends the neuraxis.

Disclosures: O.J. Lutz: None. **K.H. Long:** None. **S.J. Bensmaia:** None.

Poster

P417. Stimulus Features, Responses, Coding: Non-Human Primates

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P417.08

Topic: D.04. Somatosensation – Touch

Title: Development of a non-human primate experimental setup to investigate the neural basis of visuo-haptic 3D shape perception

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Abstract: Accurate shape perception is crucial to identifying and manipulating objects in our environment. Among our senses, touch and vision are unique in that they both convey 3D geometric shape. In order to recognize shapes across sensory modalities, the brain must transform haptic and visual sensory inputs into modality-independent representations. How the brain generates modality-independent representations of shape remains unknown. Human fMRI studies have identified the lateral occipital complex (LOC) as a key area for visuo-haptic integration, yet the low spatial resolution of fMRI has failed to reveal the neural code used for representing object shape. To address this knowledge gap, we propose to study the representations of haptic and visual shapes in macaque inferotemporal cortex (IT), the putative homologue of human LOC. Specifically, we will test the hypothesis that IT utilizes the same coding scheme to represent haptic and visual shapes. To this end, we have developed an experimental setup in which macaques are presented with both haptic and visual stimuli. In this setup, a robotic arm delivers 3D-printed haptic objects within reach of the macaques, and a visual display shows images of visual objects. By matching the position and size of the haptic and visual objects, we will be able to compare shape encoding between touch and vision within the same reference frame. Furthermore, we have developed a comprehensive stimulus set to probe the neural representation of surface curvatures and medial-axis structure. Future studies will combine this experimental setup and stimulus set with neural recordings to identify visuo-haptic brain regions and explore the neural basis of visuo-haptic shape representations in IT.

Disclosures: W.G. Snider: None. **C.E. Connor:** None. **D.H. O'Connor:** None.

Poster

P417. Stimulus Features, Responses, Coding: Non-Human Primates

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Program #/Poster #: P417.09

Topic: D.04. Somatosensation – Touch

Support: NINDS Grant R01NS101325
NINDS Grant F31NS110402

Title: Texture coding in higher order somatosensory cortices

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Abstract: Our sense of touch confers to us a sensitivity to texture that spans six orders in magnitude in spatial scale, from tens of nanometers to tens of millimeters. In early stages of processing, from the nerve through somatosensory cortex (SC) - including Brodmann's areas 3b, 1, and 2 - individual neurons respond promiscuously to most textures and, as a population, carry a large shared signal that drives the perceived roughness, the dominant perceptual dimension of texture. Nothing is known about how natural textures are encoded in higher order cortices, including secondary somatosensory cortex (S2) and parietal ventral cortex (PV). To fill this gap, we monitored single-unit activity in S2/PV as non-human primates performed a texture discrimination task, in which they indicated whether pairs of naturalistic textures were the same or different. We then characterized the neuronal responses at the single-cell and population levels and compared these to their counterparts in SC. First, we found that only a small fraction of S2/PV neurons respond to textures, in contrast to SC neurons, which typically do. Second, even neurons in S2/PV that do respond to textures only respond to a small subset of them, in contrast to SC neurons, which typically respond to most textures. As a result, the representation in S2/PV is higher dimensional than is its counterpart in SC. Finally, S2/PV neurons carry cognitive signals about task variables such as trial type (same, different) and the animals' eventual decision, unlike their SC counterparts. The present study constitutes the first attempt to characterize the sensory representations in S2/PV using a complex, naturalistic stimulus set.

Disclosures: K.H. Long: None. **A.M. Van Driesche:** None. **S.J. Bensmaia:** None.

Poster

P417. Stimulus Features, Responses, Coding: Non-Human Primates

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Program #/Poster #: P417.10

Topic: D.04. Somatosensation – Touch

Support: NIA Grant R01AG069227
NIDCR Grant R01DE027236

Title: Sensory loss affects multi-region neural network dynamics in primate sensorimotor cortex

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Abstract: The number of people living with Alzheimer's Disease (AD) doubles every five years after the age of sixty-five. In addition to cognitive impairment, most patients with AD have difficulties in chewing and swallowing. Pathological changes in large-scale neural activity and connectivity in the orofacial sensorimotor cortex (OSMcx) beyond those expected in healthy aging may serve as neural biomarkers for AD. To identify changes in cortical activity due to diminished sensation, we evaluated the effects of absent tactile sensation in the mouth as a young and healthy non-human primate (*Macaca mulatta*) engaged in feeding behavior. Local anesthetics to sensory branches of the trigeminal nerve were used to block tactile inputs from the palate, teeth, and tongue. We recorded neuronal activity from multiple regions of OSMcx (area 3a/3b, 1/2, rostral and caudal M1) simultaneously with 3D tracking of tongue and jaw kinematics using X-Ray Reconstruction of Moving Morphology (XROMM). Neurons in OSMcx modulated their firing rates to the rhythmic tongue and jaw movements during chewing. In the absence of tactile sensation, we observed a ritardando of firing rate oscillations consistent with longer chew cycles. To evaluate the dynamics of multi-region functional connectivity, we used a data-constrained recurrent neural network (RNN) model to infer the directed interactions within and across regions (CURBD, Perich et al., 2020). By training two hundred RNNs independently, we found CURBD to show differential changes in neural network interactions following nerve block. In the absence of tactile inputs, out-degree inhibition from all regions increased significantly ($p<0.01$), except area 1/2 which significantly decreased inhibition both within area and to rostral M1 ($p<0.01$). Across all regions, area 3a/3b showed the highest increase in both excitation and inhibition with rostral M1 inhibition and within area excitation accounting for most of the increase. Lastly, influences to area 3a/3b increased; excitation to area 1/2 decreased while inhibition increased. The increased inhibition to area 3a/3b and 1/2 following nerve block came primarily from area 3a/3b. Overall, the results suggest that a broadening of multi-region network interactions may accompany diminished sensation in healthy aging. Further studies will include investigating these changes in healthy aged non-human primates and those with AD-like symptoms.

Disclosures: L. Sheridan: None. **J.D. Laurence-Chasen:** None. **F.I. Arce-McShane:** None.

Poster

P451. Behavioral Effects by the Vestibular and Proprioceptive Systems

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P451.01

Topic: D.06. Auditory & Vestibular Systems

Title: Leveraging Markerless Computer Vision to Screen for Circling in Mutant Mice

Authors: *O. R. STANLEY[1], A. SWAMINATHAN[1], K. E. CULLEN[1,2,3];
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Abstract: The vestibular system provides critical contributions to behavioral stability and spatial navigation. Mouse models for studying varied causes of vestibular impairment have advantages including consistent genetic strains and homology between the mouse and human vestibular systems. Mice with vestibular peripheral dysfunction commonly display an irregular locomotion pattern characterized by repetitive spinning behavior, termed "circling". Screening mice for circling can help identify mutants of interest, which may then be more extensively tested for vestibular dysfunction. However, current methods for definitively identifying animals exhibiting this behavior are limited by inconsistent ethological definitions in the literature and the expense of specialized animal-tracking tools. Here, we present a video analysis pipeline we have developed to identify instances of circling in the paths of freely exploring mice tracked using the DeepLabCut (DLC) Python package. We compare the labeling performance of several DLC neural network models trained on different amounts of manually labeled data. In addition, for each of those neural networks, we compare the performance of multiple detection algorithms as measured by their ability to match human-consensus labeling. These detection methods incorporate different geometric or temporal features of the labeled mice paths, such as the major and minor axes of putative circling instances or how long those instances take. In general, we find that surprisingly small datasets enable behavioral detection as effective as our largest dataset. In addition, we find that in our specific use case, incorporating temporal features and allowing small deviations from a strict definition enable better overall performance in detecting circling behavior. Overall, our results indicate our tool provides a convenient, quantitative screening method for mouse models of vestibular dysfunction.

Disclosures: O.R. Stanley: None. **A. Swaminathan:** None. **K.E. Cullen:** None.

Poster

P451. Behavioral Effects by the Vestibular and Proprioceptive Systems

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P451.02

Topic: D.06. Auditory & Vestibular Systems

Support: CIHR (Canadian Institutes of Health Research): Neurosciences, Mental Health and Addiction

Title: Optogenetics in non-human primates: Targeted expression in key central vestibular system brain regions

Authors: *T. JIMENEZ[1], M. ST-LOUIS[2], A. RIBEIRO-DA-SILVA[2], J. CARRIOT[1], M. J. CHACRON[1];
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Abstract: The vestibular system is essential to ensure that self-motion information is processed, giving rise to vital reflexes alongside perception. Here we investigated the feasibility of using viral vectors to express opsins within neurons in central vestibular pathways of non-human primates (NHPs). Importantly, we chose our vector constructs while considering the functional relationships and anatomical differences between our regions of interest, which span from the brainstem to the cerebral cortex. Using optogenetics would allow for fine manipulation of vestibular pathways *in vivo*, increasing the potential of our understanding of how vestibular neurons represent sensory input. Our study induced optogenetic expression through injection of two opsin adenoviruses (serotype: AAV9) specific for different central vestibular brain regions of a cynomolgus macaque: the vestibular nuclei (VN), situated in the brainstem, and the parietoinsular vestibular cortex (PIVC). Specifically, in VN, we injected a virus with a promoter specific to neurons (hSyn) containing the gene from the red-shifted opsin Jaws. In PIVC, we injected a virus with a promoter specific to cortical GABAergic interneurons (mDlx) containing the gene for Channelrhodopsin-2. Using immunohistochemical techniques, fluorescence microscopy, and image processing, we evaluated and quantified optogenetic expression. Our results demonstrate robust specificity and sensitivity of our chosen viruses to their respective target regions and neurons. With the histological confirmation of optogenetic expression, our study will set in motion the application of optogenetic techniques to future NHP studies investigating coding and information feedback along central vestibular pathways.

Disclosures: T. Jimenez: None. **M. St-Louis:** None. **A. Ribeiro-da-Silva:** None. **M.J. Chacron:** None.

Poster

P451. Behavioral Effects by the Vestibular and Proprioceptive Systems

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Program #/Poster #: P451.03

Topic: D.06. Auditory & Vestibular Systems

Support: NIH Grant 90079498

Title: Vestibular loss alters postural responses to platform tilts in a nonhuman primate model

Authors: *O. M. E. LEAVITT, K. E. CULLEN;
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Abstract: Individuals with bilateral vestibular loss (BVL) experience dizziness, postural instability, and gait ataxia even following rehabilitation. Vestibular prostheses aim to replace vestibular function in patients with damage to the peripheral vestibular organs by bypassing the impaired neuroepithelium and stimulating vestibular afferents. In animal models such as nonhuman primates, the vestibular prosthesis can also be used experimentally to apply direct perturbations to the vestibular system to further our understanding of how the brain processes vestibular signals during natural behaviors. In the present study, we leveraged recent developments in vestibular prostheses to improve our understanding the contribution of the vestibular system to postural control.

We first established that postural responses in normal nonhuman primates are similar to what has been reported in humans. Two rhesus monkeys, one normal and one with BVL, were habituated to a chamber mounted on a hexapod motion platform. Animals were trained to place one limb each on four force plates, head position was tracked with an IMU, and poses were estimated from video via DeepLabCut. Ramp-and-hold transient platform tilts of 10 degrees were delivered at 3 accelerations and 3 velocities in pitch and roll. Both animals began actively applying limb forces in response to platform motion within 60 ms of motion onset, and the head began to move 80 ms after onset. Thus, as in humans an initial displacement of the support surface below the subject is followed by an automatic, active response returning the subject to a stable posture that orients the head vertically with respect to gravity by tilting against the direction of platform tilt.

Second, we show that the contribution of the vestibular system is similar in monkeys and humans. In the BVL animal, limb forces and head motion were the reverse of the normal animal's. The BVL animal's head motion responses were also hypometric to normal, suggesting reluctance to move the head. This mirrors studies of postural responses in human subjects, where similarly in contrast to normal subjects, patients with BVL tend to actively destabilize posture during platform tilts by tilting with the direction of platform tilt.

Taken together our results suggest that monkeys have postural responses similar to other animals including humans, and abnormal postural responses caused by BVL affirm the necessity of vestibular signals for maintaining upright posture. Notably, this indicates that replacement of these signals via stimulation of the vestibular system by prosthesis has the potential to improve balance and allow further exploration of the mechanisms of postural control.

Disclosures: O.M.E. Leavitt: None. **K.E. Cullen:** None.

Poster

P451. Behavioral Effects by the Vestibular and Proprioceptive Systems

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P451.04

Topic: D.06. Auditory & Vestibular Systems

Support: faculty research grant from Yonsei University College of Medicine (6- 2018-0107)

Title: 3d virtual enhancement as a potential confounding factor in computerized dynamic posturography

Authors: *E. SON, S. LEE, Y. KO, J. KIM;
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Abstract: The sensory organization test (SOT) of a computerized dynamic posturography (CDP) is widely used to investigate a subject's use of the sensory systems for postural control. The latest technology utilizes 3D virtual reality environment to achieve visual reference sway during SOT, compared to conventional CDP which uses sagittal plane movement. In this study, we aimed to evaluate the effect of 3D virtual reality on SOT measurements.A retrospective study included CDP data of 30 consecutive dizzy patients with normal composite scores for their age groups. Comprehensive review of medical history and vestibular function test results were performed with the focus individual performance in VR conditions. Inter-trial differences were analyzed in each SOT condition. Comparison of vaiance among the CDP conditions were performed. Interestingly, condition 3 using VR visual sway showed increased inter-trial variability compared to conditions 1 and 2. 3D virtual reality technology can be a versatile tool for visual input manipulation compared to conventional CDP hardware that provides only sagittal plane movements. Yet, addition of 3D virtual enhancement may be a potential confounding factor in SOT, and further studies are needed to optimize its clinical implementation.

Disclosures: E. Son: None. **S. Lee:** None. **Y. Ko:** None. **J. Kim:** None.

Poster

P451. Behavioral Effects by the Vestibular and Proprioceptive Systems

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P451.05

Topic: D.06. Auditory & Vestibular Systems

Support: NIH R01 DC2390

Title: Vestibular prosthesis stimulation linearly evokes eye movements over the stimulation frequency range above the maximum endogenous afferent firing rate: implications for prosthesis dynamic range

Authors: *K. WIBOONSAKSAKUL[1], C. C. DELLA SANTINA[2], K. E. CULLEN[1];
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Abstract: Bilateral vestibulopathy can lead to debilitating visual and postural instability in addition to constant dizziness. One approach to improve the quality of life in this patient population is a vestibular prosthesis that senses head rotation and transforms it into vestibular afferent stimulation, substituting for the damaged periphery. Early results from clinical trials, while encouraging, have shown only partial functional improvements. Specifically, the evoked vestibulo-ocular reflex (VOR) fails to fully stabilize the gaze (i.e., gain < 1). Current strategies to improve the VOR gain have notable downsides; increasing stimulation current level can lead to non-specific activation, while modulating the stimulation rate more strongly for a given head velocity can lead to earlier saturation and narrower dynamic range. One key parameter that remains uninvestigated, however, is the maximum stimulation rate. This is typically set at the maximum endogenous afferent firing rate (~350 Hz) based on the assumption that each stimulation pulse will reliably evoke a spike in the afferents. Accordingly, here we investigated the linearity of prosthesis-evoked VOR eye movements across a large stimulation frequency range. We first re-analyze a previous dataset from two monkeys implanted with the prosthesis. Though the dataset included saturations in the VOR gain, by plotting the maximum evoked eye velocity as a function of maximum stimulation rate, a linear relationship was observed up to 500 Hz (maximum in the dataset). In one monkey, we further investigated this linear range by acutely stimulating the afferents up to 1200 Hz and found a linear relationship up to 800 Hz. These results indicate that the observed saturations in VOR gain did not arise from a plateau in afferent firing rate, but rather resulted from the sigmoidal nonlinearity within the velocity-to-simulation-rate mapping that assumed a 1:1 efficacy between stimulation rate and afferent spiking. Thus, finally, we examined this efficacy assumption using a model of the VOR pathway to predict eye velocity based on stimulation rate. The model yielded accurate predictions only when stimulation efficacy was reduced to 25%, a value similar to that from afferent recordings during stimulation previously published by our group. Taken together, our results suggest that, by accounting for the actual physiological efficacy of afferents responses, the vestibular prosthesis can better utilize the range of afferent firing rates. We suggest a complementary approach combining eye movement recording and modeling can be used to optimize the prosthesis gain and dynamic range for prosthesis users in the clinic.

Disclosures: K. Wiboonsaksakul: None. **C.C. Della Santina:** None. **K.E. Cullen:** None.

Poster

P451. Behavioral Effects by the Vestibular and Proprioceptive Systems

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Program #/Poster #: P451.06

Topic: D.06. Auditory & Vestibular Systems

Support: NIDCD R01-DC018287-01

Title: How vestibular damage changes velocity storage

Authors: *F. KARMALI[1], **A. MADHANI**[2], R. LEWIS[3];
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Abstract: The velocity storage mechanism is a centrally mediated mechanism that processes peripheral vestibular cues. One prominent aspect of velocity storage is the dynamic response to yaw rotation. Specifically, when the body is accelerated to constant velocity rotation, eye movement and perceived velocity decay with a time constant of circa 15-30 s for humans with no known vestibular pathology. The velocity storage time constant is a key diagnostic parameter for peripheral loss, with a range of 6-12.7 s suggesting unilateral hypofunction. However, despite its clinical utility, a mechanistic explanation underlying this important diagnostic parameter is lacking. It has been hypothesized that Bayesian optimization determines ideal velocity storage dynamics based on statistics of vestibular noise and experienced motion. Specifically, while a longer time constant would make the vestibulo-ocular reflex (VOR) more accurate over time, it has been hypothesized that it would simultaneously amplify neural *noise* (variability) and consequently, compromise vestibular precision. Thus, when computing the velocity storage time constant, the brain may be resolving a tradeoff between being accurate and reducing noise, *i.e.*, being precise. In this study, we applied a Bayesian optimal Kalman filter to determine the ideal velocity storage time constant for unilateral hypofunction. Modeling predicted velocity storage time constants that were substantially lower than for normal subjects and comparable to the diagnostic range for unilateral damage. Results suggested that even after peripheral damage, ipsilateral noise originating in the periphery or early central processing remains relevant in neurocomputations, which is important because little is known about the effects of peripheral damage on vestibular noise. Modeling also predicted that age-related hair cell loss and unilateral lesions interact when impacting velocity storage. Our findings suggest that the reduced velocity storage time constant seen clinically results from the brain optimizing velocity storage in response to changes in peripheral signal-to-noise ratios.

Disclosures: F. Karmali: None. **A. Madhani:** None. **R. Lewis:** None.

Poster

P451. Behavioral Effects by the Vestibular and Proprioceptive Systems

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P451.07

Topic: D.06. Auditory & Vestibular Systems

Title: Influence of caloric vestibular stimulation on tactile temporal processing.

Authors: *A. CÉDRAS[1], A. SHARP[2], B.-A. BACON[3], F. CHAMPOUX[1], M. MAHEU[1];
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Abstract: Influence of caloric vestibular stimulation on tactile temporal processing. Recent evidences suggest that binaural vestibular stimulation affects tactile temporal processing.However, because of the stimuli used, it remains a matter of debate whether these results are bestaccounted for by a shift in covert attention or by an asymmetrical cortical activation. Providingunilateral vestibular stimulation could help to answer this question. The objective of the presentstudy was to examine tactile temporal processing using a temporal order judgment task followingselective stimulation of the right and left vestibular system with bithermal caloric vestibularstimulation (CVS). A toatal of 24 right-handed participants received bithermal CVS either in theright ear (n=12) or the left ear (n=12). Participants held vibrators in both hands which delivered asignal temporally separated by a variable asynchrony. Participants had to report the hand wherethe vibration was perceived first. The task was performed in three different CVS conditions: 1)baseline, 2) warm CVS, and 3) cold CVS. Analysis of the logistics curves parameters - justnoticeable difference (JND) and point of subjective simultaneity (PSS) - for each participant ineach CVS conditions revealed an increase in JND greater following warm CVS. A significantincrease in JND following warm CVS was measured bilaterally. However, cold CVS increasedJND only when CVS was applied in the left ear, but not in the right ear. Finally, no influence ofCVS on PSS was observed.

Disclosures: A. Cédras: None. **A. Sharp:** None. **B. Bacon:** None. **F. Champoux:** None. **M. Maheu:** None.

Poster

P451. Behavioral Effects by the Vestibular and Proprioceptive Systems

Location: Virtual Only

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Program #/Poster #: P451.08

Topic: D.08. Visual Sensory-motor Processing

Support: NSERC 1145753

Title: Subject differences in the influence of cervical muscle vibration on neck and elbow joint position sense

Authors: S. L. THOMSON, *W. E. MCILROY;
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Abstract: Background: Altered cervical proprioception from injury, or experimental manipulation such as neck muscle vibration, has been shown to impact spatial and postural orientation perception at the neck, but also in upper limb joints. Anecdotally, individual responses to altered cervical proprioception and its impact on joint position sense appears to vary considerably, but the nature of this variability is not well described. The current study focused on advancing understanding of the variability observed between individuals in response to neck muscle vibration. Specifically, the study explored between subject differences in the association between neck muscle vibration and joint position error (JPE) measured at the neck and the elbow. **Methods:** Young healthy adults (n=14, age 24.2 ± 6 years) participated in tests of cervical and elbow joint position sense with, without, and immediately following unilateral dorsal neck muscle vibration. JPE at the neck and elbow were compared across and within participants across the 3 vibration conditions (with, without and post vibration). **Results:** Absolute cervical JPE ($p<.0001$) and elbow JPE ($p<.0001$) increased significantly under vibration conditions across participants. Within participants, under no vibration conditions, there were moderate to strong positive relationships in JPE mean and variability (SD) during both the cervical ($r= .63$, $p=.02$) and elbow repositioning tasks ($r=.87$, $p<.0001$). There was a moderate negative relationship between JPE and the impact of vibration on JPE during the cervical ($r=-.57$, $p=.03$), but not the elbow ($r=-.17$, $p=.57$) repositioning task. Within participants there were no significant associations between cervical JPE and elbow JPE performance both with ($r=.15$, $p=.63$) and without ($r=-.24$, $p=.45$) neck vibration. **Conclusions:** The current study revealed that while neck vibration significantly impacts both cervical and elbow JPE across participants, the impact of vibration on JPE was not consistent within individuals across different joints (neck vs elbow). Importantly, the association between cervical JPE and the impact of vibration does suggest that individuals with better cervical joint position sense may be impacted to a greater extent when cervical proprioception is altered. This work highlights the need to further explore the individual determinants of cervical proprioception in order to better understand potential differences in the weighting of cervical proprioceptive information for use in postural and spatial orientation.

Disclosures: S.L. Thomson: None. **W.E. McIlroy:** None.

Poster

P451. Behavioral Effects by the Vestibular and Proprioceptive Systems

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Program #/Poster #: P451.09

Topic: D.06. Auditory & Vestibular Systems

Title: An animal model of third window syndrome: vestibular-cochlear based explorations of dehiscence induced impairments in decision making.

Authors: O. VAN OSCH[1], B. MORRIS[1], A. TAMAKLOE[1], *J. GAY[2], A. WACKYM[1], T. MOWERY[1];
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Abstract: Background: Third window syndrome is a vestibular-cochlear disorder in humans in which a third mobile window of the otic capsule creates changes to the flow of sound pressure level energy through the perilymph/endolymph. The nature and location of this third mobile window can occur at many different sites (or multiple sites); however, the most encountered third window is congenital or acquired superior semi-circular canal dehiscence. The primary physiological symptoms include sound induced vertigo, headaches, pseudo conductive hearing loss, tinnitus, autophony while speaking, and visual problems (nystagmus, oscillopsia). At the same time, individuals experience measurable deficits in basic decision-making, short-term memory, concentration, spatial cognition, and anxiety. The cause of many of these symptoms have been well-studied; however, without an animal model a fundamental understanding of the central processing changes will not be possible with our current technologies. **Methods:** Adult Mongolian gerbils (N=18) received surgical fenestrations to the superior semi-circular canal of the left inner ear. Auditory brain stem responses were carried out prior to surgery and over 10 or more days of recovery. Some animals were trained on a basic decision-making task (amplitude-modulation (AM) rate discrimination) using an appetitive reinforcement operant conditioning procedure, prior to 3rd Window procedure (N=5). A sensitivity measure, d', was computed for each animal across training days. Histological examination of the fenestration was carried out for each animal post-hoc and compared to both ABRs and behavior. **Results:** The fenestration created a significant elevation in hearing thresholds of the left ear; especially in the lower frequency domain (1 to 4kHz). In a subset of animals, left (EXP)/right ear(CTL) comparisons via ABR show significant threshold increases at the same frequency representations. For behavior, there was a significant effect on attenuated behavior over 5 additional days of testing. That is, post-surgery animals produced more false alarms and less correct rejections but maintained the ability to perform the task (~100% hit rate) compared to pre-surgery performance. **Conclusion:** The central basis of cognitive impairments associated with these kinds of peripheral injuries in humans is unknown. These preliminary results suggest that our animal model will provide a good basis for explorations into the central etiology of some of CNS based symptoms and decision-making impairments associated with 3rd window syndrome in humans.

Disclosures: O. Van Osch: None. **B. Morris:** None. **A. Tamakloe:** None. **J. Gay:** None. **A. Wackym:** None. **T. Mowery:** None.

Poster

P451. Behavioral Effects by the Vestibular and Proprioceptive Systems

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P451.10

Topic: D.08. Visual Sensory-motor Processing

Support: NSF IOS 1353845
NIH R01 NS076558

Title: Gap junction gene regulates choice between two temperature-dependent behaviors in C. elegans

Authors: *A. ALMORIL-PORRAS, A. C. CALVO, E. M. WISDOM, I. REN, A. ALJOBEH, J. D. HAWK, D. A. COLÓN-RAMOS;
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Abstract: Animals perform behaviors under a specific, relevant set of conditions that encompass external stimuli and internal cues experienced by the organism. In the case of plastic behaviors, prior experience can also instruct which behavior to perform, even when animals are presented with an invariant set of environmental stimuli. How animals select and switch between behavioral strategies given a set of conditions and how neuronal circuits underlie this selection is not well understood. To identify molecular underpinnings regulating this choice, we used the thermotactic behavior of the nematode *Caenorhabditis elegans*, defined as the migration to a preferred temperature instructed by previous training exposure to a given temperature. Thermotaxis can be further broken down into two behavioral strategies: wild-type animals will migrate across a temperature gradient (orthothermal migration) when placed at conditions different from the trained temperature, and they will perform long bouts of forward movement along isotherms (isothermal tracking) when they are near their preferred temperature. Here we show that gap junction genes play a key role in regulating the switch between these two behavioral strategies. Using a combination of forward and reverse genetics, we identified a gap junction mutant that fail to effectively switch strategies as they persistently perform isothermal tracking in an unfavorable context, far from their preferred temperature. We used cell-specific tools to ablate, probe and inspect the activity patterns of neurons known to play a role in thermotactic behavior in the two different contexts and determined some cellular requirements for isothermal tracking behavior. This study sets up a behavioral paradigm to probe how two different behavioral strategies might compete within a circuit and are selected to be executed by prior experience. Our findings also highlight the importance of gap junctions and electrical synapses present in neuronal circuits in mediating behavior selection.

Disclosures: A. Almoril-Porras: None. **A.C. Calvo:** None. **E.M. Wisdom:** None. **I. Ren:** None. **A. Aljobeh:** None. **J.D. Hawk:** None. **D.A. Colón-Ramos:** None.

Poster

P455. Retinal Circuitry II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P455.01

Topic: D.07. Vision

Support: NIH R01 EY029323
NIH R01 EY026555
NIH P30 EY026878

Title: Biophysical characterization of wide-field amacrine cells in the mouse retina

Authors: *W. LEI[1], S. J. PARK[2], L. M. SNYDER[5], D. A. CLARK[3], S. E. ROSS[5], J. B. DEMB[4];
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Abstract: Wide-field amacrine cells (WACs) represent a diverse class of interneurons that can extend neurites over one millimeter from the cell body in the mammalian retina. However, the biophysical properties, connections, and computations of WACs are poorly understood due to the difficulties of systematically targeting individual WAC types. Here we employed an intersectional genetic strategy to identify cell types in mouse retina within a diverse, genetically defined family of WACs expressing the transcription factor Bhlhb5 (B5 WACs). By combining B5-Flpo and Kappa Opioid Receptor(KOR)-cre alleles with a dual fluorescent reporter, we targeted B5-KOR WACs *in vitro* with two-photon microscopy for whole-cell patch clamp recording and dye filling. B5-KOR WACs stratify primarily in S1 and S4/5 levels of the inner plexiform layer (IPL). These WACs mostly have 5-12 neurites that branch near the soma and extend straight across the retina within a defined level of the IPL. B5-KOR WACs depolarize to either contrast increments (ON cells; S4/5) or decrements (OFF cells; S1). Depolarization was accompanied by small spikelets in some cases. We focused on an OFF type with a soma in the ganglion cell layer that stratified neurites in S1. These cells have four to six dendrites extending in two opposing directions. Somatic receptive field mapping by reverse-correlation technique revealed an anisotropic center-surround receptive field. Voltage-clamp recording further revealed that synaptic excitation and inhibition have similar anisotropic receptive fields. Subsequent pharmacological experiments showed that excitation is driven by the OFF pathway, whereas inhibition is driven by the ON pathway, i.e., cross-over inhibition, via glycine release. These experiments characterize the biophysical and physiological properties of a novel type of OFF WAC, and reveal a strategy for studying additional WAC types in the future.

Disclosures: W. Lei: None. **S.J. Park:** None. **L.M. Snyder:** None. **D.A. Clark:** None. **S.E. Ross:** None. **J.B. Demb:** None.

Poster

P455. Retinal Circuitry II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P455.02

Topic: D.07. Vision

Support: NIH Grant EY028542

Title: Light-adaptation Clamp: a biophysical tool minimizing photoreceptor adaption for studying visual processing

Authors: *Q. CHEN[1], F. RIEKE[2];
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Abstract: Efficient encoding of visual environments requires adaptational mechanisms that match the statistics (mean, contrast, etc.) of visual inputs to the limited dynamic range of visual neurons. Classic work shows that in steady state, adaptation takes place across visual systems, starting from the first steps in visual processing in the photoreceptors. However, steady state conditions rarely apply during natural vision; instead, the light levels encountered by visual neurons change dynamically as gaze shifts from one location to another within a single scene. We lack an understanding of the location and properties of the adaptational mechanisms that operate during dynamic natural viewing. One technical obstacle is the lack of tools to manipulate specific adaptational machinery along the visual pathways. Here, we use a biophysical model of phototransduction that captures the responses of photoreceptors to a wide range of stimuli. This model comprises a set of differential equations that describe the biochemical components of phototransduction. We verified the accuracy of this model by recording photoreceptor (rod and cone) responses to novel stimuli. We then developed a reverse-engineering paradigm (Light-adaptation Clamp, or LaC) to design stimuli which had predictable effects on the photoreceptor responses - e.g. negating the impact of nonlinearities to make the photoreceptor response depend linearly in light input. This approach allows us to alter the photoreceptor responses several ways, including: (1) predictably manipulating the kinetics of the photoreceptor responses, e.g, to compensate for differences in the kinetics of responses of different cone types; (2) removing asymmetries in the photoreceptor responses to light increments and decrements so that On and Off visual pathways receive equivalent input; (3) negating the impact of changes in light level on the gain and kinetics of the photoreceptor responses. We verify the accuracy of these designed stimuli with direct recordings of photoreceptor responses. This approach provides a new tool to manipulate inputs to the visual system in specific, predictable ways and separate photoreceptor and post-photoreceptor contributions to responses of downstream neurons and to perception.

Disclosures: Q. Chen: None. **F. Rieke:** None.

Poster

P455. Retinal Circuitry II

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Program #/Poster #: P455.03

Topic: D.07. Vision

Support: NIH Grant 1DP2EY022584
NIH Grant 5T32E025202-07
Brain Research Foundation Fay/Frank Seed Grant

Title: Evaluating genetic tools for the identification of ipRGCs

Authors: *M. A. FLORES-NARVAEZ, K. C. MIGUEL, R. H. IBRAHIM, T. M. SCHMIDT; Neurobio., Northwestern Univ., Evanston, IL

Abstract: Intrinsically photosensitive retinal ganglion cells (ipRGCs) are a class of retinal ganglion cell that express the photopigment melanopsin, allowing them to respond to light even in the absence of rod and cone input. ipRGCs are a heterogeneous cell population that comprises six characterized subtypes (M1-M6). The melanopsin expression varies greatly amongst the different subtypes, with some being too low to detect using immunohistochemistry or a transgenic line expressing GFP under the melanopsin promoter ($Opn4^{GFP}$). To account for this, researchers have taken advantage of the highly efficient Cre-LoxP system to account for this limitation and maximize the labeling of ipRGCs. However, the number of cells labeled by commonly used genetic reporter lines differs greatly. Therefore, we aimed to quantify and characterize the cell types labeled in conventionally used reporter lines: $Opn4^{Cre/+}$ x Z/EG (~3000 labeled cells), $Opn4^{Cre/+}$ x $ROSA^{YFP}$ (~6000 labeled cells) and $Opn4^{Cre/+}$ x $ROSA^{tdTomato}$ (~10,000 labeled cells). To do so, we performed immunohistochemistry to quantify and describe the different types of retinal cells labeled in these reporter lines. Additionally, we performed intravitreal injections of a viral Cre-dependent reporter (pAAV-hSyn-DIO-mCherry) into adult $Opn4^{Cre/+}$ mice, in which we quantified ~4,000 labeled cells. Altogether our data shows that $Opn4^{Cre/+}$ x Z/EG labeled the fewest retinal cells of the four genetic reporters studied. Moreover, we observed a substantial overlap amongst the different genetic reporters, suggesting that the cells labeled by $Opn4^{Cre/+}$ x $ROSA^{YFP}$, $Opn4^{Cre/+}$ x $ROSA^{tdTomato}$, and pAAV-hSyn-DIO-mCherry, but not by $Opn4^{Cre/+}$ x Z/EG, do express Cre. Collectively, these results indicate that the $Opn4^{Cre}$ line may manipulate a much wider population of retinal cells than the M1-M6 ipRGCs and suggests that there are potentially uncharacterized melanopsin-expressing ipRGCs in the mammalian retina.

Disclosures: M.A. Flores-Narvaez: None. **K.C. Miguel:** None. **R.H. Ibrahim:** None. **T.M. Schmidt:** None.

Poster

P455. Retinal Circuitry II

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Program #/Poster #: P455.04

Topic: D.07. Vision

Support: NSF DGE-1842165
NIH grant 1DP2EY022584

Title: Role of the ipRGC circadian clock in visual perception

Authors: *K. C. MIGUEL[1], J. D. BHOI[1], C. P. RIBELAYGA[2], T. M. SCHMIDT[1]; [1]Neurobio., Northwestern Univ., Evanston, IL; [2]Ophthalmology & Visual Sci., Univ. of Texas Hlth. Sci. Ctr. at Houston, Houston, TX

Abstract: Circadian rhythms, set by cellular clocks found throughout the body, are regular, repeated biological fluctuations that function over the 24-hour day to allow animals to anticipate daily changes in their environment. One of the largest changes to an animal's environment over the course of the day is the amount of illumination, from the direct sunlight at midday to the dark of a moonless night, and the visual system must function across this broad range. Mice are known to perform better on visual tasks at certain times of day, suggesting that their visual perception is under circadian control. However, it is unclear where in the visual system and by what mechanism these changes occur. The retina, the light-sensitive tissue of the eye where light stimuli are first encoded, undergoes many cellular and molecular changes throughout the day, and so it is possible that circadian changes in visual perception may stem from the retina. Within the retina, intrinsically photosensitive retinal ganglion cells (ipRGCs) are an ideal population of cells to use to examine how cell-autonomous circadian clocks influence visual function, as they are genetically tractable and contribute to at least two visual behaviors known to be under circadian control: the pupillary light reflex (PLR) and contrast sensitivity (CS). To test the role of cellular circadian clocks in visual function, we generated a mouse line where the clock gene Bmal1 is conditionally knocked out in ipRGCs ($Opn4^{Cre}$; $Bmal1^{fx/fx}$, or Bmal1cKO mice). We found that clock-controlled gene expression was altered in ipRGCs while rhythmicity of total retinal gene expression was largely maintained. Moreover, Bmal1cKO mice showed deficits in daytime CS, suggesting that the cellular clock in ipRGCs may be responsible for driving rhythmicity in this behavior. Overall, this work will provide insight into the role of cellular clocks in multiple visual behaviors.

Disclosures: K.C. Miguel: None. **J.D. Bhoi:** None. **C.P. Ribelayga:** None. **T.M. Schmidt:** None.

Poster

P455. Retinal Circuitry II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P455.05

Topic: D.07. Vision

Support: NIH Grant EY032230

Title: Tbr2 is required for the maintenance but not survival of ipRGCs

Authors: *S. ABED, D. FELDHEIM;
Univ. of California, Santa Cruz, Santa Cruz, CA

Abstract: Retinal ganglion cells (RGCs) expressing the photopigment melanopsin are intrinsically photosensitive (ipRGCs) and are important for subconscious visual behaviors such as circadian rhythm entrainment, pupil constriction, and light-mediated mood regulation. Previously, we and others showed that the transcription factor Tbr2 is required during retinal development for the formation of ipRGCs. Tbr2 is also expressed in the adult retina after development, suggesting that Tbr2 may play a role in the maintenance of ipRGC function. To test this, we removed Tbr2 in adult mice and found that loss of Tbr2 results in the loss of melanopsin expression in ipRGCs but does not lead to morphological changes to their dendritic or axonal terminations or affect their survival. We next ectopically expressed Tbr2 in RGCs and found that it is not sufficient for induction of melanopsin expression in non-ipRGCs. Lastly, we found that Meis2, a transcription factor that is expressed in GABAergic cells, is expressed in almost all Tbr2-expressing displaced amacrine cells and also in a subset of Tbr2-expressing RGCs. These findings demonstrate that Tbr2 is necessary but not sufficient for melanopsin expression and that Tbr2-expressing amacrine cells are GABAergic.

Disclosures: S. Abed: None. **D. Feldheim:** None.

Poster

P455. Retinal Circuitry II

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Program #/Poster #: P455.06

Topic: D.07. Vision

Support: NIH Grant 1DP2EY027983
Sloan Research Fellowship
Klingenstein-Simons Fellowship
Baker Program in Undergraduate Research

Title: Validating a New FlpO Line to Manipulate ipRGCs

Authors: *M. LUCE[1], E. CONTRERAS[3], T. M. SCHMIDT[2];
[2]Neurobio., [1]Northwestern Univ., Evanston, IL; [3]Northwestern Univ. IBIS, Evanston, IL

Abstract: Validating a new FlpO Line to Manipulate ipRGCs**Madison Luce,** Ely Contreras[b], Tiffany M. Schmidt (PI)[aa] *Department of Neurobiology, Northwestern University, Evanston, IL, 60201*[b] *Northwestern IBIS, Evanston, IL, 60201*madisonluce2022@u.northwestern.edu | tiffany.schmidt@northwestern.eduLight regulates the mammalian vertebrate visual system to affect both image-forming and non-image forming behaviors. Light is detected by the canonical rod and cone photoreceptors, as well as intrinsically photosensitive retinal ganglion cells (ipRGCs). ipRGCs express the photopigment, melanopsin, and are a diverse population, consisting of six types (M1-M6), which differ in morphology, electrophysiological properties, central brain projections, and how they influence visual behavior. For example, M1 ipRGCs modulate the pupillary light reflex, while non-M1 ipRGCs are involved in contrast sensitivity. However, the extent to which these ipRGC subtypes differ in their gene expression and how each individual subtype contributes to visual behavior is unknown. To address this gap in knowledge, the Schmidt Lab has developed a new $Opn4^{FlpO}$ mouse line that can be used in an intersectional genetic strategy for the genetic isolation and targeting of subsets of ipRGCs. We use immunohistochemistry in the $Opn4^{FlpO}$ mouse line, crossed to multiple reporters, and find that ipRGCs are specifically labeled in this line. Moreover, our data indicate that multiple subtypes of ipRGCs express FlpO, though M4s, M5s, and M6s are labeled at lower rates in this line than the other ipRGC subtypes. Currently, we are testing AAV-based genetic reporters to further manipulate subsets of ipRGC subtypes in the $Opn4^{FlpO}$ mouse line in hope to eventually isolate these subsets. Overall, our preliminary data suggest that the $Opn4^{FlpO}$ line labels ipRGCs and can, therefore, be used effectively to target and manipulate this RGC population, which will allow us to gain better insight into the visual behaviors evoked by particular populations.

Disclosures: M. Luce: None. **E. Contreras:** None. **T.M. Schmidt:** None.

Poster

P455. Retinal Circuitry II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P455.07

Topic: D.07. Vision

Support: SERB(SR/SO/AS-27/2012,TCN)

Title: Horizontal cell responses to light of variable photoperiod and intensities in Sprague-Dawley rats

Authors: *S. GUPTA, T. C. NAG;
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Abstract: Retinal degenerative diseases like AMD and retinitis pigmentosa are characterized by photoreceptor death, followed by changes in the inner retina, where one of the principal neuronal

type is horizontal cells (HC). We examined the morphological and neurochemical modifications in HC after light induced retinal damage (LIRD). The objective was to find out how the HC respond to outer retinal changes, in order to maintain the retinal circuitry. We exposed Sprague-Dawley rats to light of variable photoperiods and intensities and created an experimental situation that mimics retinal degenerative diseases where photoreceptor cell death is common. Adult rats (N= 30) were acclimated in 300 lux at 12 hour light: 12 hour dark (12L: 12D) photoperiod for 7 days and then exposed to 3000 lux at 12L: 12D for 2 days, followed by exposure to constant light (24L: 0D) for 2 days. Rats were then brought to 300 lux and reared for 15 days in 12L: 12D cycle. They were sacrificed at different days to see retinal HC remodeling. The identification of known markers of HC, namely calbindin D-28k (Calb) and doublecortin (DCX) immunohistochemistry and their ultrastructural changes allowed us to understand the retinal responses to light stress. In toluidine blue stained sections, swollen HC with their elongated neurites in outer plexiform layer (OPL) were seen under constant light, followed by a decrease in swelling of soma and dendritic retraction upon reverting back to ambient light of 300 lux. Calb was localized in HC and their number counted in 280 µm length of central inner nuclear layer (INL). Calb+ HC decreased in number with constant light but increased after light intensity was reversed (from 6 to 8/ 280 µm length of INL; $p<0.05$), whereas abnormal Calb+ HC increased in number with constant light and decreased with light reversal. There was an increase in total number of Calb+ HC under constant light (13/ 280 µm length of INL; $p<0.05$). DCX immunoreactivity (IR) was seen in HC, OPL and INL across all groups. Increase in IR in HC was seen in constant light group, which was still present in rats reverted back to 300 lux under 12L: 12D cycles for 5 and 15 days. Our result showed that after constant light exposure, the retina undergoes remodeling in HC soma and dendrites. This remarkable plasticity of the HC is integral to remodel the retinal circuitry altered after photoreceptor death in light stress. The increase in the number of calb+ HC may be due to their inherent property to divide under stressful conditions, which can be explained by their enhanced expression of DCX.

Disclosures: S. Gupta: None. **T.C. Nag:** None.

Poster

P455. Retinal Circuitry II

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Topic: D.07. Vision

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Title: Dendrosomatic synapses violate receptive field and connectivity rules in the mammalian retina

Authors: M. SEDLACEK[1], W. N. GRIMES[1], M. MUSGROVE[2], A. NATH[1], H. TIAN[1], M. HOON[4], F. M. RIEKE[5], J. H. SINGER[3], ***J. DIAMOND**[1];
[1]NINDS, NIH, Bethesda, MD; [2]Dept. of Biol., [3]Biol., Univ. of Maryland, College Park, MD; [4]Dept. of Ophthalmology and Visual Sci., Univ. of Wisconsin Madison, Madison, WI; [5]Univ. of Washington, Seattle, WA

Abstract: In the retina, neuronal morphology strongly influences visual response features. Ganglion cell (GC) dendrites ramify in distinct strata of the inner plexiform layer (IPL) so that GCs responding to light increments (ON) or decrements (OFF) receive appropriate excitatory inputs. This vertical stratification prescribes response polarity and ensures consistent connectivity between cell types, whereas the lateral extent of GC dendritic arbors typically dictates receptive field (RF) size. A2 amacrine cells are interneurons understood to mediate "cross-over" inhibition by relaying excitatory input from the ON layer to inhibitory outputs in the OFF layer. During night (scotopic) vision, A2s relay input from rod bipolar (RB) cells to ON and OFF cone bipolar (CB) cell axon terminals that contact GCs. ON signals from RBs are preserved via electrical synapses to ON CBs and inverted via inhibitory, glycinergic synapses to OFF CBs (Famiglietti and Kolb, 1975; Pourcho and Goebel, 1985; Strettoi et al., 1992; Tsukamoto et al., 2001). This circuitry reliably transmits scotopic signals to ON GCs (Smeds et al., 2019), but it remains unclear how visual signals are transmitted most reliably to OFF GCs at night.Here, we present ultrastructural analysis of mouse retina indicating that A2s make chemical synapses deep in the IPL and ganglion cell layer (GCL) onto the proximal dendrites, soma, and axon initial segments of sustained and transient OFFα (OFF_sα and OFF_tα) GCs. These synapses are made by a small subset of A2s whose distal, arboreal dendrites encounter OFFα GC somas. The synaptic outputs arise from the distal tips of the A2 arboreal dendrites, close to where A2s receive excitatory inputs from RBs, ensuring efficient signal transfer from input to output. Fluorescence imaging and electrophysiology experiments indicate that depolarizing stimuli elevate [Ca^{2+}] in A2 arboreal dendritic compartments and elicit glycinergic inhibitory postsynaptic currents (IPSCs) in OFFα GCs. Scotopic light-evoked IPSCs in OFF_sα GCs exhibit narrow RFs that do not correlate with dendritic arbor dimensions. Together, these results show that OFFα GCs receive powerful somatic inhibition that influences RF properties and that A2 synaptic organization is heterogeneous and depends on proximity to OFFα GC somas. This OFF pathway, avoiding entirely the OFF region of the IPL, challenges several tenets of retinal circuitry.

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Poster

P455. Retinal Circuitry II

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P455.09

Topic: D.07. Vision

Support: NIH EY030737

Title: Depolarization block shapes the contrast sensitivity of a newly identified retinal ganglion cell

Authors: *S. R. WIENBAR[1], G. W. SCHWARTZ[2];
[2]Ophthalmology, [1]Northwestern Univ., Chicago, IL

Abstract: The output of spiking neurons depends both on their synaptic inputs and on their intrinsic properties: principally their complement of ion channels. Retinal ganglion cells (RGCs), the spiking projection neurons of the retina, comprise over 40 different types in mice and other mammals, each tuned to different features of visual scenes. The circuits providing synaptic input to different RGC types to drive feature selectivity have been studied extensively, but there has been substantially less research aimed at understanding how the intrinsic properties of RGCs differ and how those differences impact feature selectivity. Here, we introduce an RGC type in the mouse whose contrast selectivity is critically controlled by its ion channels. We compare this new RGC type with a different RGC that receives similar synaptic input but produces qualitatively different spike output. Pharmacological experiments, imaging, and compartment modeling suggest that differences in sodium channel types and their localization allow these two RGC types to provide different information to the brain despite similar inputs.

Disclosures: S.R. Wienbar: None. **G.W. Schwartz:** None.

Poster

P455. Retinal Circuitry II

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Program #/Poster #: P455.10

Topic: D.07. Vision

Support: JSPS Grant-in-Aid for Young Scientists (B) (KAKENHI No. 17K16990)
JSPS Grant-in-Aid for Scientific Research (C) (KAKENHI No. 19K09939)

Title: Contribution of P2X3 receptors to visual information processing in the retina

Authors: *T. ISHII[1], A. SHIMOHATA[1], C. SUZUKI[1], T. SHIMOGORI[2], M. KANEDA[1];
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Abstract: Adenosine triphosphate (ATP) acts as a neurotransmitter in the nervous system. We have previously reported that all subtypes of P2X receptors (P2X1 - P2X7) are expressed in the mouse retina, and PPADS, an antagonist of P2X1, P2X2, P2X3 and P2X5 receptors, modulates the responses of bipolar and amacrine cells to light flash and the firing pattern of retinal ganglion cells. However, the types of P2X receptors contributing the signal processing in these neurons have not been well elucidated. In the present study, we focused on P2X3 receptors. First, we investigated the specific localization of the P2X3 receptor by *in situ* hybridization and immunohistochemistry. In the adult mouse retina, P2rx3 mRNA was expressed in the ganglion cell layer (GCL), and the immunoreactivity was found in the inner plexiform layer (IPL) and GCL, suggesting the expression of P2X3 receptors in amacrine or retinal ganglion cells in the GCL. We next examined the physiological function of P2X3 receptor in the retina using an antagonist of P2X3 receptor, A317491, In the electroretinogram, intravitreal injection of A317491 had no effects on the amplitudes of both a- and b-waves, which reflect the activity of photoreceptors and bipolar cells, respectively, whereas significantly decreased the amplitude of oscillatory potentials, which reflects the activity of amacrine cells. In multielectrode array, application of A317491 modulated the firing rate of ON and OFF-retinal ganglion cells. These results suggest that the P2X3 receptors physiologically work for the signal processing in the mouse retina.

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Poster

P526. Reaching Control: Action and Sensation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P526.01

Topic: E.04. Voluntary Movements

Support: University of Georgia Research Foundation

Title: Object motion dynamics influence both feedforward and feedback motor responses during virtual catching

Authors: *T. SINGH[1], A. M. GÓMEZ-GRANADOS[2], D. A. BARANY[2], I. KURTZER[3]; [1]Kinesiology, Pennsylvania State Univ., University Park, PA; [2]Kinesiology, Univ. of Georgia, Athens, GA; [3]Biomed. Sci., New York Inst. of Technol. Col. of Osteo. Med., Old Westbury, NY

Abstract: An important window into sensorimotor function is how we catch moving objects. Studies that examined catching of free-falling objects report that the timing of the motor response is independent of the momentum of the projectile, whereas the motor response amplitude scales with projectile momentum. However, this pattern may not be a general strategy

of catching since objects accelerate under gravity in a characteristic manner (unlike object motion in the horizontal plane) and the human visual motion-processing system is not adept at encoding acceleration. Accordingly, we developed a new experimental paradigm using a robotic manipulandum and augmented reality where participants stabilized against the impact of a virtual object moving at constant velocity in the horizontal plane. Ten participants needed to apply an impulse that mirrored the object momentum to bring it to rest and received explicit feedback on their performance. In different blocks, object momentum was varied by an increase in its speed (Speed Blocks) or mass (Mass Blocks). In contrast to previous reports on free falling objects, we observed that increasing object speed caused earlier onset of arm muscle activity and limb force relative to the impending time to contact. Also, arm force increased as a function of target momentum linked to changes in speed or mass. Our results demonstrate velocity-dependent timing to catch objects and a complex pattern of scaling to momentum.

Disclosures: T. Singh: None. **A.M. Gómez-Granados:** None. **D.A. Barany:** None. **I. Kurtzer:** None.

Poster

P526. Reaching Control: Action and Sensation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P526.02

Topic: E.04. Voluntary Movements

Support: European Research Council ERC-2015-CoG-682422
Human Frontiers in Science Program long term fellowship LT359/2018-L
DFG Grant SFB1315
DFG Grant FOR2134

Title: Sensory processing of passive limb movement in mouse primary forelimb somatosensory and motor cortex

Authors: *I. E. STEWART, C. J. WHITMIRE, J. F. A. POULET;
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Abstract: Maintaining an accurate sense of limb position during movement is a key function of the somatosensory system, but the cortical circuits and coding principles of proprioception are not yet resolved. Prior work with primates has implicated both primary somatosensory (fS1) and motor (fM1) cortex in the representation of directional passive limb movements (Delhaye et al., 2018; Fetz et al., 1980; London & Miller, 2013). In contrast, far less is known in the mouse model system, which offers an array of molecular tools to investigate circuit mechanisms of sensory coding and perception. To address this, we developed a motorised passive limb movement stimulator for head-fixed mice. Mice were trained to hold a post mounted on linear

motors that generated 8 directions of horizontal linear motion comparable to classic centre-out directional movement paradigms. Behavioural tracking with contact sensors, filming, and EMG confirmed mice held the post during passive movement trials. Extracellular laminar probe recordings from fS1 and fM1 in anaesthetised and awake mice identified sensory evoked responses to directional passive limb movements as well as to vibrotactile stimulation of the forepaw. Our preliminary recordings in mouse cortex show single units that are tuned to specific directions, suggesting comparability between the encoding of passive movement in rodents and primates. Use of single unit recordings in combination with intersectional transgenics in mice promises advances in the understanding of circuit and cellular mechanisms of proprioception.
References:
Delhaye, B. P., Long, K. H., & Bensmaia, S. J. (2018). Neural Basis of Touch and Proprioception in Primate Cortex. Comprehensive Physiology, 8(4), 1575-1602.
Fetz, E. E., Finocchio, D. V., Baker, M. A., & Soso, M. J. (1980). Sensory and motor responses of precentral cortex cells during comparable passive and active joint movements. Journal of Neurophysiology, 43(4), 1070-1089. https://doi.org/10.1152/jn.1980.43.4.1070
London, B. M., & Miller, L. E. (2013). Responses of somatosensory area 2 neurons to actively and passively generated limb movements. Journal of Neurophysiology, 109(6), 1505-1513.

Disclosures: I.E. Stewart: None. **C.J. Whitmire:** None. **J.F.A. Poulet:** None.

Poster

P526. Reaching Control: Action and Sensation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P526.03

Topic: E.04. Voluntary Movements

Support: JSPS KAKENHI Grant 19K11460

Title: The effect of the memory-guided task on the MFR amplitude

Authors: *K. NUMASAWA[1], T. MIYAMOTO[2], T. KIZUKA[2], S. ONO[2];
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Abstract: It is well established that the arm trajectory during reaching movement is corrected by a target jump, which is called an automatic pilot. Moreover, a manual following response (MFR) is also known as a similar motor response, which is rapid modulation of arm movement induced by large-field visual motion during reaching a stationary target. Since the correction of arm trajectory is induced with short latency (150 ms on automatic pilot and 125 ms on MFR) from the target jump or the onset of visual motion, these are thought of as implicit motor responses. Previous studies have suggested that one of the potential mechanisms involved in this motor

response is the internal forward model where the consequence of motor command predicted by efferent copy is compared with the current state of motor action. The target jump or the visual motion generates an error signal between the efferent copy and the actual sensory consequence, it thereby corrects the arm trajectory. However, it is still uncertain whether the accuracy of motor command affects the amount of the alternation of arm trajectory. Therefore, we attempted to clarify whether the accuracy of motor command affects the arm trajectory of memory-based reaching movement by two conditions (the normal and delay condition) and MFR which is induced by a grating motion stimulus. Several previous studies using delay conditions have demonstrated that the magnitude of delay, which attenuates the accuracy of the motor command based on a memory of the target's position, affects the performance of memory-guided reaching movements and the amplitude of EEG components involved in the motor preparation. In our task, participants performed reaching movements toward a memorized-target location with a beep cue which was presented 0 or 3 s after the target disappeared (0-s delay and 3-s delay conditions). The leftward or rightward visual grating motion was applied 400 ms after the cue. We evaluated a lateral velocity of arm movements and an event-related potential (ERP) recorded during the reaching task, which reflects the motor command accuracy. Our results showed that the N170 ERP amplitude in the posterior parietal electrodes and the MFR amplitude were significantly larger for the 3-s delay condition than the 0-s delay condition. These results suggest that the motor planning accuracy affects the amount of the implicit visuomotor response. Furthermore, there was a significant within-subjects correlation between the MFR and the N170 amplitude, which could corroborate the relationship between the implicit motor response and the motor planning accuracy.

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Poster

P526. Reaching Control: Action and Sensation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P526.04

Topic: E.04. Voluntary Movements

Title: The Sensorimotor System Increases Muscular Co-contraction, but not Visuomotor Feedback Gains, in Response to Visually Amplified Movement Variability

Authors: *J. A. CALALO[1], R. LOKESH[1], A. ROTH[1], S. SULLIVAN[1], J. D. WONG[3], J. A. SEMRAU[2], J. G. CASHABACK[1];
[1]Biomed. Engin., [2]Kinesiology and Applied Physiol., Univ. of Delaware, Newark, DE; [3]Univ. of Calgary, Calgary, AB, Canada

Abstract: Humans have a remarkable ability to perform precise and accurate motor actions, such as threading a needle or throwing a fastball. Naturally occurring movement variability poses a

significant challenge to the sensorimotor system when attempting to perform precise actions. Further challenging control, sensorimotor delays may allow movement variability to propagate and remain uncorrected for a period of time. Two differing, but potentially complementary views, are that the sensorimotor system regulates feedback gains or muscular co-contraction to mitigate movement variability. To our knowledge, no one has directly manipulated movement variability to support either of these differing views. We hypothesized that the sensorimotor system modulates both feedback gains and muscular co-contraction in response to movement variability and time delays. With visually amplified movement variability, we predicted greater muscular co-contraction and visuomotor feedback gains. Participants were seated with their arm placed on an exoskeleton robot. We collected surface electromyography of six upper-limb muscles. Participants moved their arm to control a cursor on a screen. They kept the cursor within the bounds of a narrow visual channel while reaching towards a target. Participants performed four conditions where we manipulated movement variability (amplified or non-amplified) and or time delay (delay or no-delay). Movement variability was visually amplified by doubling the cursor's distance from the channel centerline. Time delays were imposed by delaying cursor feedback 200ms in the lateral direction, perpendicular to movement along the visual channel. Muscular co-contraction was calculated as the minimum activation between muscle pairs. To estimate visuomotor feedback gains, on probe trials we laterally shifted the cursor and recorded applied hand force against a simulated mechanically stiff channel that prevented lateral movements. With amplified movement variability, we found greater elbow and shoulder muscular co-contraction. We found greater muscular co-contraction was related to decreased hand movement variability at the end of the reach. Time delays did not influence muscular co-contraction. Unexpectedly, we found no changes in visuomotor feedback gains in response to amplified movement variability and time delays. Our results support the idea that the sensorimotor system uses muscular co-contraction, but not visuomotor feedback gains, to regulate movement variability.

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Poster

P526. Reaching Control: Action and Sensation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P526.05

Topic: E.04. Voluntary Movements

Title: Force control in young adults: effects of visual feedback length of time

Authors: *E. CHRISTOU[1], S. DELMAS[1], M. WEINTRAUB[1], Y. CHOI[1], B. YACOUBI KEYHANI[2];
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Abstract: Visual feedback is an essential component for force control. Indeed, with amplification of visual feedback (increased gain), young adults exhibit less force variability during constant force contractions performed in a "pursuit" mode. Pursuit visual tasks allow the participant to see their force cursor and trajectory relative to the target. The display of the past force trajectory, therefore, creates a "history" effect, with longer lengths providing more visual information and ability to anticipate and correct future force perturbations. Here, we focus to understand how the length of the history effects force variability in young adults, an area that remains entirely unexplored. We hypothesized that young adults will exhibit lower force variability when given a history of 10 s, which contains the most influential oscillation to force variability (0.1 Hz). Ten young adults maintained a steady force at 10% MVC with ankle dorsiflexion for 30 s. They performed 3 trials for four different history conditions. Participants maintained their steady force output while they were able to see the past 1 s, 2 s, 5 s, and 10 s of their force trajectory history. The gain of visual feedback was 5º for the four history conditions, which were presented in a random order. For each contraction, we quantified the mean force, standard deviation of force, and coefficient of variation of force. In addition, we examined the power spectrum of the force output from 0-1 Hz, using a 0.05 Hz resolution. The mean force was not significantly different for the four history conditions ($p>0.3$). In contrast, the standard deviation and coefficient of variation of force were significantly greater for the 10 s condition compared with the 1 s, 2 s, and 5 s conditions ($p=0.001$), which did not differ in any of the measurements among them ($p=0.5$). Similar to the force variability, the force power spectra from 0-1 Hz was different for the 10 s condition relative to the other three conditions ($p=0.001$). Specifically, the 10 s condition exhibited greater power anywhere from 0-0.7 Hz. In contrast to our hypothesis, this study provides novel evidence that 10 s force history increases force variability in young adults by amplifying the oscillations in force output below 0.7 Hz. Although the exact reason remains unclear, we hypothesize that this exacerbation in force variability may relate to increased visual information processing and voluntary corrections derived from the 10 s history.

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Poster

P526. Reaching Control: Action and Sensation

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Program #/Poster #: P526.06

Topic: E.04. Voluntary Movements

Support: Swedish Research Council (VR Starting Grant 2019-01909)
Torsten Söderbergs Stiftelse
Göran Gustafssons Stiftelse

Title: Predictive attenuation of touch and tactile gating are distinct perceptual phenomena

Authors: *K. KILTENI, H. EHRSSON;
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Abstract: In recent decades, research on somatosensory perception has led to two important observations. First, self-generated touches that are predicted by voluntary movements become attenuated compared to externally generated touches of the same intensity (attenuation). Second, externally generated touches feel weaker and are more difficult to detect during movement than at rest (gating). Researchers today often consider gating and attenuation to be the same suppression process; however, this assumption is unwarranted because, despite more than forty years of research, no study has combined them in a single paradigm. We quantified how people perceive self-generated and externally generated touches during movement and rest. We demonstrate that whereas voluntary movement gates the precision of both self-generated and externally generated touch, the amplitude of self-generated touch is selectively attenuated compared to externally generated touch. We further show that attenuation and gating do not interact and are not correlated, and we conclude that they represent distinct perceptual phenomena.

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Poster

P526. Reaching Control: Action and Sensation

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Topic: E.04. Voluntary Movements

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Title: Modulation of tactile feedback for the execution of dexterous movement

Authors: *J. M. CONNER, A. BOHANNON, M. IGARASHI, J. TANIGUCHI, N. BALTAR, E. AZIM;
MNL-E, Salk Inst. for Biol. Studies, La Jolla, CA

Abstract: While dexterity relies on the constant transmission of sensory information, unchecked feedback can be disruptive to behavior. How somatosensory feedback from the hands is regulated as it first enters the brain, and whether this modulation exerts any influence on movement, remain unclear. Leveraging molecular-genetic access in mice, we find that tactile afferents from the hand recruit neurons in the brainstem cuneate nucleus whose activity is modulated by local inhibitory neurons. Selective manipulation of these local inhibitory circuits can bidirectionally modulate cuneate neuron responses to tactile inputs and can suppress or enhance the transmission of tactile information to downstream targets (VPL thalamus), thereby affecting behaviors that rely on movement of the hands. Further investigating whether these local circuits are subject to top-down control, we identify distinct descending cortical pathways that innervate the cuneate region in a complementary pattern. Somatosensory cortical neurons target the core tactile region of cuneate, while a large rostral cortical population drives feed-forward inhibition of tactile transmission by targeting an inhibitory shell. These findings identify a circuit basis for tactile feedback modulation, enabling the effective execution of dexterous movement.

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Poster

P526. Reaching Control: Action and Sensation

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Program #/Poster #: P526.08

Topic: E.04. Voluntary Movements

Title: Proprioceptive acuity for location of points on the hand and digits

Authors: *W. DARLING[1], B. ZUCK[1], L. MIKHAIL[2], J. ADHIKARI[2];
[1]Hlth. and Human Physiol., [2]Univ. of Iowa, Iowa City, IA

Abstract: Several studies have suggested that proprioceptive awareness of hand dimensions and locations of prominent structures on the hand such as digit tips and knuckles are quite inaccurate (e.g., Ingram et al. 2019 Plos One 14: e0210911). For example, proprioceptive estimates of hand width (distance between knuckles of the index and little finger) assessed by visually guided pointing with the index-tip of one hand to the knuckles of the unseen hand (covered by an opaque screen) resulted in a perceived hand width about 61% greater than actual width. However, visually guided reaches to proprioceptively specified external targets may result in large radial distance errors relative to the pointing arm shoulder (e.g., Darling and Miller 1995

Exp Brain Res 93:534), perhaps explaining these large errors observed previously. Here we tested accuracy of proprioceptively guided reaches to touch right index-tip to the knuckles and digit tips of the left hand in varied hand locations in 3D space. Right index-tip and target locations were measured in 3D using a magnetic tracker with small (1.5 mm dia) sensors. In the first experiment ten blindfolded right-handed subjects were instructed to reach at comfortable speed in a single movement to touch the right index tip to the specified targets on the left hand after first providing visually guided practice for each target. In a second experiment 4 naïve subjects received no visually guided practice prior to performing proprioceptively guided comfortable speed reaches to the verbally specified target. Visually guided reaches to each target were performed only after proprioceptively guided reaches to all targets had been completed. Reaches to each target while blindfolded were accurate from the first trial in both experiments, with average errors being in the range of 2-3 cm and less than 1 cm greater than when vision was allowed (note that the sensors cannot be co-localized, meaning that minimal errors are in the range of 1-1.5 cm). Variable errors were also small, averaging less than 1 cm. Final right index-tip locations were distributed around the hand targets rather than exhibiting large biases as in the previous work using visually guided pointing to the unseen hand in a fixed position. In contrast with previous findings, proprioceptive estimates of hand width were similar to or slightly smaller than actual hand width. Overall, we observed that in proprioceptively guided movements to touch the right index-tip to targets on the left hand were accurate. In contrast with previous studies using visually guided reaching to unseen structures on the hand, there was no evidence of proprioceptive overestimation of hand width.

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Poster

P526. Reaching Control: Action and Sensation

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Program #/Poster #: P526.09

Topic: E.04. Voluntary Movements

Support: VCU Wright Center Endowment Fund

Title: Establishing somatosensory milestones achieved during childhood

Authors: *V. W. T. CHU[1], S. DAVIS[2], N. AMIN[2];
[1]Occup. Therapy, [2]Virginia Commonwealth Univ., Richmond, VA

Abstract: Pediatricians and therapists are familiar with motor milestones during early childhood development. Milestones are useful in detecting motor delays, allowing for early intervention. On the other hand, we do not have similarly established milestones for somatosensory development. To begin documenting somatosensory milestones, we designed two pilot studies to

examine somatosensory development in children from 6 months to 12 years.
20 children between 6 to 60 months participated in the early childhood study. In this study, early somatosensory development was assessed in two areas: spatial awareness (reaching for visible vs unseen tactile targets), force perception and control (engaging with thin crayons). While children were able to reach for visible targets by 12 months and demonstrated object permanence by 10 months, children under 15 months did not attempt to reach for unseen targets on their body. By 24 months, most children were able to reach for unseen targets on their body and continued to refine the accuracy. In the area of force perception and control, by 12 months, most children were able to manipulate thin crayons in their hands without breaking them. However, children broke the crayons more between 24-36 months as they are exploring coloring and drawing with the crayons. By 48 months, most children mastered adequate force control to color with thin crayons.
18 children between 4 to 12 years and 11 adults participated in the school age study. In this study, somatosensory development was assessed in five activities: elbow joint position matching, index finger end-point position matching, spatial movement shape recognition, active resistive force control, and weight change detection threshold. Based on our observation, we saw that children's accuracy in performance on the elbow joint position matching and index finger end-point position matching tasks are relatively developed by 7 years old. Development of spatial movement shape recognition occurs later as the response accuracy stabilize by 10 years old to adult level. The ability to hold still resisting against a subtly changing force also develops around the same time, as the 10-12 years old group performed similar to the adults. The ability to detect changes in weight of an object in the hand appear to develop after 12 years old as the detection threshold drops significantly from the 10-12 year group to the adult group.
These two studies allowed us to establish a preliminary timeline of somatosensory development in children. In both studies, we saw that motor abilities develop prior to the related somatosensory perception, and spatial awareness appears to develop before force perception.

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Poster

P526. Reaching Control: Action and Sensation

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Program #/Poster #: P526.10

Topic: E.04. Voluntary Movements

Support: Israeli Science Foundation (ISF-1801/18)
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Title: Blockage of the cerebellar outflow to the motor cortex during multi-joint movements impairs the compensation for upper limb intersegmental interaction torques in the monkey

Authors: *N. SINHA[1,2], S. ISRAELI[1], J. DEWALD[3], Y. PRUT[1];
[1]Hebrew Univ. of Jerusalem, Jerusalem, Israel; [2]Interdepartmental Neurosci., [3]Physical Therapy and Human Movement Sci., Northwestern Univ., Chicago, IL

Abstract: During multi-joint movements, the central nervous system (CNS) uses predictive control to compensate for and take advantage of dynamically generated intersegmental coupling torques. The neural substrate for the inverse dynamics model, which facilitates this control, is likely distributed across the cortico-cerebellar loop. The objective of this study was to investigate the role of cerebellar outflow to the motor cortex in establishing this distributed model. We hypothesized that blockage of the cerebello-thalamo-cortical (CTC) pathway leads to incorrect compensation for coupling torques in the cortically generated muscle torque commands during planar reaching. To test this hypothesis, we trained a monkey (*M. fascicularis*) to wear an exoskeleton (KINARM) and perform center-out reaching movements in the horizontal plane to eight equidistant center-out reaching targets. The CTC pathway was reversibly blocked by high-frequency stimulation (HFS, 130 Hz) of the superior cerebellar peduncle. Kinematic data was obtained during 892 control and 310 HFS trials. Using inverse dynamics analysis, we computed the components of the net torque (i.e., muscle and interaction torques) acting at the shoulder and elbow joints during center-out reaching task. The covariance between muscle and interaction torques was significantly higher in control as compared to HFS trials for both the joints. Furthermore, the muscle to interaction angular impulse ratio (MI-IR) during movement initiation was significantly higher in HFS as compared to control trials at the shoulder joint Thus, the muscle torques incorrectly accounted for the upper limb coupling torques during HFS. Finally, we correlated MI-IR during movement initiation (response variable) with the peak tangential velocity of the hand (predictor variable). We found significant correlation between the two variables in HFS. This shows that the amount of correction for interaction torques increases with movement velocity during HFS. Overall, these results show that the CTC pathway is essential for facilitating accurate feedforward control of movement that takes into account multi-joint coupling torques.

Disclosures: N. Sinha: None. **S. Israeli:** None. **J. Dewald:** None. **Y. Prut:** None.

Poster

P553. Neurophysiology: Decoding and Neural Processing III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P553.01

Topic: E.05. Brain-Machine Interface

Support: UCF COM and CECS 2021 Pilot Awards Program

Title: Design and Validation of a Robust and Adaptive Brain-Machine Interface Algorithm for Control of Brain States

Authors: *H. FANG[1], S. A. BERMAN[3], Y. YANG[1,2];
[1]Dept. of Electrical and Computer Engin., [2]Disability, Aging and Technol. Fac. Cluster, Univ. of Central Florida, Orlando, FL; [3]Med., Univ. of Central Florida Col. of Med., Orlando, FL

Abstract: Brain-machine interfaces (BMIs) for controlling brain states hold promise for providing new therapies for neurological and neuropsychiatric disorders. Such a BMI adjusts brain stimulation based on real-time brain activity as feedback, forming a closed-loop brain stimulation system. Many groups have proposed model-based closed-loop brain stimulation systems and tested them in simulations. Most such systems assume linear time-invariant (LTI) models for brain activity. Brain networks, however, frequently involve nonlinear, uncertain, and time-varying dynamics, which can degrade control performance or even cause instability of LTI controllers during real-time control. Here, we develop a robust and adaptive BMI control algorithm with two main innovations. First, we design a new adaptive algorithm that can track a general form of nonlinear model uncertainty and time variations. Second, we improve the robustness of the closed-loop system by designing and applying a low-pass filter on the control input, which cancels the nonlinear model uncertainty only within the bandwidth of the closed-loop system. We next provide rigorous theoretical analyses of the stability margin and the transient bounds of the BMI control algorithm. We prove that there exists a tradeoff between the stability margin and transient boundary as we vary the bandwidth of the low-pass filter. We further develop a new method for choosing the optimal filter bandwidth based on the tradeoff. We next conduct two sets of simulations to validate the proposed algorithm. First, we use deep neural networks to simulate general nonlinear and uncertain brain network activity and systematically change the network parameters to validate our theoretical analyses. Second, we use a well-established nonlinear neurophysiological model of Parkinson's disease to simulate pathological brain activity for validating the entire robust and adaptive BMI control algorithm. The neural network simulation results validate our theoretical analyses, showing the stability and robustness of the BMI control algorithm. Moreover, the BMI control algorithm successfully regulates the pathological brain activity to a therapeutic regime in the neurophysiological simulations, which shows the potential of the BMI control algorithm in developing closed-loop treatments of brain disorders. Our robust and adaptive BMI control algorithm has implications for enabling future precise-tailored and safe treatments of a wide range of neurological and neuropsychiatric disorders.

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Poster

P553. Neurophysiology: Decoding and Neural Processing III

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P553.02

Topic: E.05. Brain-Machine Interface

Support: NSF Award 1910526

Title: Mechanisms of decreased performance from automated artifact rejection algorithms in P3 speller brain-computer interfaces

Authors: *T. SWEET, C. SCHMITZ, T. BRANDT, D. E. THOMPSON;
Electrical and Computer Engin., Kansas State Univ., Manhattan, KS

Abstract: Brain-Computer Interfaces (BCIs) can be a tool to aid in communication of people with severe paralysis. The P3 Speller BCI, based on electroencephalogram (EEG) is a noninvasive BCI for communication; unfortunately, EEG signals are susceptible to noise that is often much larger than the underlying signal. Automated artifact removal (AAR) algorithms have been developed with the intent to improve the quality of the EEG signal. In a previous study[1], we investigated the effects of several AAR algorithms on P3 Speller BCI performance and found that applying the algorithms typically resulted in lower performance. Consequently, we proposed five specific mechanisms that may contribute to the performance decrease: 1) signal sources may be renumbered or inconsistently treated during the artifact rejection process, 2) the P3 signal may be classified as noise by the AAR algorithms and removed, 3) the algorithms may lead to an ill-conditioned observation matrix, making the classifier less stable, 4) the classifier may rely on the large magnitude of eye movements in the raw signal, resulting in better performance compared to the signal with the algorithms applied, and 5) the classifier may similarly rely on the large magnitude of blink artifacts. Here, we investigate each of the five mechanisms: 1) The AAR algorithms we investigated should recombine the signal back into the time-domain before classification, eliminating the possibility of source renumbering; however, this is still under investigation, 2) While P3 amplitudes are reduced, morphology appears similar, and noise is similarly reduced, 3) While the condition numbers vary between algorithms, we find no consistent increase that could explain the performance difference, 4) early analysis shows that it is possible to track eye movement using EEG, but not with the resolution that would be required to achieve the observed BCI performance, and 5) we do find evidence that most participants typically blink in the window where the P3 would normally be present, particularly during early runs. We interpret this as evidence that original classifiers may have been partially relying on the blinks to discriminate between targets and non-targets. The AAR algorithms remove these blinks, which could reasonably explain the decrease in performance.

References:
1.Thompson DE, Mowla MR, Dhuyvetter KJ, Tillman JW, Huggins JE. Automated artifact rejection algorithms harm P3 Speller brain-computer interface performance. *Brain-Computer Interfaces*. 2019;6(4):141-148.

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Poster

P553. Neurophysiology: Decoding and Neural Processing III

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Program #/Poster #: P553.03

Topic: E.05. Brain-Machine Interface

Support: GE-2-2-023A (REXO)
IT-2-2-023 (VAFES).

Title: Implication of bi-domain representation of neural signals in non-invasive brain-computer interface (BCI) decoding applications using deep learning

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Abstract: Brain-computer interfaces (BCIs) establish a path of communication between humans and machines by translating the neural signals to control commands. Electroencephalography (EEG) is one of the most common sources of neural signals because of its inexpensive and non-invasive nature. They are often contaminated with noise and artifacts and have low spatial resolution. Henceforth, accurate decoding and translation of EEG signals into control commands are non-trivial. Mostly, important patterns for classifying EEG signals are deeply hidden. Raw time-series is either mapped to frequency domain, or time domain features are extracted. The purpose of feature extraction is to highlight the critical information for discrimination of data for different intended actions. In BCI, often the raw signals are mapped to frequency domain. Sometimes, this mapping compromises time-frequency resolution. Therefore, a better way would be to fuse the time domain and frequency domain information. Currently, deep-learning algorithms have demonstrated success in learning hidden and significant patterns with domain fusion. However, the performance of the deep learning algorithms relies upon the quality and quantity of the data. Consequently, a better domain representation technique and an attention-based method to generate better-quality data can empower deep learning algorithms. In this study, bi-domain representation of the EEG signals is employed to learn meaningful hidden patterns. Raw EEG signals show the time-domain representation of the EEG signals; however, frequency-domain representation is obtained by a novel input formation method. Additionally, an attention-based data augmentation method is proposed to generate new training examples. The frequency domain inputs are formed using the proposed modified version of Short Time Fourier Transform (STFT), called anchored-STFT. Anchored-STFT is inspired by wavelet transform and it tries to minimize the time-frequency resolution tradeoff displayed by STFT. As a result, it forms the inputs with better time-frequency resolution compared to standard STFT. Secondly, we propose a new attention-based method to generate new training inputs. The

proposed method finds the similarity score between different features of an input and utilizes the score to generate new training inputs. We used the publicly available BCI competition IV dataset 2b for the evaluation of our methods. Preliminary evaluation on dataset of two subjects (subject 5 and subject 6) shows that our methods improve the classification accuracy by 1.2% for subject 5 and 1.5% for subject 6 compared to state-of-the-art results.

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Poster

P553. Neurophysiology: Decoding and Neural Processing III

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Computing resources from the research computing data core at the University of Houston

Title: Detection of balance loss from electroencephalography: a novel approach to prevent falls implementing robotic orthosis

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Abstract: Falls remain one of the leading causes of death, injury, and hospital admission particularly in the elderly and people with locomotion difficulties. Wearable robotic systems are gaining popularity to assist or rehabilitate gait in these populations. If these powered robotic orthoses (exoskeletons) are to be able to assist with preventing the fall, early detection of balance loss is required. Studies exploring the prediction of perturbations from scalp electroencephalography (EEG) are rare and there exists a major gap in our understanding of how the brain detects and processes balance perturbations to try to avoid a fall. To fill in the above knowledge gaps and advance neurotechnologies for fall prevention in exoskeletons, we conducted, to the best of our knowledge, the first neural decoding investigation of perturbations during standing with an exoskeletal suit in healthy able-bodied adults. Specifically, we acquired high-density EEG, electromyography (EMG), and kinematic (center of pressure (COP)), data from 7 healthy able-bodied participants during a standing balance perturbation task with and without subjects wearing an exoskeleton device. The subjects were exposed to fixed and random

perturbations during standing in both conditions. The timing of perturbations was randomized across trials. We found components in the perturbation evoked cortical potentials (PEPs) as early as 50-90 ms after the onset of the mechanical external perturbation, which preceded both the peak in EMG activity (~190 ms) and the peak in COP (~ 350 ms). We later used a convolutional neural network (CNN) to detect the perturbations from single trials and obtained a 5 fold cross-validation test F score of 74.23 ± 5.2 %. The CNN model was implemented with emphasis on explaining the model decisions using gradCAM. We demonstrated that the model was driven by relevant components in the PEP to infer the predictions and not artifacts. The model's explanations further aligned with the dynamic functional connectivity measure estimated using the phase difference derivative. Specifically, the nodal connectivity was higher in the occipital frontal region in the early stage of the perturbations, shifting to parietal, then to the motor, and back to the parietal channels. Moreover, we also demonstrated the feasibility of decoding continuous COP values from EEG using a gated recurrent unit model. Overall our findings suggest that the EEG signals contain short-latency neural information related to an incoming fall which may be useful for developing brain-machine interface (BMI) systems for fall prevention in neurally-controlled robotic exoskeletons.

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Poster

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Topic: E.05. Brain-Machine Interface

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Title: Pyramidal-tract neurons are better and faster than intra-telencephalic neurons at neuroprosthetic learning due to intrinsic cell-type differences and local circuitry

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Abstract: During learning, communication between cortex and striatum is essential. Two different population of cortical neurons, intra-telencephalic (IT) and pyramidal tract (PT), project into striatum. Although a large amount of evidence demonstrates contrasting functional organization among both populations, it is unclear to what extent the different characteristics of each cell-type influence learning. To answer this question, we labeled different cell-types in two groups of mice and trained them to modulate the activity of either IT or PT neurons to control a brain-machine interface (BMI). We found that PT neurons can learn to control a BMI faster and reach better performance than IT neurons. Moreover, after disambiguating the impact of cell identity from other experimental features, our findings show that this is a result of characteristics inherent of being PT neurons as well as characteristics of their local circuitry in proximity to striatum.

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Poster

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Topic: E.05. Brain-Machine Interface

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Title: Decoding user task engagement in a long-term ECoG BCI

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Abstract: For individuals with paralysis, brain computer interfaces (BCIs) have the potential to improve motor function by translating movement-related neural signals into control signals for external devices such as cursors and prosthetic anthropomorphic upper-limb devices. Control of these devices can enable users to regain upper limb functionality, autonomy and improve their quality of life. Our lab has demonstrated that stable "plug-and-play" control of a 2D cursor is possible by leveraging the stability of electrocorticography (ECoG) and long-term closed-loop decoder adaptation. While ECoG neuroprosthetic cursor control enables patients to communicate by typing, we have found that the control can sometimes be error prone. Due to neural fluctuations in attention and maintenance of cognitive engagement, cursor control can vary from proficient to poor within a single session and even within single trials. We hypothesize that such transient errors and inconsistent control results from periods when the user is not directly engaged in the task. Thus, if we can explicitly decode user engagement and modulate direct neural control of the neuroprosthetic device, cognitive load would be reduced, and performance would improve. To study user task engagement, we looked at the spatial changes and temporal dynamics of beta frequency (13-30Hz) during a four state, four target 2D discrete cursor BCI control task. Beta frequency oscillations have been shown to play a critical role in motor functions such as reaching behaviors. Specifically, during stationary holds or moments prior to movement, beta frequencies synchronize and increase in power. In contrast, during movement initiation and execution, beta frequencies desynchronize and decrease in power. The four states of the task correspond to observation, movement preparation, active control, and idleness respectively. In this task structure we were able to capture intentional moments of engagement and disengagement. Our subject, a 38-year-old man with severe spastic tetraparesis, was implanted with a 128-channel chronic ECoG array over sensory and motor cortices. Data was collected at least once a week, with a testing day producing approximately 200-400 trials. We conducted signal magnitude and phase analysis using the Hilbert Transform as well as Singular Vector Decomposition for spatial-temporal analysis of beta with respect to the ECoG grid. We found that changes in beta frequency amplitude and beta burst rate can be used to detect movement intention immediately prior to the onset of direct BCI control.

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Poster

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Title: Eeg channel selection for ssvep bci speller

Authors: *H. KIM[1], K. WON[2], S. C. JUN[3];
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Abstract: Title: EEG channel selection for SSVEP BCI speller**Authors:** Heegyu Kim, Kyungho Won, and Sung Chan Jun****Abstract.*** The steady-state visual evoked potential (SSVEP) represents a stable neural response evoked by repetitive visual stimuli at specific frequency around the occipital area, which is applicable to high-speed brain-computer interface (BCI) speller. EEG channels for SSVEP BCI speller are commonly chosen around parieto-occipital areas to extract features. In this work, we proposed a channel selection method for SSVEP BCI speller based on CCA (Canonical correlation analysis) coefficients, which may increase user comfort as well as reduce preparation time.Our approach for optimal channel selection is follows: first, for each channel, we performed CCA method between single-channel EEG and reference signal (sinusoidal waves) per class. This estimated CCA coefficient represents the strength of linear relationship with the reference signal. Then EEG channels were sorted in decreasing order of the averaged CCA coefficients. Finally, the five channels with the top five coefficients were selected. We used a benchmark SSVEP dataset (Wang et al., 2017) that is publicly available. In this dataset, 64-channel EEG were collected from 35 participants; they performed 6 blocks of 40-class SSVEP based BCI speller, and each block contained 40 trials (single trial per class). We applied the channel selection method to 28 EEG channels around the central, parietal, and occipital areas, and compared to the whole EEG channels around parieto-occipital areas in this dataset (‘PO7', 'PO5', 'PO3', 'POZ', 'PO4', 'PO6', 'PO8', 'O1', 'OZ', and 'O2'). The performance was evaluated using standard CCA by 6-fold cross-validation.As a result, the average detection accuracy using our channels chosen by the proposed selection approach and the whole channels were 85.40±17.22% and 88.70±9.89%, respectively. In terms of information transfer rate (ITR), our channels chosen and the whole channels achieved 82.18±24.99 bits/min and 85.45±14.86 bits/min, respectively. In addition, we observed that there was no statistical significance between our channels chosen and the whole channels around parieto-occipital areas with p=0.5998 for detection accuracy, and p=0.8442 for ITR (by Wilcoxon signed-rank test). The proposed channel selection may reduce the number of EEG channels by up to 50%, while maintaining the performance of SSVEP speller.

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Poster

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Title: Predicting finger movements in a brain-machine interface across task contexts

Authors: *M. J. MENDER[1], S. R. NASON[1], H. TEMMAR[1], J. T. COSTELLO[2], D. M. WALLACE[3], M. S. WILLSEY[4], N. GANESH KUMAR[5], T. A. KUNG[5], P. G. PATIL[4], C. A. CHESTEK[1];
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Abstract: A key factor in the clinical translation of brain-machine interfaces (BMI) for restoring motor function will be robustness to changes in a task. This poses a significant challenge for applications such as brain-controlled functional electrical stimulation (FES), where the patient's hand may produce the same movements with varying postures and loads. Here we investigate the impact of task changes on performance in a BMI for restoring finger control.
To address this challenge, one rhesus macaque was implanted with a Utah microelectrode array targeted to the hand area of primary motor cortex, and 10 chronic bipolar electromyography (EMG) electrodes targeting muscles of the forearm and hand. He performed a 2-degree of freedom finger task to control a virtual hand using his index finger and middle-ring-small fingers to push doors in a manipulandum. During the task, the monkey would flex or extend one or both finger groups to match virtual targets while finger flexion, electrode threshold crossing rate (TCR), and EMG were simultaneously recorded. Context changes were introduced by adding torsion springs to the manipulandum or changing the static wrist angle during the task.
Using ridge regression, models mapping TCR to kinematics and to EMG activity were trained on normal trials, i.e. trials without the context changes, and tested on off-context trials, i.e. trials with the context changes, as well as a held-out set of normal trials. Across all kinematic variables and days, using the model to predict kinematics in off-context trials resulted in a 37% (SD =

30%) increase in mean-squared error (MSE) relative to predicting kinematics in normal trials. Predicting finger flexor and extensor EMG in off-context trials also resulted in large increases in MSE relative to predicting EMG in normal trials. The magnitude of the increases in error was larger for flexor muscles than extensors. These results suggest that neither kinematics nor EMG maintain a linear relationship with neural activity across these contexts. Lastly, decoder performance was tested in real-time using a Kalman filter trained on normal runs to control the virtual hand while context changes were added. Interestingly, the context changes in online control resulted in no significant changes in target acquisition time, time orbiting the target, or path efficiency.
These results highlight that modifying a task leads to a change in motor cortex activity which reduces kinematic and EMG prediction accuracy. However, subjects may be able to adapt to this change in online control. Future work will seek to replicate these results in a second monkey and evaluate changes to BMI performance in the context of brain-controlled FES.

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Poster

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Topic: E.05. Brain-Machine Interface

Support: NIH U01NS098975
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Title: Decoding grasp and speech signals from the cortical grasp circuit in tetraplegic human

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Abstract: Tetraplegia from spinal cord injury leaves patients paralyzed below the neck and unable to perform many activities of daily living. Brain-machine interfaces (BMIs) could give tetraplegic patients more independence by reading signals out of the brain directly and using these signals to control external devices such as robotic arms or hands. The cortical grasp network has been of particular interest because of its potential to facilitate the restoration of dexterous object manipulation. However, a network that involves such high-level cortical areas

may also provide additional information, such as the encoding of speech.
Towards understanding the role of different brain areas in the cortical grasp network, neural activity related to motor intentions for grasping was recorded in a C5-C6 tetraplegic patient in the supramarginal gyrus (SMG), the ventral premotor cortex (PMv), and the somatosensory cortex (S1). Each motor imagery trial began with a brief inter-trial interval, followed by a visual cue (picture of grasp shape) to one of five specific grasps. Then, after a delay period, the subject was instructed to imagine performing the cued grasp. In a second version of the experiment, the subject was instructed to say the name of the grasp out loud instead of motor imagery. We hypothesized that due to SMG's involvement in visual word recognition and phonological processing (Oberhuber, *Cerebral Cortex*, 2016), it would show higher neuronal modulation to speech than PMV and S1.
We found that in high-level brain areas SMG and PMV, grasps were well represented by firing rates of neuronal populations already at visual cue presentation. During motor imagery, grasps could significantly be decoded from all brain areas. A neuron-dropping analysis highlighted that at identical neuronal population sizes, SMG and PMV achieved similar decoding abilities, demonstrating their potential for grasp BMIs. During speech trials, SMG demonstrated highly significant classification accuracies, in contrast to PMV and S1, which were not able to significantly decode speech.
To conclude, these findings suggest that grasp signals can robustly be decoded from the cortical grasping circuit in human. PMV indicates a specialized role in grasping, while SMG's role is broader and extends to speech. Together, these results indicate that brain signals from high-level areas of the human cortex could be exploited for a variety of different BMI applications.

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Poster

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Title: Validation of a non-invasive, real-time, human-in-the-loop model of intracortical brain-computer interfaces

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Abstract: The opportunity to develop and test invasive brain-computer interface (iBCI) decoders in closed-loop with human users is very limited, yet essential to scientific and clinical progress. To address this challenge, we are developing a new machine learning-based iBCI encoder model that produces neural firing rate signals from finger movements, allowing neurologically intact humans to perform iBCI tasks in real time. The iBCI model combines an artificial neural network (trained on non-human primate cortical firing rates) with real-time healthy human subject input (captured non-invasively using a data glove) to generate emulated M1 firing rates. The emulated neural activity can be input to any iBCI decoder and used to operate an external device. A human subject controlling a cursor with a high-dimensional (19 DoF), non-intuitive input-space of finger joint angles is conceptually analogous to that of an iBCI system controlled from an unfamiliar, non-intuitive space of motor cortex activity. The great advantage of our approach is that readily-recruited, healthy human subjects can interact with decoders in closed loop, preserving the relevant learning dynamics between humans and adaptive decoders.
We validated the proposed iBCI model using both offline measures of the emulated neurons and performance measures of subjects during closed-loop cursor control. We found that emulated neural signals representing reaching movements in different directions maintain the spatial clustering patterns of actual M1 neurons when projected into a PCA plane, as well as the rotational dynamics commonly observed with jPCA. We used a steady state Kalman filter to decode cursor velocity from the emulated firing rates, allowing subjects to perform a center-out target acquisition task to facilitate direct comparisons to human subjects in published iBCI studies. We ran 25 healthy human subjects, each on four separate days. Our subjects' performance was consistent with that of the paralyzed, electrode-implanted, human iBCI users in studies by Kim et al., 2008, Kim et al., 2011, and Simeral et al., 2011 across seven different behavioral measures. These encouraging results suggest that this real-time firing rate emulation process is a useful means to provide statistically robust sample sizes for rapid prototyping and optimization of decoding algorithms, and the study of bi-directional learning in iBCIs, ultimately improving iBCI control.

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Poster

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Title: Developing a Spinal Cord Model that Can Acquire and Maintain Multiple Behaviors

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Abstract: The spinal cord, like the rest of the CNS, changes through life. When a new behavior is acquired, its CNS substrate or *heksor* (see Wolpaw et al. abstract) is created and, in addition, pre-existing heksors adapt to maintain their own behaviors. The new and old heksors negotiate the properties of the CNS neurons and synapses they share (Neuroscientist 16:532-549, 2010; J Physiol 596:3469-3491, 2018). For motor behaviors (behaviors with heksors that include spinal properties), this process changes the spinal cord. For example, EMG and kinematic details of locomotion may change, but its key features (e.g., right/left symmetry, posture, balance) are maintained. We are developing a model of a spinal cord-like neural network that can acquire and maintain several simple motor behaviors. We created a simple single degree-of-freedom musculoskeletal model of the elbow that approximates the normal relationship between motoneuron activity and kinematic output, i.e. behavior. Motoneuron activity is driven by a spinal-like neural network that incorporates spinal circuitry, reflexes, and a simple descending corticospinal input. The neural network architecture is based on the known circuitry of the spinal cord and has physiologically-inspired leaky integrate-and-fire neurons. In initial testing, simulated kinematics appear to be stable with no oscillatory or chaotic behavior, despite the model complexity. Additionally, this network exhibits several physiologically-appropriate characteristics, e.g. agonist motoneuron activity increases in response to muscle lengthening. Future development will optimize the neural network to complete two simple behaviors: 1) sine-wave elbow flexion/extension oscillation; 2) square-wave elbow flexion/extension oscillation. We will assess the ability of this network to learn and maintain these two behaviors by calculating for each behavior the normalized root-mean-squared error of the generated output (elbow angle) compared to the desired trajectory. Specifically, we will determine the minimum redundancy and network characteristics that are required to complete and maintain these behaviors, e.g., the minimum number of neurons or the need for signal-dependent noise. This model should provide a computational framework to: 1) assess how changes in the spinal cord affect multiple behaviors; 2) make predictions that can then be tested experimentally; and 3) suggest potential therapeutic targets for rehabilitation.

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Poster

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Stratton VA Med Center

Title: Heksor - CNS substrate of an adaptive behavior: testing the concept

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Abstract: Over the past half-century, the hardwired CNS of 1970 has gradually become the ubiquitously plastic CNS of today, in which change is the rule not the exception. This transformation requires a new paradigm for understanding how adaptive behaviors are acquired and maintained. The core of this paradigm is a previously undefined CNS entity, here given the name *heksor* from the Greek *hexis*. *A heksor is a distributed network of neurons and synapses that produces an adaptive behavior and changes as needed to maintain the key features of the behavior, the features that make the behavior satisfactory.* Muscle activity and kinematics may change; key features are maintained. Because heksors share the CNS, their concurrent changes constitute a negotiation; they negotiate the properties of neurons and synapses that they all use. They keep the CNS in a state of negotiated equilibrium that allows each heksor to maintain the key features of its behavior (Neuroscientist 16:532-549, 2010; J Physiol 596:3469-3491, 2018). We are testing the new paradigm. The strategy is straightforward. Acquisition of a new behavior changes many CNS areas. The paradigm predicts that this plasticity comprises the new heksor underlying the new behavior, plus changes in old heksors that maintain the key features of their behaviors despite the changes in neurons/synapses they share with the new heksor.
This poster presents new human and animal data that confirm and extend earlier studies. The studies are approved by the MUSC & Stratton IRBs and the Stratton IACUC. In normal rats, a new heksor created by soleus H-reflex (HR) operant conditioning in one leg does not impair key features of locomotion (e.g., right/left symmetry in step-length & hip height); but it does change how these features are produced. Changes in the locomotion heksor (e.g., in other reflex pathways) maintain symmetry. In rats with incomplete spinal cord injury (iSCI) that impairs locomotion, a new HR heksor that improves locomotion enables negotiation among the heksors that leads to widespread beneficial change in the locomotion heksor. Studies in humans with iSCI match these results: a new HR heksor that improves locomotion (e.g., by eliminating footdrop or clonus) enables practice leading to overall improvement in locomotion. Furthermore,

in rats with iSCI, an inappropriate new HR heksor does not further impair locomotion; changes in the locomotion heksor prevent this. Finally, the long-term spinal plasticity produced by cortical stimulation appears to comprise the direct effects of stimulation plus heksor adaptations to these direct effects. These results further support the new paradigm, and may guide therapeutic development.

Disclosures: J.R. Wolpaw: None. **A. Kamesar:** None. **A.K. Thompson:** None. **Y. Chen:** None.

Poster

P581. Sensory Motor Systems III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P581.03

Topic: F.01. Neuroethology

Support: CIFRE grant from ANRT
SEFA grant from PIA
ANR chaire Quantico

Title: From embodiment to enmachinment

Authors: *D. KERIVEN SERPOLLET *[1,2], A. ROQUES *[2], M. NASROUNE[2], Y. JAMES[2], P.-P. VIDAL[1,3,4];
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Abstract: * : these authors contributed equally to this work
Eye-trackers and head-trackers are robust tools to investigate how individuals interact with their environment. In the field of aeronautics, gaze control is generally used as a way to assess a pilot's performance or as a way to predict the mental workload (to prevent catastrophes from happening for instance). To the best of our knowledge, only few studies have been led on the eye-tracking data of pilots during flights. Here, we propose a study involving nine male professional helicopter pilots (age: 39 ± 5.7) equipped with an eye-tracking and a head-tracking devices in a full-flight level-D helicopter simulator in order to explore their ocular activity and gaze control during two realistic scenarios designed by expert pilots. We derived multiple gaze-related metrics that account for the pilots' current activity, experience or style. The movements of the aircraft relatively to the ground and the movements of the pilots' head were analyzed in the frontal plane (roll movements). The different roll movements of the aircraft above 10° were identified using a rupture-detection algorithm. It appeared that the movements of the aircraft and of the head were correlated, as we computed the Pearson correlation coefficient for every pilot

for both scenarios and found a positive correlation. This indicates that when the aircraft rolls to the left, the pilot's head also rolls to the left. Furthermore, a coherence analysis showed that those two signals were very coherent in the frequency range of the aircraft roll movements. Another secondary result we found is that the more experienced the pilots were, the more they relied on their flight instruments (rather than their own sensory cues) during critical flight phases. Our results suggest that the helicopter can be integrated by the pilots as extensions of their body representations, a phenomenon renewing the classical concepts of embodiment and distalization that we call 'enmachinment'. Indeed, we found, for every pilot, a pilot-specific linear relationship between the motion of the head and the motion of the helicopter. In this vision, the pilot and the machine are unified as a human-machine hybrid in which the pilot has integrated not only their own corporeal internal model but also the internal model of the helicopter (called "law of flight"). We suggest that this enmachinment will have important consequences on the training of helicopter pilots and more globally in human-machine interactions.

Disclosures: D. Keriven Serpollet *: None. **A. Roques *:** None. **M. Nasroune:** None. **Y. James:** None. **P. Vidal:** None.

Poster

P581. Sensory Motor Systems III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P581.04

Topic: F.01. Neuroethology

Support: Simons Foundation
Howard Hughes Medical Institute

Title: Decisions in an innate behavioral sequence

Authors: *K. M. CURY, R. AXEL;
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Abstract: Innate behaviors are comprised of ordered sequences of component actions that progress to satisfy drives. We have characterized the structure of egg-laying behavior in the fly and demonstrate that it consists of a sequence of component actions analogous to Nikolaas Tinbergen's "reaction chain." Tinbergen portrayed innate behaviors as a "reaction chain" in which each component action of the sequence enhances the probability of encountering "sign stimuli" that promote progression to a subsequent component. The organization of component actions into a "reaction chain" provides decision points at the junctures of component behaviors that ensure the successful progression towards the consummate act that satisfies the drive. We have identified a cluster of internal sensory neurons that provide interoceptive information about

the progression of the egg during ovipositor burrowing, the terminal behavioral component that is necessary for the subterraneous deposition of the egg. These neurons impart sensory feedback that allows burrowing to continue to egg deposition or to abort in favor of further exploration. Diminished activity of these neurons upon completed egg expulsion acts as a "sign stimulus" that results in the transition to the final phase of egg-laying, ovulation, allowing the cycle to repeat. Flexibility is therefore imposed on an innate and stereotyped sequence of motor actions to optimize the deposition and successful development of the egg.

Disclosures: K.M. Cury: None. **R. Axel:** None.

Poster

P581. Sensory Motor Systems III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P581.05

Topic: F.01. Neuroethology

Title: Myotomal Spread Pattern in Intraoperative Neurophysiological Monitoring of Selective Dorsal Rhizotomy

Authors: *K. KIM, S. LEE, J. LEE, K. KIM, S. HYUN, J. CHOI;
Seoul Natl. Univ. Hosp., Seoul, Korea, Republic of

Abstract: Introduction: Selective dorsal rhizotomy (SDR) is a surgery assuming that the cause of spasticity in cerebral palsy is related to hyperactive sensory-motor reflex arc and attempts to cut off part of the dorsal roots to reduce the afferent input. We aimed to reveal the myotomal spread pattern in which the lower extremity muscles contract during electrical stimulation of the dorsal roots or rootlets during intraoperative neurophysiological monitoring of SDR.
Methods: A total of 48 patients with cerebral palsy, who underwent SDR between April 2016 to June 2021 were included in the analysis. During SDR, recording of electromyography (EMG) was conducted in the external anal sphincter and bilateral adductor magus, vastus medialis, tibialis anterior, gastrocnemius, abductor hallucis, and biceps femoris muscles. To determine which dorsal rootlets to be sectioned, motor responses to a single stimulation of roots and tetanic stimulation of rootlets are evaluated. We present a visualization of myotomal spread pattern; distribution of muscles activated in a single and tetanic stimulation of dorsal roots and rootlets.
Results: When stimulating L2, L3, and L4 roots, there were myotomal spread patterns of co-activating adductor magnus, vastus medialis, and biceps femoris muscles in ipsilateral and contralateral sides, whereas stimulation to L5, S1, and S2 roots showed a relatively even distribution of all ipsilateral muscles (Fig 1). On the contralateral side, stimulation to L5, S1, and S2 roots produced the most activation in the biceps femoris, followed by adductor magnus and vastus medialis. There was no difference in the myotomal spread pattern in the analysis of subgroups divided by Gross Motor Function Classification System (GMFCS) and the spasticity.

Conclusion: In intraoperative neurophysiological monitoring of SDR, different myotomal spread patterns were observed in the upper and lower lumbosacral roots, irrelevant of GFMCS or spasticity. We propose that the results will help identify the function and anatomy of human locomotor patterns.

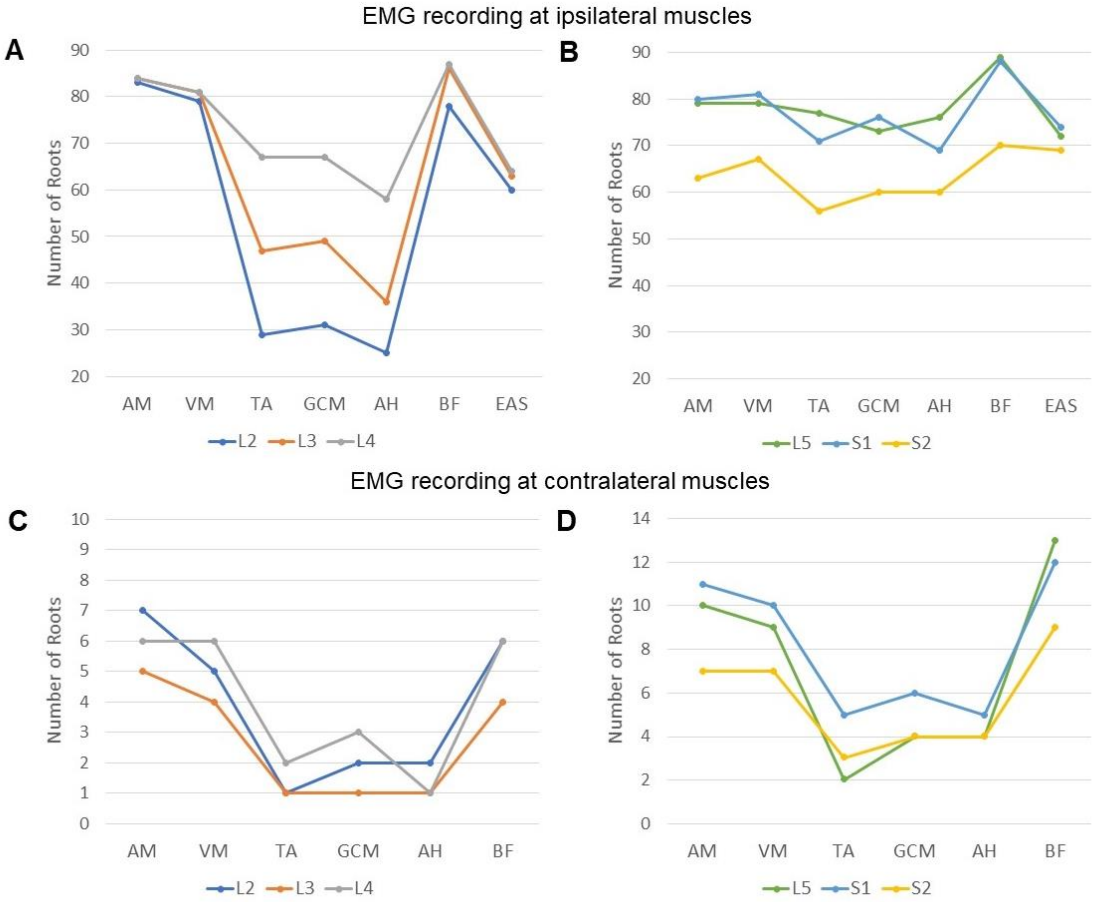


Figure 1. Myotomal spread pattern to a single stimulation of dorsal roots in ipsilateral L2-L4 (A), L5-S2 (B), contralateral L2-L4 (C), and L5-S2 (D) roots.

Disclosures: K. Kim: None. **S. Lee:** None. **J. Lee:** None. **K. Kim:** None. **S. Hyun:** None. **J. Choi:** None.

Poster

P622. Functional Imaging: I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P622.01

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: ZIAMH002783

Title: Monitoring vigilance during resting-state fMRI using ultra-slow fluctuations in the 4th ventricle

Authors: I. S. FERNANDEZ[1], J. GONZALEZ-CASTILLO[1], D. A. HANDWERKER[1], *P. A. BANDETTINI[1,2];
[1]Section on Functional Imaging Methods, [2]FMRI Core, Natl. Inst. of Mental Hlth., Bethesda, MD

Abstract: Fluctuations in wakefulness and vigilance during resting-state fMRI scans can affect the estimates of functional connectivity (FC) and, if ignored, can become a substantial confound in the data. This is important to keep in mind, especially in clinical studies where fluctuation in vigilance can range across populations. However, monitoring vigilance during fMRI requires concurrent recordings, such as eye tracking or EEG, or training of complex models with potentially limited ability to generalize to other samples. Because of this, simple methods for monitoring vigilance that rely solely on fMRI data are in great demand. Here we evaluate the efficacy of an alternative, simple way of monitoring vigilance based on frequency analysis of the fMRI signal in the 4th ventricle (FV). This method is motivated by recent research suggesting that descent into sleep is accompanied by a reversal of CSF flow that translates into inflow waves at approx. 0.05Hz observable in the FV. In this project, we first test if these CSF traces can be detected in datasets with limited inflow weighting such as the 7T resting-state Human Connectome Project dataset. To aid in our analysis we relied on concurrent eye tracking data as a secondary method of monitoring vigilance. We found that despite these data not being optimized to capture inflow effects in the FV, the above-mentioned 0.05 Hz CSF inflow-related fluctuations were clearly detectable in subjects with long eye-closure periods. Not only was the signal present in these data, but the temporal evolution of its spectral power could be used to predict individual periods of eye closure with up to 70% accuracy. Further analysis uncovered similar fluctuations in large portions of gray matter that severely influence estimates of FC, leading to significant losses in sensitivity and specificity of connectivity changes across vigilance states. In summary, our results add evidence that the value of 0.05Hz fluctuations in the FV is a marker of vigilance in fMRI data. It also highlights the strong modulatory effects of wakefulness FC estimates derived from the 7T HCP sample, an important, often re-analyzed, publicly available dataset.

Disclosures: I.S. Fernandez: None. **J. Gonzalez-Castillo:** None. **D.A. Handwerker:** None. **P.A. Bandettini:** None.

Poster

P622. Functional Imaging: I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P622.02

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: Y1AA3009

Title: Correspondence between changes in the BOLD signal and relative cerebral blood flow after one night of total sleep deprivation

Authors: *E. SHOKRI KOJORI[1], S. B. DEMIRAL[1], D. TOMASI[1], G.-J. WANG[1], N. D. VOLKOW[2];
[1]NIH, Bethesda, MD; [2]NIH/NIDA, Rockville, MD

Abstract: Sleep deprivation (SD) has been associated with marked changes in measures of brain activity and metabolic supply. Several fMRI studies have shown SD-related increases in low frequency (LF, < 0.1 Hz) fluctuations in the BOLD signal in the occipital, dorsal attention, and sensorimotor regions. Imaging of cerebral blood flow and glucose metabolism have also shown increases in metabolic supply in similar regions, particularly, in the visual cortex. It is not known whether SD-related changes in measures of brain activity and metabolic supply are coupled. To address this, a group of 20 healthy individuals (10 females, 22-72 years old) underwent brain imaging to index resting-state brain activity with fMRI (10 min) and to measure R1, an index of relative cerebral blood flow (rCBF), with FBB-PET (using a cerebellar reference region). The fMRI and PET data were collected once after rested wakefulness (RW) and once after one night of total SD (order of RW and SD days were randomized across participants). One male participant was excluded due to issues with the PET data. As expected, SD resulted in significant increases in the fractional amplitude of LF fluctuations (fALFF) within the visual, dorsal attention, and sensorimotor regions ($p_{FWE}< 0.05$). We have previously shown that these regions exhibit a strong coupling with the peripheral sympathetic tone and were labeled as an autonomic network (AN). SD also resulted in significant increases in rCBF primarily in the middle occipital gyrus ($p_{FWE} < 0.05$). However, SD-related changes in rCFB and fALFF were not associated in the visual regions where both measures significantly increased ($r(17) = 0.26$, $p = 0.28$). We performed a follow up analysis to characterize the relative metabolic cost (rCST): the extent that rCFB measure of metabolic supply exceeded fALFF measure of brain activity in a given region relative to the rest of brain. We found that SD significantly decreased rCST in most areas of the AN including sensorimotor and dorsal attention regions ($p_{FWE} < 0.05$). In sum, SD concurrently increased rCBF and fALFF, particularly, in the occipital cortex, yet these changes were not associated across participants. Furthermore, reductions in rCST suggested that SD-related increases in LF fluctuations within sensorimotor and dorsal attention regions were not accompanied by proportional increases in rCBF. The findings are relevant for delineating the vascular versus neuronal origin of SD-related increases in LF fluctuations in fMRI signal within the AN.

Disclosures: S.B. Demiral: None. **D. Tomasi:** None. **G. Wang:** None. **N.D. Volkow:** None.

Poster

P622. Functional Imaging: I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P622.03

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIH Grant HD069381

Title: The differential effects of adiposity on functional connectivity in preadolescent children

Authors: *N. E. LOGAN[1], D. R. WESTFALL[1], L. B. RAINE[2], S. ANTERAPER[3,4], L. CHADDOCK-HEYMAN[5,1], S. WHITFIELD-GABRIELI[1], A. F. KRAMER[1,5], C. H. HILLMAN[1,2];
[1]Psychology, [2]Physical Therapy, Movement, & Rehabil. Sci., Northeastern Univ., Boston, MA; [3]MIT, Cambridge, MA; [4]Carle Illinois Advanced Imaging Ctr. (CIAIC), [5]Beckman Inst., Univ. of Illinois Urbana-Champaign, Urbana-Champaign, IL

Abstract: Childhood obesity is a global health concern, with >340 million youth considered overweight or obese. In addition to contributing greatly to health care costs, excess adiposity associated with obesity is considered a major risk factor for premature mortality from cardiovascular and metabolic diseases. Adiposity is also negatively associated with cognitive and brain health. Thus, considerable efforts should be taken to reduce the negative health outcomes associated with childhood obesity. A complimentary line of research highlights the importance of cardiorespiratory fitness, a byproduct of engaging in physical activity, on an abundance of positive physical and mental health outcomes across the lifespan, including cognitive and brain health. Notably understudied, however, is the influence of adiposity and fitness on functional brain connectivity in preadolescent children, warranting further investigation. This study investigated the relationship among excess adiposity (visceral adipose tissue [VAT], subcutaneous abdominal adipose tissue [SAAT]), Body Mass Index (BMI), and fat-free cardiorespiratory fitness (FF-VO_{2max}) on resting-state functional connectivity (RSFC) in 121 (f = 68) children (7-11 years) using a data-driven whole-brain multi voxel pattern analysis (MVPA). MVPA models revealed two clusters associated with VAT and BMI measures, controlling for FF-VO_2. These clusters were then used for post-hoc whole-brain seed-to-voxel RSFC analysis. Yeo's (2011) RSFC-based 7-network cerebral cortical parcellation was used for labeling the results. Post-hoc seed-to-voxel analyses accounting for FF-VO_{2max}, IQ, sex and socioeconomic status, found robust associations of VAT and BMI with areas involved in the visual (VN), somatomotor (SMN) and frontoparietal (FPN) networks. Specifically, anticorrelations were observed within the FPN, and between the FPN and VN and SMN, for both the VAT and BMI models. No significant results were found for SAAT. These novel findings indicate that BMI and excess VAT in childhood may be selectively associated with a range of RSFC networks. The data support previously published research demonstrating a negative association between childhood obesity and brain function, and the selective association of VAT with neuroelectric

indices of cognition. Lastly, research demonstrates the FPN has also been associated with cognition in children. Consequently, these novel results extend the current literature to suggest that BMI and adiposity are differentially associated with functional connectivity in children, and these networks may also relate to cognition.

Disclosures: N.E. Logan: None. **D.R. Westfall:** None. **L.B. Raine:** None. **S. Anteraper:** None. **L. Chaddock-Heyman:** None. **S. Whitfield-Gabrieli:** None. **A.F. Kramer:** None. **C.H. Hillman:** None.

Poster

P622. Functional Imaging: I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P622.04

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIH Grant R01MH120174
NIH Grant R56AG066656

Title: Mapping glutamate in somatomotor and visual networks using novel GluCEST imaging

Authors: *M. K. PECSOK[1], J. JEE[2], V. J. SYDNOR[1], R. P. R. NANGA[5], T. D. SATTERTHWAITE[3], D. R. ROALF[4];
[1]Neurosci. Grad. Group; Dept of Psychiatry, [2]Dept of Psychiatry, [4]Neuropsychiatry, [3]Univ. of Pennsylvania, Philadelphia, PA; [5]Dept of Radiology, Perelman Sch. of Med., Philadelphia, PA

Abstract: Glutamate (Glu), the main excitatory neurotransmitter in the brain, is particularly important within the motor system. Perturbations in cortical and subcortical Glu levels been found in a range of disease states, including schizophrenia, depression, and Parkinson's Disease. While there is emerging evidence that the motor system is implicated in psychopathology, studies focusing on Glu in the motor cortex, and possible correlations with psychiatric symptoms, are lacking. Ultra high-field Glutamate Chemical Exchange Saturation Transfer (GluCEST), a novel imaging method, offers an innovative approach to examine glutamate *in vivo*. In this study, we explored Glu levels in the motor cortex as compared to other cortical areas. Furthermore, we correlated Glu levels in the motor cortex with clinical symptoms. Drawing upon data from a cohort of 47 adults who were healthy, depressed, or on the psychosis spectrum, we compared GluCEST MRI within the somatomotor network (SomMot), as defined by the Schaeffer100 atlas (Schaeffer et al., 2018), with all non-motor cortical regions (Non-Mot) and with the visual network (Vis). SomMot Glu (7.57; SD=0.72) was lower than Non-Mot (8.21; SD=0.47) or Vis (9.50; SD=1.22) (units: %GluCEST contrast; $p<.0001$; one-way ANOVA). There was greater variance in Vis than SomMot. SomMot Glu was nominally negatively correlated with PROMIS depression scores, wherein participants with higher depression scores

had lower measured Glu in SomMot. Given the robust glutamatergic inputs from the motor thalamus, we expected SomMot Glu to be higher than Vis Glu. However, the elevated Vis GluCEST signal aligns with a previous GluCEST study conducted in a different cohort, which reported high Glu in the occipital lobe. The present finding also fits with extant spectroscopy literature, which generally reports higher Glu concentrations for Vis than SomMot areas. The substantially higher Glu within the Vis may reflect the higher baseline activity in this region as is reported in previous studies of regional cerebral blood flow (e.g., Pantano et al., 1984). Slower rates of astrocyte clearance of Glu or the dense layer IV, containing glutamatergic spiny neurons, may also contribute to higher Vis Glu. Though weak, the negative correlation between SomMot GluCEST and depression scores is particularly intriguing in light of recent work drawing connections between motor system perturbations and psychopathology. Future analyses in this dataset will explore variations in baseline Glu in other functional networks, as well as the relationship between Glu levels in the SomMot and Vis and clinical or behavioral measures.

Disclosures: M.K. Pecsok: None. **J. Jee:** None. **V.J. Sydnor:** None. **R.P.R. Nanga:** None. **T.D. Satterthwaite:** None. **D.R. Roalf:** None.

Poster

P622. Functional Imaging: I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P622.05

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

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R01 MH111447
NIH S10 RR026783
P30 NS076408
WM KECK Foundation
NIBIB P41 EB027061

Title: Long-scale noise correlations support accurate localization of gray matter in ultra-high field fMRI with sub-millimeter resolution

Authors: *K. B. WELDON, L. T. DOWDLE, *C. A. OLMAN;
Univ. of Minnesota, Minneapolis, MN

Abstract: Achievements in ultra-high field (UHF, 7+ Tesla) technology have made it possible to demonstrate the ability of fMRI to reveal cortical columns and layer-dependent neuronal processes in humans. However, acquisition and analysis decisions at UHF have greater potential to degrade spatial accuracy than decisions for data acquired at lower field strengths. Thus,

accurate delineation of gray matter (GM) from white matter (WM) in functional images acquired at UHF is crucial in order to support spatial localization of the BOLD signal. Since spatial autocorrelations of residual noise in fMRI are tissue-dependent, we propose that fitting the spatial autocorrelation of residual noise in high-resolution gradient-recalled echo-planar imaging (GE EPI) images is a promising tool for independently localizing GM at UHF strengths. Seven healthy adults participated in one fMRI scan with submillimeter voxel resolution (0.8 mm isotropic) and one fMRI experiment with 1.4 mm isotropic resolution at 7T. The spatial autocorrelation function (ACF) maps and parameters were generated from single scans without any of the typical fMRI pre-processing steps to avoid unnecessary blurring. Functional data analysis was performed in order to model and subtract task-related signal from each dataset. We used AFNI's 3dLocalACF function to estimate the Gaussian plus mono-exponential spatial autocorrelation in the residual noise in both datasets. The c parameter, which characterizes signal not modeled by the analyses, was larger in GM regions than in WM regions in both datasets, which is consistent with vascular and neural processes. While this finding is not a universal truth that can be expected in all high-resolution datasets, the tissue-dependence of spatial autocorrelations in noise is a feature of thermal noise-limited datasets which may be useful in guiding analyses as we push the limits of fMRI resolution for neuroscience applications.

Disclosures: K.B. Weldon: None. **L.T. Dowdle:** None. **C.A. Olman:** None.

Poster

P622. Functional Imaging: I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P622.06

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: JP21dm0207001

Title: Functional MRI in awake marmosets for longitudinal studies

Authors: *F. SEKI, T. YURIMOTO, T. INOUE, E. SASAKI;
Central Inst. for Exptl. Animals, Kanagawa, Japan

Abstract: Resting-state network dysfunction can serve as a relatively early indicator of neurodegenerative disorders such as Alzheimer's disease (AD). Precise evaluation using non-human experimental animals such as common marmosets is important as longitudinal studies are the possible to analyze the onset of disease. To achieve this, fMRI assessment in awake marmosets should be established. Thus, we aimed to develop a non-invasive head and body restraint suitable for small bore-size MRI to perform longitudinal fMRI in the marmosets. The restraint for marmoset MRI suitable for 7.0-T Biospec 70/16 scanner system with an inner diameter of 72 mm volume coil was produced. Fulfilled conditions were 1. Size of body/head

restraint less than 72 mm diameter, 2. fixing the head non-invasively, 3. head movement less than 0.5 mm, and 4. use of non-magnetic materials. The restraint was designated by 3D CAD and printed using a 3D printer.Functional MRI was performed for normal marmosets. The parameters were as follows: repetition/echo time; 2000/15 ms, field of view; 38.4 × 38.4 mm, image matrix; 64 × 64, slice thickness; 1 mm,21 slices,repetitions; 200. The face of the marmoset was monitored using an MRI-compatible camera throughout the experiment. The head restraint enabled less than 0.5 mm movement of the marmosets' head using only non-magnetic materials non-invasively during the scan. We analyzed BOLD responses evoked by visual and auditory stimuli as preliminary data. Parts of the primary and secondary visual cortex and auditory cortex were activated during visual and auditory stimulation, respectively. In resting-state fMRI, the brain network including the parts of the posterior cingulate cortex and intraparietal cortex was confirmed. This study developed a non-invasive awake fMRI for small bore-size MRI, less than 10 cm. Fixing the head without a post head was challenging to achieve non-invasively. Although the non-invasive fMRI platforms have been available for awake marmosets, the MRI bore size must be more than 10 cm to acquire the head data. The non-invasive awake fMRI developed does not require head data individually as it fits all marmoset heads. U-type pin used for fixing the head permitted conducting fMRI for many marmosets. Further, the presented fMRI platform could introduce any size since this was designed for minimum size MRI. As a complementary tool of existing restraints for marmosets, this would also be valuable in certain conditions, including longitudinal evaluation. Recently, we have developed the AD marmosets. This awake fMRI does not require the attachment of a head post and enables cautious longitudinal evaluation of these disease model marmosets over the years.

Disclosures: F. Seki: None. **T. Yurimoto:** None. **T. Inoue:** None. **E. Sasaki:** None.

Poster

P622. Functional Imaging: I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P622.07

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: Stanford neuroscience
Simons Foundation

Title: Causal coupling between neural activity, metabolism, and behavior across the Drosophila brain

Authors: *K. MANN, S. DENY, S. GANGULI, T. CLANDININ;
Stanford Univ., Stanford, CA

Abstract: Coordinated activity across networks of neurons is a hallmark of both resting and active behavioral states in many species, including worms, flies, fish, mice and humans. These global patterns alter energy metabolism in the brain over seconds to hours, making oxygen consumption and glucose uptake widely used proxies of neural activity. However, whether changes in neural activity are causally related to changes in metabolic flux in intact circuits on the sub-second timescales associated with behavior, is unclear. Moreover, it is unclear whether differences between rest and action are associated with spatiotemporally structured changes in neuronal energy metabolism at the subcellular level. Here, we combine two-photon microscopy across the fruit fly brain with sensors that allow simultaneous measurements of neural activity and metabolic flux, across both resting and active behavioral states. We demonstrate that neural activity drives changes in metabolic flux, creating a tight coupling between these signals that can be measured across large-scale brain networks. Further, using local optogenetic perturbation, we demonstrate that even transient increases in neural activity result in rapid and persistent increases in cytosolic ATP, suggesting that neuronal metabolism predictively allocates resources to meet the energy demands of future neural activity. Finally, these studies reveal that the initiation of even minimal behavioral movements causes large-scale changes in the pattern of neural activity and energy metabolism, revealing unexpectedly widespread engagement of the central brain. As the relationship between neural activity and energy metabolism is likely evolutionarily ancient and highly conserved, these studies provide a critical foundation for using metabolic proxies to capture changes in neural activity.

Disclosures: K. Mann: None. **S. Deny:** None. **S. Ganguli:** None. **T. Clandinin:** None.

Poster

P622. Functional Imaging: I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P622.08

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

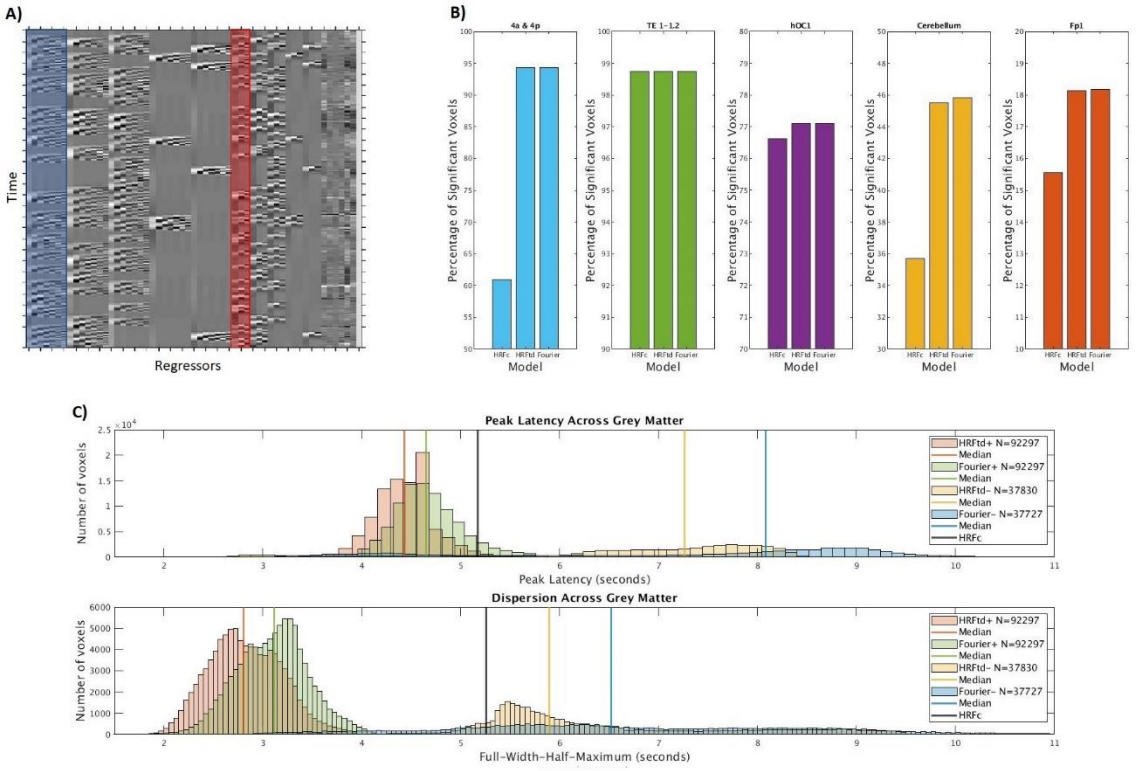
Support: BBSRC Grant BB/S009477/1
Royal Holloway University of London College Scholarship

Title: Modelling the BOLD response: On the insufficiency of conventional modelling approaches

Authors: *M. DANVERS, M. LONGLEY, N. RAMNANI;
Psychology, Royal Holloway Univ. of London, Egham, United Kingdom

Abstract: In event-related functional MRI (fMRI), short bursts of neuronal activity result in transient evoked haemodynamic responses (EHRs). The most common approach for detection and localisation is to construct General Linear Models (GLM) in which regressors model

expected EHR time-courses. These are typically based on a ‘canonical’ haemodynamic response function with a set of rigid assumptions about it’s timecourse. The utility of this approach depends upon the extent to which EHRs conform to these assumptions. How appropriate is this model given that EHR timecourses vary across tasks, brain areas, and individuals? We analysed EPI scans from 558 participants (CamCAN dataset: ages 18-88, 53% female; sensorimotor task involving auditory and visual stimuli and manual responses; SPM12/Matlab2019a). Three random-effects GLMs were tested, using either the canonical model (HRFc), the canonical with temporal and dispersion derivatives (HRFtd) to increase flexibility, or a more flexible Fourier basis set. Our approach also investigated experimental variance explained by one flexible model over and above the other by incorporating GLM regressors from both HRFtd and Fourier models (Figure 1A; only one condition was analysed, the seven Fourier regressors highlighted in blue, and three HRFtd regressors in red). First, consistent with our hypotheses the two flexible models explained experimental variance in more voxels within cytoarchitectonically defined primary motor cortex (4a and 4p), primary visual cortex (hOC1), anterior prefrontal cortex (Fp1), and cerebellar cortex, demonstrating the benefits of flexible modelling (Figure 1B). Second, we found that the timing of reconstructed EHRs from the flexible models did not correspond well to the canonical model (Figure 1C). Third, unexpectedly, the HRFtd model explained experimental variance more efficiently than the more flexible Fourier basis set in most regions. Our results suggest that flexible modelling is important, and that the HRFtd model is more optimal than the other approaches in most regions.



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Poster

P622. Functional Imaging: I

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P622.09

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: NR-017435
HL135562

Title: Acute effect of breathing awareness meditation on breathing rate and prefrontal cortex oxygenation as measured by functional near-infrared spectroscopy

Authors: *A. PAL[1], F. MARTINEZ[1], B. KRAUSE-SORIO[1], D. G. GHAHREMANI[2], A. MALHOTRA[4], R. M. HARPER[3], H. LAVRETSKY[3], P. MACEY[1];
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Abstract: Introduction: Mindfulness-based breathing awareness meditation (BA) is associated with reduced breathing rate (BR). Increased BR at the expense of tidal volume has often been associated with higher stress; BA technique could reduce stress by reducing BR. Additionally, activation of higher regulatory centers, like the prefrontal cortex (PFC), control the onset of stress associated emotional responses; thus, reduction of PFC activity would signify reduction in stress-associated emotional responses. In this study, we quantified the neural activity in response to BA meditation at left, medial and right PFC using functional near-infrared spectroscopy (fNIRS) and correlated that with changes in BR, in order to assess the stress-reduction effects of BA. **Materials and Methods:** In 18 participants (10 males, age=34±2years, BMI=25±1kg/m^2), BR was measured using a thoracic respiratory belt during the last 5 minutes of a 10-minute BA meditation task compared to 5 minutes of rest. Simultaneously, fNIRS was used to assess PFC oxygenation that correlates with neural activity. Changes in left, medial, and right PFC oxygenation during BA relative to rest were estimated and compared using repeated-measures ANOVA. In addition, changes in left, medial and right PFC oxygenation were correlated with changes in BR using Pearson's correlation. **Results:** With BA meditation, BR declined (mean±SEM, -18±9% (p<0.01)), right PFC oxygenation dropped by 1.4±0.7 times (p<0.01), medial PFC oxygenation declined by 1.2±0.4 times (p<0.05) and left PFC oxygenation non-significantly dropped by 0.7±0.3 times (p=0.16). BA task-related changes in BR significantly correlated with left PFC oxygenation at R= -0.7 (p<0.05), with no significant correlations at medial PFC (R=0.3) or at right PFC (R= -0.1).**Conclusions:** We found a decline in BR with BA meditation, consistent with previous studies. The most significant extent of BA-related changes was found in the right PFC; however, changes in the left PFC oxygenation correlated more with changes in BR. Breathing awareness meditation may reduce PFC neural activity as well as slow respiratory efforts. **Acknowledgements: NR-017435, HL135562.**

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Poster

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Program #/Poster #: P622.10

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIH Grant R42CA173976

Title: Exploring the Capacity of Resting-state fMRI vs. Breath-hold Cerebrovascular Reactivity Maps for Revealing Foci of Impaired Vasoreactivity

Authors: *N. J. FESHARAKI[1], W. E. HUDDLESTON[2], E. A. DEYOE[3];
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Abstract: Exploring the Capacity of Resting-state fMRI vs. Breath-hold CerebrovascularReactivity Maps for Revealing Foci of Impaired VasoreactivityAuthors
***N. J. Fesharaki[1,2]**, W. Huddleston[1], and E. A. DeYoe[21]College of Health Sciences, University of Wisconsin-Milwaukee, Milwaukee, WI; [2]Dept.Radiology, Medical College of Wisconsin, Milwaukee, WI.**Disclosures**
N.J. Fesharaki: None. **W. Huddleston:** None. **E.A. DeYoe:** None.
AbstractPathology-induced dropout in fMRI signals caused by local disruption of neuro-vascular coupling can compromise fMRI brain maps used for neurosurgical guidance. If not detected, such neuro-vascular uncoupling (NVU) can lead to inadvertent surgical resection of viable brain tissue. Potential NVU can be detected by mapping cerebrovascular reactivity (CVR) to blood CO2 changes induced actively by a breath-hold (BH) challenge or passively during resting-state (RS) fMRI. Holes in the CVR map that match fMRI dropouts can potentially identify sites of NVU. However, random holes do commonly occur in CVR maps even when thresholded to optimize unbiased coverage of gray/white matter. Thus, distinguishing random holes from NVU-related holes is a potential problem. To establish a normative baseline for differentiating random vs. NVU holes, we measured the spatial sampling statistics of different-size CVR holes in 9 healthy individuals. For BH fMRI, each subject's personalized respiratory response waveform was used to construct a regressor that was fit to the fMRI data for each brain voxel and the resulting amplitude was used as the CVR metric. For RS fMRI, signal power within a 0.01 - 0.08 Hz frequency band was used as the CVR metric. The spatial patterns of both metrics were individually thresholded to optimize gray vs. white matter coverage/segmentation. The frequency of occurrence of different-size holes in the resulting spatial patterns was then computed. The probability distributions of different-size holes were statistically indistinguishable

for the BH and RS CVR maps. From the frequency distributions, we determined a limiting hole size above which larger holes would be unlikely to randomly occur by chance (<5%). The BH and RS metrics had an identical hole-size limit, equal to 0.31 cm^3 (approx. 10 imaging voxels). Clinically, this establishes a potential baseline for CVR hole size that should be suspected of being NVU-related. Moreover, our results suggest that BH and RS CVR maps are comparable for assessing cerebral vasoreactivity and for identifying areas of NVU-impaired vasoreactivity.

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Poster

P627. Functional Anatomy of Gut Regulation

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Program #/Poster #: P627.01

Topic: F.07. Autonomic Regulation

Support: IndianaUniversity/Purdue University, Signature Center Initiative – Center for Spinal Cord and Brain Injury Research

Title: Sex difference in dendritic morphology of motoneurons innervating the external urethral sphincter in rats

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Abstract: A variety of sex differences are present in the anatomy and physiology of lower urinary tract, including differences in the bladder, urethra, and striated muscle of the external urethral sphincter muscle (EUS). As in humans, the EUS in rats is significantly smaller in females than in males. The motoneurons that innervate the rat EUS are located in the dorsolateral nucleus in the lumbosacral spinal cord. Although the number of motoneurons innervating the rat EUS does not differ across sexes, the size of the motoneuron somata is sexually dimorphic, being smaller in females than in males (McKenna and Nadelhaft, 1986). In this study, we examined another feature of EUS motoneuron morphology, the dendritic arbor, for evidence of sexual dimorphism.
Motoneurons innervating the external urethral sphincter in young adult male and female rats were labeled with cholera toxin-conjugated horseradish peroxidase (1 µL/side, 0.2%), and stereological assessments were made of motoneuron number (optical disector method) and soma size (Nucleator method) with Stereo Investigator. Dendritic arbors were reconstructed in three dimensions using Neuroluicda. Consistent with previous findings, the number of EUS motoneurons did not differ across the sexes, but somal areas of BHRP-labeled EUS motoneurons in female rats were significantly smaller (32%) that those of males. The dendritic arbors of EUS

motoneurons were strictly unilateral, with dense bundling dorsally along the lateral edge of the gray matter extending into lamina VI and VII as well as deep into the lateral funiculus; a second, medially-projecting array of dendrites coursed along the ventral margin of the gray matter and into lamina X. Although the distribution of dendrites was similar across sexes, total dendritic length in female rats was significantly shorter (34%) than that of males. Female rats had significantly less EUS motoneuron dendritic material in both the dorsolateral and medial projections, areas of important afferent drive to EUS motoneurons. The sex difference in soma size could reflect the sex difference we observed in dendritic length, with the larger dendritic arbor of EUS motoneurons in males requiring a larger soma size to support it. Furthermore, these differences in the EUS motoneurons of males and females could contribute to sex differences in urinary function or dysfunction.

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Poster

P627. Functional Anatomy of Gut Regulation

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Program #/Poster #: P627.02

Topic: F.07. Autonomic Regulation

Support: NIH Grant R01AI151552

Title: Unraveling the mechanism of spontaneous catecholamine release in the mesenteric lymph node

Authors: *R. BRANTLEY[1], A. E. ROSS[2];
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Abstract: AbstractUnraveling the mechanism of spontaneous catecholamine release in the mesenteric lymph nodeRebekah A. Brantley and Ashley E. RossThere is a growing interest to understand neurochemical-regulated communication between the gut, gut immune system, and brain. The mesenteric lymph node (MLN) is a secondary immune organ that resides in the gastrointestinal tract to help regulate immunity in the gut. The sympathetic nervous system (SNS) innervates the T-cell rich zones MLN to facilitate pro- and anti-inflammation responses by releasing neurotransmitters which modulate immune signaling. More specifically, the neurotransmitters norepinephrine and ATP, are known to be co-released from sympathetic neurons to function as immunomodulators. Previously, our lab has discovered spontaneous rapid catecholamine release using fast-scan cyclic voltammetry (FSCV); however, the mechanism of rapid catecholamine release in the MLN remains unknown. Here, we use FSCV at carbon-fiber microelectrodes in the MLN to probe the mechanism of norepinephrine release. We show that the rapid catecholamine transients are partially dependent on calcium signaling and are regulated

by the α2-adrenergic receptor. Overall, this work will advance understanding of catecholamine signaling in the MLN and will facilitate future studies investigating the mechanism of catecholamine immunomodulation.

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Poster

P627. Functional Anatomy of Gut Regulation

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Program #/Poster #: P627.03

Topic: F.07. Autonomic Regulation

Title: Large-scale tissue clearing of mouse and human whole gut for three-dimensional visualization and quantification of enteric nervous system

Authors: *Y. YUN[1], D. CHEON[1], K. CHUNG[1], T. SUNG[1], S.-J. KOH[1], Y. PARK[2], S.-B. RYOO[2], K. PARK[2], H. CHOI[1];
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Abstract: Nowadays, state-of-the-art tissue clearing methods enable to image thicker tissue section, or even whole organs imaging, by increasing tissue transparency and enhancing antigen-antibody binding. The goal of this research was to develop a 3D imaging method for the whole gastrointestinal(GI) tract that yields more information and insight about the enteric nervous system(ENS) than traditional 2D tissue section imaging. Visualization and quantification of the digestive organs(e.g. esophagus, stomach, small intestine, and colon) in the mouse and human were carried out through various techniques. A method encompassing tissue clearing, immunohistochemistry(IHC), confocal microscopy, light sheet fluorescence microscopy(LSFM), and quantitative analysis of full-thickness bowel without tissue sections has been developed for 3D imaging at high resolution. Furthermore, using surface rendering, volume rendering for all channels, fluorescence thresholding, and background subtraction, tools from in IMARIS, I was able to obtain an accurate 3D structure that can be measured volume. The multiscale structural decomposition of mouse and human ENS was clearly visualized in 3D. Tissue clearing method, which enables immunostaining of intact organs and visualizing of large volumes for 3D-reconstruction, could image the complex ENS network structure of myenteric plexus, submucosal plexus, and mucosal nerves. Similarly, the 3D ENS network structure of the esophagus(16 × 14 × 5.3 mm) and colon(1.2 × 1.3 × 1.4 mm) tissue samples were visualized in mouse and human, respectively. I investigated cholinergic ENS structure through the whole GI tract using mouse(n=3) and quantified the number of cell bodies and cell bodies per ganglion in two major layers. To identify the hubness of the myenteric plexus, I measured the number of ganglia and bridges without cell bodies that connect the ganglion. Quantitative data for

myenteric plexus and submucosal plexus showed relatively different aspects. I further succeeded in staining inhibitory neurons and excitatory neurons in mouse, respectively. This method can be adapted to mouse and human tissue clearing, which will enhance its applicability to the study of ENS structure and neural circuits. In particular, I provide a validation of this method for quantifying the number of cholinergic neurons. A quantitative analysis method of structure phenotypes in mouse will illuminate the potential usefulness of this technology as a diagnostic marker. For GI motility disorders, this novel technology will unravel the extensive spatial 3D network structure of neuro-immune interaction.

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Poster

P627. Functional Anatomy of Gut Regulation

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Topic: F.07. Autonomic Regulation

Support: NIH SPARC OT2 grant OD024899

Title: Transcriptomic Profiling of the Enteric Nervous System (ENS) in the pig and human colon: similarities and functional implications for the pig as a model in the translational research on the colon

Authors: T. LI[1,2], M. MORSELLI[1,3], T. SU[1,4], M. MULUGETA[1,2], M. LARAUCHE[1,2], *L. WANG[1,2,5], M. PELLEGRINI[1,3], Y. TACHE[1,2,5], P.-Q. YUAN[1,2,5];
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Abstract: The ENS controls a variety of gastrointestinal functions. We evaluated the translational relevance of the pig model for human colonic ENS by laser capture microdissection coupled with bulk RNA sequencing (RNA-seq) to cross-compare the transcriptomes in the myenteric and inner submucosal ganglia (MG, ISG) collected from the proximal, transverse and distal regions in 6 naïve pig (3 of each sex), and in parallel from the ascending, transverse and descending colon resected from the healthy margins of 12 patients (4 males and 8 females). The differentially expressed genes (DEG) enriched significantly ($p < 0.05$) by Gene Ontology terms with functional evidence were selected for comparisons among the segments, between MG and ISG and two species. The functional linkages (FLs) related to the orthologous DEG were extracted to reflect functional similarities across species. Nine and three pig orthologous genes

identified in the MG of human ascending and descending colon were involved in the regulatory networks in the MG of the corresponding pig colonic segments. The FLs related to these genes achieved 100% linkage coverage and represented all the characteristics of MG in the pig proximal and distal colon. These FLs accounted for 93 and 96% of all FLs of the regulatory networks responding to abdominal vagal nerve stimulation (VNS) in MG of the pig proximal and distal colon, suggesting that these two segments could serve as predictors in the translational research. The gene expression modules in the ISG and MG along the axis of pig proximal and distal colon were also characterized by bulk and single-cell RNA-seq. Effects of nitrergic neurons and glutamate-mediated synaptic plasticity were greatly enriched in the MG, which were involved in motility, while acetylcholine-dopamine balance enriched in the ISG played crucial roles in maintaining mucosal integrity. Compared with the proximal, the MG in the distal region contributed more to motility via combination of nitrergic and dopaminergic neurons, whereas MG in the proximal had greater complex neuronal wiring and cholinergic neurons played a leading role. Nitrergic neurons prevailed to regulate blood vessels with higher enrichment in ISG of distal colon. VNS affected primarily MG, especially in the proximal through cholinergic neurons and glia in the proximal and glutamatergic neurons in the distal. These data reveal for the first time the transcriptomic similarities in the colonic ENS of pig and human and regional heterogeneities in the pig, which helps to establish the use of pigs as a potential translational model for understanding of the human colonic functions and neuromodulation.

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Poster

P627. Functional Anatomy of Gut Regulation

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Topic: F.07. Autonomic Regulation

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Knut and Alice Wallenberg Foundation
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Title: Three-dimensional imaging reveals gradual degeneration of liver sympathetic nerves in human and experimental non-alcoholic fatty liver disease

Authors: *C. ADORI[1], T. DARAIO[2], R. KUIPER[2], S. S. BARDE[2], L. HORVATHOVA[3], T. YOSHITAKE[2], R. IHNATKO[4], I. VALLADOLID-ACEBES[2], P. VERCRUYSSE[2], A. M. WELLENDORF[5], R. GRAMIGNOLI[2], B. BOZOKY[2], J. KEHR[6], E. THEODORSSON[4], J. A. CANCELAS[5], B. MRAVEC[7], C. JORNS[8], E. ELLIS[2], J. MULDER[2], M. UHLEN[2], C. I.

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Abstract: Hepatic nerves have a complex role in synchronizing liver metabolism. We applied the 3D immuno-imaging technique iDISCO to explore the integrity of the hepatic nervous system in experimental and human non-alcoholic fatty liver disease (NAFLD). We demonstrated parallel signs of mild degeneration and axonal sprouting of sympathetic innervations in early stages of experimental NAFLD and a collapse of sympathetic arborization in steatohepatitis. The gene expression of several adrenergic receptors and the gap-junction protein CX32 were decreased, supporting a global sympathetic nerve-mediated communication impairment in experimental NAFLD livers. Human fatty livers displayed similar pattern of sympathetic nerve degeneration, correlating with the severity of NAFLD pathology. We showed that chronic sympathetic hyper-excitation is a key factor in the axonal degeneration, here genetically phenocopied in mice deficient of the Rac-1 activator Vav3. In experimental steatohepatitis, 3D imaging revealed a severe portal vein contraction, spatially correlated with the extension of the remaining nerves around the portal vein, enlightening a potential intra-hepatic neuronal mechanism of portal hypertension. These fundamental alterations in liver innervation and vasculature uncover significant novel neuronal components in NAFLD pathomechanism.

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Poster

P627. Functional Anatomy of Gut Regulation

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Topic: F.07. Autonomic Regulation

Support: NIH R01 DA043690
NIH R01 MH116026

Title: Functional Neuroimaging of the Autonomic Brain: A Meta-Analysis

Authors: *J. R. LOVELL[1], E. I. MURPHY[2], J. F. THAYER[3], R. LANE[4], M. A. LINDQUIST[5], P. J. GIANAROS[6], H. KOBER[7], T. D. WAGER[2];
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Abstract: Communication between the brain and the autonomic nervous system is integral to health, and dysregulated communication is a potential transdiagnostic factor across disorders. Human neuroimaging provides a unique opportunity to study the central brain circuits involved in autonomic regulation. However, brain-autonomic correlations have generally been studied in small samples, with heterogeneity across the autonomic measures collected in different studies. We conducted a meta-analysis of N = 151 contrasts that directly correlated brain (PET or fMRI) activity with autonomic measures from 112 studies (representing 1,543 total participants) using Multi-Level Kernel Density Analysis (MKDA). Included contrasts were weighted by their respective sample sizes, treated as random effects, and corrected at a family-wise error rate (FWER) of p&It;0.05. The results confirmed brain-autonomic relationships in both subcortical and cortical regions identified in previous studies and meta-analyses, as well as regions that were not previously reported. Analyses limited to pro-parasympathetic nervous system relationships revealed activations in previously identified regions including the basolateral amygdala and the default mode network (e.g., ventromedial prefrontal cortex and precuneus). We also found other regions that have not been consistently identified, including the putamen and the ventral attention network. Analyses limited to sympathetic nervous system relationships identified additional key areas of the diencephalon, particularly the habenula, which have been important in nonhuman experiments but not consistently identified in previous neuroimaging studies. By addressing prior methodological limitations and incorporating nearly a decade's worth of new brain imaging findings, the present meta-analysis provides an updated, comprehensive, and rigorous summary of human brain-autonomic correlations, which will serve as the basis for many future analyses and as a resource for the neuroimaging community.

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Poster

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Topic: F.07. Autonomic Regulation

Support: SPARC Program
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Title: Sparc: towards automatic detection and classification of propagating vagal nerve activity from transcutaneous recording for autonomic neuromodulation

Authors: S. AMIN[1], R. COIMBATORE[1], M. MCKINNON[3], A. GUPTA[4], T. H. EVERETT[4], T. L. POWLEY[2], T. V. NOWAK[4], I. P. CLEMENTS[3], *M. P. WARD[5];
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Abstract: Our previous work showed that gastric electric stimulation (GES) in human subjects with intractable gastroparesis evokes compound nerve action potentials (CNAPs) that can be measured using skin-mounted sensors overlying the mid-cervical vagal nerves (Ward et al., 2020). In this work, we collected body surface potential (BSP) recordings from human subjects in response to 12 combinations of GES parameters, using custom 32-channel multi-electrode arrays (MEAs) coupled to a specialized acquisition system for low-noise biopotential measurement (BioCircuit Technologies, Inc.). We used these data, along with median nerve (MN) recordings collected with the same design, to develop an algorithm that detects and classifies stimulus-evoked, propagating CNAPs from BSP recordings. The MEA recordings were analyzed as a 3D stack of 2D frames, where the third dimension is time, enabling the propagating CNAPs to be viewed as a video. Automating this process involves Dynamic Time Warping (DTW) to determine the latency, direction and speed of the propagating CNAP in 2D space. MEA recordings from MN stimulation (n = 13 sets) and a template containing distinct CNAPs were used to develop the process. Frame-by-frame DTW between the template signal and an MEA recording form a DTW matrix. Propagating is detected by thresholding the matrix data and analyzing the distribution using a histogram. The data in the lowest bin correspond to matching activity. Values corresponding to a propagating CNAP volley within the DTW matrix show sloping. We observed that for a particular polarity, a positive slope indicates that the CNAP was propagating in the same direction as the template, whereas a negative slope indicates the opposite. Once functionality was verified with MN data, we tested the algorithm on GES-evoked BSP data using a vagal CNAP template and recordings obtained from a human subject in response to a range of stimulus strengths. For the vagal template, a distinct afferent CNAP was manually segmented from a recording in response to a 10 mA stimulus. For each combination, we observed multiple instances of propagating CNAPs. The mean differences between visually and automatically detected CNAPs for the 12 GES parameter combinations were calculated, resulting in a mean and standard deviation of 1.28ms and 0.52ms. Automating the classification of propagating CNAPs (e.g., latency, speed and direction) could accelerate the development of

less invasive, more effective bioelectronics for autonomic and other disorders, especially in wearable bioelectronics. Additional verification and validation of algorithm functionality is required to fully define the potential of our approach.

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Poster

P627. Functional Anatomy of Gut Regulation

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Title: Multisegmental spinal sympathetic preganglionic projections to mouse paravertebral chain ganglionic neurons: anatomical considerations on neural unit size

Authors: *A. SOKOLOFF[1], M. SAWCHUK[1], C. LUGO[1], Y. LI[1], M. HALDER[1], M. L. MCKINNON[1], A. A. PRINZ[2], S. HOCHMAN[1];
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Abstract: Spinal cord sympathetic preganglionic neurons (SPNs) are the final arbiters of CNS sympathetic output. Many SPNs innervate the paravertebral sympathetic chain. Their axonal projections branch to issue divergent multisegmental projections on ganglionic neurons (GNs) across many segments. GNs are thought to enable amplification of CNS sympathetic commands. The ratio of GNs/SPNs has been reported to be 14:1 in mouse with each GN receiving convergent synaptic input from an average of 4.5 SPNs arising from many spinal segments. This organization has been compared to the concept of motor unit in motoneuron innervation of muscle fibers. Accordingly, the sympathetic neural unit size has been estimated to be 64 [(GNs/SPNs) x mean #SPNs synapses/GN] (Purves et al 1986 J Neurosci 6: 158-163). As synapse number was obtained with electrophysiology, recruitment assumes faithful spike propagation and transmission across all synapses in highly branched axons with modifiable conduction, so may be greatly underestimated. Individual GNs contain 100-200 anatomically-identified synapses in guinea pig (Gibbins et al 1998, JCN 402: 285-302) but this has not been examined in mouse thoracic paravertebral chain. We evaluated anatomical properties of SPN-to-GN convergence and divergence in whole mounts of adult mouse thoracic ganglia. TdTomato

reporter mouse crosses selectively labeled SPN axons (ChAT) and sparsely labeled GNs for morphologic reconstruction (TH). Additional IHC staining was used to estimate all GNs (TH) and all SPN synapses onto GNs (synapsin) as well as the fraction of synapses that originated from an individual spinal segment (after Neurobiotin [NB] injection into T7 ventral root). T5 ganglia contained 771±214 GNs (mean±SD; n=5) that had 1-7 primary dendrites (3.4±2.1; excluding GNs lacking processes). Synapsin^{+}/ChAT-Td^{+} synapses were estimated to be 116/GN. Conversely, T7 spinal SPNs labeled via NB injection had an average of 3.5, 3.7 and 2.3 synapses/GN (counted in T7, T6, and T5 ganglia, respectively). However, axonal divergence extended from T3-T12 ganglia, conversely suggesting that convergent input from other spinal segments would increase total synapse #/GN considerably. In sum, GNs in thoracic chain ganglia are innervated by many more SPNs than the 4.5 previously reported electrophysiologically. While this value is likely less than the 116 synapses/GN counted here, neural unit size and capacity for gain amplification is likely much greater than assumed. This is further supported by the wide range in spontaneous synaptic current amplitudes recorded in these GNs and their impact on synaptic gain (Prinz et al 2020, BMC Neurosci, 21(Suppl 1):P79).

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Poster

P646. Ingestive Behavior: Neuroendocrine Regulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P646.01

Topic: F.09. Food and Water Intake and Energy Balance

Support: DGAPA-PAPIIT IA204121
DGAPA-PAPIIT IN221819

Title: Metabolic effects of intranasal AdipoRon administration in obese male wistar rats

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Abstract: Signals indicating metabolic status, such as; ghrelin, leptin, insulin, and glucose are integrated in the hypothalamic arcuate nucleus. These metabolic cues can activate either the orexigenic Neuropeptide Y (NPY) and Agouti Related Peptide (AgRP) or the anorexigenic Proopiomelanocortin (POMC) expressing neurons. Thus, having regulatory effects on energy balance Although, there are other areas outside the hypothalamus that can also integrate this type

of information.The olfactory system including the olfactory epithelium and the olfactory bulb also characterizes for its ability to respond to metabolic hormones, which in these particular regions this signaling modulates olfactory sensitivity. For example, intranasal insulin administration decreases olfactory responses in humans; conversely, when ghrelin was administered, olfactory perception increases. These effects in olfactory sensitivity suggest the involvement of the olfactory system in metabolism.There is a multi synaptic connection between the olfactory bulb and the hypothalamus. Furthermore, the sensory, either visual or olfactory, perception of food rapidly changes the electrical activity of the ARC neurons, suggesting that the modulation of the responsiveness of the olfactory system modifies the activity of the hypothalamic circuits involved in the daily regulation of energy balance.The aim of the present study is to determine whether a pharmacological intervention in the olfactory system of obese rats may revert the effects of a high fat diet (HFD). For this we administered AdipoRon and agonist of the AdipoR1 and 2 adiponectin receptors. This particular hormone is an adipokine known to increase insulin sensitivity. Since AdipoR1 is highly expressed in the olfactory epithelium and the olfactory bulb we hypothesized that the intranasal administration of AdipoRon will modify odor sensitivity and enhance energy expenditure.Our results so far suggest that the intranasal administration of AdipoRon reduces olfactory sensitivity in both HFD and Control fed adult wistar rats. In addition, intranasal AdipoRon significantly decreases the glucose tolerance test of animals fed a 54% HFD for 10 weeks. Finally, we have observed changes in the expression of the proteins involved in Insulin response in the olfactory bulb and the hypothalamus of the animals treated with intranasal AdipoRon, suggesting that the intranasal administration of AdipoR1 agonists might be a potential therapeutic approach to revert the adverse metabolic effects of obesity.

Disclosures: A. Lagunes-Cruz: None. **M.A. Guzmán-Ruiz:** None. **R. Guevara-Guzman:** None.

Poster

P646. Ingestive Behavior: Neuroendocrine Regulation

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Program #/Poster #: P646.02

Topic: F.09. Food and Water Intake and Energy Balance

Support: NSF Grant IOS-1656626
UofSC SPARC Award
SCINBRE SIRP Award

Title: Role of serotonergic receptors in the leptin mediated raphe control of food intake

Authors: *N. D. MAXWELL[1], A. G. ROBU[1], A. T. SADEK[1], L. P. REAGAN[1,2], J. R. FADEL[1], C. A. GRILLO[1];

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Abstract: Leptin is an adipocyte-derived hormone that controls a myriad of homeostatic functions. Most notably, leptin acts on neurons in the hypothalamus through the long form of the leptin receptor to control energy homeostasis and appetite. Leptin has not only been shown to act on hypothalamic neurons, but also on other areas of the brain such as the dorsal raphe nucleus (DRN), the primary source of brain-derived serotonin (5-HT). Previously, we have described an anatomical connection between leptin sensitive neurons in the DRN and the arcuate nucleus of the hypothalamus. Additionally, we have shown that serotonin may be primarily involved in leptin's ability to reduce food intake through the DRN. Although these connections have been described anatomically and have shown to be important in the regulation of food intake, the mechanism of this signaling has yet to be fully explored. The serotonin 2C (5HT2C) receptor has been strongly implicated in the control of food intake, including as a potential target for the treatment of obesity. However, the link between the 5HT2C receptor and leptin responsive serotonergic neurons in the raphe has yet to be elucidated. In the present study, our objectives were to 1) characterize the expression and distribution of the 5HT2C receptor subtype in the hypothalamic nuclei, and 2) show the functional importance of this receptor subtype in the reduction of food intake when leptin is infused into the DRN. To complete these objectives, we used RNAScope to label 5HT2C receptor mRNA expression across the hypothalamic nuclei, as well as immunohistochemistry to further characterize the neurons that express these receptors. To show the involvement of this receptor on food intake behavior, we infused SB 242084, a 5HT2C inhibitor, intracranially followed by leptin into the DRN and measured food intake and activity. Through this work, we aim to further characterize this leptin-dependent pathway between the DRN and the hypothalamus, as well as how the leptin-mediated activation of this circuit regulates appetite.

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Poster

P646. Ingestive Behavior: Neuroendocrine Regulation

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

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Topic: F.09. Food and Water Intake and Energy Balance

Support: Samsung Science and Technology Foundation (Project Number SSTF-BA2001-09)
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NRF (National Research Foundation of Korea) Grant funded by the Korean Government (NRF-2019-Global Ph.D. Fellowship Program)

Title: A neural circuit mechanism for mechanosensory feedback control of ingestion

Authors: *M. KIM[1], D.-Y. KIM[1], G. HEO[1], H. KIM[1], H. KIM[1], J. JIN[1], S. JUNG[1], M. AN[1], B. H. AHN[1], J. PARK[1], H.-E. PARK[1], M. LEE[1], J. LEE[1], G. J. SCHWARTZ[3], S.-Y. KIM[2]; [2]Dept. of Chem., [1]Seoul Natl. Univ., Seoul, Korea, Republic of; [3]Medicine/ Neurosci., Albert Einstein Col. of Med., Bronx, NY

Abstract: Mechanosensory feedback from the digestive tract to the brain is critical for limiting excessive food and water intake, but the underlying gut-brain communication pathways and mechanisms remain poorly understood. Here we show that, in mice, neurons in the parabrachial nucleus that express the prodynorphin gene (hereafter, PB^{Pdyn} neurons) monitor the intake of both fluids and solids, using mechanosensory signals that arise from the upper digestive tract. Most individual PB^{Pdyn} neurons are activated by ingestion as well as the stimulation of the mouth and stomach, which indicates the representation of integrated sensory signals across distinct parts of the digestive tract. PB^{Pdyn} neurons are anatomically connected to the digestive periphery via cranial and spinal pathways; we show that, among these pathways, the vagus nerve conveys stomach-distension signals to PB^{Pdyn} neurons. Upon receipt of these signals, these neurons produce aversive and sustained appetite-suppressing signals, which discourages the initiation of feeding and drinking (fully recapitulating the symptoms of gastric distension) in part via signaling to the paraventricular hypothalamus. By contrast, inhibiting the same population of PB^{Pdyn} neurons induces overconsumption only if a drive for ingestion exists, which confirms that these neurons mediate negative feedback signaling. Our findings reveal a neural mechanism that underlies the mechanosensory monitoring of ingestion and negative feedback control of intake behaviors upon distension of the digestive tract.

Disclosures: M. Kim: None. **D. Kim:** None. **G. Heo:** None. **H. Kim:** None. **H. Kim:** None. **J. Jin:** None. **S. Jung:** None. **M. An:** None. **B.H. Ahn:** None. **J. Park:** None. **H. Park:** None. **M. Lee:** None. **J. Lee:** None. **G.J. Schwartz:** None. **S. Kim:** None.

Poster

P646. Ingestive Behavior: Neuroendocrine Regulation

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Program #/Poster #: P646.04

Topic: F.09. Food and Water Intake and Energy Balance

Support: DK107500 (DD)

Title: Using vasopressin-deficient Brattleboro rats to elucidate the role of glucagon-like peptide-1 in the control of fluid intake

Authors: *D. J. BRAKEY[1], K. C. SCHATZ[1], M. J. PAUL[1], D. DANIELS[2];
[1]Dept. of Psychology, Univ. at Buffalo-SUNY, Buffalo, NY; [2]Dept. of Psychology and Ctr. for Ingestive Behavior Res., Univ. At Buffalo - SUNY, Buffalo, NY

Abstract: Central glucagon-like peptide-1 (GLP-1) has been shown to play a role in the control of fluid intake; however, the sites of action are unknown. One problem in furthering our understanding of GLP-1's role in the control of fluid intake is that GLP-1 affects both drinking and feeding and these behaviors are intertwined. Our lab has begun using a rat with a genetic mutation that causes extreme polydipsia without affecting food intake (Brattleboro; BB) in order to disassociate these normally intertwined behaviors. We previously found that BB rats were hypersensitive to the fluid intake suppression by a GLP-1 receptor agonist, Exendin-4 (Ex4), without any differences from wild-type (WT) rats in food intake suppression by the same drug. In contrast, BB rats do not show a hypersensitive response to the GLP-1R antagonist, Exendin 9-39 (Ex9); only WT males drank more after a LV injection of Ex9 compared to vehicle. The absence of increased Ex9-induced drinking does not appear to be due to a ceiling effect, because further stimulation with hypertonic saline increased drinking in all groups. Under these stimulated conditions, Ex9 treatment did not impact drinking in any group. In addition to these behavioral experiments, we have begun to assess Ex4-induced Fos in areas commonly associated with food and fluid intakes in order to disassociate areas that may be important for the control of food, fluid, or both. In some brain regions, both sex and genotype impacted the number of Ex4-induced Fos-immunoreactive cells, although the pattern was not consistent across brain regions. For example, in the *organum vasculosum* of the *lamina terminalis* and median preoptic nucleus, Ex4 did not alter the number of Fos-immunoreactive cells in WT male or BB female rats, but there was an Ex4-induced decrease in BB male and WT female rats. In the subfornical organ, there was an Ex4-induced increase in male WT and female BB rats, but an Ex4-induced decrease in male BB and female WT rats. Lastly, in the paraventricular nucleus, there was an Ex4-induced increase in WT male rats, but a decrease in all other groups. In other regions there were no effects of sex or genotype - in the *nucleus tractus solitarius* and area postrema, there was an Ex4-induced increase in all groups. These experiments continue to provide important data that will be used to guide future studies to test for eating and drinking specific effects of GLP-1.

Disclosures: D.J. Brakey: None. **K.C. Schatz:** None. **M.J. Paul:** None. **D. Daniels:** None.

Poster

P646. Ingestive Behavior: Neuroendocrine Regulation

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Topic: F.09. Food and Water Intake and Energy Balance

Support: NARSAD Young Investigator
NIH DK121883
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NIH DA032750
NIH DA038168
NIH P30DA048736

Title: Fasting and feeding hormones differentially impact activity dynamics of transcriptionally distinct lateral hypothalamic glutamate neurons projecting to the lateral habenula and ventral tegmental area

Authors: *M. A. ROSSI[1,2], M. L. BASIRI[3,4], Y. LIU[4], K. HASHIKAWA[4], Y. HASHIKAWA[4], L. E. FENNO[5], Y. KIM[5], C. RAMAKRISHNAN[5], K. DEISSEROTH[5], G. D. STUBER[4]; [1]Rutgers RWJMS, New Brunswick, NJ; [2]Child Hlth. Inst. of NJ, New Brunswick, NJ; [3]Univ. of North Carolina At Chapel Hill, Chapel Hill, NC; [4]Univ. of Washington, Seattle, WA; [5]Stanford Univ., Stanford, CA

Abstract: The lateral hypothalamic area (LHA) regulates feeding and reward related behavior, but because of its molecular and anatomical heterogeneity, the functions of defined neuronal populations are largely unclear. Glutamatergic neurons within the LHA (LHA^{Vglut2}) negatively regulate feeding and appetitive behavior. However, this population comprises transcriptionally distinct and functionally diverse neurons that project to diverse brain regions, including the lateral habenula (LHb) and ventral tegmental area (VTA). To resolve the function of distinct LHA^{Vglut2} populations, we systematically compared their projections to the LHb and VTA using viral tracing, single-cell sequencing, electrophysiology, and *in vivo* calcium imaging. LHA^{Vglut2} neurons projecting to the LHb or VTA are anatomically, transcriptionally, electrophysiologically, and functionally distinct. While both populations encode appetitive and aversive stimuli, LHb projecting neurons are especially sensitive to satiety state and are bidirectionally modulated by the feeding hormones leptin and ghrelin. These data illuminate the functional heterogeneity of LHA^{Vglut2} neurons, suggesting that reward and aversion are differentially processed in divergent efferent pathways.

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Poster

P646. Ingestive Behavior: Neuroendocrine Regulation

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Program #/Poster #: P646.06

Topic: F.09. Food and Water Intake and Energy Balance

Support: NINDS R15NS095317 (Brower, CS)
TWU Research Enhancement Program (Na ES, Brower, CS)
TWU Startup funds (Na ES)

Title: Leptin effects on ATE1 expression

Authors: J. MANZANO[1], M. ALKHATATBEH[2], P. FRAYRE[2], *E. NA[2], C. BROWER[2];
[1]Biol., [2]Texas Woman's Univ., Aubrey, TX

Abstract: Arginyltransferase 1 (ATE1) is highly expressed throughout the mammalian brain and is essential for the Arg/N-degron dependent breakdown of proteins bearing acidic N-terminal amino acids. Loss of *Ate1* gene function in mice was shown to have effects on metabolism with ATE1-deficient mice expressing a lean phenotype and increased metabolism despite being hyperphagic. Leptin is an anorectic hormone produced by adipocytes. Given the metabolic phenotype of ATE1-deficient mice, we wanted to determine the effects of leptin on ATE1 expression levels in wild type mice. ATE1 expression was quantified in the hypothalamus, prefrontal cortex, hippocampus, and the cerebellum using qRT-PCR. Overnight fasted mice were injected with either PBS or leptin and samples were collected one hour post-leptin/PBS injection. Preliminary data suggest that leptin plays a role in the regulation of ATE1 expression; however the mechanism through which this occurs is not known. Future experiments will examine leptin effects on ATE1 expression in specific neuronal populations, and will explore mechanistically how the *Ate1* promoter is regulated.

Disclosures: J. Manzano: None. **M. Alkhatatbeh:** None. **P. Frayre:** None. **E. Na:** None. **C. Brower:** None.

Poster

P646. Ingestive Behavior: Neuroendocrine Regulation

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Program #/Poster #: P646.07

Topic: F.09. Food and Water Intake and Energy Balance

Support: Texas Woman's University Research Enhancement Program Award (Na, E.S, Brower, C.S)
Texas Woman's University Startup Funds (Na, E.S)

Title: Loss in ATE1 may affect energy metabolism through leptin

Authors: *P. FRAYRE[1], M. A. ALKHATATBEH[1], C. S. BROWER[1], E. NA[2];
[1]Biol., [2]Psychology & Philosophy, Texas Woman's Univ., Denton, TX

Abstract: Arginyl-tRNA protein transferase 1 (ATE1) regulates protein degradation by Arg/N-end rule pathway. Previous studies have shown that deletion or downregulation of ATE1 can lead to neurodegeneration, behavioral abnormalities, hyperphagia and lower body weight. To determine how loss of ATE1 affects energy metabolism and food intake, ATE1 deficient and ATE1 containing mice were given high fat diet for 6-8 weeks. Based on our current data, we found significant sex differences in body weight and food intakes, with ATE1 deficient mice less sensitive to the obesogenic effects of high fat diet even while consuming the same amount of high fat diet as controls. Interestingly, ATE1 deficient mice exposed to high fat diet displayed lower plasma leptin levels even after normalizing to white adipose tissue (WAT) weight. We also show sex differences in ATE1 deficient female mice given acute leptin treatment. Taken together, these data indicate that loss of ATE1 affects leptin levels independent of WAT but may alter leptin sensitivity in ATE1 deficient mice.

Disclosures: P. Frayre: None. **M.A. Alkhatatbeh:** None. **C.S. Brower:** None. **E. Na:** None.

Poster

P646. Ingestive Behavior: Neuroendocrine Regulation

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Program #/Poster #: P646.08

Topic: F.07. Autonomic Regulation

Support: NIH Grant

Title: Neuroendocrine mechanisms for social buffering of autonomic responses to stress

Authors: *R. OYAMA, I. CARCEA;
Rutgers Brain Hlth. Inst., Newark, NJ

Abstract: Social interactions can buffer the negative effects of stress, and the oxytocin hormone has been previously implicated, however the mechanisms remain poorly understood. We find that the oxytocin receptor is expressed in the posterior hypothalamus, a pre-autonomic structure that can modulate heart rate in rodents and in people. Other neurons in this area, particularly those expressing the receptor for the corticotropin-releasing factor, have been shown to project to brain stem structures modulating cardiac function, and their activation increased heart rate. We predict that neurons in the posterior hypothalamus expressing the oxytocin receptor will counteract these effects on heart rate, and that their activation during stress could buffer the effects of stress on heart rate.

Disclosures: R. Oyama: None. **I. Carcea:** None.

Poster

P646. Ingestive Behavior: Neuroendocrine Regulation

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Program #/Poster #: P646.09

Topic: F.09. Food and Water Intake and Energy Balance

Title: High salt intake increases RhoA-mDia1 signalling and triggers cytoskeleton reorganization in magnocellular vasopressin neurons

Authors: *B. LI, S. JACOB-THOMAS, M. PRAGER-KHOUTORSKY;
Dept. of Physiol., McGill Univ., Montreal, QC, Canada

Abstract: High dietary salt (HDS) strongly correlates with cardiovascular diseases and is a major factor contributing to the pathogenesis of hypertension. Recent studies show that HDS leads to neurogenically-mediated increases in sympathetic activity, vascular resistance, and water and sodium retention, revealing a possible involvement of central sodium detection mechanisms in salt-dependent hypertension. Changes in plasma sodium are detected by specialized osmosensory neurons located in the supraoptic and paraventricular nuclei (SON and PVN). Under normal physiological conditions, increased plasma sodium activates SON and PVN magnocellular neurons releasing vasopressin (VP), antidiuretic hormone causing renal water retention and vasoconstriction, to achieve body fluid homeostasis. Chronic exposure to HDS is associated with excessive activation of VP neurons, leading to a VP-mediated increase in blood pressure. The mechanisms underlying excessive VP secretion in HDS are not fully understood. Magnocellular VP neurons harbor unique cytoskeletal networks comprised of a subcortical actin layer, an array of actin comet-like structures, and somatic scaffold of interweaved microtubules, regulating the sensitivity of neuronal activation. Chronic exposure to HDS increases the density of actin and microtubule networks in magnocellular VP neurons. mDia1 is the major direct RhoA effector, mediating its effects on actin and microtubule polymerization and stability. Our data suggest that RhoA and mDia1 are elevated in the SON and PVN following HDS. We hypothesize that chronic exposure to HDS causes an activation of the RhoA-mDia1 pathway to increase the cytoskeletal density and osmosensitivity of magnocellular VP neurons, leading to excessive VP secretion, volume expansion, elevated blood pressure, and hypertension.

Disclosures: B. Li: None. **S. Jacob-Thomas:** None. **M. Prager-Khoutorsky:** None.

Poster

P682. Animal Behavior: Social Behavior and Cognition in Mouse Models

Location: Virtual Only

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Program #/Poster #: P682.01

Topic: H.06. Social Cognition

Support: NRF Grant 2020R1A2C4002156
DGIST Grant 21-CoE-BT-03
DGIST Grant-21-IJRP-01

Title: Interbrain synchrony across the brains of multiple mice

Authors: *J. LEE[1], G. LEE[2], D. KWAK[2], J. CHOI[3], S. LEE[4], H. CHOE[2];
[1]Brain and Cognitive Sci. Res. Ctr., [2]Dept. of Brain & Cognitive Sci., Daegu Gyeongbuk Inst. of Sci. and Technol., Daegu, Korea, Republic of; [3]Korea Inst. of Sci. and Tech. (KIST), Seoul, Korea, Republic of; [4]Electronics Telecommunications Res. Inst., Daejeon, Korea, Republic of

Abstract: Recent neural evidence indicates that there is interbrain synchrony across animals interacting in a naturalistic social environment. However, due to technological constraints, most studies reported synchronous neural activities from up to two animals. In this study, we used CBRAIN (collective brain research platform aided by illuminating neural activity), a minimized edge-computing solution with wireless communication capabilities, to record electrophysiological signals from multiple mice simultaneously. Four mice were allowed to interact freely with each other or were individually separated, while their behaviors and electrophysiological signals were recorded for three hours. The device implants were well-tolerated after habituation sessions as mice returned to the baseline huddling behavior, a well-known example of prosocial behavior exhibited by mice in which they spontaneously aggregate in a group. We analyzed whether the huddling behavior affects interbrain synchrony across a group of mice. Power spectral density (PSD) estimates of local field potential (LFP) recordings in the dorsomedial prefrontal cortex (dmPFC) showed differential patterns of powers at low- and high- frequencies between huddling and non-huddling groups of mice. The huddling-specific patterns were distinct from the differences arising from active/inactive disparity. Furthermore, the mice in the group condition showed correlated patterns of LFP powers in the time domain, compared to the mice in the single-mouse condition, which showed no correlation, suggesting the existence of the interbrain synchrony. To our knowledge, this is the first study that demonstrated the interbrain synchrony in a larger group of animals up to four in a naturalistic social situation.

Disclosures: J. Lee: None. **G. Lee:** None. **D. Kwak:** None. **J. Choi:** None. **S. Lee:** None. **H. Choe:** None.

Poster

P682. Animal Behavior: Social Behavior and Cognition in Mouse Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P682.02

Topic: H.06. Social Cognition

Support: NRF-2020R1A4A1018714

Title: The effects of chronic social isolation in adulthood on social behaviors

Authors: *H. LEE, Y. JEONG;
Korea Advanced Inst. of Sci. and Technol., Daejeon, Korea, Republic of

Abstract: Social isolation (SI) is a global problem that can lead to an aversive emotional state in humans and influences health across the life course. Since the global impact of the COVID-19 pandemic has increased the consequences of social isolation, it is important to investigate the effects of chronic social isolation on the ability to engage in social interaction. However, the aspects of social behaviors and related neuronal communications among social brain networks that are more sensitive to chronic social isolation in adulthood have not been fully investigated. Here, we evaluated social behaviors in chronic single-housed adult mice for 8-10 weeks to recapitulate the impacts of being socially isolated in adults. We also performed local field potential (LFP) recordings in several brain regions including the medial prefrontal cortex (mPFC) and nucleus accumbens (NAc), which are related to social cognition and behaviors. In addition, anterior insula (AI), which is important to social emotion, was also recorded and evaluated the relationships between social networks during social avoidance and interaction. We found isolation-induced social withdrawal behaviors in the chronic adult SI model during direct interaction with the juvenile conspecifics. Furthermore, chronic SI mice were divided into two groups based on the level of withdrawal behaviors: susceptible and resilient SI mice exhibited high and low social withdrawal behaviors, respectively. Although resilient SI mice showed similar sociability as those of the control group housing (GH) mice, susceptible SI mice exhibited significantly reduced sociability compared with GH or resilient SI mice. Reduced sociability in resilient SI mice is associated with decreased low theta (4-8 Hz) coherence between left AI and left mPFC or right NAc, which was not visible in resilient SI mice. In addition, decreased low theta coherence between left mPFC and right NAc was also observed. These results may not only facilitate an understanding of specific neural circuit dynamics for social behavioral changes in chronic individual housing in adulthood but may also uncover the important role of AI in representing the susceptibility to negative social experiences.

Disclosures: H. Lee: None. **Y. Jeong:** None.

Poster

P682. Animal Behavior: Social Behavior and Cognition in Mouse Models

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P682.03

Topic: H.06. Social Cognition

Support: NSERC Grant 400212

Title: Effects of oxytocin receptor antagonist on estrogens' rapid enhancing effects on social recognition in the medial amygdala

Authors: *C. SEXTON, E. CHOLERIS;
Univ. of Guelph, Guelph, ON, Canada

Abstract: Social recognition (SR) is a critical cognitive skill required for animals' successful participation in its social group and is widely regarded as necessary for individual survival. Estrogens and three of their receptors, ERα, ERβ, and GPER1 have been shown to impact SR performance. Estrogens have also been indicated to interact with other neurochemicals such as oxytocin (OT). Previous research has shown infusions of a subeffective dose of an oxytocin receptor antagonist (OTRA) into the medial amygdala (MeA) prior to an infusion of 17-β estradiol (E2) into the PVN prevents E2 rapid facilitating effects on SR. E2 and agonists for all 3 ERs rapidly facilitate SR even when infused directly in the MeA. Whether an E2/OT interaction occurs also within the MeA, is unknown. If such interactions exist, then the OTRA will block the enhancing effects of E2, at a dose that does not block the natural occurrence of SR. 10nM and 25nM doses of E2 were employed to examine possible dose differences in their ability to be blocked by the OTRA. The focus is on female mice, as estrogens have been shown to have more consistent and significant enhancement effects on SR than males. Ovariectomies are conducted to reduce circulating estrogen levels to those of diestrus, and cannulae are surgically implanted into the MeA. The test mouse is initially administered 75nM OTRA, which was previously shown to be a subeffective dose, or a vehicle of artificial cerebrospinal fluid (aCSF) into the MeA. After two minutes the test mice then receive either 25nM or 10nM of E2 or aCSF into the MeA. Next, the test mouse receives a "difficult" social recognition paradigm. Additionally, an object recognition paradigm is employed to determine whether the facilitating effects of 25nM E2 in the MeA previously found are specific for SR. It is predicted that the infusion of the OTRA will prevent the enhancing effects of E2 in the SR paradigm. Furthermore, we anticipate E2 will not facilitate object recognition in the MeA, as this region has shown to be highly specific for social information processing. Preliminary results indicate E2 when paired with aCSF facilitates SR, but when paired with the OTRA does not, supporting our hypothesis. Combined, this research program elucidates the mechanisms of estrogens, specifically those of E2, rapid enhancing effects on female SR in the MeA, which is a key component of the social brain.

Disclosures: C. Sexton: None. **E. Choleris:** None.

Poster

P682. Animal Behavior: Social Behavior and Cognition in Mouse Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P682.04

Topic: H.06. Social Cognition

Support: Harris Research Endowment
Drake Undergraduate Science Collaborative Institute

Title: Performance of 3xTg-AD mice in a social task: identification of confounds in the behavioral phenotype

Authors: A. K. KLEIN, N. J. ANDERT, B. A. SULKOWSKI, *C. C. WRENN;
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Abstract: Behavioral and psychological symptoms of dementia include mood disturbances, psychosis, aggression, and inappropriate social behavior. These problems occur in patients with Alzheimer's Disease (AD) and are detrimental to overall health. The 3xTg-AD mouse model of AD has been reported to have social deficits. Our study sought to explore this phenotype along with determining whether any observed social impairments result from alterations in other behavioral domains. Our first experiment tested 3xTg-AD mice in the three-chamber social task. We found that, unlike wild-type (WT) controls, 3xTg-AD mice lacked a preference for social novelty. To determine whether this impairment was specific to the social domain, we repeated the experiment using objects as stimuli rather than conspecifics. We found a similar lack of preference for a novel object in the 3xTg-AD mice showing that the social test results are generalizable to other stimuli and not specifically social. We hypothesized two possible explanations for this generalized lack of preference for novelty: a neophobic, anxiety-like phenotype or impaired habituation to stimuli. After testing these hypotheses, we found an anxiety-like phenotype in 3xTg-AD mice in the elevated-plus maze. Additionally, habituation experiments found impaired habituation to both social and non-social stimuli. These findings challenge the conclusion that observed impairments in the three-chamber social test are due to social deficits *per se.* In total, this work highlights the importance of addressing the contribution of non-social confounds to behaviors in the three-chamber social task.

Disclosures: A.K. Klein: None. **N.J. Andert:** None. **B.A. Sulkowski:** None. **C.C. Wrenn:** None.

Poster

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Program #/Poster #: P682.05

Topic: H.06. Social Cognition

Title: Male Mice Demonstrate Helping Behavior Releasing a Trapped Conspecific.

Authors: *G. L. PARRISH[1], A. PORRAS[1], A. E. HOWARD[1], M. N. RODRIGUEZ[1], S. L. LIPPI[1], C. M. KREITLER[1], J. A. OLIVAREZ[2];
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Abstract: Research on prosocial rodent behavior is growing within the fields of cognitive psychology and social neuroscience. There remain essential questions about variables influencing this empathetic behavior. There are discussions and disagreements on whether this behavior is empathetically motivated and correctly labeled as prosocial or whether the behavior is motivated by social contact and curiosity. The present study utilized a helping behavior paradigm and apparatus analysis on 16 adolescent male mice offspring expressing the genotypes tau (with no promoter) or promoter (and no tau). The apparatus in this research is a modified version of the apparatus used by Sato and colleagues (2015), who demonstrated helping behavior in soaked rats. In this paradigm, a mouse can release a familiar cage mate and a non-familiar mate from a restricted water box. The number of friend and food door touches and latency to make a decision, either free the trapped conspecific or choose the food box, was recorded for each mouse. The first six days were stress-free, alternating days testing cagemates and non-cagemates. A damp bedding stressor was implemented for the last six days an hour before mice were evaluated in the apparatus. This research study sought to investigate whether empathy, sympathy, and prosocial behavior can be displayed in a mouse model. Repeated measures ANOVAs revealed several significant results were found regarding stress and empathetic behavior. Findings may aid in understanding the process of empathy and have diagnostic and therapeutic implications in understanding social and emotional impairments seen in individuals with Autistic Spectrum Disorder and substance abuse disorders.
Keywords: prosocial, empathy, apparatus, mouse, cagemate, non-cagemate

Disclosures: G.L. Parrish: None. **A. Porras:** None. **A.E. Howard:** None. **M.N. Rodriguez:** None. **S.L. Lippi:** None. **C.M. Kreitler:** None. **J.A. Olivarez:** None.

Poster

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Program #/Poster #: P682.06

Topic: H.06. Social Cognition

Support: Seaver Autism Center at Mount Sinai
NIH Grant

Title: Studying the impact of Shank3-deficiency on neural circuits of social reward

Authors: *M. BARBIER[1], K. THIRTAMARA RAJAMANI[1], S. NETSER[2], S. WAGNER[3], H. HARONY-NICOLAS[1];
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Abstract: Background: Social deficits are a core symptom of autism spectrum disorder (ASD). Clinical studies have implicated the mesoaccumbens reward circuit in autism spectrum disorder. However, the causality between alterations in this system and social deficits has not been established. The ventral tegmental area (VTA), a core node of the mesolimbic circuit, is interconnected with the nucleus accumbens (NAc) via the VTA dopaminergic projections. Despite the role of the reward system in social interaction, little is known about the impact of ASD associated mutations on processing social reward and on the functional integrity of this pathway. In this work, we study the effect of a mutation in an ASD high-risk gene, *Shank3*, on the mesoaccumbens system in rats. We hypothesize that *Shank3* mutation impacts neural activity in the mesoaccumbens system, causing abnormalities in accumbal dopamine transmission and leading to impairments in processing social reward. **Methods:** To identify abnormalities in dopamine transmission in *Shank3*-deficient rats that correlate with deficits in processing social reward, we used fiber photometry to record in the VTA in combination with a dopamine sensor in the NAc during a social reward paradigm. In this paradigm we introduced two rewarding stimuli, social and food, during satiety and food deprivation and examined investigation time for each reward during the two conditions. To control for attentional deficits, we used the same paradigm, but replaced the social stimuli with a moving toy rat. To rule out reduced motivation to food or impairment in food consumption, we assessed food consumption. **Results**: We found that *Shank3*-deficient rats have deficits in processing social reward that are associated with perturbation in VTA neural activity and an intact attention and food consumption. **Conclusions**: Our study demonstrates that *Shank3*-deficient rats have a specific deficit in processing social reward and provides a first step toward understanding the role of *Shank3* in the reward system, and how *Shank3*-deficiency may leads to social deficits.

Disclosures: M. Barbier: None. **K. Thirtamara Rajamani:** None. **S. Netser:** None. **S. Wagner:** None. **H. Harony-Nicolas:** None.

Poster

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Program #/Poster #: P682.07

Topic: H.06. Social Cognition

Support: NIH Grant RO1MH107535

Title: Postnatal environment modulates social dominance

Authors: *K. LOPEZ[1], F. TAKI[2], J. G. TOTH[2], M. TOTH[2];
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Abstract: In social living groups, members compete for access to resources and mating opportunities and their repetitive interactions create a social order with dominant and subordinate individuals. Little is known about the causative factors that drive the formation of dominance hierarchies and the eventual social rank of an individual. Genetic factors can play a role, but hierarchy develops even in a genetically homogenous group of animals and under controlled laboratory conditions. This raises the possibility that an individual's social dominance is established by early environmental influences. Indeed, we found that voluntary wheel running of dams during the lactation period increased the social dominance of offspring compared to offspring from sedentary mothers, as measured by the tube test. In agreement with these findings, I found that pyramidal neurons in layer 2/3 in the prelimbic region of the medial prefrontal cortex (mPFC) of these offspring exhibited increased length and number of nodes in apical dendrites at the rostral region. This increased arborization suggests increased input to the area. Thus, the effects of physical activity by the mother extended to the non-exercising offspring. However, although runner offspring are primed for an elevated rank, its manifestation is dependent on the post-weaning environment. Post-weaning in a socially deprived state led to the offspring of runner mothers exhibiting subordination. However, their elevated rank was restored with a bout of sexual interaction in adulthood. This suggests that dominance can be programmed by the mother during the postpartum period, but this enhancement in rank develops through exposure to a social environment. Future studies aim to understand the mechanism underlying this primed socially dominant state by profiling mPFC nuclei to determine genes and molecular pathways targeted. This research may help to better understand how the maternal and the offspring environment, individually and in combination, determine social status.

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Poster

P682. Animal Behavior: Social Behavior and Cognition in Mouse Models

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Topic: G.03. Motivation

Support: Hungarian National Research, Development and Innovation Office via NKFIH-4300-1/2017-NKP_17 (National Brain Research Program)
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Title: Preoptic GABAergic neurons control parenting and depression

Authors: *D. DIMEN[1,2], G. PUSKA[2], V. SZENDI[2], E. SIPOS[3], D. ZELENA[4], A. DOBOLYI[2]; [1]Indiana Univ. Program in Neurosci., Bloomington, IN; [2]MTA-ELTE Lab. of Mol. and Systems Neurobiology, Dept. of Physiol. and Neurobio., Eötvös Loránd Res. Network and Eötvös Loránd University, Budapest, Hungary, Budapest, Hungary; [3]Dept. of Behavioral and Stress Studies, Inst. of Exptl. Med., Budapest, Hungary; [4]Ctr. for Neuroscience, Szentágothai Res. Centre, Inst. of Physiology, Med. Sch., Univ. of Pécs, Pécs, Hungary

Abstract: Parenting is a critically important behavior in mammals associated with anxiety and occasionally with depression. Here, the role of preoptic GABAergic inhibitory neurons was addressed in parenting, anxiety and depression. Pup exposure and forced swimming resulted in similar c-Fos activational pattern in neurons expressing vesicular GABA transporter in the preoptic area in females. In males, the number of pup exposure-activated neurons was generally less, with the highest density in the central part of the medial preoptic nucleus. Chemogenetic stimulation of preoptic GABAergic cells resulted in elevated maternal motivation and caring behavior in females and mothers but aggression towards pups in males. Behavioral effects were the opposite following inhibition of preoptic GABAergic neurons suggesting their physiological relevance. In addition, increased anxiety- and depression-like behaviors were found following chemogenetic stimulation of the same neurons in females while previous pup exposure increased only anxiety-like behavior. A sexually dimorphic projection pattern of preoptic GABAergic neurons was also identified, which could mediate sex-dependent parenting and associated emotional behaviors.

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Poster

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Program #/Poster #: P697.01

Topic: G.05. Mood Disorders

Support: Intramural Research Program at the National Institute of Mental Health, National Institutes of Health

Title: Rates of temporal discounting and lateral intraparietal sulcus gamma power are associated in patients with mood disorders.

Authors: *G. E. ANDERSON, D. K. GREENSTEIN, C. R. BURTON, J. R. GILBERT, E. D. BALLARD, C. A. ZARATE, Jr.;
Section on the Neurobio. and Treatment of Mood Disorders, Natl. Inst. of Mental Hlth., Bethesda, MD

Abstract: Impulsivity, reward processing, and psychopathology can be understood using delay discounting tasks, a neuroeconomics metric that quantifies the decline in the current value of a reward in relation to the delay of its receipt. Patients diagnosed with major depressive disorder, bipolar disorder, and suicidal ideation have been shown to value a lower current reward over a higher delayed reward (i.e., increased delay discounting) compared to healthy control participants (HCs). In addition, both human and primate studies have demonstrated that activity within the lateral intraparietal sulcus is associated with individual-specific rates of temporal discounting. Here, we extend these findings by examining electrophysiological correlates of temporal discounting in patients with a mood disorder diagnosis (MDs, *n*=28) and HCs (*n*=19), focusing on the gamma (30-58 Hz) band, a proxy measure of excitation-inhibition balance, which is often dysregulated in mood disorders. In this ongoing study, a delay discounting task was administered behaviorally alongside resting state magnetoencephalography (MEG) scanning. MDs included patients with a recent suicide attempt (i.e., within two weeks, AS, *n*=8), patients with a history of suicide attempt but no attempt or ideation within the last year (HS, *n*=20), and HCs. Prior to scanning, participants completed the the 27-item Monetary Choice Questionnaire (MCQ). An overall *k* value was calculated from the MCQ, with higher *k* values indicating higher discounting rates. Up to one week later, participants completed MEG scanning, during which they rested with their eyes closed. Synthetic aperture magnetometry was used to project gamma power using a realistic head model and covariance matrix regularization on a 5-mm grid over the entire rest period. Analysis of variance was then used to evaluate group (AS, HS, and HC) and discounting rate effects on gamma power. While there was no effect of diagnostic group on gamma power, there was an effect of discounting rate, such that individuals who demonstrated a higher *k* value (i.e., higher discounting rate of the delay) showed reduced gamma power in regions of the lateral intraparietal sulcus and frontal lobe. These findings align with previous studies, implicating this region in delay discounting, and suggest that alterations in gamma power are associated with discounting rates in both MDs and HCs. Future work will explore the temporal dynamics of gamma band changes in this region, as well as the relationship between *k* and other frequency bands. Further, we aim to evaluate the relationship between *k* and other clinical measures in our MD sample, such as impulsivity, anhedonia, and symptom severity.

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Poster

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P697.02

Topic: G.05. Mood Disorders

Title: Optimizing analytic pipelines of reliable diagnostic biomarkers for major depressive disorder using resting state fMRI

Authors: *Y. TAKAHARA[1,3], Y. KASHIWAGI[3,1], T. TOKUDA[1], J. YOHSHIMOTO[5,1], Y. SAKAI[1], A. YAMASHITA[6,7,8], T. YOSHIOKA[2], S. MOMOSAKI[9], K. OGAWA[4], M. KAWATO[1,10], O. YAMASHITA[1,10];
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Abstract: Most of psychiatric disorders, including major depressive disorder (MDD), are generally diagnosed by medical interviews and visible signs, but objective diagnostic and stratification biomarkers developed with resting-state functional magnetic resonance imaging (rs-fMRI) data is expected to contribute to more effective treatment. There is, however, no widely accepted biomarkers due to the absence of optimal analytic pipelines for the development of such biomarkers with sufficient generalization capabilities. In this study, we conducted a comprehensive evaluation of combining analytic methods in order to identify optimal pipelines for developing MDD diagnostic biomarker with generalization performance. We considered 4 processes of developing biomarkers: brain parcellation, estimation of functional connectivity (FC), site difference harmonization, and machine learning. We constructed 360 different MDD diagnostic biomarkers using a large-scale multi-site rs-fMRI dataset (138 MDD patients and 545 healthy subjects from 4 imaging sites) and evaluated classification performances with nested 10-fold cross validation method similarly to Yamashita et al. (*PLOS Biology*, 2020). To confirm the reliability of constructed biomarkers, we also conducted validation tests with the independent Japanese dataset and Russian open-source dataset respectively, and identified several biomarkers with high classification performances (AUC: 0.71~). Among these biomarkers, FCs between subcortical and somatomotor area were commonly identified as important connectivity for classification despite the differences of parcellations and FC combination. Besides, some of these biomarkers have shown the fluctuation with treatment and showed potential for monitoring biomarkers. Lastly, using the analytic pipelines for developing generalizable MDD diagnostic biomarkers, we also succeeded in the development of diagnostic biomarkers with high classification performances for the other psychiatric disorders (autism spectrum disorder and schizophrenia, AUC: 0.88~). This result suggested that our identified pipelines possess generalizability across several psychiatric disorders. Furthermore, combining constructed multiple disorder biomarkers potentially could contribute to the understanding of psychiatric disorders, not bound by conventional categorical disease classification.

Disclosures: **Y. Takahara:** A. Employment/Salary (full or part-time):; SHIONOGI & CO., LTD.. **Y. Kashiwagi:** None. **T. Tokuda:** None. **J. Yohshimoto:** None. **Y. Sakai:** None. **A. Yamashita:** None. **T. Yoshioka:** None. **S. Momosaki:** None. **K. Ogawa:** None. **M. Kawato:** None. **O. Yamashita:** None.

Poster

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Topic: G.05. Mood Disorders

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Title: Psilocybin therapy enhances cognitive and neural flexibility in patients with major depressive disorder.

Authors: *M. K. DOSS[1], M. POVAŽAN[4], M. D. ROSENBERG[5], N. D. SEPEDA[1], A. K. DAVIS[6], P. H. FINAN[2], G. S. SMITH[2], J. J. PEKAR[7], P. B. BARKER[3], R. R. GRIFFITHS[1], F. S. BARRETT[1];
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Abstract: Psilocybin has shown promise for the treatment of depression, a family of disorders often accompanied by cognitive rigidity. One mechanism by which psilocybin could reduce depressive symptomology is by increasing cognitive and neural flexibility. In an open-label study of 24 patients with major depressive disorder, we tested the enduring effects of psilocybin therapy on depression, cognitive flexibility (perseverative errors on a set-shifting task), and neural flexibility (dynamics of functional connectivity or dFC via functional magnetic resonance imaging). Psilocybin therapy reduced depression and increased cognitive flexibility for at least four weeks post-treatment, though these improvements were not correlated. One-week after psilocybin therapy, glutamate and N-acetylaspartate concentrations (via magnetic resonance

spectroscopy) were decreased in the anterior cingulate cortex (ACC), and dFC was increased between the ACC and the posterior cingulate cortex (PCC). Surprisingly, these increases in dFC between the ACC and PCC were associated with less improvements in cognitive flexibility after psilocybin therapy. Connectome-based predictive modeling demonstrated that baseline dFC emanating from the ACC predicted improvements in cognitive flexibility. In these models, greater baseline dFC was associated with better baseline cognitive flexibility but less improvements in cognitive flexibility. These findings show that psilocybin therapy in patients with depression can enhance cognitive flexibility and neural flexibility, especially of the ACC, but suggest a nuanced relationship between neural flexibility and depression symptomology. Whereas some enduring increases in neural dynamics may allow for shifting out of a maladaptively rigid state, larger persisting increases in neural dynamics may be detrimental to the efficacy of psilocybin therapy.

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Poster

P697. Human Imaging and Behavioral Studies: Focus on Depression

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Topic: G.05. Mood Disorders

Support: National Institutes of Health Predoctoral Training Grant (T32-GM108540)
Kiwanis Neuroscience Research Foundation
NIMH (1 P50 MH094258)

Title: Depression symptoms in neurological patients: Large-scale survey and lesion symptom mapping of patients with focal brain lesions

Authors: *E. BRANDT[1], N. TRAPP[1], J. BRUSS[1], O. ABU-HALAWA[1], S. LI[1], F. ALBAZRON[1], K. MANZEL[1], J. GRAFMAN[2], A. BOES[1], D. TRANEL[1];
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Abstract: Background. Examining depression following neurological injury is useful for our understanding of post-lesion depression and depression more generally. However, the extant literature shows inconsistent findings when it comes to the demographic and neural correlates of depressive symptoms following a neurological event. Thus, the present study aims to

characterize depression symptoms in a large sample of individuals in the chronic phase of recovery from a focal brain lesion. **Methods.** We sampled 526 individuals across two locales. The patients had focal, stable brain lesions and were in the chronic phase of recovery (> 3 months post onset). First, mean Beck Depression Inventory - Second Edition (BDI-II) scores were examined and individual demographic (gender, years of education), time (age at lesion onset, time since lesion onset), and lesion (lesion laterality, lesion etiology) factors were used to predict depressive symptoms measured by the BDI-II. Second, multivariate lesion-symptom mapping was used to identify lesion locations and networks associated with depression symptoms in order to identify lesion locations that might be associated with "risk" or "resilience" for developing depression post-lesion. **Results.** We found that on average, individuals in the chronic phase of recovery from focal brain lesions do not exhibit depressive symptoms that are appreciably different from the general population. Individual characteristics were not predictive of depressive symptoms, with the exception of age at lesion onset, where younger age at lesion onset was associated with elevated depressive symptoms. Peak "risk" regions for post-lesion depression include the bilateral insula and left prefrontal cortex. Peak "resilience" regions - viz., areas where damage was associated with lower depression - included the right ventromedial prefrontal cortex, right orbitofrontal cortex, and right inferolateral temporal cortex. Seed-based network analysis demonstrated peak "risk" region overlap with the ventral attention/salience network and the peak "resilience" region overlap with the default mode network. **Conclusion.** Depression is a heterogeneous disorder, but our results suggest that some individuals may be at increased risk for developing depression post-lesion while others are protected against the development of depression. This may be mediated by individual factors such as age at lesion onset or lesion location. These findings have important implications for the diagnosis, prognosis, and treatment of depression.

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Poster

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Topic: G.05. Mood Disorders

Support: National Institute of Mental Health (K23MH112769)
Brain and Behavior Research Foundation

Title: Alterations in Cortical Gray Matter Associated with Cardiac Vagal Tone and Remitted Major Depressive Disorder

Authors: *S. GUPTA[1], L. M. JENKINS[3], M. KOUCHAKIDIVKOLAEI[4], K. POPURI[4], J. WU[2], Z. YE[5], L. WANG[6], S. A. LANGENECKER[7], J. P. STANGE[8];
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Abstract: Major Depressive Disorder (MDD) is a highly debilitating disorder that affects at least 16% of adults in the US over their lifetime. Only a few studies have examined structural brain differences in early course remitted MDD (rMDD) and even fewer have determined whether these changes are associated with biomarkers of emotion regulation capacity. We aimed to identify structural brain differences in rMDD and determine how these differences relate to cardiac vagal tone, a measure of parasympathetic nervous system activity and regulatory capacity. T1 weighted MRI was obtained on a 3T GE scanner for 26 young adults with rMDD (mean=27.7 years, SD=4.1) and 18 healthy controls (HC) (mean=26.1 years, SD=3.8). Diagnosis was confirmed based on the DSM-5 criteria for lifetime MDD. Physiological self-regulation was measured using a Hexoskin, which collected participant biometric data, and ecological momentary assessments for 7-days. Resting respiratory sinus arrhythmia (rRSA), a measure of heart rate variability in the respiration cycle indicative of vagal tone associated with emotion regulation capacity, was measured using a Biopac MP160 and analyzed with AcqKnowledge software using the peak-valley method. Imaging data were processed through Freesurfer to calculate cortical measures with the Desikan-Killiany atlas. The groups were compared using a GLM and smoothing was performed using a kernel size of 5 mm. Multiple comparisons were corrected using the Monte Carlo Null-Z simulation with a vertex-wise cluster threshold of 1.3. Relative to HC, individuals with rMDD had a smaller surface area in the right rostral middle frontal gyrus (rRMF) (rMDD mean=6265 mm^2, SD=686; HC mean=6884 mm^2, SD=776; p=0.004). Pearson correlation found that decreased volume in rRMF was associated with lower rRSA (r=0.36, p=0.020). Our findings build on previous research identifying cortical reductions in MDD patients in the dorsolateral prefrontal cortex, which is critical for executive functioning including inhibitory control, and is hypothesized to contribute to depression via its role in top-down cognitive regulation of emotion. Decreased surface area could be indicative of neurodevelopmental pruning or experience-related reshaping. Previous studies have established that rRSA reflects the capacity for emotion regulation. Our finding suggests that the neuroanatomical underpinning for this could be the rRMF. Elucidating relationships between neuroanatomical and physiological biomarkers may have utility for understanding the etiology of MDD and may inform future preventative interventions targeting these biomarkers.

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Poster

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Program #/Poster #: P697.06

Topic: G.05. Mood Disorders

Support: USC Department of Psychology Doctoral Research Grant Award for Academic Year 2019-2020
First-Year Dornsife PhD Academy Psychology Department Research Award

Title: Dopamine Dependent: examining the link between learning and treatment-resistant depression

Authors: *X. LIU, S. J. READ;
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Abstract: Treatment resistant depression (TRD) is a subtype of Major Depressive Disorder (MDD) that is not responsive to typical antidepressants such as SSRIs. There is some evidence that treatment responsiveness may be related to dopamine neuron dysfunction, specifically in the mesolimbic pathway. To address the gap in understanding of how this translates to behavioral and personality differences in TRD, we used several self-report scales in conjunction with a reinforcement learning task administered online to objectively measure individual variation in learning from positive and negative feedback. In Experiment 1, we found participants (N = 141) on average were better at learning the negative stimuli than the positive. Response bias during the test block was significantly correlated with the Inventory of Depression and Anxiety Symptoms-II (IDAS-II) for General Depression subscale, such that greater depression severity was related to a more negative response bias (r = -.18, p = .030). In Experiment 2, we empirically defined three underlying facets of anhedonia by factor analyzing items of the Motivation and Energy Inventory (MEI) and Temporal Experience of Pleasure Scale (TEPS) administered online to a sample of 137 participants with and without depression. The major latent factors were found to be motivation or energy, wanting or anticipation, and consummatory liking. We found that TRD was associated with significantly lower anticipation scores, whereas MDD overall was associated with significantly lower consummatory pleasure. Additionally, the TRD group had significantly lower Extraversion scores than responders. After controlling for depression severity, antidepressant responders demonstrated a significantly less negative learning bias than both the TRD group (p = .047) and control group (p = .017) on our reinforcement learning task. We postulated this was due to the effect of SSRIs; however, further research is needed to determine a causal effect.

Disclosures: X. Liu: None. **S.J. Read:** None.

Poster

P697. Human Imaging and Behavioral Studies: Focus on Depression

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P697.07

Topic: G.05. Mood Disorders

Support: School of Arts & Sciences, University of Pennsylvania

Title: Parsing depression across the socioeconomic spectrum using neuroanatomy

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Abstract: Socioeconomic status (SES) is a potent risk factor for depression, with symptoms increasing by a factor of two or more between poor and middle class adults (DJ Brody, 2018). Here we ask: Is the "surplus" of depression symptoms at low SES just like those at higher levels, or does it differ? We addressed this question using data from the Human Connectome Project, with a sample of 881 healthy young adults (22 - 37 yo), with neuroanatomical, psychological and SES (income and education) data. Symptom load was the summed ratings of depression-typical symptoms from the ASEBA (LA Rescorla, 2018) scale. The anatomical feature of a priori interest was amygdala volume, which is associated with SES and depression. We used multiple regression controlling for age, gender and race to test the relations among SES, amygdala volume, and depression symptom load. Gender, age and race did not moderate any reported effects. We tested 3 different hypotheses, with different implications for the heterogeneity of depression across levels of SES. A diathesis-stress model, which describes the interaction of risk factors in psychopathology, was tested by adding an SES-amygdala volume interaction term in the regression model and was disconfirmed. A mediation model, based on previous proposals that amygdala volume accounts for the effect of SES on depression (EC Merz, 2018), was also disconfirmed, despite significant pairwise relations among SES, amygdala volume and depression symptom load in this sample. In sum, amygdala volume and SES neither synergize nor account for common variance; rather they have additive and independent effects on depression. Hence the excess of depression observed at lower levels of SES differs in its relation to amygdala volume from depression that is evenly distributed across the SES continuum. Two more tests were then undertaken to validate this difference. First, the 14 specific depression symptoms endorsed by participants were examined in relation to the two risk factors (FDR corrected). SES and amygdala volume were associated with largely nonoverlapping symptoms. Second, the role of perceived stress (NIH Toolbox) in mediating the relations between the two risk factors and depression was assessed. Stress accounts for SES effects on depression but has no relation to amygdala volume. In sum, by using amygdala volume as a marker for depression risk, we found it to be independent of SES, hence SES is a distinct risk factor, associated with different symptoms and is, in contrast to amygdala volume, accounted for in part by perceived

stress. Understanding different pathways to depression can help develop new treatment strategies on psychotherapy and antidepressant medicines.

Disclosures: Y. Hao: None. **M.J. Farah:** None.

Poster

P697. Human Imaging and Behavioral Studies: Focus on Depression

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P697.08

Topic: G.05. Mood Disorders

Support: NIMH K23MH125018
P&S Fund/BBRF A136828
R25 MH060482
FFOR P0548058

Title: Identification of a Personalized Intracranial Biomarker of Depression and Response to DBS Therapy

Authors: A. C. FRANK[1], K. SCANGOS[2], P. S. LARSON[3], A. T. LEE[3], ***A. LEE**[2];
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Abstract: Deep brain stimulation (DBS) for obsessive-compulsive disorder provides improvement in symptom burden for many people with treatment-refractory OCD. However, the stimulation parameter space remains large and stimulation is currently open-loop, without responsiveness to patient's symptoms states. Here we attempted to identify neural correlates of symptom burden and assess how DBS stimulation impacts these neural signals, with consideration toward developing responsive, closed-loop DBS systems. We utilized the Medtronic Percept device to both stimulate and chronically record neural signals in the home and office setting. Data were acquired from leads placed in the BNST of a patient with comorbid depression and treatment-refractory OCD. The patient recorded symptom burden over several months by completing visual-analog surveys. DBS stimulation improved both OCD symptom burden (Y-BOCS reduction of 29%) and depression (PHQ-9 reduction of 48%). Further, we find that in the ambulatory setting, LFP power in the gamma frequency band is strongly, inversely correlated with patient ratings of depression severity (i.e. low gamma power is associated with high levels of depression). During in-office recordings, we find that DBS stimulation broadly increases LFP power, including within the gamma range. In conclusion, LFP gamma power in the BNST is strongly, inversely correlated with ratings of depression and DBS stimulation broadly increases power, including in the gamma frequency range. These results suggest a neural biomarker of depression that could be utilized to develop adaptive, closed-loop DBS.

Disclosures: A.C. Frank: None. **K. Scangos:** None. **P.S. Larson:** None. **A.T. Lee:** None. **A. Lee:** None.

Poster

P697. Human Imaging and Behavioral Studies: Focus on Depression

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Program #/Poster #: P697.09

Topic: G.05. Mood Disorders

Support: The research grant (ASH9/2562) from the Faculty of Allied Health Sciences, Burapha University, Thailand
Thailand Research Fund, the Royal Golden Jubilee Ph.D. Program
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Travel grant from International Brain Research Organization (IBRO)

Title: Association of brain-derived neurotrophic factor (BDNF) and glutamate ionotropic receptor NMDA type subunit 2B (GRIN2B) polymorphisms with major depressive disorder susceptibility

Authors: S. IAMJAN[1], B. THAWEETHEE-SUKJAI[2], S. SUTTAJIT[3], S. THANOI[4], G. P. REYNOLDS[5], S. NUDMAMUD-THANOI[4];
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Abstract: Major depressive disorder (MDD) is a psychiatric disorder that relates in part to dysfunction of the glutamatergic system (Miladinovic et al., 2015). Previous study has reported that abnormal expression of glutamate receptor, NR2B subunit of N-Methyl-D-aspartic acid (NMDA) can promote depression-like behaviors in a rat epilepsy model (Peng et al., 2016). Additionally, brain-derived neurotrophic factor (BDNF) is a member of neurotrophin family which plays crucial roles in regulation of neurotransmitter systems and glutamatergic synaptic strength (Carvalho et al., 2008). Deficits of BDNF have been reported in the pathology of MDD (Phillips, 2017). Therefore, the functional polymorphisms (SNPs) of *BDNF* rs6265 and *GRIN2B* rs3764030, in genes for BDNF and NR2B proteins respectively, may be candidate risk factors for MDD. The objectives of this study were to determine the associations of functional *BDNF* rs6265, *GRIN2B* rs3764030 and its interaction with MDD in the Thai sample. The rs6265 and rs3764030 genotyping were performed in blood DNA samples of 100 MDD patients and 100 matched controls using TaqMan SNP genotyping assays. The results showed that genotype of *GRIN2B* is significantly associated with MDD (p= 0.038) with T carriers having an increased frequency (p=0.013; Odds ratio=2.07). No association was observed with *BDNF* genotype.

However, taken together, *BDNF* and *GRIN2B* risk genotypes showed a slightly stronger association with MDD (p=0.008; Odds ratio=2.31). The findings demonstrate a significant association of a single *GRIN2B*, but not *BDNF*, variant with MDD in the Thai population. The result supports the involvement of the glutamatergic system in the neurobiology of MDD. A possible interaction between the rs6265 in *BDNF* and a glutamate receptor variant in a psychiatric disorder is consistent with a previous study (Iamjan et al., 2018) in which we identified such an effect - between *GRIA3* and *BDNF* - on susceptibility to methamphetamine psychosis (Iamjan et al., 2018). Therefore, the result may suggest a relationship between BDNF and glutamate receptor function on the pathology of MDD. Alteration of BDNF activity may impair synaptic plasticity at the glutamatergic synapse (Carvalho et al., 2008), and contribute to the development of MDD. The present study provides evidence that a functional variant of *GRIN2B,* possibly interacting with *BDNF* rs6265, contribute genetic vulnerability for MDD in the Thai population; further studies with larger samples are need to confirm these findings.

Disclosures: S. Iamjan: None. **B. Thaweethee-Sukjai:** None. **S. Suttajit:** None. **S. Thanoi:** None. **G.P. Reynolds:** None. **S. Nudmamud-Thanoi:** None.

Poster

P699. Human Studies and Therapeutic Approaches: Mood and Anxiety

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P699.01

Topic: G.05. Mood Disorders

Support: NIH NIMH K01MH120433

Title: Positive self-bias increases with age and is reduced in patients with mood and anxiety disorders

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Abstract: The sense of agency, or the belief of action ownership, is an elusive construct that impacts day-to-day experience and decision-making. Despite its relevance in a range of neuropsychiatric disorders, it is widely under-studied and remains difficult to measure objectively in patient populations. We developed and tested a novel cognitive measure of valence-modulated agency perception in 53 healthy control subjects and 20 subjects with mood or anxiety disorders (MAD), including major depressive disorder and generalized anxiety disorder. The task consisted of a simple computerized effort task for monetary rewards with performance-based trial-by-trial positive or negative feedback. Throughout the task, the visual representation of progress during effort was ambiguously but consistently perturbed by computer

interference. Trial-by-trial ratings of self-versus-other agency were collected. In line with previous work, healthy subjects demonstrated higher self-agency after receiving positive feedback, compared to negative feedback when the level of computer inference was kept constant ($p=7.3 \times 10^{-6}$). This "positive self-bias" was associated with increasing age ($R=0.29$, $p=0.02$). Patients with MAD showed reduced positive self-bias compared to healthy controls ($p=0.048$), and this reduction was associated with worse self-reported symptoms of anhedonia (Snaith-Hamilton Pleasure Scale, SHAPS) at trend level ($R=0.42$, $p=0.06$). Together this work suggests how positive and negative environmental information impacts the sense of self-agency in healthy subjects, and how it is perturbed in patients with mood and anxiety disorders.

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Poster

P699. Human Studies and Therapeutic Approaches: Mood and Anxiety

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Program #/Poster #: P699.02

Topic: G.05. Mood Disorders

Support: Natural Science and Engineering Research Council of Canada (NSERC)
UBC Djavad Mowafaghian Centre for Brain Health (DMCBH) Innovation Fund
Kickstart Research Grant

Title: Levels of depressive and anxiety symptoms in humans predict distinct patterns of reward-seeking: A cross-species translation of an appetitive go/no-go task.

Authors: *I. D. DALY[1], R. J. TOMM[1], B. J. FORYS[1], A. R. TERPSTRA[1], L. CLARK[1,2], T. CHAKRABARTY[1,2], S. B. FLORESCO[1,2], R. M. TODD[1,2];
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Abstract: Traditional methods used to classify neuropsychiatric disorders, such as depression and anxiety, are based on heterogenous symptom clusters and can often fall short of reliably predicting treatment efficacy. In part, this may be attributable to a reliance on self-report symptoms and observable signs which fail to capture the underlying pathophysiologies- and may not account for differences amongst disorder subtypes and comorbidities (i.e., anxious depression). Depression and anxiety are often characterized by impaired reward-seeking and avoidance behaviors, respectively. However, few studies investigate whether anxiety symptoms predict reward-seeking behaviors, especially when accounting for comorbidity with depressive symptoms. Furthermore, it is unclear how symptoms of depression and anxiety predict biases towards active versus inhibitory reward-seeking strategies. Here, a cross-species translation of an

appetitive go/no-go task was used to differentiate between active and inhibitory reward seeking. In this study, undergraduates with depressive and anxiety symptoms ranging from minimal to severe completed a modified go/no-go reward-seeking task (translated from a rodent paradigm). In this uninstructed learning task, visual cues signaled the response required to obtain a monetary reward. The task involved three stages: 1) An acquisition stage, where participants learned an active response; 2) An intermixed stage, where participants learned to discriminate between active and inhibitory responses; and 3) A reversal stage where the previous response rules were reversed. Participants were grouped into the highest and lowest quartiles for depressive and anxiety symptoms. Participants with higher depressive symptoms were impaired on both active and inhibitory reward-seeking trials during the reversal stage, compared to those with low depressive symptoms. In addition, those with high anxiety symptoms had increased accuracy on active reward-seeking trials compared to low anxiety participants in the intermixed stage. Most notably, these results differ from those observed in an analogous avoidance task that assessed the relationship between depressive and anxiety symptoms in active and inhibitory avoidance. This task, alongside analogous tasks that assess various aspects of reward-seeking and avoidance behaviours, may aid in our understanding of the heterogeneity and interaction of symptoms of depression and anxiety, two highly comorbid conditions.

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Poster

P699. Human Studies and Therapeutic Approaches: Mood and Anxiety

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Topic: G.05. Mood Disorders

Support: Natural Science and Engineering Research Council of Canada (NSERC)
UBC Djavad Mowafaghian Centre for Brain Health (DMCBH) Innovation Fund Kickstart Research Grant

Title: Depressive and anxiety symptom clusters in humans are related to distinct avoidance profiles in a cross-species translation of a go/no-go avoidance task.

Authors: *R. J. TOMM[1], B. J. FORYS[1], I. D. DALY[1], A. R. TERPSTRA[1], L. CLARK[1,2], T. CHAKRABARTY[1,2], S. B. FLORESCO[1,2], R. M. TODD[1,2];
[1]Dept of Psychology, [2]Djavad Mowafaghian Ctr. for Brain Hlth., Univ. of British Columbia, Vancouver, BC, Canada

Abstract: Depression and anxiety show notable heterogeneity in symptomatology and commonly co-exist. Comorbid anxiety in depression has been consistently associated with worse

treatment outcomes than depression without anxiety, indicating that this may be a more severe and perhaps pathophysiologically distinct subtype. Depression and anxiety are often characterized by impaired reward-seeking or avoidance behaviors, respectively. However, few studies investigate how depressive symptoms relate to avoidance behaviors, especially when accounting for comorbidity with anxiety symptoms. Additionally, it is not clear whether symptoms of depression and anxiety predict active versus inhibitory forms of avoidance. In this study, undergraduates (N_{Total} = 174) with a range of depressive (Beck Depression Inventory) and anxiety (Beck Anxiety Inventory) symptoms from low to severe completed a modified go/no-go avoidance task (reverse translation of a rodent paradigm). In this uninstructed learning task, cues were used to signal the response type (go/active vs. no-go/inhibitory) required to avoid an aversive sound. The task consisted of three-stages: 1) An acquisition stage required learning of an active avoidance response; 2) An intermixed stage required learning to flexibly discriminate between active and inhibitory avoidance responses; 3) In a reversal stage the response rules were reversed. Compared to participants with lower depressive (Low_{DEP}, *N* = 39; *M* = 4.38, *Range* = 0-7) and anxiety (Low_{ANX}, *N* = 41; *M* = 2.73, *Range* = 0-5) symptoms, participants with higher depressive ($High_{DEP}$, *N* = 44; *M* = 28.02, *Range* = 21-55) and anxiety ($High_{ANX}$, *N* = 41, *M* = 33.93, *Range* = 25-63) symptoms were impaired in active avoidance during the acquisition stage. However, this effect was driven mostly by a subset of participants that scored high in both depressive and anxiety symptoms ($High_{DEP}$ + $High_{ANX}$, *N* = 22). $High_{DEP}$ participants showed increased active avoidance accuracy in the intermixed stage compared to Low_{DEP} participants. Conversely, $High_{ANX}$ participants were impaired in active avoidance, showing decreased accuracy on Go trials during all 3 task stages compared to Low_{ANX} participants. Depressive and anxiety symptoms were not associated with differences in inhibitory avoidance during any task stage. This translational avoidance task revealed distinct avoidance profiles that were dependent on relative levels of depressive and anxiety symptoms, suggesting that it might be particularly useful in discriminating differences in avoidance behaviors in clinical populations of depression and anxiety - especially those diagnosed with comorbidities.

Disclosures: R.J. Tomm: None. **B.J. Forys:** None. **I.D. Daly:** None. **A.R. Terpstra:** None. **L. Clark:** None. **T. Chakrabarty:** None. **S.B. Floresco:** None. **R.M. Todd:** None.

Poster

P699. Human Studies and Therapeutic Approaches: Mood and Anxiety

Location: Virtual Only

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Support: Natural Science and Engineering Research Council of Canada (NSERC)
UBC Djavad Mowafaghian Centre for Brain Health (DMCBH) Innovation Fund
Kickstart Research Grant

Title: Gender selectively moderates relationships between mood disorder symptoms and performance of effort related avoidance and reward-seeking tasks

Authors: *B. J. FORYS[1], R. J. TOMM[1], D. STAMBOLIYSKA[1], A. R. TERPSTRA[1], L. CLARK[1,2], T. CHAKRABARTY[3,2], S. B. FLORESCO[1,2], R. M. TODD[1,2];
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Abstract: We must often decide how much effort to exert to avoid undesirable outcomes or obtain rewards. Past work suggests that dopamine (DA) modulates active avoidance and reward-seeking while serotonin (5HT) modulates inhibitory avoidance. Moreover, DA dysregulation in depression may limit effective effort deployment for obtaining rewards. Yet understanding the neuromodulation of such dimensional behaviours in depression is hampered by outstanding questions about the links between motivated action and inhibition and depressive symptoms, and whether these differ between males and females or with comorbid anxiety. The present study used a task back-translated from rodent studies to examine whether effort deployment on active and inhibitory avoidance (Study 1) vs. reward seeking (Study 2) predicts depression symptoms, and whether these relationships are moderated by gender. Undergraduates and paid participants (N_{Avoid} = 540, N_{Reward} = 308; N_{Female} = 376, N_{Male} = 472, M_{Age} = 23.10, SD_{Age} = 5.79) were assessed on Beck Depression Inventory (BDI-II) as well as the Beck Anxiety Inventory (BAI) to control for anxiety comorbidity, and performed an online instructed avoidance or reward-seeking task. Participants had to make multiple presses on active trials and withhold presses on inhibitory trials to avoid an unpleasant sound (Study 1) or obtain points towards a monetary reward (Study 2). Physical effort on active trials (number of presses) increased every 20 trials on a progressive ratio schedule. Participants ranged in depressive symptom levels; many scored above the threshold (>13) for moderate depression (M = 16.14, *Range* = 0-58, $N >$ *clinical threshold* = 475). In the avoidance task, gender moderated the relationship between depressive symptoms and active trial accuracy. Males had lower accuracy overall on active trials; females with more depressive symptoms had higher active trial accuracy. This pattern emerged as effort requirements increased. We found no effects of gender or depressive symptoms on inhibitory trials. In the reward-seeking task, females with higher depressive symptoms were more likely to exert effort for reward. To examine depression-effort interactions, we simulated each task with a computational model and evaluated the output's fit to our results. Our results illuminate gender differences in the relationship between depressive symptoms and motivation to actively and effortfully respond to achieve positive and avoid negative outcomes. Future studies can examine how sex/gender moderates the role of DA and 5HT in effortful avoidance and reward-seeking. Targeting goal-directed effort deployment may also be a useful clinical intervention.

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Poster

P699. Human Studies and Therapeutic Approaches: Mood and Anxiety

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P699.05

Topic: G.05. Mood Disorders

Support: ANII, Uruguay
PEDECIBA, Uruguay
C-SIC, UdelaR, Uruguay
Santiago Achugar Diaz fund, PEDECIBA, Uruguay

Title: Approach avoidance decision making in depression and social anxiety

Authors: *A. ACUÑA[1], S. MORALES[1], L. URIARTE-GASPARI[1], N. AGUIRRE[1], E. CUÑA[2], M. GARCÍA FONTES[2], A. PÉREZ[1], V. GRADÍN[3,1], Á. CABANA[3,1], N. HUART[4];
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Abstract: Approach-avoidance conflict (AAC) occurs when we make decisions that can lead to both rewarding and aversive outcomes. AAC has been implicated in the pathophysiology of Major Depression (MD) and Social Anxiety (SA), but few studies have investigated this relationship on a neural level. When AAC includes social threat and rewards, it is called social AAC (sAAC). Given that people with MD and SA suffer from social impairments, sAAC is of interest in these populations.
We used fMRI to study brain activity during sACC, in participants with MD, SA, or both disorders (MD/SA, n=52) and matched healthy controls (n=41). In our task, players are thought to be categorized as a one/two/three/four or five star players, depending on how good they are responding trivia questions. Participants are actually categorized as three star players. On every trial, participants choose between two possible options, with which category of co-player they would like to make a team. From a material point of view, choosing the higher category (approach) is better than choosing the lower one (avoidance), since this makes a stronger team. However, conflict arises because approach decisions increase the chances for the participant being the worst one in the team, which relates to social comparisons and guilt processes.
The MD/SA group reported higher negative emotions, such as embarrassment and guilt, during the task. At a neural level, for the avoidance > approach contrast, we found across participants activations spanning the anterior cingulate cortex (ACC) and medial prefrontal cortex (mPFC). These regions have been associated with the detection of conflict and with deliberate and effortful decision making. Therefore we believe their activity is related to avoidance decisions encompassing a more thoughtful and less automated process than approach decisions. Supporting this idea, we found that avoidance decisions took longer to make than approach decisions. The avoidance > approach contrast also showed between group differences in the ACC and mPFC. This difference was related to controls showing higher activity than the MD/SA group during avoidance decisions in these regions. This suggests that controls experienced more conflict and

had to put more effort when choosing to play with lower categories, while for the MD/SA group this was a more automatic decision. This idea is backed up by an interaction between group and type of decision found in reaction times, showing that between group differences were larger when choosing the avoidance option (with controls taking longer to choose) compared to choosing the approach option.
Findings highlight neural substrates underlying sAAC biases in MD and SA.

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Poster

P699. Human Studies and Therapeutic Approaches: Mood and Anxiety

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P699.06

Topic: G.05. Mood Disorders

Title: Neural substrates of repetitive negative thinking: an fMRI analysis of overlapping worry and rumination signals in the brain

Authors: *N. PUCCETTI[1], E. K. MORAES[1], C. A. STAMATIS[2], K. R. TIMPANO[1], A. S. HELLER[1];
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Abstract: Depression and anxiety disorders are detrimental and often co-occur. To mitigate the financial, physical, and psychological costs of this comorbidity, it is critical to understand the risk factors that explain shared and unique features of these disorders. One shared risk factor is repetitive negative thinking (RNT). We still lack a clear neurobiological model of RNT, however, because neuroimaging studies mainly 1) examine RNT's content-specific components (worry and rumination) 2) use general prompts rather than idiographic stimuli to induce RNT, and 3) conduct group-wise comparisons rather than examining the full dimension of RNT. To this end, we used idiographic worry and rumination statements to induce RNT during an fMRI scan in 37 young adults with dimensionally assessed RNT. During imaging, participants reflected on their personal worries and ruminations, as well as neutral prompts, for 16s each. We characterized RNT through a contrast of the worry and rumination conditions minus the neutral condition. This contrast showed that RNT was related to greater activity in the inferior parietal lobule, insula, putamen, thalamus, and midcingulate cortex. During the neutral condition there was greater activity in default mode network (DMN) structures, including the posterior cingulate cortex, the medial prefrontal cortex (mPFC) and the hippocampus. In addition, to determine unique features of worry and rumination, we examined a worry minus rumination contrast. Here,

rumination was associated with mPFC and anterior cingulate cortex, precuneus, and middle temporal gyrus activation. Results from the broad RNT contrast highlight that negative, perseverative thoughts are not simply a product of DMN-supported self-referential processes, but may require regions within the salience network (i.e., the insula and cingulate cortex). Moreover, the rumination-specific relations to both DMN and salience network structures suggest that worry and rumination are distinguishable from one another in the brain. These results extend the literature by characterizing both content-specific and -independent RNT neural substrates, which could ultimately inform etiological and treatment models of depression and anxiety.

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Poster

P699. Human Studies and Therapeutic Approaches: Mood and Anxiety

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Program #/Poster #: P699.07

Topic: G.05. Mood Disorders

Support: NIMH Intramural Research Grant ZIA-MH002957-01

Title: Passage-of-time dysphoria: a highly replicable decline in mood during rest and simple tasks that is moderated by depression

Authors: *D. C. JANGRAW[1,2], H. KEREN[2], R. L. BEDDER[5], R. B. RUTLEDGE[5], F. PEREIRA[3], A. G. THOMAS[4], D. S. PINE[2], C. Y. ZHENG[3], D. M. NIELSON[2], A. STRINGARIS[2];
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Abstract: Does our mood change as time passes, and is this change different in people with depression? These questions are central to affective neuroscience, yet they remain largely unexamined. Here we demonstrate that rest and simple task periods lowered participants' mood, an effect we call “passage-of-time dysphoria.” This finding was replicated in 15 cohorts totaling 27,882 adult and adolescent participants, including preregistered analyses. Interestingly, the dysphoria was reduced in people with higher depression risk.
We first recruited 40 adults online using Amazon Mechanical Turk (MTurk) and asked them to rate their mood during a rest period before a gambling game. We observed a gradual, apparently linear decline in mood. This result was replicated in 5 other cohorts. The mean dysphoria was relatively large (13.8% after 7.3 minutes, Cohen's d=0.574). However, it was quite variable across individuals and showed only moderate stability within individuals (ICC(2,1)=0.411 across 2 weeks).

Using modified versions of the task, we showed that the decline in mood ratings was not due to these methodological confounds: the aversive nature of rating one's mood, the mood rating method's susceptibility to fatigue, the expected rest period duration, the aversive nature of multitasking, or regression to the mean. We showed that the decline also occurs in simple tasks, including a visuomotor task and a gambling game with random zero-mean outcomes.
To test the effect's generalizability, we collected similar data from 116 adolescent participants recruited in person. This group showed a mood decline not significantly different from the MTurk cohort. We also analyzed a large dataset (n=26,896) of mood ratings during a mobile app gambling task. Even in this engaging game, mood decreased with time on task. But the dysphoric effect was smaller in this group ($p= 8.10e{-}45$).
An individual's level of dysphoria was inversely related to depression risk and a computationally estimated reward sensitivity parameter, supporting the idea that depressed people feel that rest better aligns with their reduced expectations for task rewards. Rest also impacted behavior: participants were less likely to gamble at the start of a task if it was preceded by rest.
Our findings call into question neuroscience's long-held assumption of a constant affective background by demonstrating spontaneous, seemingly linear drifts in mood that occur over time. This has theoretical implications for the nature of mood and its aberrations, and methodological consequences for the design and interpretation of experiments, particularly those in which neuroimaging results might be affected by mood and confounded by time.

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Poster

P699. Human Studies and Therapeutic Approaches: Mood and Anxiety

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P699.08

Topic: G.05. Mood Disorders

Support: NRF 2017M3C7A1048040

Title: Subjective Pain, Emotion Regulation Function, and Regional Brain Volume Reduction in Nonsuicidal Self-injury

Authors: *S.-E. LEE, H. SHIN, G. KIM, H. MOON, J.-W. HUR;
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Abstract: Background: Investigating the dynamics of pain during nonsuicidal self-injury (NSSI) is crucial to understanding why individuals choose and maintain self-destructive behaviors. Little is known, however, whether there are structural changes in the brain regions

responsible for the pain-processing of NSSI. This study aimed to examine gray matter volume changes in the NSSI group and its associations with subjective pain, frequency, versatility, and functions of NSSI. **Methods:** Individuals who reported five or more NSSI episodes in the past year (n = 63) and age- and sex-matched controls (n = 63) participated in the study. T1-weighted MRI data were obtained from all participants. Voxel-based morphometry (VBM) was done using Computational Anatomy Toolbox version 12 (CAT). To explore the brain structural differences between groups, two-sample *t*-tests, adjusting for age, sex, total intracranial volume (TIV), and scan site, were applied. Within the NSSI group, multiple regression analyses were used to examine the relationships between gray matter volumes and variables of NSSI. **Results:** We found regional gray matter volume decrements in the left opercular part of the inferior frontal gyrus (IFG), right insula, and bilateral secondary somatosensory cortex (S2). Gray matter volumes of the opercular part of the IFG showed significant negative correlations with subjective pain and toughness function of NSSI. Moreover, the volumes of the insula and S2 were negatively associated with the emotion regulation function of NSSI. **Conclusions:** Our findings highlight regional brain volume reductions among individuals who engage in NSSI in relation to pain-processing. Future studies are needed to further clarify the relationship between structural brain abnormalities, subjective pain, and emotion regulation function of NSSI.

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Poster

P699. Human Studies and Therapeutic Approaches: Mood and Anxiety

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Program #/Poster #: P699.09

Topic: G.05. Mood Disorders

Support: NSERC Postgraduate Scholarship (Doctoral)
Ontario Brain Institute Integrated Discovery Program Grant
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Title: Brooding and reflection as trait predictors of mentation rate measured via wholebrain network activity transitions during resting-state fMRI: A CAN-BIND-4 analysis

Authors: *S. D. SQUIRES[1], J. TSENG[1], M. ZAMYADI[2], S. HASSEL[3], S. C. STROTHER[2], G. MACQUEEN[3], S. H. KENNEDY[4], R. V. MILEV[1], K. L. HARKNESS[1], J. POPPENK[1]; [1]Queen's Univ., Kingston, ON, Canada; [2]Baycrest Hlth. Sci. Ctr., Toronto, ON, Canada; [3]Univ. of Calgary, Calgary, AB, Canada; [4]St. Michael's Hosp., Toronto, ON, Canada

Abstract: Most of us are familiar with dwelling on our feelings and problems when we are sad. The tendency to do this is called trait depressive rumination (DR) and it is known to increase risk of psychiatric disorder in vulnerable individuals. Two kinds of DR have been proposed:

Reflection, which involves understanding one's feelings and problems, and Brooding, which involves abstract, "why me?" thoughts about obstacles and negative social comparisons. Mentation rate (MR) is the number of discrete thoughts had over time and intuition suggests it is linked to DR due to the sustained and repetitive nature of ruminative thoughts. This association's direction has been impossible to ascertain, however, due to limitations with self-report measures of MR. Does DR involve a fast MR as one rapidly changes thoughts to understand oneself and situation, or a slow MR due to fixation on one's feelings and problems? Does this depend on the kind of DR? To address these questions objectively, we turned to participants' brain network dynamics, leveraging cutting-edge neuroimaging methods that enable us to use co-occurring network activity changes to detect the moments when each participant transitions between thoughts. We calculated this network transition rate (TR) during resting-state fMRI (rsfMRI) and examined its relations with Reflection and Brooding scores from the Ruminative Responses Scale. We used archival data from the Canadian Biomarker Integration Network for Depression: Stress and Reward in Anhedonia (CAN-BIND-4) study, which were collected from 63 participants (ages 18-65, 68% female, 67% white) with no history of psychiatric disorder. Higher Reflection and Brooding scores were associated with faster and slower TR, respectively. A significant Reflection X Brooding interaction was also observed, such that higher scores in both variables predicted a faster TR than was predicted by those variables separately. This analysis is the first to use an objective approximation of MR, TR, to demonstrate how different forms of DR, Reflection and Brooding, may be linked respectively to diverse and restricted thought content over time. Additionally, we speculate that the higher TR in those who both brood and reflect may represent a greater number of network transitions due to switching among these and other modes of thought, but this must be verified with further research. These observations elucidate how different kinds of repetitive thought defined in the psychology literature can be differentiated based on objective neuroscience metrics like TR. By leveraging both fields in such a way, we inch closer towards identifying a biopsychologically supported taxonomy of repetitive thought.

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Poster

P699. Human Studies and Therapeutic Approaches: Mood and Anxiety

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Program #/Poster #: P699.10

Topic: G.05. Mood Disorders

Support: McDonnell Center for Systems Neuroscience, Small Grants Program

Title: Longitudinal Disruption of Sleep Structure by Electroconvulsive Therapy

Authors: *M. KAFASHAN, A. K. LABONTE, O. HYCHE, T. NGUYEN, S. SUBRAMANIAN, N. B. FARBER, B. J. PALANCA;
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Abstract: Sleep plays an essential role in maintaining homeostasis of cognitive function through synaptic plasticity. Slow oscillations are hallmarks of electroencephalographic (EEG) during slow-wave sleep and define non-rapid eye movement sleep (NREM) stage N3. Sleep slow-wave activity (SWA) is associated with memory formation and synaptic plasticity. A decrease in sleep SWA has been observed in patients with major depressive disorder. Electroconvulsive therapy (ECT) is an effective therapy for treatment-resistant depression (TRD), defined as the lack of response to at least two antidepressant pharmacologic agents. Therapeutic effects of ECT stem from the induction of generalized seizures during general anesthesia; however, underlying mechanisms remain unclear. We hypothesize that ECT alters the expression of sleep slow waves to facilitate synaptic reconfiguration in pathologic neural circuits underlying psychiatric illnesses. In this study, we assessed sleep macrostructure and microstructure longitudinally. Wireless wearable devices, Dreem (Rhythm, New York USA), were used to acquire sleep EEG data and to address previous technical impediments to the study of longitudinal changes in SWA in the outpatient setting. Participants provided overnight sleep EEG the day after each ECT session. Sleep EEG data were also acquired for a minimum of one night prior to the first ECT session, providing a true baseline measure. Sleep records were scored as rapid eye movement (REM) and non-rapid eye movement (NREM) stages N1-N3, using modified American Academy of Sleep Medicine guidelines. Severity of depression was quantified utilizing the Quick Inventory of Depressive Symptomatology-16 (QIDS) before each ECT session, serially up to a 7-week index course of the treatment. SWA was quantified as the average 0.5-4 Hz frontal EEG power per minute during N3 sleep. We acquired 111 sleep records from 10 subjects in the first year of this study, in which 81 records were scorable. The proportion of N3 sleep was linearly correlated with treatment number (linear mixed-effects model with QIDS score as a covariate, $R^2=0.44$, $P=0.003$). Similarly, SWA increased over the course of therapy (linear mixed-effects model with QIDS score as a covariate, $R^2=0.41$, $P=0.035$). This study confirms the feasibility of using wireless wearable devices for the longitudinal assessment of sleep structure over the course of ECT in TRD patients. Furthermore, our findings might shed light on other non-invasive interventions targeting sleep structure in patients with psychiatric illnesses.

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Poster

P699. Human Studies and Therapeutic Approaches: Mood and Anxiety

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Program #/Poster #: P699.11

Topic: H.05. Working Memory

Title: Effects of bullying on anxiety symptoms in adolescents: Parental warmth as a potential buffer

Authors: *S. PAULISIN[1], B. BORG[2], J. EVANSKI[2], H. SCHACTER[2], H. MARUSAK[2];
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Abstract: *Effects of bullying on anxiety symptoms in adolescents: Parental warmth as a potential buffer*
Shelley Paulisin, Breanna Borg, Julia Evanski, Hannah Schacter, Ph.D. and Hilary Marusak, Ph.D.
Anxiety disorders are the most common psychiatric disorder, affecting nearly one in three, and commonly begin during adolescence. Peer victimization, or bullying, is common (10-30%) among adolescents and has been shown to increase risk of anxiety disorders. Prior research suggests that a healthy parent-child relationship may protect adolescents from anxiety. Here, we examine whether peer victimization is associated with anxiety symptoms in early adolescents, and whether parental warmth can buffer the effects of peer victimization on anxiety. One-hundred and fifty early adolescents (M±SD = 12±1.5, 45% female) and one parent/guardian completed online surveys. Adolescents self-reported on peer victimization, anxiety symptoms, and parental warmth using validated questionnaires, including the Revised Personal Experiences Checklist (De Los Reyes & Prinstein, 2004), Screen for Child Anxiety Related Disorders (Birmaher et al., 1997; Monga et al., 2000), and the Parent Child Relationship Scale (Pianta, 1992), respectively. Correlation analyses were used to examine bivariate associations among peer victimization, anxiety, and parental warmth, and hierarchical regression was used to test for interactive effects of peer victimization and parental warmth on anxiety. Peer victimization was positively associated with anxiety symptoms, and negatively associated with parental warmth. Parental warmth was also negatively associated with anxiety. There was no significant interaction between peer victimization and parental warmth on child anxiety symptoms. All results remained consistent when controlling for age and gender. These results suggest that peer victimization may increase risk of adolescent anxiety. Although parental warmth was associated with lower anxiety, we did not find evidence that parental warmth can buffer the negative effects of peer victimization on anxiety. These findings highlight the need to improve mental health outcomes in victims of bullying. They also suggest that parents, in general, can play an important role in supporting adolescents' mental health.

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Poster

P708. Neural Mechanisms Underlying Emotional Liability and Stress Responsivity

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Program #/Poster #: P708.01

Topic: G.05. Mood Disorders

Support: NIDA U01DA043098
Office of Naval Research (ONR) 00014-19-1-2149;
The Hope for Depression Research Foundation (HDRF): The Pritzker Neuropsychiatric Research Consortium

Title: Neural and Behavioral Predictors of Chronic Social Defeat Stress

Authors: *D. MURRA[1], A. FITZPATRICK[2], K. L. HILDE[2], H. KHALIL[2], H. AKIL[2], S. J. WATSON, Jr.[2];
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Abstract: Chronic Social Defeat Stress (CSDS) has been widely used as an animal model that captures individual variation following a social stressor. In response to repeated psychological and physical stress, mice diverge into separate populations of social reactivity post-stress: resilient (socially interactive) and susceptible (socially avoidant) animals. A rich body of work reveals distinct neurobiological and behavioral consequences of this experience that map onto the resilient and susceptible phenotypes. By contrast, we have less insight into the factors that drive the eventual split in stress response; we cannot predict which group of mice will emerge as either susceptible or resilient to CSDS. To address this challenge, we focused on behaviorally characterizing resilient and susceptible mice prior to, during, and following CSDS. We found that behavioral coping strategies used by the mice during the initial stress encounter distinguish resilient and susceptible animals. Based on these findings, we asked whether these coping styles that were deployed early on by the two groups were reflective of distinct pre-existing brain-wide neural activity patterns. Our hypothesis is that brain-wide activity patterns and behavioral coping mechanisms that emerge during the initial defeat encounter can be used to predict the eventual social reactivity phenotype. Together, our work aims to provide an integrative characterization of the neural and behavioral signatures associated with resiliency and susceptibility in this social stress model.

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Poster

P708. Neural Mechanisms Underlying Emotional Liability and Stress Responsivity

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Topic: G.05. Mood Disorders

Support: NIH Grant MH100583
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NIH Grant AG064895

Title: Activation of hypothalamic POMC neurons is sufficient and necessary to induce depression-related behaviors

Authors: X. FANG, ***Y. CHEN**, J. WANG, Z. ZHANG, Y. BAI, K. DENNEY, M. GUO, L. GAN, N. WEINTRAUB, Y. LEI, X.-Y. LU;
Med. Col. of Georgia At Augusta Univ., Augusta, GA

Abstract: How distinct neuronal populations participate in mediating maladaptive changes induced by stress exposure and contribute to the onset and development of depression remain unclear. Here, we examined the activity of hypothalamic proopiomelanocortin (POMC) neurons in brain slices from control mice and mice exposed to chronic unpredictable stress (CUS). POMC neurons spontaneously generate action potentials at relatively low rates under control conditions. Following exposure to 10 days of CUS, POMC neuron firing became more frequent and regular. This high-frequency firing correlated with reduced GABA-mediated spontaneous inhibitory postsynaptic currents (IPSCs) and increased intrinsic neuronal excitability in POMC neurons. Under non-stressed conditions, while acute chemogenetic activation of POMC neurons alone failed to alter depression-related behaviors, repeated activation of POMC neurons reduced sucrose preference after 3 days and induced the full spectrum of depression-like behavioral deficits after 10 days, mimicking the effects of chronic exposure to unpredictable stress. In mice subjected to subthreshold levels of unpredictable stress, acute activation of POMC neurons decreased sucrose preference and increased despair behavior. Importantly, inhibition of POMC neurons was sufficient to reverse CUS-induced anhedonic and despair behaviors. Collectively, these results indicate a causal relationship between hyperactivity of POMC neurons and depressive-like behaviors and suggest that dampening POMC neuronal activity can be used as a novel strategy to alleviate depression symptoms.

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Poster

P708. Neural Mechanisms Underlying Emotional Liability and Stress Responsivity

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Support: United States Army Medical Research Acquisition Activity Grant W81XWH-14-0390
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Title: Identification of a novel cell-type specific mood-modulating protein, Neurensin-2

Authors: *G. UMSCHWEIF[1,2], L. MEDRIHAN[2], W. WANG[2], A. GUILLÉN-SAMANDER[3], Y. SAGI[2], P. GREENGARD[2];
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Abstract: Major depressive disorder (MDD) is a leading cause of disability. Despite the high prevalence of MDD and its devastating impact on society, the pathophysiology of this condition is poorly understood. One of the hallmarks of MDD is molecular, functional, and morphological impairments in the hippocampus. Recent studies suggest that two subtypes of hippocampal GABAergic interneurons, the cholecystokinin (CCK) and parvalbumin (PV) interneurons, play key roles in both depression and its treatments. Nonetheless, little is known about the underlying molecular mechanisms by which these cells mediate emotional behaviors. By applying Translating Ribosome Affinity Purification (TRAP) from these cells, we discovered a new cell-type specific signaling pathway that regulates emotional behavior. We found that in CCK and PV interneurons, the chromatin modulator SMARCA3 inhibits the expression level of a novel depression-associated gene, Nrsn2. This gene encodes Neurensin-2, a vesicular protein which is highly and selectively expressed in hippocampal CCK and PV interneurons. We found that Neurensin-2 expression in the hippocampus is dynamic and bidirectionally responds to chronic stress and to chronic treatment with antidepressants, suggesting that Neurensin-2 is a cell-type specific mood modulator. Indeed, we show that overexpression of Neurensin-2 in CCK interneurons results in depressive-like behaviors, whereas down-regulation of Neurensin-2 in PV interneurons is essential for the behavioral response to chronic treatment with antidepressants. Moreover, Neurensin-2 knockout mice are resilient to chronic stress, indicating that Neurensin-2 inhibition is sufficient to prevent depression and anxiety. Mechanistically, we show that Neurensin-2 associates with endosomes and postsynaptic proteins and regulates the α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptors signaling. Together, we introduce a novel cell-type specific protein which directly and bidirectionally affects both cellular function and emotional behavior. These findings implicate Neurensin-2 in the pathophysiology and future treatments of neuropsychiatric disorders such as MDD.

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Poster

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Topic: G.05. Mood Disorders

Support: NIDA U01DA043098; Office of Naval Research (ONR) 0014-19-1-2149
The Hope for Depression Research Foundation (HDRF): The Pritzker Neuropsychiatric Research Consortium

Title: Neurobiological mechanisms that influence emotional reactivity: a developmental study of the hippocampus

Authors: *S. COLEMAN[1], K. L. HILDE[2], P. M. MARAS[2], M. H. HAGENAUER[1], A. V. STEFANOV[1], I. BIRT[1], V. KUMAR[2], E. K. HEBDA-BAUER[1], S. J. WATSON, Jr.[1], H. AKIL[1]; [2]Mol. and Behavioral Neurosci. Inst., [1]Univ. of Michigan, Ann Arbor, MI

Abstract: Debilitating psychiatric disorders such as major depression and chronic anxiety have a major personal and societal impact. Both prevention and treatment strategies require a better understanding of the relationship between genetic predisposition and brain function. For instance, while it is known that there is a genetic component to emotional reactivity, the neurobiological mechanisms through which gene expression patterns shape these behavioral traits remain largely unknown. Using a selective breeding model in rodents, this study aims to elucidate potential cellular mechanisms that link genetic predisposition and the emergence of vulnerable or resilient behavioral traits.

Our laboratory has selectively bred rats for over 60 generations in order to select for locomotor responses to novelty, ultimately generating a robust behavioral model of emotional reactivity and temperament. Bred low responder (bLR) animals and bred high responder (bHR) animals represent extreme ends of emotional reactivity. bLRs represent an internalizing phenotype, are behaviorally inhibited, with a high level of anxiety- and depressive-like behaviors. In contrast, bHRs represent an externalizing phenotype, with low behavioral inhibition and high levels of impulsivity.

Our lab has characterized genetic variants as well as hippocampal gene expression changes in the bLR/bHR model and identified candidate genes that differentiate between the lines. A top candidate gene is the Bone Morphogenetic Protein 4 (Bmp4), which has diverse roles in nervous system development. Preliminary results demonstrate that differences in Bmp4 in the bLR/bHR model arise early in life and are maintained throughout the lifetime of the animals. We perform a comprehensive analysis of cell-type composition and gene expression in the bLR/bHR model using a combination of multiplexed fluorescent in situ hybridization, immunohistochemistry, and confocal imaging. We test the hypothesis that Bmp4 is important for establishing cell-type balance within the developing hippocampus, and that these differences, in turn, play a role in shaping adult behavior.

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Poster

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Topic: G.05. Mood Disorders

Support: NIH grant P01DA047233-02

Title: A stress-responsive circular RNA regulates depression-like behavioral abnormalities in mice

Authors: *V. E. LEHMANN[1], M. P. HEYER[2], *P. BALI[3], O. ISSLER[4], A. RAMAKRISHNAN[5], K. BEAUMONT[5], E. J. NESTLER[6], P. J. KENNY[7];
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Abstract: Circular RNAs (circRNAs) are formed by back-splicing of pre-mRNA transcripts to generate a circularized product, many of which may have regulatory functions in the brain. Functional deficits in medium spiny neurons (MSNs) in the nucleus accumbens (NAc) may contribute to behavioral abnormalities in depression and other stress-related disorders. Here, we investigated the role of circRNAs in the NAc in regulating stress-induced behavioral deficits. We found that expression of circRIMS2, which is derived from exons 20-22 of the RIMS2 gene, was increased in postmortem NAc from male, but not female, patients who had suffered from major depressive disorder prior to death. Similar increases in cirRIMS2 were detected in the NAc of male mice subjected to social defeat stress (SDS). circRIMS2 overexpression in the NAc, accomplished using an AAVDJ-sEF1a-ZKSCAN1-circRIMS2-sYFP vector, decreased the expression of depression-like deficits in anxiety and reward-related behaviors in male mice subjected to SDS or restraint stress. Single-cell RNA sequencing (scRNAseq) was performed using 10X Chromium on NAc tissue from stressed and unstressed male mice. Stress induced profound alterations in the expression of genes relevant to mitochondrial-mediated bioenergetics including mitochondrial solute carriers, transcriptional plasticity that was reversed by circRIMS2 overexpression. Currently, we are exploring the role of circRIMS2-regulated mitochondrial genes with respect to stress-induced behavioral abnormalities.

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Poster

P708. Neural Mechanisms Underlying Emotional Liability and Stress Responsivity

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Topic: G.05. Mood Disorders

Support: NIDA U01DA043098
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The Hope for Depression Research Foundation (HDRF)
The Pritzker Neuropsychiatric Research Consortium

Title: Hippocampal differential gene expression converges across animal models of mood disorder: Results from an interactive meta-analysis pipeline encompassing five animal models

Authors: *Y. SANNAH[1], H. KHALIL[1], M. H. HAGENAUER[1], J. MARROCCO[2], C. AYDIN[1], Q. WEI[1], F. MENG[1], C. NASCA[2], E. J. NESTLER[3], M. MEANEY[4], S. J. WATSON[1], H. AKIL[1];
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Abstract: The Hope for Depression Research Foundation (HDRF) focuses on the neurobiology of mood disorders and brings together several research groups which have been conducting transcriptomic studies within different animal models used for the study of depression. Each animal model typically represents one dimension of a plethora of changes that might be occurring within the brains of depressed individuals, so integrating the results from across these studies is essential to gain a better understanding of mood disorders. To facilitate comparison, we created an interactive meta-analysis pipeline that allows the user to identify consistent gene expression signatures across custom-defined sets of animal models, conditions, transcriptional platforms, and brain regions. To illustrate utility, we tested this pipeline using thirteen microarray and RNA-seq data sets derived from either the hippocampus or dentate gyrus of five animal models: selectively-bred high responder (HR) and low responder (LR) rats, Flinders sensitive and resistant rats, mice with glucocorticoid receptor overexpression (early life and lifetime), chronic social defeat stress (CSDS) mice, and chronic corticosterone-treated mice (total n=146). To ensure comparability, the raw data from each study were first run through a standardized pipeline for annotation, quality control, and data preprocessing. We calculated the effect size (Hedge's *g*) for each gene from each study from the moderated t-statistic obtained as the result of a differential expression analysis conducted within the limma or limma-voom

pipeline. This effect size was then used within a random effects meta-analysis model. Interestingly, we found 27 candidate genes (FDR < 0.1) which showed similar differential expression in the hippocampus across these animal models of mood disorder despite the fact that a superficial comparison indicated minimal intersection between the top results from each individual study. These differentially expressed genes were highly skewed towards down-regulation (89%) and especially enriched within two particular hippocampal cell types: 63% enriched within astrocytes, 30% within ependymal cells. Gene Set Enrichment Analysis similarly indicated a particular enrichment within cell type related gene sets, including a down regulation within gene sets specific to the choroid plexus, ependyma, astrocytes, interneurons, polydendrocytes, and oligodendrocytes, and upregulation within gene sets specific to CA1 pyramidal neurons. These results illustrate the power of integrating results from numerous distinct models to understand the neurobiology of mood disorder.
In memory of Bruce McEwen.

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Poster

P708. Neural Mechanisms Underlying Emotional Liability and Stress Responsivity

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Title: Potential role of DCC-positive neurons in the adult prefrontal cortex projecting to the nucleus accumbens shell in susceptibility to social defeat stress

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Abstract: A consistent trait of depression-like behavioural states in humans and mice is elevated levels of the guidance cue receptor gene *DCC* in the prefrontal cortex (PFC). In fact, DCC receptors in the PFC play a causal role in susceptibility and resilience to chronic social defeat stress (CSDS) in adult mice, but the underlying mechanisms remain to be discovered. DCC receptors refine neuronal connectivity and plasticity throughout the lifespan, even in adulthood, by controlling axon arborization, dendritic arborization, and synapse formation. Here we

assessed whether DCC receptors play a role in stress susceptibility by modifying the architecture and connectivity of selective PFC pyramidal neuronal circuits. Using the retrograde tracer cholera toxin subunit B in adult male and female mice, we show that DCC-positive neurons in infralimbic (IL) and prelimbic (PrL) subregions of the PFC prominently innervate the nucleus accumbens core (NAcC) and nucleus accumbens shell (NAcS). We then examined CSDS-induced changes in dendritic spine density in DCC-positive neurons in the PFC projecting to the NAcS, by bilaterally microinfusing the AAVrg-CAG-GFP virus in the NAcS of adult male mice 3 weeks before CSDS exposure. Compared to control and resilient mice, susceptible mice show a reduction in thin and mushroom spine density in apical dendrites, but not in basal dendrites of NAcS-projecting DCC-positive pyramidal neurons. Moreover, compared to control and resilient mice, susceptible mice show reduced mushroom spine density in apical dendrites, but not in basal dendrites of PFC-NAcS DCC-positive projecting neurons. To evaluate the role of DCC receptor in PFC-NAcS pyramidal neurons in CSDS susceptibility, we deleted DCC in these neurons using $DCC^{lox/lox}$ mice and a Cre-dependent retrograde virus. Our preliminary findings show that deletion of DCC in PFC-NAcS circuitry induces resilience to CSDS. Our findings suggest that DCC receptors in adult PFC pyramidal neurons projecting to the NAcS play a causal role in susceptibility to social defeat stress in adulthood by inducing changes in their dendritic spine architecture.

Disclosures: A. Mahmud: None. **G.A. Hernandez:** None. **C. Flores:** None.

Poster

P708. Neural Mechanisms Underlying Emotional Liability and Stress Responsivity

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P708.08

Topic: G.05. Mood Disorders

Title: Gene expression changes in serotonin neurons due to Chronic Stress

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Abstract: Major Depressive Disorder (MDD) is a serious medical issue in the United States with over 16 million adults suffering from one or more major depressive episodes at any given time. Current antidepressant therapy is inadequate; with delayed efficacy and undesirable side effects, there is a clear unmet medical need for those requiring fast acting and continuous antidepressant therapy. Susceptibility to depression is related to decreased serotonin release, alterations in receptor levels, or impaired signal transduction. Serotonin neuron circuity can be studied by measuring neuronal firing, serotonin synaptic release, and serotonin receptor occupancy at presynaptic terminal regions. Another approach to identifying neurobiological mechanisms affected by chronic stress is to determine gene expression activity and functional networks and

pathways affected by stress. We utilized a mouse expressing enhanced green fluorescent protein (eGFP)-tagged ribosomes specifically in serotonergic neurons, which enabled visualization and isolation of mRNA being actively translated into protein using the translating ribosomal affinity purification (TRAP) method. An unpredictable chronic mild stressors (UCMS) protocol was conducted for 14 days to establish chronic stress, and the behavioral effects of stress were confirmed using the novelty suppressed feeding test and sucrose consumption test, and stress-induced weight loss was also observed. In order to explore gene expression changes in serotonin neurons due to chronic stress, TRAP was conducted on the dorsal raphe nucleus and actively translating mRNA were isolated. We identified expression of serotonin neuron-specific transcripts and differentially expressed genes due to chronic stress using TRAP-qPCR. Next Generation Sequencing will be conducted to expand our preliminary findings and identify differentially expressed transcripts, gene families, and networks affected by chronic stress in serotonin neurons. This work could determine novel underlying mechanisms important in the physiological and behavioral response to stress, and determine gene expression signatures in serotonin neurons.

Disclosures: A. Zhang: None. **M. Manners:** None.

Poster

P708. Neural Mechanisms Underlying Emotional Liability and Stress Responsivity

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Program #/Poster #: P708.09

Topic: G.05. Mood Disorders

Support: 5R01MH111918
5T32MH015174

Title: Role of pre-existing connectomic differences in driving susceptibility and resilience to chronic social defeat stress in mice

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Abstract: Stress is unavoidable, but there is inherit variability in stress responses across individuals. Some are susceptible while others display natural resilience. Stress is often a precipitating factor in the development of many preventable disorders, including depression and mood disorders. Understanding the neurobiological basis of the individual variability in stress can inform novel interventions and pathways to prevention. Many studies are accumulating evidence that suggests stress susceptibility and resiliency are not opposing responses but

represent distinct neural mechanisms. However, it is unclear if this neural circuitry is different prior to stress and if these differences pre-dispose individuals towards susceptibility or resiliency. In this study, we aim to uncover the pre-existing neural circuity that mediates individual differences in divergent stress responses. We utilize a chronic social defeat stress (CSDS) paradigm, a highly validated model of rodent stress that produces divergent responses. We expanded our paradigm to include females. In CSDS, aggressor male urine is applied to both male and female mice and a pair is exposed to an aggressor for 10 minutes followed by co-housing with an aggressor. This process repeats for 10 days total and is followed by a social interaction (SI) test. There is a reliable population spilt (n= 106) with 60% of females (SI res 1.61 ± 0.19, SI sus 0.62 ± 0.082) and 50% of males displaying resilient behavior (SI res 2.06 ± 0.29, SI sus 0.77 ± 0.44). To uncover pre-existing circuity, we previously validated an acute social defeat stress (ASDS) model. In ASDS, mice are exposed to 3 consecutive 5-minute bouts of defeat followed by SI 1-hour later. Here, we utilize both paradigms along with a TRAP2 mouse model that allows for temporal-targeting of activated neurons following an injection with tamoxifen with an indelible marker in combination with immunohistochemistry for a marker of cellular activation. We have validated the optimal trapping timeline through mass spectrometry of tamoxifen uptake into the brain (n=142, 14.42 ± 2.7 & 10.69 ± 1.07 μg/mLs at 120 & 150 mins for males & females) and western blot analysis of the Cre-ER protein in the amygdala following a defeat session (n=58, 86.4 +/-7.2 & 83.4 ± 2.9 μg at 90 mins for males & females). Using this timeline, we TRAP neurons during an acute stressor to capture initial stress-activated ensemble and compare it to cFos-activated neurons in the same individual after CSDS. By comparing these two time points in stress exposure, we provide evidence on how pre-existing connectivity patterns contribute to the consolidation of a chronic stressor in resilient versus susceptible mice.

Disclosures: K. Anderson: None. **A. Lipshutz:** None. **A. Manganaro:** None. **P. Rogu:** None. **V. Saltz:** None. **J.Y. Zhang:** None. **Y. Grossman:** None. **D. Dumitriu:** None.

Poster

P785. Neural Mechanisms of Decision-Making and Risk-Taking Behavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P785.01

Topic: H.03. Decision Making

Title: Effects of reproductive experience on cost-benefit decision making in females

Authors: *M. FARAJI[1], J. L. BIZON[2], B. SETLOW[1];
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Abstract: Effects of reproductive experience on cost-benefit decision making in females
Mojdeh Faraji[1,3], Jennifer L. Bizon[2,3], Barry Setlow[1,3]

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After decades of neglect, a growing body of behavioral neuroscience research is including female subjects, with significant findings that underscore the importance of this inclusion. These efforts, however, have largely neglected the variable of reproductive experience, in that almost all studies employ reproductively naïve animals. In particular, the majority of women undergo reproductive/maternal experience at some point in their lives, and pregnancy and childbirth are associated with alterations in risks of several psychiatric disorders. Research in rodents shows that maternal experience affects spatial learning and other aspects of hippocampal function. In contrast, there has been little or no investigation of how reproductive experience affects cost-benefit decision making, despite the relevance of this aspect of cognition for psychiatric disorders. To begin to address these issues, female Long-Evans rats (nulliparous, n=8; parous, n=8) were evaluated in tests of risky and impulsive decision making in standard operant chambers. Rats in the parous group were mated, gave birth, and nursed for 21 days until pup weaning, whereas rats in the nulliparous group were unmated. One week after weaning, rats began behavioral testing. Rats were initially tested in a delay discounting task, in which they chose between a small immediate food reward and a large food reward delivered after a variable delay period ranging from 0 to 60 s. There were no differences between parous and nulliparous rats in their preference for the large, delayed reward, suggesting no effects of reproductive experience on impulsive choice. In contrast, in a risky decision-making task, in which rats chose between a small, “safe” food reward and a large food reward accompanied by variable risks of mild shock punishment, parous rats showed significantly greater choice of the large, risky reward compared to the nulliparous rats. These latter data suggest that reproductive experience enhances risk-taking behavior. To determine whether enhanced risk taking in parous rats is due to greater food motivation, rats were tested on a progressive ratio schedule of reinforcement. There were no differences between groups, however, indicating comparable motivation to obtain the food rewards in the risky decision-making task. Together, these results show selective effects of reproductive experience on cost-benefit decision making.

Disclosures: M. Faraji: None. **J.L. Bizon:** None. **B. Setlow:** None.

Poster

P785. Neural Mechanisms of Decision-Making and Risk-Taking Behavior

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Program #/Poster #: P785.02

Topic: H.03. Decision Making

Support: RF1AG060778
R01DA036534
K99 DA041493

Title: Temporally-specific inhibition of ventral tegmental area dopamine neurons during decision making under risk of punishment.

Authors: *W. PYON, S. BLAES, C. ORSINI, J. BARRETT, H. HOLIK, S. SINGHAL, C. FRAZIER, B. SETLOW, J. BIZON;
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Abstract: Decision making represents an incredibly complex behavior that necessitates numerous different brain regions working in tandem to obtain a desired outcome. As our own understanding of dopamine's role in the brain improves, it is also becoming clear that the ventral tegmental area (VTA) plays a fundamental role in dictating how we make these decisions. To further elucidate the functional role of the dopamine in decision making, male and female tyrosine hydroxylase-cre rats were utilized to selectively drive halorhodopsin expression localized to dopaminergic neurons of the VTA. Rats were then trained on a risky decision-making task (RDT) in which they were tasked with repeatedly selecting between one of two levers: a small reward lever that dispensed one food pellet or a large reward lever that dispensed two food pellets accompanied by variable risks of mild footshock delivery (0%, 25%, 75%). After reaching stable choice performance, rats were implanted with optic fibers targeting the VTA through which 560 nm laser light was delivered to selectively inhibit dopaminergic neurons of the VTA during discrete timepoints during the choice process (during deliberation prior to lever selection, during receipt of the small reward, during receipt of the large reward with or without punishment, or during the intertrial interval). Preliminary findings show that inhibition of VTA dopamine neurons selectively during receipt of the large reward in the absence of punishment causes rats to become more likely to choose the small safe reward on subsequent trials. This shift toward risk-averse behavior is supported by our current understanding of reward prediction error signaling, with inhibition of dopamine neuron activity appearing to mimic a reduction of tonic firing as a result of negative prediction error.

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Poster

P785. Neural Mechanisms of Decision-Making and Risk-Taking Behavior

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Topic: H.03. Decision Making

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Alfred P. Sloan Fellowship

Title: Hormonal regulation of dopaminergic signaling and value-based decision-making

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Abstract: Value-based decision-making behavior varies across the reproductive cycle in women, but the underlying neural mechanisms of these effects are not well-understood. In reinforcement learning models of decision-making, a subject learns the value of an option from experience and updates that value based on reward prediction errors (RPE), or the difference between what was expected and what actually happened. Dopamine released in the nucleus accumbens core (NAcc) represents RPEs. Estradiol and progesterone activate dopaminergic signaling, suggesting that this modulation may be responsible for the influence of the reproductive cycle on value-based decision-making. We examined value-based decision-making in female rats over their estrous cycle with a novel task wherein they deliberate between waiting for an unspecified amount of time for a reward or moving on to the next trial. The length of time they wait provides an analog readout of subjective value. We manipulated rats' reward expectations over trials by varying the magnitude of rewards they recently received (local reward context). During estrus, rats' subjective value is more sensitive to local reward context. Mass spectrometry identified the downregulation of two proteins (Smpd3 and Sl6a4) involved in dopamine reuptake in the NAcc during estrus. Given this data, we hypothesized that dopamine release would be enhanced during estrus, perhaps leading to larger RPEs, which could account for behavioral differences in estrus. To estimate trial-by-trial RPE, we generated a behavioral model that includes principles of reinforcement learning. Using fiber photometry and a dopamine sensor, we observed dopaminergic encoding of RPE after the reward-predicting cue. This encoding of RPE was stronger during estrus compared to other stages, as suggested by our proteomic results. Altogether, these findings suggest that gonadal hormones promote the downregulation of proteins that mediate dopamine reuptake in the NAcc in estrus, leading to enhanced encoding of RPEs and learning. Moreover, we have identified novel molecular targets of hormonal regulation of dopamine signaling. Finally, we emphasize that this computational, decision theoretic approach provides a principled framework for interpreting behavioral, neural, and molecular profiling data, and represents a powerful approach for studying endocrine systems in the brain.

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Poster

P785. Neural Mechanisms of Decision-Making and Risk-Taking Behavior

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Program #/Poster #: P785.04

Topic: H.03. Decision Making

Support: CIHR Grant (PJT-162312)

Title: Gambling cues facilitate ropinirole-induced deficits in decision making by blocking protective adaptions in midbrain dopamine neurons and the prelimbic cortex

Authors: *S. RAMAIAH, L. MORTAZAVI, T. HYNES, C. CHERNOFF, H. BRODIE, B. A. HATHAWAY, B. RUSSELL, S. KAUR, C. WINSTANLEY;
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Abstract: The alluring lights and sounds of modern electronic gambling machines may contribute to their addictive potential. Indeed, the addition of casino-like cues to laboratory-based gambling tasks increases risk-taking in both humans and rats. These heavily-cued probabilistic schedules of reinforcement may drive the mesolimbic dopamine system, causing neuroadaptations that promote risk-taking. Supporting the idea that repeated agonism of the dopamine system can promote risk-taking and problem gambling is evidence that the D2/3 receptor agonists ropinirole and pramipexole (used in the treatment of Parkinson's) can induce impulse control and addiction disorders in a significant minority of patients. We, therefore, asked whether chronic administration of ropinirole could amplify cue-induced risk-taking and impulsivity in a translationally valid model of gambling-like behaviour - the rat gambling task. We furthermore asked whether the presence of cues in the task would exacerbate the deleterious effect of ropinirole treatment. To this end, we implanted rats with osmotic minipumps to chronically deliver ropinirole or saline. The instrumented rats were then trained in either the cued or uncued rat gambling tasks. In both versions of the task, chronic ropinirole increased impulsivity, but only in the cued task was risk-taking exacerbated. Following cessation of ropinirole treatment, enhanced risk-taking persisted in the cued task and, surprisingly, emerged in the uncued gambling task. Ex-vivo analyses of neuronal activity patterns (i.e., c-Fos immunohistochemistry) provided evidence as to why, in the absence of cues, rats were spared from ropinirole-induced deficits in decision making. Specifically, the activity of midbrain dopamine neurons was reduced in ropinirole-treated animals trained in the uncued task, likely in compensation to pharmacological dopamine agonism. Ropinirole furthermore provoked an increase in prelimbic activity in the uncued task, which may indicate the compensatory recruitment circuits that titrate dopamine release at the terminal level. In the absence of ropinirole, these adaptions could lead to a hypodopaminergic (i.e., reward-deficient) state, explaining the increased risk-taking following cessation of ropinirole treatment.

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Poster

P785. Neural Mechanisms of Decision-Making and Risk-Taking Behavior

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Topic: H.03. Decision Making

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Title: Zebrafish capable of generating future state prediction error show improved active avoidance behavior in virtual reality

Authors: *M. TORIGOE[1], T. ISLAM[1,3], H. KAKINUMA[1,3], C. A. FUNG[4], T. ISOMURA[2], H. SHIMAZAKI[5], T. AOKI[1], T. FUKAI[4], H. OKAMOTO[1,3];
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Abstract: Selecting optimal actions in decision making according to the current sensory input is essential for animals. One prevailing model underlying this behavioral process is based on the idea that the ultimate aim of choice is to maximize utility or reward. In addition to this, adaptive behavior requires animals to generate the internal model of their environment and to take actions to minimize surprise (*i.e.* improbability or prediction error) about the state they encounter in comparison with the state predicted from the internal model. How these principles are actually adopted by animals and are reflected in their behavior remains unknown. We addressed this question using the closed-loop virtual reality-2photon calcium imaging system, in which the scenery presented to adult zebrafish by the surrounding displays moved backwardly in response to the tail beat of the fish. In the virtual reality, fish was trained for active and passive avoidance. The activity of excitatory neurons expressing G-CaMP7 in the dorsal pallium of the telencephalon during the training was imaged. The adult zebrafish can learn various adaptive behaviors and has the dorsal pallium-basal ganglia circuit implicated in behavioral selection. In the active avoidance, fish had to escape from the region surrounded by blue walls to the region in their front surrounded by red walls to avoid electric shock. In the passive avoidance, fish had to stay in the red wall region where the tasks started. Analysis of the dorsal pallium's neural activity by non-negative matrix factorization revealed neural ensembles assigning rules to the colors of the surrounding walls (e.g. blue color is dangerous, or red color is safe). Furthermore, conversion

of the virtual reality system to the open-loop condition, where the scenery did not respond to the fish tail beat, revealed that one-third of learner fish generated another ensemble encoding the prediction error of the perceived scenery's status compared with the predicted favorable status. Intriguingly, the fish with both ensembles escaped more efficiently than the fish with the former ensemble alone, implicating that the latter ensemble in fact guided zebrafish to take action to minimize this prediction error.Our results suggest that some zebrafish, if not all, can use both of the two principles for achieving safe escape in active avoidance, *i.e.* value maximization and surprise minimization. Further, under limited time constraint of the trials, zebrafish using both principles exhibit optimized escape behavior compared with zebrafish behaving with only the reward-based principle.

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Poster

P785. Neural Mechanisms of Decision-Making and Risk-Taking Behavior

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P785.06

Topic: H.03. Decision Making

Title: A new assay to study the genetic and neuronal process behind decision making in fruit flies

Authors: *C. E. LADD, J. H. SIMPSON;
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Abstract: Decision making is a vitally important, but poorly understood part of human and animal life. Humans with Parkinson’s disease, Conduct Disorder, and Alcohol Use Disorder show defects in decision making, so a better understanding of the mechanisms could eventually have clinical impact. To study the underlying neuronal and genetic process of decision making, I have established a new assay using the fruit fly, *Drosophila melanogaster* that presents pairs of competing stimuli to induce flies to choose between grooming, courting, and feeding. The assay does not require training, reward, or punishment. Since all of these behaviors are innate, natural, and advantageous under different circumstances, the assay creates three decisions that lack a “correct” answer. I use varying contextual conditions to determine how the behavior changes with response to increasing and decreasing drive strengths. I found that there is no simple hierarchy of needs between the three behaviors. The flies alternate between behaviors to satisfy competing drives. As competing drives become closer in strength, the incidences of alternations increase until the fly is rapidly and continuously switching between behaviors. I also found that the three behaviors are not mutually exclusive acts (e.g. a hungry and dirty fly will groom while

eating). As the respective drive strengths become closer, an increase in moments of simultaneous behaviors occurs.
Previous fly decision-making assays have identified genes and neurons that affect sensory detection thresholds or contextual states (such as hunger levels or mating drive). I will test the candidates from these assays (FoxP and Leukokinin receptor mutants and mushroom body and central complex neurons) to see if decision-making in general is impaired, or if the process is specific to each decision. The advantages of the fly for genetic screens and neural circuit mapping, employed in this the new assay, will help illuminate the neural basis for decision making.

Disclosures: C.E. Ladd: None. **J.H. Simpson:** None.

Poster

P785. Neural Mechanisms of Decision-Making and Risk-Taking Behavior

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Program #/Poster #: P785.07

Topic: H.03. Decision Making

Support: NIH Grant R15DA046797

Title: Dopaminergic modulation of decision-making guided by delayed punishment

Authors: *G. L. MINNES[1], A. J. WIENER[1], A. E. LILEY[1], D. B. GABRIEL[2], N. W. SIMON[2]; [2]Psychology, [1]Univ. of Memphis, Memphis, TN

Abstract: Effective decision-making requires evaluation of the rewards and consequences associated with each available option. Critically, negative outcomes often occur later in time, leading to underestimation, or “discounting”, of these delayed consequences. To investigate sensitivity to immediate vs delayed punishment during cost/benefit decision-making in rats, we developed the Delayed Punishment Decision-making Task (DPDT). In brief, rats choose between two levers, one resulting in a single-pellet reward, and the other resulting in a three-pellet reward accompanied by a mild foot shock punishment. The shock is initially delivered immediately following a decision; as the session progresses, punishment is preceded by a systematically increasing delay (0, 4, 8, 12, 16 s). On average, rats increase choice of the punished option as delay increases, indicative of discounting of delayed punishment. Here, we tested the effects of systemic administration of dopaminergic drugs on sensitivity to delayed punishment in male and female adult rats. We observed that cocaine, a dopamine and norepinephrine reuptake inhibitor, caused a dose-dependent reduction in choice of delayed but not immediate punishment. Interestingly, this effect was more pronounced in females than males. Selective D1 dopamine receptor activation or blockade did not affect decision-making making, whereas D2 dopamine receptor activation reduced punishment discounting comparably to

cocaine. D2 dopamine receptor blockade had no effects on punishment driven decision-making. Collectively, these data demonstrate that dopamine transmission modulates sensitivity to delayed punishment, and this is likely regulated by D2 dopamine receptor activation.

Disclosures: G.L. Minnes: None. **A.J. Wiener:** None. **A.E. Liley:** None. **D.B. Gabriel:** None. **N.W. Simon:** None.

Poster

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Topic: H.03. Decision Making

Support: NIH Grant R15DA046797

Title: Orbitofrontal Cortex and Basolateral Amygdala Regulate Decision-making Guided by Delayed vs. Immediate Punishment

Authors: *A. E. LILEY[1], L. M. CLARK[2], E. D. EDDINS[3], S. E. CARTWRIGHT[1], D. B. GABRIEL[1], N. W. SIMON[1];
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Abstract: Undervaluation of negative consequences is observed in multiple psychiatric disorders, and often engenders detrimental health, financial, and social outcomes. The majority of the preclinical research on punished reward-seeking utilizes punishment that occurs immediately after a decision; however, real-world punishment often occurs long after a decision has been made. We developed the rat Delayed Punishment Decision-making Task (DPDT), which reveals that rats avoid rewards associated with immediate punishment, but increase choice of the same reward/punishment combination when punishment is delayed. Therefore, as in humans, rats underestimate, or "discount", delayed punishment. Here, we investigated the role of lateral orbitofrontal cortex (lOFC) and basolateral amygdala (BLA) in the discounting of delayed punishment. Male and female rats (n=45) were trained in DPDT with either ascending (0,4,8,12,16s) or descending delays (16,12,8,4,0s), then lOFC or BLA were pharmacologically inactivated prior to task performance. We observed that lOFC inactivation did not affect choice of rewards associated with immediate punishment, but decreased choice of delayed punishment. Interestingly, this effect was observed in the standard DPDT with ascending delays, but did not occur with reversed contingencies. BLA inactivation evoked complex effects on decision-making, driving increased choice of punished rewards with short delays, and reduced choice of punishment with longer delays. These data suggest that lOFC conditionally regulates the

discounting of delayed punishment, and may serve as therapeutic target in disorders characterized by insensitivity to delayed consequences.

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Poster

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Topic: H.03. Decision Making

Support: NIH Grant F31DA050458
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Title: Neuronal activity in lateral orbitofrontal cortex predicts risk of punishment during decision-making in rats

Authors: *D. B. K. GABRIEL[1], F. HAVUGIMANA[1], A. E. LILEY[1], I. AGUILAR[1], G. MINNES[1], G. R. JOHNSON[1], M. YEASIN[2], N. W. SIMON[1];
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Abstract: Risky decision-making is defined as preference for rewards associated with the risk of negative consequences. The rat risky decision-making task (RDT) captures this phenotype by presenting subjects with choice between either a small food reward or a large reward potentially accompanied by a mild footshock. Lateral orbitofrontal cortex (lOFC) has been identified as a brain region that regulates punishment-based risky decision-making; however, the functional neuronal activity in lOFC that encodes risk level and risky choice remains unknown. Here, we employed single-unit electrophysiology to record extracellular neuronal activity from lOFC of rat during a version of RDT optimized for electrophysiology. In this task, rats were required to hold a 1 sec response, which we define as a "deliberation hold", before levers were extended for reward choice. Following this deliberation period, subjects chose first between small (1 pellet) and large (3 pellet) rewards, then in a second block progress to small rewards vs. large rewards with 50% risk of footshock. We recorded from 148 units in lOFC and observed that subsets of units were either activated or inhibited by specific task events, with this selectivity being altered by risk level and reward magnitude. Notably, during the "pre-decision" period (.5 sec immediately before choice), units were activated prior to large reward choice during safe but not risky trials, suggesting that lOFC signals information about reward safety immediately prior to a decision. Additionally, a subset of units was suppressed during post-decision anticipation of small rewards but was unaffected during anticipation of large rewards regardless of risk level, suggesting that these neurons encode information about impending reward magnitude. Finally,

we applied a parameter-optimized Random Forest classifier to predict the presence of risk of punishment during any given trial using neural features from different events in the decision-making process. We observed that neuronal activity during the deliberation hold displayed the highest classification accuracy of all task events (96.53%), suggesting that lOFC activity encodes risk level prior to a decision. Furthermore, combining lOFC neuronal activity across all relevant task events was more effective at predicting risk than any individual event (98.2% accuracy). In summary, lOFC neurons encode multiple events during risky decision-making in rats, and lOFC activity predicts both presence/absence of risk and reward magnitude. This suggests that manipulating lOFC activity may serve as a promising treatment to increase awareness of risky outcomes during decision-making.

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Poster

P785. Neural Mechanisms of Decision-Making and Risk-Taking Behavior

Location: Virtual Only

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Program #/Poster #: P785.10

Topic: H.03. Decision Making

Support: Wellcome Trust Sir Henry Dale Fellowship 206207/Z/17/Z (FG)
Taiwan Ministry of Education (F-Y H)

Title: Nutrient-sensitive reinforcement learning in rhesus monkeys

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Abstract: Animals make adaptive food choices to acquire essential nutrients for survival. In reinforcement learning (RL), animals choose by assigning values to objects and update these values with experience. This framework has been instrumental in identifying neural mechanisms of learning and decision-making. However, canonical RL models do not address how learning depends on biologically critical constituents of food rewards, such as nutrients, and how internal physiological states modulate adaptive food choices. Here, we addressed these questions in rhesus monkeys (*Macaca mulatta*) choosing nutrient-defined food rewards under varying reward probabilities. We trained two adult male monkeys in a probabilistic reversal learning task involving four nutrient-defined liquids (monkey Ya: 17-19 kg, age: 6 years, N = 4,684 trials; monkey Ym: 12-13 kg, age 6 years, N = 5,321 trials). Specifically, these liquids were prepared with 2 x 2 fat and sugar levels to examine the effects of fat and sugar on learning and decision-making. All liquids were cued by fixed but novel visual stimuli within each testing session and

were matched in flavor (blackcurrant or peach), temperature, and other ingredients (protein, salt, etc). Therefore, differential learning and choice patterns could only be attributed to the specific nutrient content of the liquids, rather than preference for the visual stimuli or other attributes of the liquids. We found that the nutrient composition of rewards strongly influenced monkeys' choices and learning. The animals preferred rewards high in nutrient content and showed individual preferences for fat and sugar. These nutrient preferences affected how the animals adapted to changing reward probabilities: the monkeys learned faster from preferred nutrient rewards and chose them frequently even when they were associated with lower reward probability. Although more recent experiences generally had a stronger influence on monkeys' choices, the impact of reward history depended on the rewards' nutrient composition and individual nutrient preferences. From these data, we propose a nutrient-sensitive RL model that separately updates the value of fat and sugar components based on subjective nutrient preferences and integrates them into scalar reward values to explain the monkeys' choices. Our findings suggest that preference for nutrient constituents contributes to subjective valuation, learning, and choice of food rewards. Incorporating nutrient value functions into RL models enhances their biological validity and helps reveal unrecognized nutrient-specific learning and decision computations.

Disclosures: F. Huang: None. **F. Grabenhorst:** None.

Poster

P796. Primate Prefrontal Cortex: Circuit Mechanisms

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Program #/Poster #: P796.01

Topic: H.04. Executive Functions

Support: NEI R01-EY029666

Title: The connectome of primate lateral prefrontal cortex

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Abstract: Lateral prefrontal cortex (LPFC) plays an important role in executive control, but how it interacts with the rest of the cortex remains unclear. To address this, we densely mapped the cortical connectome of LPFC, using electrical microstimulation combined with functional MRI (EM-fMRI).
Two macaque monkeys were scanned in 3T magnets under propofol anesthesia. In each animal, we stimulated ~100 sites within ~15x20 mm^2 of cortex in caudal-to-mid LPFC, which likely overlaps with areas 9, 46, 8, 45, and 12. At each stimulation site, we collected up to 34 mins of MION signals at 2-mm isotropic resolution. We delivered 500-μA current pulses at 333 Hz for

210 ms per second in 30-sec blocks. The parameters were tuned to optimize contrast-to-noise ratio.
When downsampled to the level of cytoarchitectonic areas, the dense connectome was highly consistent with a state-of-the-art inter-areal connectome mapped by tracing (Markov et al., 2014), even for long-range connections. Furthermore, the EM-fMRI data exhibited statistical traits comparable to tracing, such as 1) following log-normal distribution; 2) decaying exponentially as inter-areal distance increases; 3) showing a wide range of connection weights that are highly characteristic across areas and rather consistent across subjects.
Importantly, utilizing the high density of spatial sampling (extrapolating to ~2500 sites/hemisphere/animal if applied to the entire cortex), we were able to capture significant and systematic differences of connections across adjacent LPFC sites, whereas sparser sampling could have missed such differences, or worse, have been misleading due to aliasing. We found isomorphic mappings between LPFC and five major processing domains comprising most of the cerebral cortex except early sensory and motor areas. As one moves in an orderly fashion across LPFC sites, the bulk of their connections progress systematically on the cortical surface of each domain. Taken together, the LPFC grid of stimulation sites topographically mapped to separate grids of activation sites in the five domains, coarsely resembling how the visual cortex maps the retina. The temporal and parietal maps largely overlapped in LPFC, potentially allowing integration of the ventral and dorsal streams, and the other maps overlapped at least partially. Thus, the LPFC contains overlapping, millimeter-scale maps that mirror the organization of major cortical processing domains, supporting LPFC's role in coordinating activity within and across these domains.

Disclosures: R. Xu: None. **N.P. Bichot:** None. **A. Takahashi:** None. **R. Desimone:** None.

Poster

P796. Primate Prefrontal Cortex: Circuit Mechanisms

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Program #/Poster #: P796.02

Topic: H.04. Executive Functions

Title: Mapping of stimulus-response associations in the macaque prefrontal cortex: contribution of putative pyramidal cells and interneurons

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Abstract: Prefrontal cortex (PFC) plays a key role in several cognitive processes related to goal-directed behavior, including working memory and mapping associations between stimuli and responses. Several neurophysiological studies have elucidated the involvement of different areas

within the PFC in these processes. However, relatively little is known at the microcircuitry level, especially how local cell-types contribute to these processes. We investigated the population coding proprieties of dorsolateral and mediolateral neurons of PFC classified as broad-spiking (BS) putative pyramidal cells (390 cells) and narrow-spiking (NS) putative inhibitory interneurons (278 cells) according to the trough-to-peak duration feature of their extracellular waveforms. The recording was performed while two rhesus macaques performed a stimulus-response association task where during each trial, after a fixation period, one of the three instruction stimuli (IS) and three potential targets appeared for a delay period. Then the IS disappeared and served as go-signal to perform a saccade toward one of the three targets. After a period of maintenance of the target fixation, a reward was delivered if the correct target was chosen. We applied a decoding algorithm to quantify the ability of the BS and NS populations to encode the associations during the delay period where monkeys were required to generate the correct response associated with the stimulus and retain it in memory until the subsequent execution phase. We found that NS population encoded associations more strongly in the early phase of the delay (150-400ms) than BS population. Next, we investigated how BS and NS populations represented the associations over time according to a static or dynamic temporal coding scheme during such a memory period. For this purpose, we used a cross-temporal decoding approach in which the decoder was trained and tested using different time periods. A direct comparison of the population coding schemes during delay highlighted how the coding scheme is characterized by a dynamic coding in NS population compared to BS population, where the representation was static. These findings shed light on the cellular mechanisms underlying local processing of associative information, suggesting an essential role of interneurons through a specific coding scheme compared to pyramidal neurons.

Disclosures: F. Ceccarelli: None. **A. Genovesio:** None.

Poster

P796. Primate Prefrontal Cortex: Circuit Mechanisms

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Title: Neuronal congruency signals in macaque frontal cortex

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Abstract: Cognitive control is crucial for flexible and goal-directed behavior, allowing subjects to swiftly adapt to ever-changing environments and circumstances. Conflict tasks (e.g., Stroop, Simon, Flanker, and Wisconsin card sorting tasks) have been instrumental to study cognitive control. The hallmark of such conflict tasks is a behavioral congruency effect, whereby subjects respond faster and more accurate in congruent (e.g. blue printed in blue in the Stroop task) than incongruent trials (e.g. blue printed in red). Despite almost a century of research since Stroop's original conflict study in 1935, however, the exact neuronal mechanisms underlying congruency effects are still poorly understood. To investigate how the congruency signal developed in the frontal cortex, we trained two monkeys to perform a novel rule-based attention-conflict task, which required the monkeys to covertly attend to a target and detect its dimming by pressing a button while ignoring distractor stimuli. The target was determined by a combination of a task rule-cue (color or spatial rule), and a color-cue (pink or red). A trial was considered congruent or incongruent when the two rules indicated the same or a different target location under both rules for a given color-cue.By recording single unit activity in the frontal eye fields (FEF), an area critically involved in spatial attention and target selection, we discovered a neuronal congruency representation which was composed of a concurrent facilitation effect in task-relevant neurons (when the target was in the neurons' RF, the neurons responded higher in the congruent trials than incongruent trials) and an interference effect in task-irrelevant neurons (when the distractor was in the neurons' RF, the neurons responded higher in the incongruent trials than congruent trials). Compared to the incongruent trials, the neuronal facilitation effect increases the signal, while the interference effect decreases noise to detect the target in the congruent trials. By combining the facilitation and interference effect, a better signal to noise ratio to detect the target is achieved in congruent than in incongruent trials in FEF, which causes better behavioral performance in congruent trials. Based on our results, the neuronal facilitation and interference effect can explain behavioral congruency effects of most, if not all, conflict tasks. Meanwhile, our results explain observed imaging and low spatial-resolution electrophysiological (fMRI, EEG, EMG) data obtained using conflict tasks in humans. These results significantly advance our understanding of the neuronal mechanisms underlying conflict processing and cognitive control.

Disclosures: T. Yao: None. **W. Vanduffel:** None.

Poster

P796. Primate Prefrontal Cortex: Circuit Mechanisms

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Program #/Poster #: P796.04

Topic: H.04. Executive Functions

Support: CIHR

Title: Movements do not dominate neural dynamics in the macaque prefrontal cortex

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Abstract: Cognitive neurophysiology experiments in laboratory macaques commonly place movement restraints for practical purposes. Hence, most of what we know about the single neuron basis of cognition in the primate brain often does not account for the influence of non-task-related movements (i.e., "uninstructed") - though these movements co-occur with cognitive processing in natural settings. In parallel, evidence from mice show that uninstructed movements, when measured, dominate cortical dynamics during task performance (Musall et al. 2019, Nat. Neuro), which questions the interpretation of many primate cognitive neurophysiology experiments. Here, we addressed this concern by investigating neural activity in monkeys performing a classical cognitive task while free to move their body, head and eyes. We video-tracked instructed (i.e., task-based) and uninstructed movements while recording from Utah arrays implanted in prefrontal area 8A (N=4,414 units) - a region causally involved in the cognitive task monkeys performed. Single-neuron analyses and ensemble-based machine learning demonstrated that cognitive signals can be decoded with single-trial resolution and high accuracy despite free movement in monkeys. Moreover, replicating Musall et al.'s analytical approach, we found that uninstructed movements did not explain most of the neural variability, as opposed to what was found in mice. Instead, cognitive task variables dominated neural dynamics, though movements still explained a significant amount of variance. These findings demonstrate that single-trial neural dynamics in the monkey prefrontal cortex reliably encode cognitive processing even during free movement.

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Poster

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Title: Neuronal synchronization between the primate anterior cingulate cortex and the amygdala underlying vicarious reward

Authors: *P. PUTNAM[1], C.-C. J. CHU[1], N. A. FAGAN[1], O. DAL MONTE[2], S. W. CHANG[1]; [2]Dept. of Psychology, [1]Yale Univ., New Haven, CT

Abstract: Vicarious reinforcement powerfully impacts social learning and decision-making. Research largely supports the role of the anterior cingulate cortex (ACC) in processing vicarious reward, but its network-level coordination with other brain structures is unclear. We investigated neuronal synchrony between the gyrus of ACC and the basolateral amygdala (BLA) while monkeys delivered rewards either to themselves (self) or to a conspecific (other). To examine neural synchrony, we computed spike-field coherence from simultaneous electrophysiological recordings of the ACC and BLA. Upon delivery of the reward, coherence between the spikes in ACC and the local field potential in BLA increased in in the gamma frequency band (30-50 Hz) for rewards delivered to other, but not self. By contrast, coherence between the spikes in ACC and the local field potential in BLA increased in the alpha/beta frequency band (8-20 Hz) for rewards delivered to self, but not others. These enhancements in coherence were time-locked to the onset of reward delivery and crucially relied upon dissociated frequency bands for processing reward to self or other in an agent-specific manner. Moreover, the increase in ACC-BLA coherence for other's reward was specifically found during reward delivery following free-choice trials, in which monkeys actively expressed other-regarding preference and was not present following forced-choice trials with only a single outcome option. Our results therefore support that vicarious reward processing in the ACC-BLA circuits occurs through a frequency-selective communication channel that is distinct from processing self-reward during social decision-making. Together, these findings emphasize the role of network-level coordination between ACC and BLA facilitating vicarious reward processing following prosocial decisions.

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Poster

P796. Primate Prefrontal Cortex: Circuit Mechanisms

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Program #/Poster #: P796.06

Topic: H.06. Social Cognition

Support: R01MH110750
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Title: A causal role of the primate prefrontal cortex in social gaze monitoring: evidence from closed-loop microstimulation

Authors: *S. FAN[1], O. DAL MONTE[1,5], N. A. FAGAN[1], A. NAIR[1], C. C. J. CHU[1], M. B. ZHOU[1], P. T. PUTNAM[1], S. W. CHANG[1,2,3,4];
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Abstract: Social gaze interaction serves an important function in communicating social information. The primate prefrontal cortex has been implicated in multiple social behaviors, including social learning and decision-making, as well as higher-order face processing. Yet, how the prefrontal areas encode social information used for real-life social gaze interaction remains elusive. We recorded single-unit activity from three prefrontal cortical areas during spontaneous social gaze interaction between pairs of macaques - the orbitofrontal cortex (OFC, n=241), rostral anterior cingulate gyrus (ACCg, n=236), and dorsomedial prefrontal cortex (dmPFC, n=187). Notably, many neurons in these areas parametrically tracked the recorded monkey's gaze positions relative to the partner monkey's eyes ('Self-distance'), the partner monkey's gaze positions relative to the recorded monkey's eyes ('Other-distance'), and the distance between the gaze positions of the two monkeys ('Joint-distance'), supporting a single-cell mechanism of social gaze monitoring. To further test if these prefrontal areas causally contribute to social gaze monitoring based on tracking social gaze distance, we applied closed-loop microstimulations in each of these areas (n=15 sites per area). Microstimulations (100 Hz, 75 microA, 200 msec with a probability of 50%) were delivered contingently upon the precise moment of looking at the partner's eyes in the social condition or random dot motion stimuli in the non-social condition. Microstimulations led the monkey to look closer to the partner by significantly decreasing 'Self-distance'. Critically, this facilitation of attention was specific to the social condition and occurred in the absence of any change in saccade kinematics. Moreover, microstimulations in OFC greatly increased the likelihood of fixating on partner's face, indicated by a gain change of the social attention decay curve, as well as both increased frequency and longer duration of fixations on the face. Taken together, all three prefrontal areas, OFC, ACCg and dmPFC, were causally involved in social gaze monitoring based on tracking where one looked relative to another social agent, with OFC exhibiting the strongest effect that specifically increased fixations on partner's face. These results suggested that multiple prefrontal areas within the primate social interaction networks causally contribute to social gaze interaction.

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Poster

P796. Primate Prefrontal Cortex: Circuit Mechanisms

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Program #/Poster #: P796.07

Topic: H.06. Social Cognition

Title: Neurons in the macaque amygdala encode face animacy and self-relevant gaze shift

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Abstract: With the development of animation and computer graphics technology, people are exposed to various kinds of faces, ranging from realistic faces (high animacy) to cartoon-like faces (low animacy). Human observers are readily capable of extracting socially-relevant information (e.g., gaze direction) from both real and artificial faces. The neural basis of this cognitive capability remains unknown. To address this issue, we performed behavioral and electrophysiological experiments while monkeys were presented with face stimuli with three different degrees of animacy. A high-animacy face stimulus was a frontal face image of a real monkey. A low-animacy face stimulus was created by replacing the eyes, nose, and mouth of the high-animacy face image with simple diagrams. These two images were morphed into a middle-animacy face stimulus. Each face stimulus had one of three eye directions: direct (D), left (L), and right (R). Each trial began when a fixation point (FP) appeared at the center of a display. After the monkey had fixated at the FP for 500 ms, one of the face stimuli was presented at the display center. The monkey was then required to hold fixation at the FP for another 1000 ms. At 500 ms from the stimulus onset, the eye direction of the face shifted in either of three ways: *toward* (L to D or R to D), *averted* (D to L or D to R), or *non-related* (L to R or R to L). Successful central fixation was followed by a reward after a 500-ms delay. Single-unit activity was recorded in the amygdala, a brain region known for processing facial information. Of 1335 neurons recorded, 502 were classified as task-related. Among these, activities of 113 neurons (22%) were modulated by the face animacy. Neurons preferring the high and low animacy were predominant and equally common (n = 44/113 for both populations), while neurons preferring the middle-animacy face were in the minority (n = 25/113). Most of the neurons modulated by gaze direction (n = 39/49) or gaze shift (n = 52/58) were insensitive to the face animacy. Furthermore, the gaze shift-related neurons were modulated most strongly in the *toward* (n = 26/58) or *averted* conditions (n = 21/58). The present results indicate that there are two distinct populations in the amygdala: neurons encoding face animacy and neurons encoding self-relevant gaze shift of observed faces. The amygdala may provide a neural mechanism with which observers are capable of processing important social signals from real and artificial faces.

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Poster

P796. Primate Prefrontal Cortex: Circuit Mechanisms

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Topic: H.06. Social Cognition

Support: R01MH110750
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Title: Extending Fano Factor to naturalistic paradigms: neural variability is suppressed during spontaneous gaze interactions

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Abstract: Fano factor (FF) is a measure extensively used to characterize variability, calculated as a neuron's spike-count variance over the spike-count mean. Most previous studies assumed that FF remains mostly stable across trials (Gur et al., 1997; Carandini, 2004), but later work has suggested that stimulus onset drives a decline in the variability of neural activity in several brain regions across multiple processing domains (Churchland et al., 2010). Here, we extend this finding and show that the decline in FF is not limited to structured stimuli or task events, but also applies to naturalistic behavioral paradigms such as social gaze interaction. In this study, we recorded single-unit activity from four brain regions implicated in social behaviors, including the basolateral amygdala (BLA, n=537), orbitofrontal cortex (OFC, n=241), rostral anterior cingulate gyrus (ACCg, n=236), and dorsomedial prefrontal cortex (dmPFC, n=187) while pairs of macaques engaged in spontaneous, face-to-face, social gaze interaction, without any task constraints or reward contingencies. For each neuron, we characterized its FF trace by computing the variance of spike counts across trials and observed different types of modulation. To quantify neurons with decreasing or increasing FF modulation, we fitted a linear regression on each neuron's FF trace from 200 msec before to 350 msec after the onset of gaze events associated with looking at the conspecific's eyes, face, or non-social objects. There were greater proportions of BLA and OFC neurons that exhibited significantly decreasing (rather than increasing) firing-rate variability when looking at a partner monkey's eyes or face. In contrast, there was no difference in such proportion in ACCg or dmPFC. Crucially, this decrease in neural variability in BLA and OFC was specific to social gaze events, and not observed when looking at a non-social object. Furthermore, these findings remained robust across various time windows used to examine the decline in neural variability. These results suggest that BLA and OFC firing-rate variances can be stabilized by looking at a social agent during spontaneous social gaze exchanges. More broadly, our findings indicate that the phenomenon of neural variability

reduction in the brain is not limited to processing structured experimental stimuli or task but is also observed during naturalistic behaviors.

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Poster

P796. Primate Prefrontal Cortex: Circuit Mechanisms

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Title: Anterior cingulate cortex encodes functionally distinct error signals

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Abstract: Anterior Cingulate Cortex (ACC) is central to the adaptive control of behavior, monitoring outcomes, and comparing them against predictions. Discrepancies between outcomes and predictions can result in different forms of error signals: An internally generated one upon executing an incorrect response (Error), a prediction error signal based on an unexpected feedback cue (FPE), and a reward prediction error (RPE). These types of error computations are demonstrated in the ACC but the degree to which their neural substrates overlap remains unclear. To address this, we sampled neural activity using linear electrode arrays, from the dorsal (area 6/32) and ventral (area 24c) banks of the ACC of two macaques performing a saccade stop-signal task. Subjects were required to generate a saccade to a peripheral target, but to inhibit this planned saccade when a stop-signal appeared. The stop-signal timing was adjusted to ensure ~50% successful inhibition on stop trials. Following the response, an auditory tone indicated whether reward will be provided. Reward amount was asymmetric for the two directions with the association flipped on a block-by-block basis. Errors occurred when monkeys generated saccade despite the appearance of stop-signal. FPE occurred when a correct saccade was followed by the feedback tone indicating no upcoming reward. RPE occurred immediately after block switch when the monkey received more (positive RPE) or less (negative RPE) reward than expected.

Analysis of a preliminary sample of 538 units recorded from 992 linear electrode contacts across 18 sites, revealed units signaling Error, FPE, and RPE in ACC. These signals were commonly found in distinct neurons and although some multiplexing occurred, no systematic patterns emerged. Furthermore, some Error neurons also exhibited modulation following predictable negative feedback tone on Error trials but not following unpredictable tone on FPE trials, suggesting a role in error validation. These results suggest that different modes of error processing involve distinct but partially overlapping functional units within ACC. These findings can guide models of ACC function and contribute to interpretations of non-invasive electrophysiological indices, namely the Error- and Feedback-Related Negativity.

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Poster

P824. Prefrontal Cortex

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Support: CCTST Pilot Translational Research & Innovative Core Grant

Title: Neural mass model based study of frontal-temporal theta oscillations in human subjects during the performance of a cognitive control task

Authors: A. ROSS[1], K. FARNES[1], A. C. PAULK[2], A. S. WIDGE[3], S. S. CASH[4], J. E. TORRES-REVERON[1], ***I. BASU**[1];
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Abstract: Neuropsychiatric disorders are the number one cause of disability and health-related economic burden in the United States. This is partly because established therapies primarily focus on diagnostic labels and not necessarily brain circuits underlying functional deficits. A more robust approach would be to use objective psychological constructs that act as “ingredients” of a given patient’s dysfunction. Cognitive control, the ability to rapidly shift one's attention and behavioral strategy in response to environmental changes is often compromised across mood and anxiety disorders. One of the well validated behavior paradigms for tapping into the cognitive control circuits is a cognitive interference task, where subjects must suppress a natural response to follow a less intuitive rule, slower response on these tasks indicate difficulty exerting control to overcome response conflict. Conflict evokes robust electrophysiologic signatures, such as theta (4-8 Hz) oscillations in the prefrontal cortex (PFC). Although the cingulate and PFC are well established as being involved during tasks requiring executive control, the underlying neural mechanisms of conflict evoked theta oscillations in the PFC are

not clear. The objective of this work is to use a neural mass model (NMM) to find feasible cortical networks generating theta oscillations during conflict processing in human subjects. We used intracranial EEG (iEEG) recorded from dorsolateral PFC (dlPFC) and temporal lobe (TL) of human subjects with intractable epilepsy undergoing invasive monitoring, while they performed a multi-source interference task (MSIT). MSIT requires subjects to identify which of three numbers (0-3) is different than its neighbors. In high conflict trials, the unique number is out of position and is flanked by distractors while in low conflict trials, it corresponds to the position and is flanked by zeros. We used a dynamic causal modeling (DCM) framework to simulate dlPFC-TL theta using a Jansen-Rit NMM. We found significant evidence for a TL input into the dlPFC during the initial 500 ms of conflict processing compared to a bidirectional connection between the dlPFC and TL. We conclude that a neural mass modeling framework can be used to elucidate candidate mechanisms of neural oscillations underlying conflict resolution in human subjects. This can be used to find feasible target mechanisms for designing therapy in patients with compromised cognitive control. This framework can also be expanded to serve as an in-silico test bed for designing and testing neuromodulatory interventions such as electrical stimulation for improving cognitive control in mood/anxiety disorders.

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Poster

P824. Prefrontal Cortex

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P824.02

Topic: H.04. Executive Functions

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NIMH Conte Center Grant P50MH094258
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Title: The geometry of domain-general performance monitoring in the human medial frontal cortex

Authors: Z. FU, 91125[1,4], D. BEAM[4], J. M. CHUNG[5], C. M. REED[5], A. N. MAMELAK[4], R. ADOLPHS[2], U. RUTISHAUSER[4,3];
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Abstract: Humans can flexibly control their behaviors to achieve various goals. This process can be conceptualized as a feedback control loop: the course and outcomes of goal-directed actions are monitored for deviations from intended goals, and behavioral and physiological adaptations are initiated to compensate for such deviations. An important open question is how such behavioral optimization can generalize across many different tasks while also support specialized functions of single tasks. We study the neural representation of performance monitoring by recording single neuron activity in the medial frontal cortex (MFC) of epilepsy patients. Subjects performed the Stroop task and the Multi-source Interference Task (MSIT). The Stroop task involves Stroop conflict, while the MSIT involves Simon and Flanker conflict. The data set contains over 1000 MFC neurons from 34 patients. We found that subjects' behavior is well described by a Bayesian conflict learning model, in which the conflict probability is estimated iteratively on a trial-by-trial basis. We found neurons encoding conflict retrospectively and neurons encoding predicted demand for control, both of which are signals that are predicted by our Bayesian model. Neurons encoding the predicted demand for control, which in our model is the prior distribution for the conflict probability, appeared to be biophysically distinct as they demonstrate long-range temporal correlation in the baseline spike counts and have thinner spikes compared to other groups of neurons. Neurons encoding error, conflict and predicted demand for control in one or both tasks coexisted in the same population. Collectively, these neurons gave rise to a representational geometry that simultaneously allowed generalization between different tasks while maintaining maximal separation of within-task conditions, such as separating the four different types of conflict conditions within MSIT. Neural representations of conflict were additive: when two conflicts occur on the same trial, its neural representation approximately equals the sum of the representations of the constituent conflicts. These findings reveal how the MFC representation of evaluative signals are both abstract and specific, suggesting a mechanism for computing and maintaining estimated demand for control across trials and tasks.

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Poster

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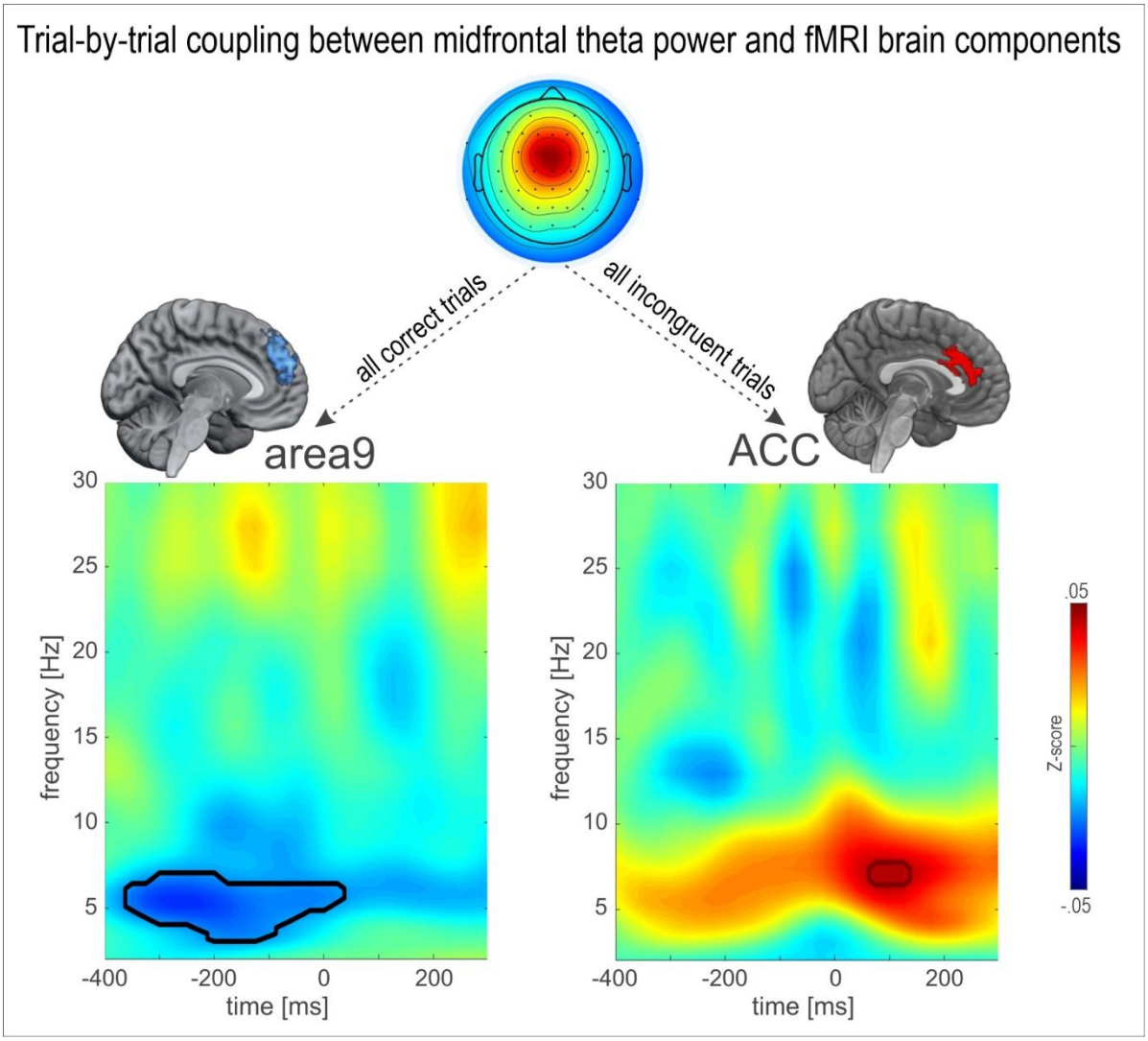
Topic: H.04. Executive Functions

Support: National Science Centre, Poland, 2016/21/D/HS6/02962

Title: Midfrontal theta increase in conflict task reflects deactivation in midline area 9

Authors: *E. BELDZIK, A. DOMAGALIK, T. MAREK;
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Abstract: Both conflict and error processing in the human brain have been linked to the midfrontal theta (4-8 Hz) increase as indicated by EEG studies and greater hemodynamic activity in the anterior cingulate cortex (ACC) as indicated by fMRI studies. Conveniently, the source of midfrontal cognitive theta was estimated in or nearby ACC. However, studies using concurrent EEG and fMRI recordings in resting-state found only a negative relationship of theta power and BOLD signal. This negative correlation was observed in brain regions typically showing task-related deactivations. In this study, we used a simultaneous EEG-fMRI technique to investigate a trial-by-trial coupling between theta power and hemodynamic activity during the performance of two conflict tasks. Independent component analysis (ICA) was applied to select individual midfrontal EEG components, whereas group ICA was applied to fMRI data to obtain a functional parcellation of the frontal cortex. Theta power was coupled with the peak of hemodynamic responses using a linear mixed-effect model. Our goal was to obtain a source of conflict- and error-related theta activity. The fMRI results revealed several brain regions showing greater activity for correct incongruent trials than congruent ones and far greater activity for erroneous trials, including pre-supplementary motor area, anterior insular cortex, and ACC. Despite the presence of these task-engaged regions, conflict theta showed only a negative correlation to BOLD signal in the midline area 9, a region exhibiting conflict-sensitive deactivation. Conversely, error-related theta showed a positive relationship to activity in the ACC. Our results provide evidence that pre-response midfrontal theta reflects cortical inhibition of area 9, and this process strongly affects the timing of the reactions and supports successful conflict resolution. In contrast, post-response theta reflects ACC activity, which signals the commitment of an error. Our findings highlight that conflict- and error-related theta oscillations represent fundamentally different processes.



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Poster

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Topic: H.04. Executive Functions

Support: Japan Epilepsy Research Foundation JERFTENKAN 19101
NIH Grant DC015260

Title: Human cingulate cortex during planning and production of natural speech

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Abstract: The role of human cingulate cortex during speech has been under investigated. Previous reports based on nonhuman primates' vocalization could give us insight about this region's role during human speech production. Some lesion studies and electrical stimulation studies in human suggest the involvement of anterior cingulate cortex in speech production in a causal manner, but the high-resolution dynamics and its relation to other canonical speech areas such as ventrolateral prefrontal cortex (vlPFC) or sensorimotor cortex (SMC) remains unclear. We here report the functional role of cingulate cortex during naturalistic question-and-answer-based dialogue task. The neural signal was recorded from depth electrode contacts (cingulate cortex) and subdural grid/strip electrode (lateral cortices) that were implanted in 11 subjects with intractable epilepsy. All the patients underwent intracranial electrode placement for the purpose of epileptic foci detection. All the subjects had above average cognitive function based on the pre-operative neuropsychological screening. The task was run in a shielded room in the epilepsy monitoring unit located at the University of Iowa Hospital. The neural activity was recorded by the Neuralynx Pegasus system, and the vocal response during the dialogue task was recorded from TASCAM recorder. Each utterance was timestamped using PRATT software. We analyzed high frequency (70-150Hz) activity power from cingulate cortex both during speech production phase (-200ms re subject's answer onset to answer offset) and speech planning phase (0ms re interviewer's question offset to -200ms re subject's answer onset). Only utterances that were correctly answered were used for production analysis, and the questions ending with critical information was used for planning analysis. The temporal dynamics of the high frequency response in cingulate cortex was compared with other canonical speech related cortices. We then compared the responses between two phases of the dialogue (planning/production) by using a production-planning index to see whether the response seen was phase specific in each cingulate subregion. Our results showed that anterior cingulate cortex (ACC) has a higher preference for production whereas posterior cingulate cortex (PCC) showed higher preference for planning. The peak patency of response in PCC preceded that of vLPFC and SMC. The findings show clear evidence of the involvement of cingulate cortex during speech planning and production.

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Poster

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The MIT Picower Institute Innovation Fund

Title: Layer-specific deactivation of prefrontal cortex as a causal test of predictive coding

Authors: *A. J. MAJOR[1], A. M. BASTOS[2], E. K. MILLER[1];
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Abstract: In conditions such as autism spectrum disorder and schizophrenia, abnormal neurodevelopment leads to altered laminar cortical architecture. The laminar organization of cortex has attracted growing interest and is critical to microcircuit function and interregional communication. For example, in a recent model of Predictive Coding, superficial cortical layers propagate sensory feedforward signals via gamma rhythms (> 35 Hz) to higher-order cortex and deep layers send feedback signals via alpha/beta rhythms (10-30 Hz) to sensory regions (Bastos et al., *PNAS*, 2020). Alpha/beta and gamma rhythms are anti-correlated. Feedback alpha/beta signals from prefrontal cortex (PFC) are thought to suppress feedforward sensory information in sensory cortex. This project is a causal test of this layer-specific model of Predictive Coding, known as *Predictive Routing*.
Using custom laminar probes, we have developed an experimental paradigm to selectivity suppress either superficial or deep PFC cortical layers using $GABA_A$ receptor agonist muscimol while simultaneously recording from all layers of multiple rhesus cortical areas. Muscimol deactivation of deep-layer PFC is hypothesized to suppress feedback alpha/beta signals and thus remove the suppressive effect of PFC onto sensory regions such as visual area V4. In support of the model, preliminary data show deactivation of deep-layer PFC resulted in heightened V4 gamma and spiking activity. This work will provide a causal test to a prevalent model of laminar cortical processing and yield specific insights into conditions related to sensory filtering, such as autism spectrum disorder and schizophrenia.

Disclosures: A.J. Major: None. **A.M. Bastos:** None. **E.K. Miller:** None.

Poster

P824. Prefrontal Cortex

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Topic: H.04. Executive Functions

Support: MRC
Wellcome Trust
BBSRC

Title: Neural dynamics of abstract rule encoding in macaque prefrontal areas: Recording simultaneously from multiple chronically implanted multi-electrode arrays

Authors: *J. M. GALEAZZI, M. AINSWORTH, C. PEDREIRA, M. J. BUCKLEY;
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Abstract: Previous studies identified different prefrontal areas contributing in different ways to rule-guided behavior in the context of a Wisconsin Card Sorting Test (WCST) analog for macaque monkey; dorsolateral prefrontal cortex (dlPFC) is necessary to maintain a specific abstract rule in working memory, ventrolateral prefrontal cortex (vlPFC) supports rule maintenance in general, and orbitofrontal frontal cortex (OFC) is needed to rapidly update representations of rule value based on reward. (Buckley et al., 2009; Mansouri et al., 2009). Furthermore, recent studies on role of the frontopolar cortex (FPC) suggest it contributes to representing the relative value of unchosen alternatives, including but not limited to rules (Mansouri et al., 2017). As neuronal activity from these different areas was not previously recorded simultaneously we do not yet understand the underlying computations and functional interactions operating within and between these regions. We present here the first study that simultaneously applies multi-area multi-electrode recording techniques to four different frontal regions implicated in rule-guided behaviour. Multi-electrode micro-arrays ('Utah arrays') were chronically implanted in dlPFC, vlPFC, OFC and FPC of two rhesus macaques, allowing us to simultaneously record single and multiunit activity and local field potential (LFP) from all four regions together while the animals performed a WCST analog. Here we present preliminary results from these recordings showing the neural activities underlying task performance. We obtained a typical daily yield of 100-200 separable units/clusters from all arrays combined, with single and multiunit activity responsive to different task-relevant components. For example, we found a variety of pre-reward and post-reward unit responses in dlPFC, vlPFC, OFC, and FPC with different onsets and durations. We present here the extent to which different aspects of the task can be accurately decoded from the neural activity recorded on each of the arrays employing a variety of commonly used machine learning classification algorithms. Lastly, we also show the differences of time evolving neural trajectories of population activity in a low dimensional state space.

Disclosures: J.M. Galeazzi: None. **M. Ainsworth:** None. **C. Pedreira:** None. **M.J. Buckley:** None.

Poster

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Program #/Poster #: P824.07

Topic: H.04. Executive Functions

Support: HHMI
NIHP50MH112491

Title: Reward-based decision making in freely-moving Marmosets

Authors: *E. SCHOENBECK[1], K. J. MASTRO[2,1], L. STANWICKS[1], B. A. STEVENS[2,1]; [1]The Broad Inst., Cambridge, MA; [2]Harvard Med. Sch. Neurobio., Boston Children's Hosp., Boston, MA

Abstract: The ability to flexibly adapt one's behavior in an ever-changing environment is necessary for survival. To maximize positive and/or reduce negative outcomes, we must balance the need to explore for new information and exploit known action-outcome relationships. From mice to humans to computers, reinforcement learning has become a fundamental area of study and provides a dynamic landscape to probe the decision making process. Moreover, to capitalize on the emergence of genetic and viral tools, the Common Marmoset has received renewed interest across neuroscience for its potential to link findings between primate and non-primates. In this study, we established a touchscreen apparatus that can be permanently affixed to the marmoset home-cage and allows for the assessment of reward-based decision making. We have trained a cohort of marmosets on simple (Match to Sample) to more complex (Multi-armed bandit) cognitive tasks. By incorporating the touchscreen apparatus into the home-cage, we found that marmosets will perform upwards to 500-800 trials per training session (3-4 hours). We demonstrate that marmosets can successfully learn a rule, increase their rate of learning with experience, and generalize their knowledge of the task to novel sets of stimuli. In the next series of experiments, marmosets will be presented with a probabilistic version of the multi-armed bandit task where the probability of being rewarded will shift within a session. Together, this data will provide important insights into the marmoset behavioral strategies employed in reward-based decision making that will set the stage to compare across conditions and species.

Disclosures: E. Schoenbeck: None. **K.J. Mastro:** None. **L. Stanwicks:** None. **B.A. Stevens:** None.

Poster

P834. Hippocampal Interactions III

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Program #/Poster #: P834.01

Topic: H.08. Learning and Memory

Support: NIH: National Institute of Diabetes and Digestive and Kidney Diseases R01DK114700 to MBP

Title: Distinct mnemonic functions of intermediate hippocampus in male rats

Authors: *K. E. WHITLEY, S. B. BRIGGS, M. B. PARENT;
Neurosci. Inst., Georgia State Univ., Atlanta, GA

Abstract: In rodents, the hippocampus is functionally divided along its longitudinal axis into dorsal and ventral poles. Generally, dorsal hippocampal (dHC) neurons are necessary for episodic and spatial memory, whereas ventral hippocampal (vHC) neurons are essential for affective and motivational processes and emotional memory. dHC and vHC have different anatomical connections, cellular and circuit properties, and patterns of gene expression that likely contribute to the different functions that they serve. Compared to dHC and vHC, relatively little is known regarding the function of the intermediate region of the hippocampus (iHC). Gene expression and anatomical connections in iHC do not overlap completely with those of dHC and vHC, suggesting that iHC may have functions that differ from dHC and vHC. In support, other researchers have shown that permanent lesions of iHC, but not dHC or vHC, impair rapid place learning in the water maze. The goal of the current project was to determine whether reversibly inactivating iHC would impair memory that is known to be dependent on dHC (spontaneous alternation; SA), vHC (temporal ordering of odors; TO-O), or both (inhibitory avoidance [IA], spatial and temporal episodic memory task; EMT). Male Sprague-Dawley rats were implanted with bilateral cannulae aimed at iHC. After at least 1 week of recovery, one cohort (n = 8-10/group) was given infusions of muscimol (0.05 ug in 0.5 ul) 15 min prior to SA testing, EMT testing, and IA training (at least 48 hrs between tests; infusions counterbalanced across behavioral tests). A second cohort (n = 8-10/group) was given these infusions prior to TO-O training and rapid place water maze training. Our preliminary data indicate that, like chronic lesions, reversible inactivation of iHC impairs rapid place learning. More importantly, the same manipulation does not appear to affect memory in dHC and vHC-dependent memory tasks. These findings support the possibility that iHC plays a distinct role in memory that is different from dHC and vHC.

Disclosures: K.E. Whitley: None. **S.B. Briggs:** None. **M.B. Parent:** None.

Poster

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Support: NIH BRAIN INITIATIVE 1R01NS109994

NIH 1R01NS109362-01
McKnight Scholar Award in Neuroscience
Klingenstein-Simons Fellowship Award in Neuroscience
Alfred P.Sloan Research Fellowship
Whitehall Research Grant
American Epilepsy Society Junior Investigator Award

Title: Local integration of long-range inputs from the lateral entorhinal cortex in hippocampal area CA3 drives dendritic non-linearities and somatic output

Authors: *V. ROBERT, R. DE LA TORRE, K. O'NEIL, J. BASU;
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Abstract: Functional interactions between the entorhinal cortex and the hippocampus are crucial for the formation and recall of episodic memories. Multimodal sensory inputs, specifically spatial from the medial entorhinal cortex (MEC) and contextual from the lateral entorhinal cortex (LEC), are projected to and processed by several parallel circuits within the hippocampus. While cortico-hippocampal interactions at the level of dentate gyrus (DG) and area CA1 are well studied, the functional connectivity and circuit dynamics between LEC and area CA3 remain unknown. By integrating direct long-range LEC inputs with local feedforward DG and feedback CA3 inputs, area CA3 is poised to gate hippocampal information flow and mnemonic function. Non-linear dendritic integration of LEC inputs in CA3 pyramidal cells could underlie the flexibility of spatial representations displayed by CA3 ensembles. Here we use dual color optogenetic circuit mapping in acute slices and behavioral manipulations in vivo to dissect the functional role of excitatory and inhibitory inputs from LEC to area CA3 and test the circuit level interactions that shape CA3 output. We found that CA3 interneurons located in the apical dendritic layers received monosynaptic connections from both LEC glutamatergic and GABAergic projections. LEC excitatory inputs drove spiking in these interneurons, whereas LEC inhibitory inputs shunted interneuron excitability. In CA3 pyramidal cells, LEC excitatory inputs evoked monosynaptic excitation, as well as disynaptic inhibition which acted to prevent excessive excitation and CA3 output. Next, we combined optical stimulation of LEC excitatory and inhibitory projections with electrical stimulation of DG mossy fibers and CA3 recurrent collaterals to assess the dendritic and somatic integration of these inputs and the resulting drive of CA3 output. Although single input stimulation did not elicit CA3 pyramidal cell spiking, LEC excitatory projections could drive CA3 output when paired with local inputs, and additional stimulation of LEC inhibitory projections further boosted CA3 output. Dendritic recordings from CA3 pyramidal cells suggest that this disinhibitory effect may facilitate the generation of dendritic spikes, which ultimately impact somatic excitability. These results demonstrate that the LEC input can drive CA3 output when coordinated with local input activity, highlighting the role of dendritic integration of LEC signals in CA3 dendrites. To assess the role of this circuit in hippocampal learning and memory functions, we are currently manipulating LEC excitatory and inhibitory inputs in vivo during hippocampal-dependent behavioral tasks.

Disclosures: V. Robert: None. **R. De La Torre:** None. **K. O'Neil:** None. **J. Basu:** None.

Poster

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Topic: H.08. Learning and Memory

Support: NIH BRAIN Initiative 1R01NS109994

Title: Automatic detection, segmentation, and de-mixing of dendritic regions of interest in 2-photon calcium data

Authors: *J. J. MOORE[1,2], D. CHKLOVSKII[2], J. BASU[1];
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Abstract: Neurons maintain large dendritic arbors which support non-linear input integration that shapes neural dynamics and can greatly boost the computational power of individual neurons. Population 2-photon calcium imaging typically focuses on the activity of neural somata, but several brain regions, such as hippocampal area CA3, superficial neocortex, and cerebellum, afford imaging of multiple dendrites at once. To analyze these data, dendritic regions of interest (ROIs) are currently manually identified, a laborious and error-prone process that takes hours of skilled work. Thus, there is a widespread need for tools to automatically detect and segment dendritic ROIs and link them to their parent soma.

Dendritic segmentation poses a number of unique challenges not faced by somatic segmentation. Variable morphology of dendrites within and across different brain regions severely restricts possible geometric assumptions. While a soma may be treated as one compartment, extended dendrites may have propagating spikes or otherwise asynchronous activity across an entire branch. Additionally, densely labeled preparations feature overlapping dendrites, necessitating robust de-mixing techniques to separate independent signals.

Sparse constrained non-negative matrix factorization (sCNMF) has been used to identify and de-mix dendrites in dense fields of view, but the results require random initialization and can be difficult to map to individual neurons. We modified the sCNMF initialization procedure to cluster regions of contiguous simultaneously active pixels. We obtained ROIs that are deterministic and traceable to the parent soma, with efficient time and memory requirements.

To demonstrate generality, we have applied our algorithm to several fields of view in mouse hippocampus in CA3, CA2, and CA1, using GCaMP6s, 6f, and 7b, with a wide range of expression densities, dendritic morphologies, and signal-to-noise ratios. The resulting ROIs are comparable to those manually drawn by a human expert and are robust to moderate levels of noise and overlap. Our approach has few tunable free parameters, which are easily interpretable and generally transferable between imaging preparations.

We present our dendritic detection algorithm as an extension of the CaImAn imaging suite, enabling easy and robust processing of dendritic data in an accessible, open-source manner. We are optimistic that this will encourage more studies of dendritic function from a diverse array of investigators, accelerating the field's understanding of the importance and utility of dendritic activity.

Disclosures: J.J. Moore: None. **D. Chklovskii:** None. **J. Basu:** None.

Poster

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Whitehall Three Year Research Grant
Sloan Research Fellowship

Title: Lateral entorhinal cortex: a cortical input that drives strong dendritic excitation, inhibition, and disinhibition onto CA1 pyramidal neurons

Authors: *O. BILASH, J. BASU;
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Abstract: The functional interaction between the entorhinal cortex (EC) and hippocampus (HC) is critical for episodic memory formation. Direct projections from the medial and lateral entorhinal cortex (MEC and LEC) provide the hippocampus with spatial and non-spatial (contextual) sensory information, respectively. Previous studies have largely focused on MEC-HC interactions, and only recently have behavioral studies started to demonstrate that LEC is involved in olfactory learning, novelty- and object-related learning, and contextual learning. Still, the circuit-level functional interactions between LEC and the hippocampus remains underexplored. Our study examines how distal dendritic input from LEC modulates hippocampal information flow through non-linear dendritic computations and excitation-inhibition balance. Using slice electrophysiology and optogenetics, we demonstrated that glutamatergic LEC projections drive excitation onto CA1 PNs, while simultaneously recruiting strong feed-forward inhibition. LEC inputs elicited subthreshold post-synaptic potentials in CA1 PNs, but were not strong enough to elicit action potentials. Interestingly, using dendritic recordings, we found that

optogenetic stimulation of LEC axons in CA1 can drive dendritic spikes in CA1 PN distal dendrites. To better understand the mechanism underlying the LEC-driven dendritic spikes, we probed the GABAergic microcircuit elements recruited by LEC inputs. Using targeted recordings from GABAergic neuron-specific transgenic mice, we found that LEC inputs recruit specific inhibitory neurons (INs) in area CA1, including VIP+ INs and CCK+ INs. Given the disinhibitory nature of VIP+ INs described in the hippocampus and neocortex literature, we proposed that LEC inputs gate dendritic spikes in CA1 PNs by engaging this local disinhibitory microcircuit. Optogenetic silencing of VIP+ INs in CA1 revealed a subtle but consistent effect on LEC-driven dendritic activity. Currently, we are developing a computational model to elucidate the details of the disinhibitory gating mechanism. Given that *in vitro* and *in vivo* studies have previously demonstrated the role of dendritic spikes in plasticity, our findings suggest that LEC plays an important role in promoting supralinear dendritic computations, thus enabling it to influence long-term representation in the hippocampus. In summary, our study provides a comprehensive overview of how a distal dendritic input from the cortex can differentially activate various local circuit elements, including pyramidal neuron somata, dendrites, and various GABAergic interneurons.

Disclosures: O. Bilash: None. **J. Basu:** None.

Poster

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Topic: H.08. Learning and Memory

Support: NWO TOP Grant 612.001.853

Title: Emergence of sensory and memory representations in the hippocampus and prefrontal cortex of mice

Authors: *B. SOUZA[1], J. KLEE[1], L. MAZZUCATO[2], F. BATTAGLIA[1];
[1]Donders Inst. for Brain, Cognition and Behaviour, Radboud Univ., Nijmegen, Netherlands;
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Abstract: The hippocampus and the medial prefrontal cortex (mPFC) are both known to modulate their activity in response to different stimuli and rewards in a variety of paradigms. Because those two areas are intimately connected to episodic and working memory, it has been hypothesized that they encode relevant features of the environment by developing neural representations that can be accessed in the absence of the stimulus. Despite some evidence endorsing this hypothesis, the nature of those representations and whether representations in the two areas interact remain unknown. In trace conditioning, animals learn to associate a

conditioned stimulus (CS) and a reward separated by a delay (trace period). Although the neural activity in both areas can be linked to memory representation formation, it is unknown how their coordinated neural activity may maintain the stimulus-action map evoked during the CS until the behavior is expressed. In this work, we address this issue using auditory trace conditioning in head-fixed mice to record simultaneous activity from hippocampal area CA1 and mPFC. First, we found that both CA1 and mPFC encoded time within the trial via the emergence of neuronal sequences at the beginning of stimulus presentation. Interestingly, this time-structured pattern appeared in PFC since early learning sessions, while it emerged in CA1 only after learning. Moreover, the temporal patterns associated with the stimulus onset in PFC were also evoked at reward onset. This echo of the stimulus representation during reward disappeared after learning, suggesting a disentangle of the representation of stimulus and reward. We also found that the representation of the stimulus identity was strong and stable in mPFC, emerging with the stimulus onset, while it was weaker in CA1, mildly increasing during the trace period. To investigate whether the weaker representation in CA1 could be due to a more dynamical activation of its representations, we used Hidden-Markov models to estimate the network states. Interestingly, we could still find states in CA1 encoding trial-type identity. Finally, we found that awake sharp-wave ripples deferentially shaped the reactivation of CA1 and PFC assemblies - increasing reactivations within and across sessions, respectively. Also, CA1 assemblies that encoded stimulus identity during trace were more reactivated during ripples. Together, those results clarify the dynamics of stimulus encoding in the mPFC-hippocampal circuit, suggesting the hippocampus is important to sustain the CS representation during the trace period (i.e., in the absence of the stimulus), while mPFC might be needed for creating the stimulus-reward association.

Disclosures: B. Souza: None. **J. Klee:** None. **L. Mazzucato:** None. **F. Battaglia:** None.

Poster

P834. Hippocampal Interactions III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P834.06

Topic: H.08. Learning and Memory

Support: Samsung Science and Technology Foundation SSTF-BA1602-11
HHMI Wellcome Trust 208468/Z/17/Z

Title: Real-time visualization of mRNA synthesis during memory formation in live animals

Authors: *B. LEE[1], J. SHIM[1], H. C. MOON[1], D. KIM[1], J. KIM[2], J. YOOK[2], J. KIM[2], H. PARK[1];
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Abstract: Memories are thought to be encoded in populations of neurons called memory trace or engram cells. However, little is known about the dynamics of these cells because of the difficulty in observing them repeatedly over long periods of time in vivo. To overcome this limitation, we present a genetically-encoded RNA indicator (GERI) for intravital chronic imaging of endogenous Arc mRNA—a popular marker for memory trace cells. We used our GERI to identify Arc-positive neurons in real time without the delay associated with reporter protein expression in conventional approaches. We found that Arc-positive neuronal populations rapidly turned over within two days in CA1, whereas ~4% of neurons in the retrosplenial cortex (RSC) consistently expressed Arc following contextual fear conditioning and repeated memory retrievals. Dual imaging of GERI and calcium indicator in CA1 of mice navigating a virtual reality environment revealed that only the overlapping population of neurons expressing Arc during encoding and retrieval exhibited relatively high calcium activity in a context-specific manner. This in vivo RNA imaging approach opens the possibility to unravel the dynamics of the neuronal population underlying various learning and memory processes.

Disclosures: B. Lee: None. **J. Shim:** None. **H.C. Moon:** None. **D. Kim:** None. **J. Kim:** None. **J. Yook:** None. **J. Kim:** None. **H. Park:** None.

Poster

P834. Hippocampal Interactions III

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Program #/Poster #: P834.07

Topic: H.08. Learning and Memory

Support: NSF T32 1829071

Title: Genesis of multi-modal representations in the entorhinal cortex during working memory

Authors: *C. DORIAN[1], J. TAXIDIS[2], S. CHEUNG[2], K. SAFARYAN[3], P. GOLSHANI[4]; [1]Univ. of California, Los Angeles Interdepartmental Ph.D. Program In Neurosci., Los Angeles, CA; [3]Departments of Physics and Astronomy, Neurology, Neurobio., [2]UCLA, Los Angeles, CA; [4]UCLA Dept. of Neurol., Los Angeles, CA

Abstract: The medial and lateral entorhinal cortices (MEC and LEC) play an important role in coding spatial features of the environment, and their projections to hippocampus are critical for driving hippocampal representations. These structures are also important for maintenance of working memory representations when working memory load increases. Yet, how the activity patterns of these entorhinal projections to hippocampus change during the learning and stable performance of non-spatial working memory tasks is still not understood. Do these projections carry distinct sensory and temporal information? To answer these questions, we have optimized methods for performing calcium imaging of LEC temporoammonic axons expressing GCaMP7s

and terminating in the stratum lacunosum moleculare layer of the dorsal hippocampal CA1 region during the performance of a non-match to sample olfactory working memory task. We find that the large majority of axons showing significant firing fields were active during the presentation of a specific odor. Rare axons had odor-specific firing during the delay period. We will compare these response properties to those from MEC to determine whether the two structures play distinct roles in coding for sensory and temporal information and how their activity patterns change with learning. We will also assay the effects of LEC and MEC inhibition on learning and stable performance. These experiments will yield insights on the distinct and complementary roles of MEC and LEC in learning and maintenance of working memory related sensory and temporal information.

Disclosures: C. Dorian: None. **J. Taxidis:** None. **S. Cheung:** None. **K. Safaryan:** None. **P. Golshani:** None.

Poster

P850. Consolidation and Reconsolidation: Behavior

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Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P850.01

Topic: H.07. Long-Term Memory

Support: NSERC RGPIN-2018-04901

Title: "Looking past the foggy ruins of time": Hippocampal and Non-Hippocampal contributions to Long-Term Memory

Authors: *K. ROBERTS[1], D. MCCALLUM[1], N. M. FOURNIER[2], R. J. SUTHERLAND[4], H. LEHMANN[3];
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Abstract: Scoville and Milner (1957) were the first to clearly state that episodic memories can transition from a hippocampal-dependent to a hippocampal-independent state, a process now termed long-term systems consolidation. Their key observation supporting this process was the temporally graded retrograde amnesia in patient H.M., meaning his loss of memories for events that occurred in the 2-4 years prior to his hippocampal resection and not those that preceded that period. However, measurements of H.M.'s retrograde amnesia changed over the following years with his memory loss progressively encompassing longer periods. By the early 2000s, a little prior to his death, H.M.'s retrograde amnesia presented with a flat gradient with perhaps only one spared episodic memory. A few hypotheses have been put forth to account for this accelerated memory decay in H.M., including that the non-hippocampal systems alone are insufficient to maintain consolidated long-term memories. The current study examined this

possibility in rats by assessing decay of a context fear memory that had become independent of the hippocampus over distributed learning. In Experiment 1, we replicate the common finding that hippocampal lesions cause retrograde amnesia for a context fear memory acquired within a single conditioning session. In Experiment 2, we show that the same conditioning experience distributed over five days mitigates the amnesic effects of the lesions and creates a hippocampal-independent memory. Retention tests in these rats, however, continued every five weeks, and by the fourth and fifth test, the lesion rats were showing memory impairments. In Experiment 3, we again examined decay of the hippocampal-independent memory but reduced the number of tests to limit the influence of extinction. During the first retention test, the lesion rats again showed intact memory, but they were impaired when tested a second time 20 weeks later. These findings could be explained by dual stores (hippocampus and non-hippocampus) normally synergistically contributing to the recall or by continued interactions between systems. Experiment 4 examined this by damaging the hippocampus 18 weeks after the distributed learning - matching a period sufficient to cause decay in Experiment 2. No memory deficits were observed following the lesions, suggesting the context fear established in the non-hippocampal system did not show any evidence of decay and ruling out a dual-store account. Combined, our findings point to an explanation for the changes in retrograde amnesia reported in H.M. across studies and provides new evidence of persistent interaction between the hippocampus and other memory systems.

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Poster

P850. Consolidation and Reconsolidation: Behavior

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Program #/Poster #: P850.02

Topic: H.07. Long-Term Memory

Support: Israel Science Foundation (ISF 526/17)
United States - Israel Binational Science Foundation (BSF 2016058)

Title: Reconsolidation of procedural learning does not operate at a rapid micro-timescale level

Authors: *J. HERSZAGE[1], M. BÖNSTRUP[2], L. G. COHEN[3], N. CENSOR[1];
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Abstract: Memory consolidation typically refers to stabilization of the encoded memory or offline gains in skill performance, a process which commonly requires hours or days from completion of training. However, recent evidence has shown that consolidation occurs also in

early stages of skill learning, at a micro-timescale (Bönstrup et al., 2019). Thus, while consolidated memories usually show offline learning gains between sessions, offline learning gains in micro-consolidated memories were evident between trials in early stages of a single session. In light of abundant evidence showing that consolidated memories are susceptible to modifications following their reactivation through reconsolidation (Nader and Hardt, 2009), here we asked whether micro-consolidated memories are also susceptible to modifications through rapid 'micro-reconsolidation', thus even at initial stages of learning. In a set of experiments, we collected crowdsourced motor sequence learning (Karni et al., 1995) data from 459 participants recruited from the Amazon Mechanical Turk platform (MTurk), to test whether post-reactivation (1) interference and (2) enhancement occur through rapid micro-reconsolidation. Since memories were shown to be susceptible to interference following reactivation (Walker et al., 2003; de Beukelaar et al., 2014), *Experiment 1* tested whether micro-consolidated memories are more susceptible to interference within a micro-reconsolidation time window. Participants underwent rapid micro-consolidation (Bönstrup et al., 2019) of the motor sequence task. Then, an interfering sequence was presented interleaved with brief reactivations of the original sequence. Retest was compared to a control group without reactivations. Since reactivations were recently shown to induce learning of motor skills (Herszage et al., 2021), *Experiment 2* tested if micro-consolidated memories can benefit from micro-reconsolidation to produce enhanced learning gains. Following micro-consolidation, the memory was reactivated repeatedly to induce learning. Retest was compared to a control group without reactivations. The results of both experiments indicate that micro-consolidated memories are not susceptible to interference nor to enhancement within a rapid micro-reconsolidation time window, relative to controls. This set of evidence suggests that memory reconsolidation might be dependent on consolidation processing occurring at the macro-scale, requiring hours or days to occur.

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Poster

P850. Consolidation and Reconsolidation: Behavior

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Topic: H.07. Long-Term Memory

Support: The Human Frontiers Science Program Organization (HFSP) Grant RGY0063/2017 to RH
The Netherlands Organisation for Scientific Research Grant NWA.1228.191.208 to RH and SR

Title: Restoring persistent accessibility to memories after sleep deprivation-induced amnesia

Authors: *Y. G. BOLSIUS[1], P. R. A. HECKMAN[1,2], F. RAVEN[1,3], E. L. MEIJER[1], M. J. H. KAS[1], S. RAMIREZ[4], P. MEERLO[1], R. HAVEKES[1];
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Abstract: Sleep deprivation is a common problem in our modern 24/7 society. Loss of sleep negatively impacts brain function and in particular cognitive processes that require the hippocampus. In fact, a brief period of sleep deprivation immediately after a hippocampal learning trial leads to memory deficits. It is unclear, however, whether sleep deprivation leads to an absolute loss of information or, alternatively, affects the retrievability of this information stored under suboptimal sleep deprivation conditions. In the current study, we addressed this question using memory engram labeling techniques in combination with optogenetics. Male c-fos tTA mice were injected with a TRE-ChR2-mCherry virus in the dentate gyrus, allowing us to specifically tag and reactivate hippocampal memory engrams. To assess the impact of sleep deprivation on spatial memory processes, we used the object-location memory task with 6 hours of sleep deprivation immediately after the training trial. In line with previous work, we found that sleep deprivation hampered the detection of spatial novelty (*i.e.* the relocated object). However, optogenetic activation of the memory engram preceding the test session resulted in a successful detection of the spatial novelty. This observation suggests that sleep deprivation does not lead to the loss of information but, rather impacts the retrievability of memories formed under sleep deprivation conditions. As a next step, we investigated whether these deficits in memory retrievability could be reversed by treatment with the clinically-approved phosphodiesterase 4 (PDE4) inhibitor roflumilast, which increases the cAMP levels. Systemic treatment with roflumilast preceding the test session rescued the sleep deprivation-induced memory retrievability impairment. Lastly, we combined optogenetic engram stimulation with PDE4 inhibitor treatment to test whether we could make object-location memories stored under sleep deprivation conditions accessible for natural retrieval again. When engram activation and roflumilast treatment were combined three days following training and subsequent sleep deprivation, it resulted in a more persistent memory trace that allowed for natural retrieval several days later. Our studies demonstrate that sleep deprivation does not necessarily cause memory loss, but instead leads to the suboptimal storage of information that is difficult to retrieve. We also provide proof of principle that these suboptimally stored memories can be made accessible again far beyond the learning episode and that the clinically-approved PDE4 inhibitor roflumilast may be used to successfully retrieve information thought to be lost.

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Poster

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Program #/Poster #: P850.04

Topic: H.07. Long-Term Memory

Support: VIEP-BUAP grant to CA in Neuroendocrinología BUAP-CA-288.
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Title: Innate anxiety influences learning strategies in Barnes maze.

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Abstract: We selected two sublines from Sprague-Dawley (SD) rats one with a high-spontaneous yawning (HY) with a mean of 20 yawns/h, and the other with a low-yawning (LY) frequency with just 2 yawns/h. LY rats are more stress-sensitive than HY when tested in the open-field arena because they ambulated less and had more fecal boli. Additionally, in the Barnes maze LY showed an impairment in their long-term memory with respect to HY and SD male rats. It is well stablished that the intensity of stressor can enhance or impair memory consolidation and influence the strategy used by subjects to solve spatial tasks. The aim of this study was to determine if LY innate anxiety and HY resilience influences learning strategies. We use SD, LY and HY male rats (8 rats/group), they were maintained under standard animal room conditions. All experiments were done between 1000 to 1300 in the Barnes maze, training consisted in six trials per day during two consecutive days, on day one with extra-maze cues subjects (Ss) learned a fixed position of the escape box (spatial strategy), on day two a black and white striped cue was attached to goal box, their position varied in each trial and extra-maze cues were removed (stimulus-response strategy). Ss learned both strategies in random order. Short-term memory (STM) was evaluated on day three, two goal boxes were present one of them with the striped cue attached, extra maze cues were present, three consecutive trials were given to the Ss and the box preference was used as criteria to evaluate the strategy. Long-term memory (LTM) was evaluated seven days later following the same procedures as in STM. The escape latency and number of errors were measured too. We obtained that in stimulus-response training LY males take more time to find the escape box with respect to HY and SD ($P<0.05$). In STM HY and SD rats used a stimulus-response strategy with respect to LY, which preferred spatial strategy ($P<0.001$). In LTM LY used more frequent stimulus-response strategy with respect to the spatial one ($P<0.01$). We conclude that anxiety promotes spatial strategy use in LY and performance of learning strategies in Barnes maze can change along time.

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Poster

P850. Consolidation and Reconsolidation: Behavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P850.05

Topic: H.07. Long-Term Memory

Title: Examining reward-enhancement of memory consolidation from earning points competitively

Authors: *E. GRANDOIT, P. J. REBER;
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Abstract: Associating reward with objects after study has been previously shown to improve human memory consolidation. Here we aimed to extend this finding to recognition memory for object locations after a reward task based on competitively earning points on a time-estimation challenge unrelated to object locations. Participants (n=45) learned a series of object-location associations for stimuli placed around a 2D ring (5 lists of 8 item-location pairs, 80 total stimuli presented twice each). After study, participants earned points-based rewards through a 3-second time estimation task. For half the object-location items, the reward was high value (750-2000 points) and for the other half, reward was low value (50 points). A competitive leaderboard, avatars, and storytelling were also introduced to increase the salience of reward. We hypothesized that the high reward value would enhance consolidation of memory for those object-location items on a delayed memory test. Memory for the object-location and item-recognition (with confidence judgments) of studied items were assessed both at the end of the learning and reward session and after a 24-hour delay. For the object-location tests, immediate memory for the high and low reward conditions was similar (high reward, M=68.4%, SE = 3.0%; low, M= 69.6%, SE = 2.6%), as were scores on the immediate memory item-recognition tests (high confidence hits-false alarms: high, M=35.6%, SE=4.9%; low, M=37.1%, SE=4.4%). For the delayed tests, object-location memory was slightly higher for the more rewarded condition (high, 62.2%, SE=3.0%; low, 59.1%, SE=3.2%), and item-recognition memory was also slightly higher for the more rewarded condition (high confidence hits-false alarms: high, M=21.9%, SE=3.8%; low, M=18.1%, SE=3.3%). However, the advantage for the high reward condition was not statistically reliable for either test. The effect of memory consolidation enhancement by the experience of high reward after study was observed to be of a very modest size here compared to prior reports. One possibility is that a points-based reward task may be less salient than a more robust incentive (e.g., monetary) and reduce the effect on memory consolidation. Another possible explanation for the reduced memory-enhancing effect observed here could be in the methodological details such as the inter-item spacing of the reward trials following memory study. Understanding the key methodological criteria that influence how and when the experience of reward enhances memory consolidation will eventually allow for this idea to be integrated into effective learning and educational interventions.

Disclosures: E. Grandoit: None. **P.J. Reber:** None.

Poster

P850. Consolidation and Reconsolidation: Behavior

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Program #/Poster #: P850.06

Topic: H.07. Long-Term Memory

Title: Interactions among different types of saliences explain the change in habitual gaze behavior after long-term retention time

Authors: *Y. RA, S. HWANG, S. PAENG, H. F. KIM;
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Abstract: Remembering values for decision-making is critical for survival. However, memory is often forgotten, affecting animals' behaviors. Although primates including humans and monkeys are known to show habitual gaze toward high-valued objects, it remains unclear how value memory changes over periods of time, and how this memory change alters habitual behavior. To address these questions, we trained human subjects to learn object values for 5 days, and their value memory was examined with habitual gaze task more than a month after the last learning. We found that subjects showed habitual gaze bias toward learned good objects 1 day after learning, as previously described (Kang et al., 2021). Interestingly, this gaze preference changed into the preference for both good and bad objects 30 days of retention time after the last learning. A computational model with reward value-driven salience is difficult to address this habitual gaze preference 30 days after learning. We therefore hypothesized that the brain processes both positive and negative values-driven salience to guide the preference for both good and bad objects after 30 days of retention (Madan et al., 2012). Indeed, we found that the selective change of habitual gaze was best explained by a three-layer model, which includes perceptual salience (PS), reward value-driven salience (RS), and both positive and negative value-driven salience (BS). We additionally found that the contribution of BS increases while that of RS decreases after long-term retention period. To further investigate a possible advantage of the habitual gaze driven by BS, we designed a simulation of the Bayesian learning under forgetting the value memory. Our simulation results suggest that BS-driven gazes facilitate re-learning the forgotten values of objects compared to the other gaze strategies (i.e. RS-driven gazes). Our data provide an insight into how the interaction among different components of saliences contribute to habitual gaze behaviors across time and its possible advantages for re-learning. This work was supported by NRF-2020M3E5D907990, NRF2019R1A2C2005213 , and Creative-Pioneering Researchers Program through SNU.

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Poster

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Program #/Poster #: P850.07

Topic: H.07. Long-Term Memory

Support: NSERC Discovery Grant to MK
NSERC Discovery Grant to KPO
NSERC CGS-D to IRB

Title: Peripheral lipopolysaccharide impairs the acquisition and consolidation processes of lithium chloride-induced anticipatory nausea in adult male rats.

Authors: *I. BISHNOI, M. KAVALIERS, K.-P. OSSENKOPP;
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Abstract: Introduction. Lipopolysaccharide (LPS), a cell wall component of Gram-negative bacteria, impairs learning and memory in a variety of behavioural tasks including, but not limited to, fear conditioning in rats. However, these studies use tasks that cannot readily discriminate between LPS-induced locomotor deficits, a sickness behaviour associated with LPS, and its effects on learning and memory. In order to more reliably study LPS' effects on learning and memory, the current study used the LiCl-induced anticipatory nausea paradigm (ANP). In the LiCl-induced ANP, the unconditioned stimulus (LiCl) becomes associated with a context, leading to reflexive disgust behaviours (e.g., gaping) surfacing when exposed to the context alone (conditioned stimulus). As such, this task is not influenced by LPS-induced locomotor deficits. In the current study, peripheral LPS effects on the resulting memory acquisition, consolidation processes and extinction were studied in the LiCl-induced ANP. **Methods.** *Acquisition.* 32 adult male Long Evans rats were divided into 4 groups (NaCl-NaCl, LPS-NaCl, LPS-LiCl, NaCl-LiCl). On conditioning days 1, 4, 7 and 10, rats were intraperitoneally (ip) injected with LPS (200 μg/kg) or NaCl 90 mins before LiCl (ip, 127 mg/kg) or NaCl and immediately placed into a distinct context for a 30 min trial. *Consolidation.* 32 adult male Long Evans rats were divided into 4 groups (NaCl-NaCl, NaCl-LPS, LiCl-LPS, LiCl-NaCl). On conditioning days 1, 4, 7 and 10, rats were ip injected with LiCl (127 mg/kg) or NaCl and immediately placed into a distinct context for a 30 min trial. After this, rats were immediately ip injected with LPS (200 μg/kg) or NaCl. *Extinction.* Both acquisition and consolidation conditioning days were followed with four no injections test days (13, 16, 18 & 22). **Results.** Compared to rats that were not pretreated with LPS before LiCl treatment, LPS given 90 mins before the LiCl-induced ANP impaired the acquisition of anticipatory nausea (*ps*<.05). Additionally, compared to rats that were not given LPS after the LiCl-induced ANP, LPS administered immediately after the LiCl-induced ANP impaired the consolidation processes of anticipatory nausea (*ps*<.05). After day 13, a rapid extinction of disgust behaviour was found in LiCl treated rats in both experiments. **Implications.** Immune system activation with the immunogen LPS significantly impairs both the acquisition and consolidation of LiCl-induced anticipatory nausea. Additionally, the effects of LiCl-induced anticipatory nausea extinguish

rapidly during the no injection extinction period. These findings confirm the impairing effects of LPS on learning and memory per se.

Disclosures: I. Bishnoi: None. **M. Kavaliers:** None. **K. Ossenkopp:** None.

Poster

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Program #/Poster #: P850.08

Topic: H.07. Long-Term Memory

Support: National Institute on Aging R01AG054180
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Title: Caloric Restriction and Fasting Diets have Negligible, or Potentially Damaging, Effects on Cognition in Diversity Outbred Mice

Authors: *A. OUELLETTE[1,2], N. HADAD[1], A. DEIGHAN[1], L. ROBINSON[1], A. FREUND[3], G. CHURCHILL[1], C. KACZOROWSKI[1,3];
[1]The Jackson Lab., Bar Harbor, ME; [2]The Univ. of Maine, Orono, ME; [3]Calico Life Sciences, LLC, South San Francisco, CA

Abstract: Aging remains the greatest risk factor for developing Alzheimer's disease (AD). Given the projected increase in lifespan across the globe, the risk of developing AD is expected to increase, along with significant economic and social burden. As such, the demand for interventions that increase cognitive longevity in parallel with lifespan is high. Several studies have reported that caloric-restriction (CR) or fasting increase lifespan and improve cognition in humans and models of aging, but others have reported no beneficial effect of CR on cognition. These discrepancies may be explained by uncontrolled environmental factors inherent in large human studies, and the lack of genetic diversity in previous animal studies where effects of CR or fasting on cognitive longevity studies in one inbred strain of mice may not generalize to other mouse strains, let alone to humans.
In this study, we compare the effects of Ad Lib, 1 day fast, 2 day fast, 20% CR and 40% CR on Y-maze working memory and contextual fear memory (CFM) in a genetically diverse population of mice - the Diversity Outbred (DO). We observed no benefit of CR or fasting on working memory or CFM at 24 months, despite expected increase in %live mice at 24 months. Actually, the 40% CR group exhibited impaired long-term CFM compared to groups fasted for 1 and 2 days. These results emphasize the importance of identifying therapeutics to enhance cognitive longevity, as CR enhancement of lifespan may not generalize to cognitive abilities.
We also performed QTL mapping of CFM outcomes with diet as an interactive covariate; when

comparing diets that enhance lifespan in the DO population but do not confer cognitive deficits (i.e. 20% CR, 1-day and 2-day fasting) we identified a locus on chromosome 10 that overlaps with the gene *Slc16a7*. Allele association mapping reveals a strong influence of founder allele on CFM variance, with B6 alleles associating with worsened outcomes and WSB with enhanced outcomes. Future work will involve validating the role of *Slc16a7* as an effective mediator of both life- and health span.

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Poster

P850. Consolidation and Reconsolidation: Behavior

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Program #/Poster #: P850.09

Topic: H.07. Long-Term Memory

Support: NIH Grant AG028271
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Title: Dha prevents diet-induced memory impairments and inflammatory gene expression in aged rats and cultured microglial cells

Authors: *M. BUTLER[1], N. P. DEEMS[1], S. M. MUSCAT[1], N. MASSA[1], C. M. BUTT[2], M. A. BELURY[1], R. M. BARRIENTOS[1];
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Abstract: It is known that diets high in refined carbohydrates can lead to impairments in brain aging and cognitive performance, but the underlying mechanisms of these impairments are not well understood. Our lab has previously demonstrated that aging is associated with a decrease in the omega-3 polyunsaturated fatty acid docosahexaenoic acid (DHA) in the rat brain. DHA is involved in many beneficial functions in the brain including neuronal signaling, altering membrane structure and function, lipid mediator production, and regulating immune cells to resolve inflammation. Thus, this age-dependent decrease in DHA may contribute to diet-induced memory impairments and inflammation observed previously. Here, we studied the impact of a refined-ingredients diet, with and without DHA, on learning and memory and inflammatory gene expression in aged rats. In addition, we examined the impact of DHA on microglia-mediated inflammation *in vitro*. Our data indicate that consuming a diet high in refined carbohydrates for one month led to memory impairments and increased inflammatory gene expression in the hippocampus of aged, but not young rats, relative to control rats fed the standard chow diet. In

contrast, the same processed diet supplemented with DHA produced no changes in memory or inflammatory gene expression in either young or aged rats, suggesting that DHA may mitigate the impact of diet-induced memory deficits via neuroimmune mechanisms in aged rats. It is important to note that in addition to reduced complex carbohydrates and fiber, processed foods are also often high in saturated fatty acids, (SFAs) such as palmitic acid (PA). *In vitro*, BV2 microglial cells treated with PA exhibited increased inflammatory gene expression relative to vehicle-treated cells. Furthermore, DHA pretreatment prevented these SFA-induced responses. Together, these data suggest that DHA can prevent diet-induced behavioral and cellular changes elicited by processed ingredients and SFAs, respectively.

Disclosures: M. Butler: None. **N.P. Deems:** None. **S.M. Muscat:** None. **N. Massa:** None. **C.M. Butt:** None. **M.A. Belury:** None. **R.M. Barrientos:** None.

Poster

P850. Consolidation and Reconsolidation: Behavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P850.10

Topic: H.07. Long-Term Memory

Support: BLR&D 5IK2BX004105-2

Title: Repetitive transcranial magnetic stimulation improvement of memory consolidation is time of day dependent

Authors: A. HEATH[1], J. ZEITZER[1], *M. MCNERNEY[2];
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Abstract: Brain stimulation is known to have specific effects on memory and the protein pathways involved in memory. We have previously shown the ability for 10Hz repetitive transcranial magnetic stimulation (rTMS) to improve memory consolidation in mice when tested on their object recognition recall after 72 hours. This experiment also showed changes to the phosphorylation of CREB and CAMKII proteins at the synapse. These proteins are involved in sleep memory consolidation and undergo circadian cycles of their phosphorylation in the brain. As abnormal sleep and circadian rhythm can have disruptive effects on learning and memory, there is a potential that rTMS could have altered effects on memory at different times in the circadian cycle. Therefore, we hypothesized that rTMS given either during the light or dark cycle of a mouse's circadian rhythm will have differential effects on its ability to improve memory. We first implanted male B6129SF2/J mice (n=43, under a 12-hour light/12-hour dark cycle starting at 0630) with a cranial attachment to secure the rTMS coil so that the mice could be given consistent stimulation to the frontal area whilst freely moving. Mice were either experimented on at 0900 (Light-Sham n=11, Light-rTMS n=11) or at 2100 (Dark-Sham n=10, Dark-rTMS n=11).

Mice first underwent familiarization phase of the object recognition test and then given treatment +3, +24, +48 hours following the test. Treatment consisted of 600 pulses of intermittent theta burst (iTBS) rTMS stimulation or sham treatment. At +72 hours mice were tested for their exploration of the novel vs familiar object. During the light cycle, the mice which received the iTBS stimulation, not the Sham, explored the novel object more than the familiar object. Interestingly, in the dark cycle the iTBS and Sham stimulation demonstrated good object recognition memory, both exploring the novel object significantly more than the familiar object. These results show that rTMS improved memory consolidation only when it was disrupted during the light sleep phase. Additional results examining the glutamatergic receptors, synaptic pathways and circadian rhythm changes that have occurred will be discussed. These results will help us determine the circadian specific effects of rTMS that can help improve memory when it is disrupted in the sleep cycle.

Disclosures: A. Heath: None. **J. Zeitzer:** None. **M. McNerney:** None.

Poster

P854. Hormones and Cognition: Development and Modulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P854.01

Topic: F.02. Behavioral Neuroendocrinology

Support: University of California Regents
Hellman Foundation
Brain and Behavior Research Foundation

Title: Impact of Reproductive Stage on Working Memory Circuitry

Authors: *E. MURATA[1], L. PRITSCHET[1], E. JACOBS[1,2];
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[2]Neurosci. Res. Inst., Univ. of California Santa Barbara, Santa Barbara, CA

Abstract: Sex steroid hormones are neuromodulators of learning and memory (Taxier & Frick, 2020). During the menopausal transition, 17β-estradiol and progesterone production declines by ~90%, while gonadotropin [Follicle Stimulating Hormone (FSH), Luteinizing Hormone (LH)] levels rise considerably. However, cognitive neuroscience studies have largely overlooked the relationship between the brain and endocrine system despite the fact that the brain is an endocrine organ. The UCSB Healthy Midlife Aging Study, a multimodal neuroimaging study, works to fill this unexplored area by characterizing the neural and cognitive changes that unfold in midlife, as a function of endocrine aging. In this analysis, we examined the impact of menopausal status on working memory (WM) performance and task-evoked fMRI activity. Midlife adults (current *n*=88, age range: 45-55 years, mean age: 51.5 years, 65% women) from

the Santa Barbara community underwent in-person neuropsychological evaluations, an MRI scan, and serological hormone assessments. Reproductive stage (pre, peri-, post-menopause) was determined via STRAW-10 guidelines. Participants completed an n-back WM paradigm (cf. Jacobs et al., 2011) during fMRI scanning to assess sustained activity in WM circuity as a function of load (0, 2, 3-back blocks) and transient changes in response to specific trial types with an increasing demand for cognitive control (non-targets, targets, lures). Behaviorally, across all women, lower 17β-estradiol was associated with longer response times when WM demands were taxed (e.g., 2-back condition [$t(43)=-2.76$, $p<0.01$, $r=-0.39$] and lure trials [$t(43)=-3.40$, $p<0.01$, $r=-0.46$]; but not during the 0-back control condition [$t(42)=-0.83$, $p=0.41$, $r=-0.13$]). Similarly, increased FSH, an indicator of advanced reproductive stage, was associated with longer WM-related response times (e.g., 2-back [$t(43)=2.55$, $p<0.05$, $r=0.36$]; lure trials [$t(43)=2.03$, $p<0.05$, $r=0.30$]). In an exploratory analysis at $p<0.001_{unc}$ (k=10*)*, postmenopausal women exhibited greater task-evoked activity in right middle frontal gyrus (MFG; cluster coordinates: 24, 14, 34) relative to premenopausal women as well as perimenopausal women (22, 12, 34). FSH levels were positively associated with activity in a near-identical cluster in MFG (24,12, 38). Together, these findings suggest that advancing reproductive stage is associated with increased WM-related prefrontal cortex activity, perhaps as a compensatory mechanism that sustains task accuracy or as an indicator of neural inefficiency. These findings begin to address the critical knowledge gap in women's brain health.

Disclosures: E. Murata: None. **L. Pritschet:** None. **E. Jacobs:** None.

Poster

P854. Hormones and Cognition: Development and Modulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P854.02

Topic: F.02. Behavioral Neuroendocrinology

Support: CREST Project, Japan
JST Project, Japan

Title: Hippocampus-synthesized estrogen and androgen rapidly modulate dendritic spines and LTP in non-genomic manner

Authors: *S. KAWATO, M. SOMA, M. OGIUE-IKEDA;
Univ. of Tokyo, Tokyo, Japan

Abstract: We demonstrated (1) hippocampal synthesis of estrogen and androgen, and (2) non-genomic synaptic modulation by these sex-steroids. [Synthesis] We showed expression as well as neuronal/synaptic localization of essential enzymes in the adult male rat hippocampus. Mass-spectrometric analysis demonstrated that hippocampal levels of estradiol (E2), testosterone (T),

dihydrotestosterone (DHT) were around 8 nM for all, higher than in plasma. Castration significantly decreased T and DHT in the hippocampus. Castration did not decrease male hippocampal E2, indicating that E2 is synthesized from hippocampal T. Female hippocampal levels of E2 (0.5-4 nM), and T (1 nM) were less than male, but much higher than in plasma. [Synaptic Modulation] E2-induced rapid non-genomic modulation (1 h) was demonstrated by analysis of spinogenesis and LTP of adult male rat hippocampal 'acute' slices (steroid-depleted slices). Spine analysis was performed for pyramidal neurons in hippocampal slices. The density of spines and their head diameters were obtained by mathematical software Spiso-3D which identifies spines by calculating geometrical parameters. E2 at 1 nM rapidly increased the density of small-head spines, in CA1. T and DHT increased the density of small-head and large-head spines. Signaling pathways are: synaptic ERalpha or AR→LIMK, MAPK, PKA, PKC, Src →cofilin or cortactin → actin polymerization→ new spines. LTP analysis showed that 1 nM E2 induced full-LTP (E2-LTP) upon sub-threshold stimulation, although without E2 the sub-threshold stimulation did not induce full-LTP. Kinase inhibitors against MAPK, PKA, PKC blocked E2-LTP. Rapid E2 synthesis can also be measured with LTP in hippocampal slices. References: Kimoto et al., 2001 Endocrinology, Hojo et al., 2004 PNAS, Mukai et al., 2007 J. Neurochem, Hojo et al., 2009 Endocrinology, Mukai et al. 2011 Cerebral Cortex, Ooishi et al. 2011 Cerebral Cortex, Okamoto et al., 2012, PNAS, Kato et al., 2013, Frontier Neurosci. Hasegawa et al., 2015 Brain Res., Hatanaka et al., 2015 Brain Res., Murakami et al., 2015 Brain Res., Soma et al., 2018 Frontier Neurosci., Hojo and Kawato 2018 Frontier Neurosci.

Disclosures: S. Kawato: None. **M. Soma:** None. **M. Ogiue-Ikeda:** None.

Poster

P854. Hormones and Cognition: Development and Modulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P854.03

Topic: F.02. Behavioral Neuroendocrinology

Support: CIHR Project Grant 169203

Title: Androgen modulation of behavioural flexibility in male rats

Authors: *V. M. LO, C. R. KELSEY, S. B. FLORESCO, K. K. SOMA;
Psychology, Univ. of British Columbia, Vancouver, BC, Canada

Abstract: Behavioural flexibility, the ability to adapt behaviour in response to environmental changes, is regulated by the mesocorticolimbic system. Strategy set shifting is one form of behavioural flexibility, and in preclinical rodent assays, subjects initially learn to use one rule to receive a reward (e.g. select the lever denoted by a visual cue, or a "cue rule"), but then must switch to a new discrimination strategy (e.g. select the lever in one position, irrespective of the

position of the visual cue, or a “response rule”). Androgens, such as testosterone (T), are produced locally within the mesocorticolimbic system, and T treatment impairs set shifting. Our previous studies show that decreasing androgens with the androgen synthesis inhibitor abiraterone acetate (ABI) improves behavioural flexibility in male rats performing a strategy set-shifting task using a cue-response shift. However, the effect size of ABI treatment was relatively small. In the present study, we assess different set-shifting procedures intended to make the shift more difficult for control animals and examine whether this may potentially increase the effect size of ABI treatment. In so doing, we modified the strategy set-shifting task to be more difficult and determined the effect of ABI on the original (cue-response shift) and modified (response-cue shift) paradigms. In the first study, rats were assigned to one of six different set shifting tasks, which required them to perform either the cue-response shift or response-cue shift with variable numbers of minimum learning trials when learning the first discrimination, and varied whether the set-shift on the following day was preceded by 20 reminder trials prior to the shift. Rats performing the response-cue shift required a significantly greater number of trials to criterion to complete the task and made significantly more total errors, perseverative errors, and never-reinforced errors to criterion, compared to rats performing the cue-response shift. These results show that the response-cue shift is significantly more difficult for rats. In the second study, rats were assigned to (1) ABI or vehicle treatment and (2) the cue-response shift or the response-cue shift. Data analysis for the second study is ongoing. Together, the data will clarify the effects of androgens on behavioural flexibility and the mesocorticolimbic system.

Disclosures: V.M. Lo: None. **C.R. Kelsey:** None. **S.B. Floresco:** None. **K.K. Soma:** None.

Poster

P854. Hormones and Cognition: Development and Modulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P854.04

Topic: F.02. Behavioral Neuroendocrinology

Title: The effects of long-term oral tamoxifen administration on anxiety and cognition in Long-Evans Hooded female rats: a potential role for BDNF?

Authors: S. M. BLOSSOM[1], A. R. HALLIDAY[1], G. ROTH[1], K. DOMENECH[1], P. ABRAMENKO[1], K. POLLACK[1], G. PASCAL[1], L. BEHBEHANI[1], *L. BEEN[2], M. KELLY[1]; [1]Psychology, [2]Haverford Col., Haverford, PA

Abstract: Long-term administration of tamoxifen (Tam), a selective estrogen receptor modulator (SERM), is a common adjuvant therapy for estrogen receptor-positive breast cancers. Though initial protocols recommended 5 years of treatment, current protocols often recommend up to 10 years of Tam therapy to reduce the risk of cancer recurrence. Clinical studies have reported significant alterations in mood and cognition during Tam treatment. However, the impact of

long-term modulation of estrogen receptors by Tam on cognitive health has not been adequately studied. To better understand the impact of Tam on the brain and behavior, this study used custom medicated food pellets (Envigo) to model long-term oral administration of Tam to adult female Long-Evans Hooded rats. Following 10-13 weeks of Tam chow (n = 16) or standard chow (n = 16), rats underwent behavioral testing for several cognitive and affective measures, including the elevated plus maze, open field, and novel object recognition. At the conclusion of the study, animals were sacrificed by intracardial perfusion. Importantly, long-term oral Tam administration resulted in clinically-relevant levels of Tam in plasma, demonstrating that this method of administration is a robust model for long-term oral Tam therapy. Consistent with reports of increased anxiety in people prescribed long-term Tam therapy, we observed an anxiogenic influence of Tam in the open field and elevated plus tests. Specifically, rats in the Tam group demonstrated a higher tendency of thigmotaxis than control rats, spending significantly more time in the periphery versus the center of the open-field apparatus, and significantly more time in the closed arms versus the open arms of the elevated plus. To better understand the mechanistic underpinnings of TAM's influence on behavior, we are currently exploring the potential role of long-term oral Tam administration on the expression of Brain Derived Neurotrophic Factor (BDNF) in several estrogen-sensitive brain regions. We are targeting BDNF because previous research has demonstrated that BDNF-synthesizing neurons co-localize with estrogen receptors in the forebrain. Given that Tam acts as a modulator of estrogen receptors, BDNF expression may be an important readout of the effects of long-term oral TAM administration on behaviorally-relevant neuroplasticity. Brain analyses are ongoing and may point to downstream targets for further investigation.

Disclosures: S.M. Blossom: None. **A.R. Halliday:** None. **G. Roth:** None. **K. Domenech:** None. **P. Abramenko:** None. **K. Pollack:** None. **G. Pascal:** None. **L. Behbehani:** None. **L. Been:** None. **M. Kelly:** None.

Poster

P854. Hormones and Cognition: Development and Modulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P854.05

Topic: F.02. Behavioral Neuroendocrinology

Support: NIH Grant MH123768
BBRF NARSAD Young Investigator Award
NCSU CVM Intramural Research Award

Title: Ovarian hormones tune amygdala inhibition to drive anxiety-like behavior across the estrous cycle

Authors: *A. L. FARTHING, A. L. BUNCE, G. L. RYHERD, E. K. LUCAS;
Mol. Biomed. Sci., North Carolina State Univ., Raleigh, NC

Abstract: Fluctuations of ovarian hormones during the female reproductive cycle influence emotional processing and have been shown to modulate anxiety behavior in humans and rodents. In the context of anxiety disorders, higher levels of ovarian hormones are associated with less severe symptoms. Similarly, female rodents display decreased anxiety-like behavior in the proestrus phase of the estrous cycle when ovarian hormone levels are high. Currently, the cellular mechanisms driving the anxiolytic effect of proestrus remain unknown. Activity of the basolateral amygdala (BLA), the emotional switchboard of the brain, is tightly regulated by GABAergic parvalbumin-expressing interneurons (PVIs), which are also known to express ovarian hormone receptors. We therefore hypothesized that ovarian hormones regulate BLA PVI activity to drive anxiety-like behavior across the estrous cycle. Using whole-cell patch clamp electrophysiology, we first demonstrated that BLA PVIs exhibit hyperpolarized resting membrane potential and display reduced frequency, but not amplitude, of miniature excitatory postsynaptic currents in proestrus compared to diestrus. This indicates that BLA PVIs have reduced function when ovarian hormone levels are high. We next hypothesized that increasing BLA PVI activity would eliminate the anxiolytic effects of proestrus. Using the Cre-dependent excitatory DREADD hM3D, we bilaterally targeted the BLA of male and female PV-IRES-Cre mice. We first validated that systemic administration of clozapine-N-oxide induced equivalent c-fos expression in male versus female BLA PVIs. We then tested the effects of BLA PVI activation in males and females in proestrus versus diestrus on the elevated plus maze, open field, and social interaction tests. We found that activation of BLA PVIs reversed the anxiolytic effects of proestrus while having no effects on elevated plus maze or open field performance in males or diestrus females. On the social interaction test, PVI activation decreased social preference in females, independent of estrous cycle stage. Our data supports a model for decreased BLA PVI activity in females during proestrus. Contrary to current models, our data suggest that increased BLA PVI activity promotes, rather than inhibits, anxiety-like behavior. Our work implicates a role for cycling ovarian hormones in regulating the critical balance of excitation and inhibition in the BLA and its ultimate impacts on affective behavior.

Disclosures: A.L. Farthing: None. **A.L. Bunce:** None. **G.L. Ryherd:** None. **E.K. Lucas:** None.

Poster

P854. Hormones and Cognition: Development and Modulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P854.06

Topic: F.02. Behavioral Neuroendocrinology

Support: NIH Grant R21MH11909

Title: Pubertal estradiol regulates development of adult anxiety- and depressive- behavior in female mice

Authors: *E. M. WOODWARD[1], L. COUTELLIER[1,2];
[1]Dept. of Neurosci., [2]Dept. of Psychology, The Ohio State Univ., Columbus, OH

Abstract: Neuropsychiatric disorders are equally diagnosed in men and women until puberty, when the incidence of mood disorders including anxiety and major depressive disorder in women rises to over two times that of men. This suggests that puberty may be a time of sexually dimorphic neurobiological development that leaves females more vulnerable to neuropsychiatric disease. A potential mechanism leading to this sex-specific vulnerability may be reorganization of structures responsible for regulating stress responsivity, such as the prefrontal cortex (PFC). Previous work in our lab has shown that in adult mice, stress increases the activity of prefrontal parvalbumin (PV) interneurons in females only, a change that is causally responsible for increased anxiety. Sex-specific differences in sensitivity to stress of prefrontal PV+ cells may develop during puberty. Since others have found that the pubertal increase in female gonadal hormone 17β-estradiol and its receptor estrogen receptor beta (ERβ) are required for prefrontal maturation of inhibitory neurotransmission, here we propose 17β-estradiol acts on ERβ on PV+ cells to promote their maturation. We hypothesize that female vulnerability to stress-induced anxiety in adulthood develops during puberty via the organizing actions of 17β-estradiol binding ERβ on PV+ interneurons in the PFC. To test this hypothesis, female C57Bl6 mice underwent ovariectomy (OVX) or sham surgery at a prepubertal timepoint (P25) and were supplemented with 17β-estradiol or vehicle once daily throughout the end of the experiment. In adulthood, mice underwent four weeks of unpredictable chronic mild stress (UCMS) followed by assessment of anxiety- and depressive-like behaviors. OVX mice not supplemented with estradiol had reduced anxiety-like behavior in the marble burying test and increased depressive-like behavior in the splash test, effects that are independent of UCMS. These behavioral results suggest that the organizing actions of 17β-estradiol during puberty are necessary to facilitate maturation of a normal emotional phenotype regardless of stress exposure. Further analyses will determine the potential role of PV+ cells in this process.

Disclosures: E.M. Woodward: None. **L. Coutellier:** None.

Poster

P854. Hormones and Cognition: Development and Modulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P854.07

Topic: F.02. Behavioral Neuroendocrinology

Support: National Science Foundation Grant 2023985

Title: High-amplitude co-fluctuations in cortical activity are linked to concentrations of reproductive hormones over the menstrual cycle

Authors: *S. GREENWELL[1], J. FASKOWITZ[2], L. A. PRITSCHET[4], T. SANTANDER[5], E. G. JACOBS[6], R. BETZEL[3];
[1]Dept. of Psychological and Brain Sciences, Indiana Univ., Bloomington, IN, IN; [2]Psychological and Brain Sci., [3]Dept. of Psychological and Brain Sci., Indiana Univ., Bloomington, IN; [4]Psychological and Brain Sci., [5]Psychological & Brain Sci., [6]Dept. of Psychological and Brain Sci., Univ. of California, Santa Barbara, Santa Barbara, CA

Abstract: Many studies have demonstrated associations between hormones involved in the menstrual cycle and brain function, including cortical activity and functional connectivity, but the relationship between transient changes in hormone concentration and fast fluctuations in brain connectivity are not well understood. Here, we leverage fMRI "edge time series"—a novel method for estimating functional connectivity at a framewise timescale – to investigate the relationship between high-amplitude network states and daily variation in sex hormones (estradiol and progesterone) and gonadotropins (luteinizing hormone and follicle-stimulating hormone) in a "densely sampled" single individual over the course of two complete menstrual cycles, one natural and the other during a hormone regimen (60 MRI scans, in total). We find that concentrations of gonadotropins are associated with the instance of high-amplitude states of network connectivity, particularly during states of opposing activation of default-mode/control and attention/sensory systems. Further, we find that edge-level variations within these high-amplitude states are associated with concentrations of both sex hormones and gonadotropins. Our results were robust across two processing pipelines, one that included global signal regression as a processing step and the other that did not. We also found that our results were consistent when data from each menstrual cycle was analyzed separately and after systematically removing data from each to ensure that our results were not driven by outliers from a single scan session. We conclude that transient changes in hormone levels during the menstrual cycle are related to high-amplitude network states of cortical functional connectivity, indicating a possible route for these hormones to influence daily changes in cognition and behavior.

Disclosures: S. Greenwell: None. **J. Faskowitz:** None. **L.A. Pritschet:** None. **T. Santander:** None. **E.G. Jacobs:** None. **R. Betzel:** None.

Poster

P854. Hormones and Cognition: Development and Modulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P854.08

Topic: F.02. Behavioral Neuroendocrinology

Support: NIH Grant HD093907

Title: Progesterone receptor expression in developing medial prefrontal cortex of rats.

Authors: *P. GRANEY[1], C. K. WAGNER[2];
[1]Univ. At Albany, Albany, NY; [2]Univ. Albany, Albany, NY

Abstract: During development, progesterone receptors (PR) are transiently expressed in virtually all regions of the neocortex, beginning in the subplate at embryonic day 18 (E18), followed by expression in layer V at postnatal day 3 (P3) and in layers II/III by P6. PR expression is essentially absent in neocortex by P28. PR is also expressed in the developing medial prefrontal cortex (mPFC), in layer III from P6 until P30, but does not appear to be expressed in layer V. However, PR expression in the subplate of mPFC has not been well characterized. Subplate is a transient structure that emerges at E18 in rodents and receives the first afferent synapses arriving in cortex. Disruption in the development and function of subplate in rodents disrupts cortical organization and has been implicated in neurodevelopmental disorders in humans. The presence or absence of PR in the perinatal mPFC subplate could have significant implications for understanding the normal development of mPFC connectivity. mPFC was collected from neonatal rats on the day of birth (P1), P2, P5, P6, P7, and P10, sectioned on a freezing microtome and immunocytochemistry for PR was performed using primary antisera (DAKO) that recognizes both PR_A and PR_B. A semi-quantitative analysis of PR-ir levels within cells of mPFC subplate was conducted using a scale from 0-5 with 5 representing the darkest stained cells and a score of 0.50 being considered just detectable levels. Results indicate that PR expression in the subplate of frontal cortical areas was detectable at P1 with a peak mean score of 4.89. PR expression in mPFC was greatest within the anterior cingulate cortex (AC) where it was detectable at P1 (mean score = 1.44), gradually increasing until a peak at P6 (mean score = 2.86). PR within the prelimbic (PL) mPFC was detectable by P5 (mean score = 0.90), peaking at P6 (mean score = 1.71). PR within the infralimbic cortex (IC) did not exceed a mean of 0.50 for any of the days examined. PR within the subplate of all three subregions was absent by P10. While PR is expressed in the subplate of PL and IL during the first week of development, it is clearly expressed at lower levels per cell compared to the subplate of AC or the subplate of other regions of frontal cortex. PR is also at lower levels in PL and IL subplate compared to expression levels in layer 3 of these regions. These preliminary findings suggest that the role of PR in the development of mPFC may be less dependent on subplate expression than other regions and that effects of progestins on mPFC-dependent behaviors may be primarily mediated by PR activity in layer 3 and in dopaminergic afferents from the ventral tegmental area.

Disclosures: P. Graney: None. **C.K. Wagner:** None.

Poster

P854. Hormones and Cognition: Development and Modulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P854.09

Topic: F.02. Behavioral Neuroendocrinology

Support: OISE Grant #1545803

Title: Nandrolone Use During Adolescence Increases Cocaine Sensitization And Impairs Reproductive Function In Adult Female Rats

Authors: *C. J. RIVERO[1], J. A. FREIRE[1], P. SIACA[2], I. SANTIAGO[3], A. C. SEGARRA[1]; [1]Physiol., UPR Med. Sci. Campus, San Juan, Puerto Rico; [2]Biol., UPR Rio Piedras Campus, San Juan, Puerto Rico; [3]Neurosci., Case Western Reserve Univercity, Cleveland, OH

Abstract: Anabolic androgenic steroids (AAS) are drugs that mimic the chemical structure of testosterone. The most commonly AAS used by young adults is nandrolone. Use of AAS has been associated with increased risk of steroid dependence, mood disorders and polysubstance use. Indeed, cocaine use in this population is approximately 33% compared to 4.9% in the rest of the population. The effect that use of nandrolone during adolescence has on female motivational behaviors and on the reproductive system has not been investigated. Adolescent female rats were divided into two groups, one that received nandrolone decanoate (20mg/kg/sc) and another that received sesame oil (vehicle) for 10 successive days starting on day 28. On day 38 rats were tested on an Open Field and on day 39 in an Elevated Plus Maze. On day 40, the rats were then divided into 2 additional groups for a total of 4 groups: Oil-Saline, ND-Saline, Oil-Cocaine and ND-Cocaine and tested for locomotor sensitization to cocaine. From days 1-5 and at days 13 and 23 rats received an injection of cocaine (15 mg/kg/ip) or saline. Their locomotor response to cocaine was measured at days 1, 5, 13 and 23. Our data show that nandrolone treated rats have increased time spent in center of open field and augmented locomotor response to cocaine on days 5 and 13 compared to oil treated rats. This suggests that nandrolone increases risk taking behavior and the psychoactive properties of cocaine. In addition, in the ovaries it reduced weight and induced the formation of cysts. In the Uterus, it increased weight and hindered endometrial growth. These data show that exposure to supra-physiological levels of androgens during adolescence induces neurochemical changes in brain circuitry that regulate emotional and addictive behaviors increasing the psychoactive properties of cocaine. Nandrolone severely impairs reproductive system function, inducing formation of ovarian cysts and uterine dysfunction. This effect is exacerbated when paired with cocaine.

Disclosures: C.J. Rivero: None. **J.A. Freire:** None. **P. Siaca:** None. **I. Santiago:** None. **A.C. Segarra:** None.

Poster

P854. Hormones and Cognition: Development and Modulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P854.10

Topic: F.02. Behavioral Neuroendocrinology

Support: CU Foundation

Title: Effects of Mycobacterium vaccae NCTC11659 treatment on fear extinction in female rats in the fear-potentiated startle paradigm

Authors: *A. K. LUTHENS[1], A. K. LEE[1], J. E. HASSELL, JR[3], C. L. FOXX[1], A. L. ARNOLD[1], C. A. LOWRY[1,2,4,5,6];
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Abstract: Prior research in our laboratory has shown that preimmunizations with a whole-cell, heat-killed preparation of *Mycobacterium vaccae* NCTC 11659 (*M. vaccae*) in rats reduces stress-induced resistance to fear extinction in the fear-potentiated startle paradigm (FPS) (Fox et al., 2017). More recently, we demonstrated that *M. vaccae* immunizations reduced stress-induced resistance to both within-session and between-session fear extinction in the FPS (Hassell et al., 2019), which may have implications on future treatments for posttraumatic stress disorder (PTSD). These studies did not include any female rats, however. To confirm these results in both male and female populations, we measured interactions between *M. vaccae* and emotional behavior in FPS for adult Female Sprague-Dawley® rats (N=24) (strain code 400, Charles River, Wilmington, MA, USA). Fear conditioning took place over two days. For three consecutive weeks following fear conditioning, the rats received weekly subcutaneous injections of *M. vaccae* (n=12) or borate buffered saline (BBS) vehicle (n=12). One week after the final injection, there were six days of extinction training. Estrous cycle staging also took place on day 6 of extinction training. Here, we show that preimmunizations with *Mycobacterium vaccae* NCTC 11659 reduced stress-induced resistance to within-session extinction on day 1 of extinction training as in previous studies. Unlike the results of previous studies, however, *M. vaccae* preimmunization had no effect on between-session extinction in female rats. We additionally show that treatment with *M. vaccae* did not alter the distribution of estrous cycle stage. Overall, the results of the current study suggest that *M. vaccae* has stress-resilient properties and may be useful as a treatment for PTSD. However, these results suggest that sex differences exist in emotional behavioral responses to *Mycobacterium vaccae* NCTC 11659 in the FPS paradigm.

Disclosures: A.K. Luthens: None. **A.K. Lee:** None. **J.E. Hassell:** None. **C.L. Foxx:** None. **A.L. Arnold:** None. **C.A. Lowry:** None.

Poster

P931. Advances in the Application of Optical Methodologies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P931.01

Topic: I.04. Physiological Methods

Support: NIH Grant R01NS078168

Title: Understanding the origins of brain motion in behaving mice

Authors: *C. S. GARBORG[1], Q. ZHANG[2], Y.-J. KIM[1], F. COSTANZO[2], P. J. DREW[2]; [1]Dept. of Biomed. Engin., [2]Dept. of Engin. Sci. and Mechanics, Pennsylvania State Univ., University Park, PA

Abstract: The brain moves within the skull, but the origin and physiological impact of this movement are not understood. Because brain movement is a ubiquitous confound for both human and animal imaging, it is important to understand the nature and drivers of this brain motion. Furthermore, the movement of the brain may aid in the circulation of cerebrospinal fluid (CSF), which could serve to transport metabolic waste out of the brain. Despite the relevance of brain motion to many aspects of neuroscience and brain function, we know remarkably little about the drivers of this motion, or its nature. One intriguing possibility is brain motion is caused by pressure changes in the abdomen, generated by abdominal muscle contraction, that are transmitted to the brain and spinal cord. To quantify brain motion, we used high-speed, multi-plane two-photon microscopy in awake mice free to locomote on a spherical treadmill. We quantified abdominal muscle activity using electromyography (EMG) recordings in external oblique abdominal muscles, as well as the EKG and respiratory rate. During voluntary locomotion, we observed contraction of abdominal muscles and found robust, rostral displacement of the brain, along with smaller lateral movements. These abdominal contraction-linked brain movements were orders of magnitude larger than those driven by respiratory or cardiac pulsations. Our results suggest that brain motion in the behaving animal is tied to abdominal muscle contractions.

Disclosures: C.S. Garborg: None. **Q. Zhang:** None. **Y. Kim:** None. **F. Costanzo:** None. **P.J. Drew:** None.

Poster

P931. Advances in the Application of Optical Methodologies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P931.02

Topic: I.04. Physiological Methods

Title: Estimating fiber photometry collection volumes across different brain regions

Authors: *C. MONTINARO[1,2], M. PISANELLO[3], M. BIANCO[4], B. SPAGNOLO[4], F. PISANO[4], A. BALENA[4], F. DE NUCCIO[5], D. LOFRUMENTO[5], T. VERRI[5], M. DE VITTORIO[4,5], F. PISANELLO[4];
[1]Inst. Italiano di Tecnologia, Arnesano, Italy; [2]Dip. Scienze e Tecnologie Biologiche ed Ambientali, Univ. del Salento, Lecce, Italy; [3]Ctr. for Biomolecular Nanotechnologies, [4]Inst. Italiano Di Tecnologia, Arnesano, Italy; [5]Univeristà del Salento, Lecce, Italy

Abstract: Estimating fiber photometry collection volumes acrossdifferent brain regionsCinzia Montinaro[1, 2, *], Marco Pisanello[1], Barbara Spagnolo[1], Marco Bianco[1,3], Filippo Pisano[1], Antonio Balena[1,3],Francesco De Nuccio[2], Dario D. Lofrumento[2], Tiziano Verri[2],Massimo De Vittorio[1, 3, *] and Ferruccio Pisanello[1, *1]Istituto Italiano di Tecnologia (IIT)-CBN, 73010 Arnesano(LE), Italy[2]DiSTeBA, Università del Salento, 73100 Lecce, Italy[3]Dip. di Ing. Inn., Università del Salento, 73100 Lecce, Italy*Corresponding authors: cinzia.montinaro@iit.it; massimo.devittorio@iit.it; ferruccio.pisanello@iit.itThe advent of high-efficiency genetically-encoded optical indicators of neural activity has widened theuse of fiber photometry (FP) [1-4], enabling *in vivo* detection of functional fluorescence with simple optical fibers. This has found applications for monitoring calcium transients, membrane potentials, neurotransmittersconcentration as well as the intracellular biochemical state of neurons with fluorescence lifetime photometry[5-8]. FP is commonly implemented using the same fiber to excite and generate fluorescence, and bothexcitation and fluorescence photons undergo to tissue attenuation and scattering. Despite the influence of thefiber's constitutive parameters on the detection volume has been estimated [9,10], its dependence on the opticalproperties of the probed region is often over-looked. In this work, we describe a method to link the anatomyof the brain structures to the fiber photometry collection volume. We used a two-photon microscope (Fig. 1A)to measure the illumination and collection fields of a fiber optics inserted in 300 μm-thick coronal brainslices (mouse line *Thy1-GCaMP6s*). We compared light collection volume in the cerebral cortex, thehippocampus and the striatum , obtaining a quantitative estimation of the fiber photometryefficiency field (ρ) [9]. In light of these results, we report how the shape of ρ is influenced by thecytoarchitecture of the investigated regions (Fig. 1B), highlighting inter/intra-region differences, with primarydifferences on the photons detection depth.**REFERENCES**1. Lütcke, H. *et al. Front. Neural Circuits*. (2010).2. Cui, G. *et al. Nat. Protoc*. **9**(6), 1213-1228 (2014).3. Gunaydin, L.A. *et al. Cell* **157**(7), 1535-1551 (2014).4. Cho, Y.K. *et al. J. Opt*. **18**(9), 093007 (2016).5. Yang, Y. *et al. Nat. Commun*. **9**(1), 1504 (2018).6. Piatkevich, K.D. *et al. Nature* **574**(7778), 413-417 (2019).7. Patriarchi, T. *et al. Science* **360**(6396), eaat4422 (2018).8. Lee, S.J. *et al. Front. Neurosci*. **13** (2019).9. Pisanello, M. *et al. Front. Neurosci*. **13** (2019).10. Maglie, E. *et al. Opt. Lett*. **45**(14), 3856 (2020).

Disclosures: C. Montinaro: None. **M. Pisanello:** None. **M. Bianco:** None. **B. Spagnolo:** None. **F. Pisano:** None. **A. Balena:** None. **F. De Nuccio:** None. **D. Lofrumento:** None. **T. Verri:** None. **M. De Vittorio:** None. **F. Pisanello:** None.

Poster

P931. Advances in the Application of Optical Methodologies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P931.03

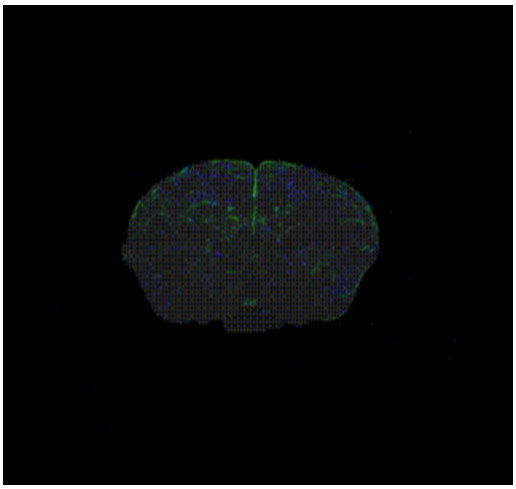
Topic: I.04. Physiological Methods

Support: Dr. Donald E. and Patsy Meier Fund for Cerebral Amyloid Angiopathy (CAA) Research
Texas Institute for Brain Injury and Repair

Title: Characterizing 3D patterns of whole brain amyloid deposition in transgenic mice using serial two-photon tomography and a machine learning pipeline

Authors: *L. L. SCHNEIDER[1], A. S. IRANI[1], A. D. AJAY[2], E. J. PLAUTZ[2], X. KONG[2], D. M. RAMIREZ[2], M. P. GOLDBERG[1];
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Abstract: Quantification and localization of vascular amyloid in three dimensions promotes understanding of spatial patterns of vascular amyloid deposition and disease progression in experimental models of cerebral amyloid angiopathy (CAA). Serial two-photon tomography (STPT) is suited to this task in murine models because it produces 3D datasets of the whole mouse brain in a matter of hours. Here, we present the pipeline we developed for machine learning classification of vascular and parenchymal amyloid, registration to standard atlases, and quantification across identified brain regions. We used traditional histological sectioning and slide mounted imaging to compare published methods for fluorescent amyloid and vascular labeling. Vascular and parenchymal amyloid deposits are labelled by an intraperitoneal injection of methoxy-x04 the day prior to euthanasia. Cerebral vessels are visualized by transcardial perfusion of Rhodamine-B isothiocyanate in gelatin. Labeled brains are next imaged using STPT and rendered in 3D to appreciate regional amyloid distribution. Volumetric image datasets are quantified using a modification of our custom designed image analysis pipeline utilizing supervised machine learning for pixel classification as vascular or parenchymal amyloid. Classified signals are registered into the Allen Institute for Brain Science Common Coordinate Framework version 3.0 (CCF v3.0). Our strategy produces images amenable to automatic classification and quantification of parenchymal and vascular amyloid deposits throughout the brain, useful for high-throughput studies on rodent models of CAA.



A 3D probability map representing the brain of a 52-week-old female Tg2576 mouse. Vascular amyloid is labelled in green and parenchymal amyloid is blue.

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Poster

P931. Advances in the Application of Optical Methodologies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P931.04

Topic: I.04. Physiological Methods

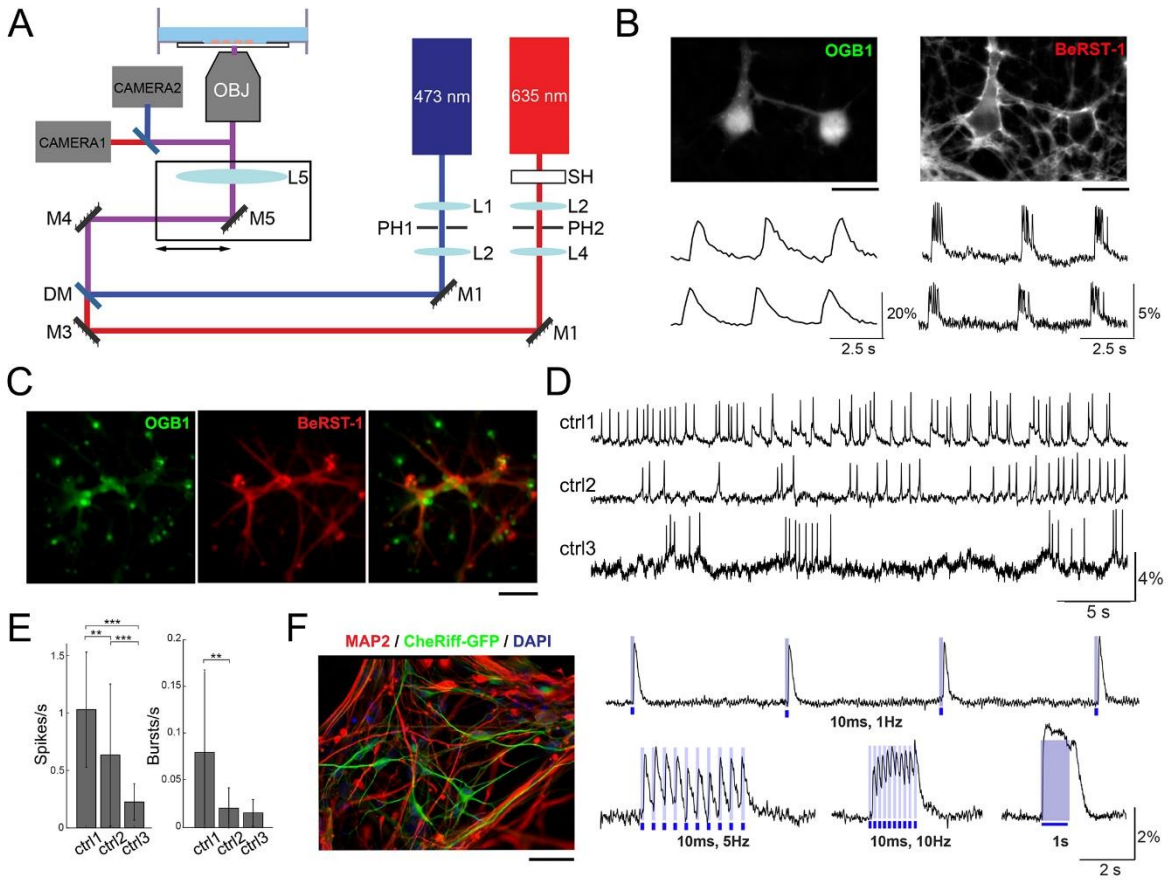
Support: NIH BRAIN R21EY030727
NIH BRAIN R01MH111359
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Research Council of Norway (223273, 248828, and 283798)
K.G. Jebsen Stiftelsen and South-Eastern Norway Regional Health Authority (#2018094)
K01 NIAAA (K01AA026911) to C.T.

Title: All-optical electrophysiology in hiPSC-derived neuronal cultures with the synthetic voltage sensor BeRST-1

Authors: F. PUPPO[1], S. SADEGH[2], C. A. TRUJILLO[1], M. THUNEMANN[6], E. P. CAMPBELL[3], M. VANDENBERGHE[7], X. SHAN[1], I. AKKOUH[10], E. W. MILLER[11], B. L. BLOODGOOD[3], G. A. SILVA[4], A. M. DALE[5], G. T. EINEVOLL[12], S. DJUROVIC[8], O. A.

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Abstract: Voltage imaging and all-optical electrophysiology in human induced pluripotent stem cell (hiPSC)-derived neurons have opened unprecedented opportunities for high-throughput phenotyping of neuronal activity in the context of the unique genetic background of individual patients. While prior all-optical electrophysiology studies relied on genetically encoded voltage indicators, here, we demonstrate a protocol that generates robust and reproducible results using the synthetic voltage sensor BeRST-1.Our imaging setup consisted of a body of an inverted epifluorescence microscope with a custom illumination light path (**Fig. 1A**). We established the protocol using primary cultures of dissociated rat neurons co-labeled with BeRST-1 and the calcium indicator OGB1 (**Fig. 1B**); co-labeling with OGB1 was critical for quick and efficient evaluation of the level of spiking activity and for choosing fields of view for subsequent voltage imaging. In parallel, we customized culture protocols to obtain healthy, active, and spatially uniform monolayer cultures of differentiated hiPSC-derived neurons. Again, co-labeling with OGB1 was critical for visual inspection providing a feedback for optimization of cell culture procedures (**Fig. 1C**). We applied our imaging protocol to three control human cell lines and observed clear differences in the firing pattern including the firing rate and bursting behavior (**Fig. 1D-E**). This finding advocates for use of isogenic controls in disease-related studies and poses a challenge to hiPSC-based studies of polygenic brain disorders. Finally, we used lentivirus to express the optogenetic actuator CheRiff in human neurons for demonstration of all-optical electrophysiology (**Fig. 1F**). In the future, we envision that the same protocol, including fixation following all-optical-electrophysiology, will be used for single-cell-resolved in situ RNA hybridization providing a link from the functional phenotype to the underlying gene expression profile.



Disclosures: F. Puppo: None. **S. Sadegh:** None. **C.A. Trujillo:** None. **M. Thunemann:** None. **E.P. Campbell:** None. **M. Vandenberghe:** None. **X. Shan:** None. **I. Akkouh:** None. **E.W. Miller:** None. **B.L. Bloodgood:** None. **G.A. Silva:** None. **A.M. Dale:** None. **G.T. Einevoll:** None. **S. Djurovic:** None. **O.A. Andreassen:** None. **A.R. Muotri:** None. **A. Devor:** None.

Poster

P931. Advances in the Application of Optical Methodologies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P931.05

Topic: I.04. Physiological Methods

Support: CIHR
BRAINSCAN, Canada
NEURONEX, NSF

Title: Examining the dynamics and time course of cortical pyramidal neuron responses to electrical stimulation with Ca^{2+}imaging in mouse and primate cortical slices.

Authors: *S. VIJAYRAGHAVAN[1], H. IGARASHI[3], V. SUKUMAR[4], W. INOUE[5], A. PRUSZYNSKI[2], J. C. MARTINEZ-TRUJILLO[6];
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Abstract: Cortical neurons integrate information to generate diverse response properties employing recurrent and persistent activity at different timescales (Spitmaan et al., PNAS (2020), 117(36), 22522-22531; Murray et al., Nat. Neurosci. (2014), 17 (12), 1661-1663). The expansion of the neocortical mantle in primates, including the prefrontal cortex, has accompanied increased capacity for cognitive control and executive functions, such as working memory, where persistent activity in cortical neurons encodes sensory, cognitive and response parameters. However, less is known about the role of intrinsic temporal profile and time constants of cortical neuron activation across mammalian species in subserving higher-order cognitive functions. Two-photon Ca^{2+} imaging provides an attractive methodology to further address the issues of diversity in cortical neuronal activation profiles. Here, we describe experiments where we transfected mouse and primate association cortex with retrograde AAV expressing gCAMP6f under the CAMKII promoter and studied activation profiles based on Ca^{2+} imaging of pyramidal neurons in cortical slices upon electrical and pharmacological stimulation. We report that pyramidal neurons in the vicinity of the electrode show diverse responses to different electrical stimulation pulses, with consistent dose response profiles. We found neurons where Ca^{2+} responses were augmented or attenuated upon stimulation. Increasing stimulation frequency with identical stimulation duration increased the Ca^{2+} response in a stimulation frequency-dependent manner, and the durations of the induced responses were more prolonged at higher stimulation frequencies, and in some cases, in the order of minutes. Moreover, pharmacologically-mediated stimulation showed the presence of both positively and inversely correlated spontaneous Ca^{2+} responses between pairs of pyramidal cells, presumably due to their connectivity in the local circuitry. Previous studies employing weaker electrical microstimulation with two-photon Ca^{2+} imaging in vivo have suggested that recruitment of neurons by microstimulation primarily occurs due to stimulation of axons in the vicinity of the electrode (Histed et al., Neuron (2009), 63, 508-522). We will discuss our results with higher current strengths in brain slices in this context.

Disclosures: S. Vijayraghavan: None. **H. Igarashi:** None. **V. Sukumar:** None. **W. Inoue:** None. **A. Pruszynski:** None. **J.C. Martinez-Trujillo:** None.

Poster

P931. Advances in the Application of Optical Methodologies

Location: Virtual Only

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Program #/Poster #: P931.06

Topic: I.04. Physiological Methods

Support: JSPS Grant-in-Aid for Scientific Research on Innovative Areas 4303
JSPS Grant-in-Aid for Scientific Research on Innovative Areas 4805
JSPS KAKENHI JP18H03662
JSPS KAKENHI 19H04984
AMED JP20dm0307021
AMED JP18dm0207003

Title: Microendoscopic calcium imaging of the primary visual cortex of behaving macaques

Authors: *M. OGUCHI[1], J. JIASEN[1], T. W. YOSHIOKA[1], Y. R. TANAKA[1], K.-I. INOUE[2], M. TAKADA[2], T. KIKUSUI[3], K. NOMOTO[4], M. SAKAGAMI[1];
[1]Brain science institute, Tamagawa Univ., Tokyo, Japan; [2]Primate Res. Institute, Kyoto Univ., Aichi, Japan; [3]Azabu Univ., Sagamihara, Japan; [4]Dept. of Physiol., Dokkyo Med. Univ. Sch. of Med., Tochigi, Japan

Abstract: Macaque monkeys have long been used as experimental models to understand the neural mechanisms of the human brain. Recently, calcium imaging with genetically encoded calcium indicator has been applied to the macaque brain. Calcium imaging allows the simultaneous recording of a number of neurons and thus is a powerful tool for understanding the function of local neural circuits at the mesoscopic scale. In primates, two-photon microscopy has been mainly used in calcium imaging studies to record neural activity from the cortical surface. In rodents, microendoscopic calcium imaging, which implants a gradient index lens into the brain and observes neural activity through a miniaturized fluorescent microscope, is also widely used, producing a number of innovative results concerning various brain areas, including deep neural nuclei. Although there also has been a report on the application of microendoscopic calcium imaging to marmosets, its application to macaque monkeys has not yet been reported. In this study, we applied microendoscopic calcium imaging to macaque monkeys targeting the primary visual cortex (V1), where neurons can be easily activated by presenting visual stimuli. We incorporated the calcium indicator GCaMP6s into a mosic adeno-associated virus vector packaged with AAV1 and AAV2 capsid protein (AAV2.1). We injected this virus into the bilateral V1 of three macaques and observed highly efficient infection in these regions. Prism lenses, which were equipped with a triangular prism at the tip of a rod lens, were implanted into the vector injection sites. Miniaturized microscopy through prism lenses revealed fluorescence changes in four hemispheres from two of the three monkeys and was able to identify dozens of neurons in three of these four hemispheres. For these two monkeys, Gabor patches with different orientations were presented in the peripheral field of view during fixation to examine the stimulus response. Observed V1 neurons had receptive fields at the corresponding positions on the retinotopic map and showed significant selective responses to specific orientations. We also successfully decoded the orientation of the visual stimulus and tracked the cells across days. In summary, we successfully recorded neuronal populations simultaneously in the macaque V1

using microendoscopy, and the observed cell populations showed response patterns consistent with the established V1 neuronal properties. These results indicate that microendoscopic calcium imaging is a powerful recording method for the macaque brain.

Disclosures: M. Oguchi: None. **J. Jiasen:** None. **T.W. Yoshioka:** None. **Y.R. Tanaka:** None. **K. Inoue:** None. **M. Takada:** None. **T. Kikusui:** None. **K. Nomoto:** None. **M. Sakagami:** None.

Poster

P931. Advances in the Application of Optical Methodologies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P931.07

Topic: I.04. Physiological Methods

Support: Howard Hughes Medical Institute

Title: In vivo voltage imaging during behavior combined with transcriptomics enables dissection of interneuron subtypes in the mouse motor cortex

Authors: *A. SINGH[1,2], T. WANG[1], A. ABDELFATTAH[3], E. SCHREITER[1], K. SVOBODA[1]; [1]Howard Hughes Med. Inst. Janelia Res. Campus, Ashburn, VA; [2]Neurosci., Johns Hopkins Univ., Baltimore, MD; [3]Brown Univ., Providence, RI

Abstract: Recent advances in single-cell transcriptomics have enabled the construction of a transcriptomic taxonomy of cortical neurons[1]. A key question in neuroscience is to what extent the behavior-related activity of individual neurons is determined by their transcriptional profiles. Can the function of a neuron be predicted by its transcriptomic type, and vice-versa? To answer this question, we need to measure the activity of particular transcriptomic types in behaving animals. We focus on GABAergic interneurons in superficial layers of the motor cortex, a transcriptionally diverse neural population whose function is not well understood. In order to obtain rich dynamics, we study mice performing a delayed-response task with sensory, motor and short-term memory components. In this task, preparatory activity during the delay epoch is maintained by a cortico-thalamocortical loop[2]. Movement initiation is triggered by ascending signals from the midbrainviathalamus[3]. Motor cortex GABAergic interneurons receive thalamocortical input and likely prevent runaway excitation in thalamocortical loops and trigger motor commands through disinhibition. We perform voltage imaging using Voltron, which allows measurement of spikes and subthreshold activity with millisecond time scale resolution in tens of neurons at a time[4]. After the imaging experiment we use multiplexed fluorescence *in situ* hybridization (mFISH) to retrospectively identify the transcriptomic types of the imaged neurons. By combining these methods, we obtain a unique dataset that includes detailed functional characterization and transcriptomic identity. Preliminary results show that superficial

GABAergic neurons are selective for trial epoch, activity of neurons within a sub-class is highly correlated, and spiking patterns are correlated with the expression of marker genes for specific transcriptomic types.
References1. Tasic, B., Yao, Z., Graybuck, L.T.*etal*.Shared and distinct transcriptomic cell types acrossneocortical areas.*Nature* 563, 72-78(2018). 2. Guo, Zengcai V., et al. "Maintenance of persistent activity inafrontal thalamocortical loop."*Nature* 545.7653(20 17): 181-186.3. 3. Inagaki, HidehikoK., et al. "A midbrain-thalamus-cortex circuit reorganizes cortical dynamics toinitiate planned movement."*bioRxiv* (2020).4. Abdelfattah, Ahmed S., et al. "Bright and photostable chemigenetic indicators for extended invivovoltage imaging."*Science* 365.6454(2019): 699-704.

Disclosures: A. Singh: None. **T. Wang:** None. **A. Abdelfattah:** None. **E. Schreiter:** None. **K. Svoboda:** None.

Poster

P940. Electrophysiology Methods: Electrode Arrays II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P940.01

Topic: I.04. Physiological Methods

Support: ONR N00014161253
NSF ECCS-2024776
NSF ECCS-1752241
NSF ECCS-1734940
NIH R21 EY029466
NIH R21 EB026180
NIH DP2 EB030992

Title: Virtual imaging of cortex-wide brain activity through decoding from locally recorded neural potentials

Authors: *X. LIU, C. REN, M. WILSON, J.-H. KIM, Y. LU, M. RAMEZANI, T. KOMIYAMA, D. KUZUM;
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Abstract: In both basic neuroscience research and clinical applications, electrical recordings of neural activity from brain surface have been widely employed to investigate neural circuit functions and treat neurological disorders. Traditionally, these surface potentials have been believed to mainly reflect local neural activity. Little is known about how informative they could be for neural activities across multiple cortical regions. To investigate that, we performed simultaneous local electrical recording and wide-field calcium imaging in awake head-fixed

mice. By developing a recurrent neural network model, we successfully decoded the mean activity of different cortical regions from locally recorded surface potentials. We also investigated the predictive power of multiple frequency bands of surface potentials and found that all the frequency bands were informative for large-scale brain activity. Despite the close spacing between adjacent recording channels, the recorded surface potentials provided complementary information about the large-scale cortical activity. Finally, we demonstrated the successful virtual imaging of whole dorsal cortex activity at pixel-level using locally recorded surface potentials. Therefore, in contrast with the prevailing view, we showed that the locally recorded neural potentials indeed contain rich information of the large-scale neural activities, which could be recovered using a neural network model.

Disclosures: X. Liu: None. **C. Ren:** None. **M. Wilson:** None. **J. Kim:** None. **Y. Lu:** None. **M. Ramezani:** None. **T. Komiyama:** None. **D. Kuzum:** None.

Poster

P940. Electrophysiology Methods: Electrode Arrays II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P940.02

Topic: I.04. Physiological Methods

Support: ONR N00014161253
NSF ECCS-2024776
NSF ECCS-1752241
NSF ECCS-1734940
NIH R21 EY029466
NIH R21 EB026180
NIH DP2 EB030992

Title: Ultra-small, fully-transparent, high-density graphene microelectrode arrays for multimodal experiments combining electrophysiology with optical imaging

Authors: *M. N. WILSON, J.-H. KIM, X. LIU, M. RAMEZANI, C. REN, C. DE-EKNAMKUL, T. KOMIYAMA, D. KUZUM;
Univ. of California San Diego, La Jolla, CA

Abstract: Optical technologies such as multi-photon microscopy and optogenetics have transformed basic neuroscience research by allowing high resolution recording and manipulation of neuronal activity. Combining optical techniques with electrophysiological recordings is of critical importance to harness high temporal resolution of electrical recordings along with the spatial resolution and cell specificity of optical techniques. Transparent microelectrode technologies were developed to integrate electrical recordings with optical methods. However,

previous demonstrations of transparent electrodes have been limited to small densities (16-electrodes) and large microelectrode dimensions (~100 μm) due to the pinhole defects in graphene wires and high quantum capacitance of monolayer graphene, respectively. Here, we overcome both limitations and present a new transparent graphene electrode technology based on double layer graphene with interlayer doping. We demonstrate that double layer graphene with interlayer doping achieves low sheet resistance and free of pinhole defects allowing patterning of thin and long transparent graphene wires to build high density arrays. That also eliminates the need for patterning metal wires surrounding transparent arrays, which are not desirable for light induced artifacts. Using this new technology, we have successfully overcome quantum capacitance using Pt nanoparticles and demonstrated fully-transparent 64-channel low impedance graphene arrays with electrodes scaled down to 20 μm, important for recording units and local high frequency activity. We performed multimodal experiments combining simultaneous electrical recordings and two-photon calcium imaging from visual cortex in transgenic mice. Our results show that ultra-small fully transparent graphene arrays can record neural activity with high signal-to-noise ratio during optical imaging. By performing two-photon imaging at two different depths, we investigated the relationship between the surface potentials and the underlying neural activity. Our high density fully transparent graphene electrode technology facilitates a more comprehensive investigation of cortical dynamics across different spatiotemporal scales.

Disclosures: M.N. Wilson: None. **J. Kim:** None. **X. Liu:** None. **M. Ramezani:** None. **C. Ren:** None. **C. De-Eknamkul:** None. **T. Komiyama:** None. **D. Kuzum:** None.

Poster

P940. Electrophysiology Methods: Electrode Arrays II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P940.03

Topic: I.04. Physiological Methods

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NSF ECCS-2024776
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NSF ECCS-1734940
NIH R21 EY029466
NIH R21 EB026180
NIH DP2 EB030992

Title: Flexible, insertable, and transparent microelectrodes (Neuro-FITM) for investigating cortical-hippocampal interactions

Authors: *J.-H. KIM, X. LIU, C. REN, Y. LU, Y. LIU, S. LEUTGEB, T. KOMIYAMA, D. KUZUM;
UCSD, La Jolla, CA

Abstract: Communications between neocortex and hippocampus are crucial for many cognitive processes. However, due to the difficulty of simultaneous recordings of these regions, the coordinations between large-scale cortical dynamics and hippocampal activity are not well-understood. Here we present a Flexible, Insertable, and Transparent Microelectrode array technology ('Neuro-FITM') to investigate the cortical-hippocampal coordinations during hippocampal sharp-wave ripples (SWRs). By performing simultaneous electrical recordings of local field potentials and neural spiking from hippocampus and wide-field calcium imaging of large-scale cortex, we found that diverse cortical activity patterns accompanied SWRs. In most cases, hippocampal SWRs happened after cortical activations. Our multimodal experiments showed that, during SWRs, hippocampal neural population exhibited selective spiking activity associated with distinct cortical activity patterns. These results suggest that the interaction between hippocampus and large-scale cortical activity is selective and diverse during SWRs. Our technology can be broadly applied to comprehensive investigations of interactions between cortex and other subcortical structures.

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Poster

P940. Electrophysiology Methods: Electrode Arrays II

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Support: NIH NINDS Award #5R0NS111028-03
Brain Initiative Award #R42NS110165
Minnesota Nano Center NSF NNCI Award #ECCS-2025124
NSF IGERT Award #DGE-1069104

Title: Esee-shells: polymer skulls with integrated transparent electrode arrays for cortex-wide opto-electrophysiological recordings

Authors: *P. D. DONALDSON, Z. S. NAVABI, R. E. CARTER, L. GHANBARI, T. J. EBNER, S. B. KODANDARAMAIAH, S. L. SWISHER;
Univ. of Minnesota, Minneapolis, MN

Abstract: Electrophysiological and optical imaging provide complementary neural sensing capabilities - electrophysiological recordings have the highest temporal resolution, while optical imaging allows for recording the activities of genetically defined populations at high spatial resolution. Combining these complementary, yet orthogonal modalities to perform large-scale, multimodal sensing of neural activity across the entire cortex would be very powerful. We have previously developed transparent polymer skulls which allow chronic optical access to over 45 mm^2 of the dorsal cortex of the mouse (Ghanbari et al., *Nature Communications* 2019). Here we show that transparent, reconfigurable, inkjet-printed electrocorticography (ECoG) electrode arrays can be seamlessly integrated into the complex 3-dimensional inner surface of the polymer skulls, resulting in a chronically implantable device (eSee-Shells) which enables multi-modal recordings across the cortex. Ten-channel electrode arrays were inkjet-printed using poly(3,4-ethylenedioxythiophene) polystyrene sulfonate (PEDOT:PSS) and encapsulated with Parylene C. Benchtop characterization indicated electrode impedances of ~20 kOhms in saline at 1 kHz and optical transparency through the electrodes of ~75% at the GFP emission wavelength (509 nm). eSee-Shells were implanted on transgenic mice expressing the calcium indicator GCaMP6f in cortical excitatory cells and provided a robust opto-electrophysiological interface for over 100 days. We have performed simultaneous mesoscale calcium imaging and ECoG recordings under anesthesia as well as in awake animals presented with sensory stimuli. Finally, eSee-Shells implanted on mice sparsely expressing GCaMP6s in a subset of L2/3 excitatory neurons show sufficient clarity and transparency to observe single-cell calcium transients directly below the electrode pads. Additionally, the discrimination of awake somatosensory evoked responses based on simultaneous whisking behavior shows behavioral-state dependent responses in both modalities, and intra- and inter- modality correlation and coherence analyses of spontaneous multi-modal recordings reveal diverse relationships during awake and anesthetized states. This ability to directly study the relationships between electrophysiological signals and optical reporters across large areas of the mouse cortex will ultimately allow more elaborate investigations of neural circuitry, aiding in the development of therapies for a variety of neurological and psychiatric disorders.

Disclosures: P.D. Donaldson: None. **Z.S. Navabi:** None. **R.E. Carter:** None. **L. Ghanbari:** None. **T.J. Ebner:** None. **S.B. Kodandaramaiah:** None. **S.L. Swisher:** None.

Poster

P940. Electrophysiology Methods: Electrode Arrays II

Location: Virtual Only

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Program #/Poster #: P940.05

Topic: I.04. Physiological Methods

Title: Evaluation of Ready-to-Use Quick-Neuron™ Plate - MEA 48

Authors: *T. NAKAYAMA[1], M. SEO[2], T. ARATANI[1], M. AZUMA[1];
[1]Biomed. Business Ctr., RICOH COMPANY, LTD., Kawasaki-city, Japan; [2]Elixirgen Scientific, Inc., Baltimore, MD

Abstract: [Background] The significant difference between animal models and humans is one of major reasons for project closures of drug discovery and development in central nervous system (CNS) field. And, it is also well known that *in vitro*/ *in vivo* animal models are often insufficient to predict drug efficacies and toxicities in human. To overcome this challenge, many assay methods with the human induced pluripotent stem cell (iPSC) derived neurons are being reported. Multielectrode array (MEA) is one of the well-known methods to evaluate nerve functions. However, MEA has some challenges. For example, it's a lot of effort to do the iPSC preparation and maintenance on MEA plate. And, it takes a long time and know-hows to optimize the conditions because it's difficult to stably evaluate drug efficacy and toxicity. To solve these challenges, we have developed the Quick-Neuron™ Plate - MEA48, which is the ready-to-use plate. [Objective] In this study, we evaluated the practicality Quick-Neuron™ Plate - MEA48 in drug discovery projects from the following viewpoints. 1. Long distance transporting, 2. Pharmacological responses, 3. Reaction to existing drugs which have anti-epileptic activity [Results] 1. Transporting test - Quick-Neuron™ Plate - MEA48 which was transported from Japan to the US showed almost the same network bust pattern as before transportation. And, the transported plate also showed sufficient response to 4-AP (K+ channel blocker). 2. Pharmacological responses - Quick-Neuron™ Plate - MEA48 showed the expected neuronal responses to representative neurotransmitters or receptor antagonists. 3. Reaction to existing drugs which have anti-epileptic activity - the anti-epileptic drugs which act on ion channels reduced the number of spontaneous neuronal firing on Quick-Neuron™ Plate - MEA48. [Conclusion] In this study, we showed Quick-Neuron™ Plate-MEA 48 has good transportability and responds to various neurotransmitters, receptor antagonists and anti-epileptic drugs. These results suggested that Quick-Neuron™ Plate-MEA 48 can be used for drug efficacy testing and/ or drug toxicity testing in CNS field such as anti-epilepsy screening or convulsive toxicity.

Disclosures: T. Nakayama: A. Employment/Salary (full or part-time):; RICOH COMPANY, LTD. **M. Seo:** A. Employment/Salary (full or part-time):; Elixrgen Scientific, inc. **T. Aratani:** A. Employment/Salary (full or part-time):; RICOH COMPANY, LTD. **M. Azuma:** A. Employment/Salary (full or part-time):; RICOH COMPANY, LTD..

Poster

P940. Electrophysiology Methods: Electrode Arrays II

Location: Virtual Only

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Program #/Poster #: P940.06

Topic: I.04. Physiological Methods

Support: NIH T-32-NS076401
NIH DA 020140-09

Title: A Novel Approach to Study Gamma-Band Coherence in Ex Vivo brain slices using perforated MEAs

Authors: *J. RODRIGUEZ[1], P. M. JENKINS[2], D. L. PRITCHETT[4], K. S. JONES[3]; [1]Univ. of Michigan, Ypsilanti, MI; [2]Dept. of Pharmacology, Dept. of Psychiatry, [3]Pharmacol., Univ. of Michigan, Ann Arbor, MI; [4]Biol., Howard Univ., Washington, DC

Abstract: Oscillations play crucial roles in many cognitive processes such as memory formation and attention. GABAergic interneurons can synchronize neuronal activity leading to gamma oscillations (30-60 Hz). Abnormalities in oscillatory activity in the hippocampus have been implicated in the pathology of some mental health disorders including schizophrenia and bipolar disorder, however the neurobiological mechanism underlying these abnormal oscillations are not yet fully understood. We set out to develop a reliable approach to study gamma oscillations in *ex vivo* hippocampal preparations using a 60-channel perforated microelectrode arrays (pMEAs). We utilized the pMEAs to record the electrical activity in multiple sites from hippocampal brain sections while allowing for rapid exchange and delivery of pharmacological agents. Bath application of kainate rapidly induced and maintained oscillatory activity in the CA1 and CA3 regions of the hippocampus. Kainate-induced oscillations were quickly abolished by the GABAA receptor antagonist, bicuculline. We demonstrated the usefulness of this approach by characterizing gamma oscillations evoked in hippocampal sections from a transgenic mouse model with Ank3-dependent inhibitory synaptic dysfunction. Sections prepared from mutant mice exhibited an increase in the coherence of gamma power within CA1 despite a reduction in gamma band power. Future studies will focus on determining how different neurotransmitters systems contribute to the induction and maintenance of kainate-induced oscillations in hippocampal sections from wild type and disease relevant animal models.

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Poster

P940. Electrophysiology Methods: Electrode Arrays II

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Program #/Poster #: P940.07

Topic: I.04. Physiological Methods

Support: JST Grant JPMJTR20UP

Title: Detailed electrical activities measurement of neural networks using 236,880 electrodes CMOS-MEA

Authors: N. MATSUDA, X. HAN, M. SHIBATA, S. KIMURA, N. NAGAFUKU, Y. ISHIBASHI, ***I. SUZUKI**;
Tohoku Inst. of Technol., Sendai, Miyagi, Japan

Abstract: Detailed electrical activity measurement of neural networks is essential for understanding brain functions and elucidating neurological disorders. CMOS-MEA is excellent in detecting detailed electrical activity patterns of brain slices and cultured neural networks due to the large number of electrodes. In this study, we investigated the electrical activity characteristics of brain slices, human brain organoids, and cultured neural networks using CMOS-MEA with 236,880 electrodes, which have the highest specifications in the world. In the measurement of brain slices, we succeeded in measuring the interregional propagation of the hippocampus and cerebral cortex in detail, and detected changes in β-wave generation patterns and propagation patterns due to drug administration. In sensory neuron measurement, calculation of axon conduction velocity in single neuron and change in propagation velocity due to drug administration were detected. In human iPS cell-derived central nervous system networks and human cerebral organoids, network activity was detected on a cell-by-cell basis, and changes in propagation patterns due to drug administration were detected. It was found that CMOS-MEA with 236,880 electrodes and a large measurement area can measure the electrical activity characteristics of ex vivo and in vitro neural networks and single neuron in detail. It was suggested that big data with high temporal resolution is effective for elucidation of neural circuit function and drug evaluation based on new neural activity information.

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Poster

P943. Neurodegenerative Diseases: Methods, Tools, and Diagnostics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P943.01

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: U19 AG062418
P01 AG066597 (Imaging Core)
U01 AG068057

Title: Parkinsons disease classification using 3d convolutional neural networks and random forest methods

Authors: *N. J. DHINAGAR[1], S. I. THOMOPOULOS[1], C. OWENS-WALTON[1], D. WEINTRAUB[2], P. COOK[3], C. MCMILLAN[2], P. M. THOMPSON[1];
[1]Imaging Genet. Ctr., USC, Los Angeles, CA; [3]Dept. of Radiology, [2]Univ. of Pennsylvania, Philadelphia, PA

Abstract: Parkinson's disease (PD) is a progressive neurodegenerative disease that affects 10 million people worldwide and over 1 million people in the United States. Structural magnetic resonance imaging (MRI) offers the potential to discover diagnostic and predictive biomarkers of PD; and can also assess cortical and subcortical brain atrophy non-invasively. Here we developed a deep learning model to classify PD based on brain MRI. We analyzed the benchmark Parkinson's Progression Markers Initiative (PPMI) dataset (N = 471; 341 PD/130 healthy age-matched controls; 308M/163F; age 30.6-84.9). We partitioned 70% of the PPMI data for training, 20% for validation and 10% as the in-distribution test set. We also analyzed balanced subsets of independent data from the University of Pennsylvania (UPenn) as the out-of-distribution test dataset (N = 117; 104 PD/13 controls; 75M/42F; age 50-84). The MRI were preprocessed with nonparametric intensity normalization (N4 bias field correction), brain extraction, linear registration to a template and isometric voxel resampling to 2mm. As a baseline approach, we implemented a machine learning (ML) pipeline to extract 93 key radiomics features [1] including textural features based on first-order statistical; co-occurrence matrices which we used to train a random forest-based classifier. Our proposed 3D convolutional neural network (CNN) model uses each 3D T1-MRI volume as input. The model was trained for 50 epochs with early stopping. We used a binary cross-entropy loss, the Adam optimizer with weight decay and dropout. The validation subset was used to optimize the deep neural network's hyperparameters. We also used image rotations for data augmentation. The CNN model achieved an average receiver-operator characteristic curve-area under the curve (ROC-AUC) of 0.606 over multiple runs on the PPMI test set compared to the ML classifier's average ROC-AUC of 0.489. In tests on independent UPenn data, the CNN yielded an average ROC-AUC of 0.583 and the ML classifier yielded only 0.471. The CNN outperformed the ML approach and generalized better to new, unseen MRI from a different center. Training and testing our models on a more balanced dataset, as well as multimodal neuroimaging data, may improve performance. The proposed model could assist in early detection of PD using T1 MRI and help to identify disease-relevant biomarkers. Reference: [1] J. J. M. van Griethuysen, A. Fedorov, et al., "Computational Radiomics System to Decode the Radiographic Phenotype. Cancer Research," 77(21), e104-e107, 2017.

Disclosures: N.J. Dhinagar: None. **S.I. Thomopoulos:** None. **C. Owens-Walton:** None. **D. Weintraub:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Michael J. Fox Foundation, Alzheimer's Therapeutic Research Initiative (ATRI), Alzheimer's Disease Cooperative Study (ADCS), The International Parkinson and Movement Disorder Society (IPMDS), The National Institute on Aging (NIA), The U.S. Department of Veterans Affairs. F. Consulting Fees (e.g., advisory boards); Acadia, Aptinyx, CHDI Foundation, Clintrex LLC (Otsuka), Eisai, Great Lake Neurotechnologies, Janssen, Sage, Scion, Signant Health, Sunovion, Vanda. Other; University of

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Poster

P943. Neurodegenerative Diseases: Methods, Tools, and Diagnostics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P943.02

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: PRC Toth Family Fellowship

Title: Neurophysiological correlates of countermanding saccade deficits in Parkinson's disease

Authors: *M. LEUNG[1], A. J. SACHS[2];
[2]Neurosurg., [1]Ottawa Hosp. Res. Inst., Ottawa, ON, Canada

Abstract: Eye movements are convenient indicators of basal ganglia (BG) function and eye movement abnormalities are common in Parkinson’s Disease (PD). The oculomotor system is an appealing model motor system because saccades are ballistic and stereotyped (each saccade is associated with a unique set of muscle contractions) allowing better correlation of movement and movement preparation with neural signals, comparison across subjects, and more trials in a limited amount of experimental time. The cortical and subcortical structures involved in the saccadic system require tight regulation in the BG-thalamocortical loop, and this regulation degenerates gradually in PD due to decreased dopaminergic innervation from the substantia nigra pars compacta (SNc). The saccadic system can be evaluated through tasks completed with prosaccades (saccades toward a target), antisaccades (saccades opposite to a cued target), and cognitive tasks involving response inhibition (stop-signal task). We developed a stop-signal task in Virtual Reality (VR) that probes the subject’s ability to recruit the neural processes involved in action selection and response inhibition, and have verified that we could identify key signature changes in the EEG profile during left and right saccade, countermand, and antisaccades in healthy subjects. Independent Component Analysis (ICA) provided spatial filters for determining source signals involved in response inhibition, and subsequently utilizing the fitting-oscillations-one-over-f (FOOOF) algorithm on these sources yielded subject-specific oscillatory ranges. These ranges were applied in the construction of subject-specific machine learning (ML) models

for differentiating between task conditions, such as saccade types, by biasing the model with a favored range that was unique between subjects. The average F1 score for all subjects from their respective ML pipelines was 0.92 when evaluated against testing data. This pipeline has been approved for use in the clinic on PD patients, towards the process of elucidating neurophysiological correlates of PD and developing an adaptive deep brain stimulation (aDBS) solution.

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Poster

P943. Neurodegenerative Diseases: Methods, Tools, and Diagnostics

Location: Virtual Only

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Program #/Poster #: P943.03

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: MOST 110-2321-B-001-012

Title: Predicting and suppressing non-stationary, pathological neural oscillation in Parkinsonian rats with high efficacy by an adaptive, wireless neuromodulator

Authors: *H. CHEN[1], B.-H. CHANG[1], C.-H. CHIEN[3], H.-H. HO[3], Y.-T. HUANG[1], R. PERUMAL[1], S.-R. YEH[2];
[1]Electrical Engin., [2]Life Sci., Natl. Tsing Hua Univ., HsinChu, Taiwan; [3]BioPro Scientific, HsinChu, Taiwan

Abstract: The high-voltage spindle (HVS) is a spike-and-wave, neural oscillation that has been identified as a pathological signature related to the dopamine depletion in Parkinsonian rats. It is of great interests to investigate whether suppressing HVSs helps to improve the motor deficits associated with the HVS, or even to delay the progression of the Parkinson's disease. As each HVS episode occurs randomly and lasts for only a few seconds, an adaptive neuromodulator is demanded for predicting the onset of HVSs and suppressing the HVSs by deep-brain stimulation at precise timing. This presentation will introduce an adaptive neuromodulator consisting of a lightweight(~4g) neuro-interfacing headstage and a wireless data hub. The proposed algorithm for detecting the HVS is based on the autoregressive modelling at interval (ARt), whose parameters are adaptable on-line by a Kalman-filter framework. In our pilot study, the algorithm is proved able to predict the onset of HVS with high precision (96%) and short latency (61ms). This study further implements the algorithm in the wireless neuromodulator for experiments with freely-moving Parkinsonian rats. The ARt model is realized in the microcontroller embedded in the headstage to predict the onset of HVS in real time, and stimulation on the subthalamic nucleus is triggered automatically whenever an HVS onset is alarmed. In parallel, the recorded neural activity is transmitted wirelessly to the data hub, wherein the Kalman-filter framework is

implemented to re-estimate optimal model parameters for detecting the HVS reliably. The re-estimated parameters are then uploaded to the headstage regularly, so as to adapt the ARt model in response to the uncertain changes of neural activities from one day to another. Such machine-learning capability is also crucial for applying the ARt model to detecting HVSs in different brain regions or different rats. On the other hand, the wireless, lightweight headstage facilitates the investigation on whether the motor deficits associated with HVSs are ameliorated at the same time. The adaptive neuromodulator is proved able to suppress HVSs with high efficacy. The efficacy refers to the finding that stimulation duration as short as 0.2s is already sufficient for suppressing the HVS effectively, even though the mean duration of HVS episodes is 4.3s. This intriguing finding indicates that precise control on the timing of brain stimulation is crucial for increasing stimulation efficacy while reducing stimulation dosage and thus its side-effects. Moreover, the proposed algorithm and adaptive neuromodulator is applicable to detecting different pathological signatures of other neural disorders.

Disclosures: H. Chen: None. **B. Chang:** None. **C. Chien:** A. Employment/Salary (full or part-time):; BioPro Scientific. **H. Ho:** A. Employment/Salary (full or part-time):; BioPro Scientific. **Y. Huang:** None. **R. Perumal:** None. **S. Yeh:** None.

Poster

P943. Neurodegenerative Diseases: Methods, Tools, and Diagnostics

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Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

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NIH TL1 TR002016
PA Department of Health Tobacco CURE Funds
Michael J. Fox Foundation for Parkinson's Research

Title: Magnetic resonance imaging and cell density correlates in the substantia nigra pars compacta of parkinsonism patients

Authors: *M. L. JOHNSON, E. W. WANG, G. L. BROWN, M. M. LEWIS, G. DU, L. C. JELLEN, C. PU, H. CHEN, L. KONG, X. HUANG;
Penn State Hlth., Hershey, PA

Abstract: Background: Previous studies have shown that quantitative MRI metrics are sensitive to microstructure and susceptibility changes in the SNc of parkinsonism patients. These are

mainly lower SNc fractional anisotropy (FA), higher apparent transverse relaxation rate (R2*), and higher quantitative susceptibility mapping (QSM). The exact pathologies underlying these signals, however, remain unclear.Methods: This study included 33 parkinsonism patients and three controls with both *in vivo* MRI scans and post-mortem neuropathologic assessment. SNc diffusion metrics of FA, mean diffusivity (MD), axial diffusivity (AD), radial diffusivity (RD), and free water fraction (FW) were determined, as were the susceptibility metrics R2* and QSM. Archival hematoxylin and eosin-stained midbrain slides were whole-slide scanned at 40× magnification. Slide scans were color- and intensity-normalized. A blinded rater then semi-automatically quantified neuromelanin-positive (NM+) neuron and glial cell densities in the SNc using ImageJ. Associations between MRI and cell densities were examined using semi-partial Pearson correlations where cell densities were regressed on the MRI-pathology interval.Results: SNc FA associated positively with NM+ neuron density residuals ($r = 0.35$, $df = 31$, $P = 0.047$). SNc R2* and QSM each associated positively with glial cell density residuals ($r = 0.71$, $df = 24$, $P < 0.0001$ and $r = 0.70$; $df = 24$; $P < 0.0001$, respectively). The FA-NM+ neuron density and R2*/QSM-glial cell density associations were not improved by additional adjustments for glial cell density and NM+ neuron density, respectively. AD, MD, RD, and FW did not associate with either cell density metric.Conclusion: This is the largest parkinsonism study to date to examine *in vivo* MRI associations with post-mortem neuropathology. The results suggest that lower FA may reflect dopaminergic neuron loss in the SNc, and higher MRI susceptibility reflects iron accumulation distributed to glial cells. Both histologic associations confirm that MRI is sensitive to key changes in the SNc associated with neurodegeneration, and validate its clinical and translational value.

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Poster

P943. Neurodegenerative Diseases: Methods, Tools, and Diagnostics

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Program #/Poster #: P943.05

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: NIH NIA Grant U54 AG065187

Title: The Emory-Sage-SGC TREAT-AD Center: Catalyzing Diversity in Alzheimer's Disease Target Discovery Through Open Science and Target Enablement Packages

Authors: *K. LEAL[1], L. M. MANGRAVITE[1], R. BETARBET[2], G. W. CARTER[3], S. V. FRYE[4], H. FU[2], A. K. GREENWOOD[1], F. M. LONGO[5], L. OMBERG[1], K. H. PEARCE[4], A. EDWARDS[6], A. I. LEVEY[7];

[1]Sage Bionetworks, Seattle, WA; [2]Emory Univ., Atlanta, GA; [3]The Jackson Lab., Bar Harbor, ME; [4]Univ. of North Carolina, Chapel Hill, Chapel Hill, NC; [5]Dept. of Neurol. and Neurolog. Sci., Stanford Univ. Med. Ctr., Stanford, CA; [6]Univ. of Toronto, Toronto, ON, Canada; [7]Dept Neurol., Emory Univ. Sch. of Med., Atlanta, GA

Abstract: The Alzheimer's disease (AD) drug development pipeline has been marked by repeated failures and a lack of portfolio diversity. With the shadow of AD on our society growing longer, it is becoming evident that clinical care for treatment and prevention of AD will be possible only when we have multiple drugs available that operate across multiple disease-causing biological pathways in order to effectively reduce the burden of AD. The Emory-Sage-SGC TREAT-AD Center was established with the aim of developing reagents for a diverse portfolio of potential AD target hypotheses and to use open science practices to speed up drug development for new therapies. The Emory-Sage-SGC TREAT-AD Center has identified a diverse set of novel AD targets derived from systems biology studies with in the Accelerating Medicines Partnership in AD (AMP-AD) consortium and further evaluated using data from multiple AD consortia and existing literature. Targets nominated through the AMP-AD consortium were mapped to mechanistic hypotheses and prioritized based on unbiased bioinformatic assessments across multiple lines of evidence. By using a systems biology and bioinformatic analyses approach, we can search for multiple ideal targets to perturb for potential AD drug development versus one target at a time. To accelerate this process, the Center is generating and openly distributing experimental tools for use in target validation to advance or reject therapeutic hypotheses. Distribution of all data and Target Enabling Packages that include expression constructs, knockout cell lines, assays, antibody validation data, and crystal structures for 20 proteins per year identified within these mechanisms will be publicly shared through standard repositories and cataloged on the ADKnowledge Portal (https://adknowledgeportal.org). The open drug discovery approach of TREAT-AD (https://treatad.org/) is aimed to de-risk potential AD therapeutics by catalyzing robust and independent evaluation of a diverse portfolio of promising yet untested AD therapeutic hypotheses.

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Poster

P943. Neurodegenerative Diseases: Methods, Tools, and Diagnostics

Location: Virtual Only

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Program #/Poster #: P943.06

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: Acute localization of maximum beta power within the subthalamic nucleus using clinic local field potential recordings reveals minimal variability in the peak frequenc

Authors: *S. LEWIS[1], L. HIRT[2], M. CASE[4], C. PULLIAM[4], J. EUBANKS[5], S. GOETZ[5], R. S. RAIKE[5], S. OJEMANN[3], D. KRAMER[3], D. KERN[3,2], J. A. THOMPSON[3,2];
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Abstract: Neuromodulation via deep brain stimulation (DBS) is a highly effective therapy for movement disorders such as Parkinson's disease (PD). A recent technological advance, *in vivo* local field potential (LFP) recording from the implanted DBS electrode using the clinical pulse generator, permits investigation of neurophysiological biomarkers of PD. These biomarkers are relevant to optimizing stimulation parameters and tailoring stimulation to address specific symptom profiles. In this study, we focused on assessing a common pathophysiological biomarker, increased beta band power, in the context of optimizing the anatomic localization and efficiency of stimulation parameters during a standard therapeutic monopolar review. In n = 3 patients diagnosed with PD, implanted with bilateral DBS targeting the subthalamic nucleus (STN), we conducted 3 differential electrode measurements (30-60 second recording, 2-5-minute interval between measurements) to quantify the relative beta power (13 - 30 Hz) along the linear array of electrode contacts of both hemispheres. Following the identification of the bipolar electrode pair with the highest beta power, we compared the mean and standard deviation of the 3 repeated recordings, across hemisphere, to assess the consistency. We observed that for this pilot cohort of patients with PD, across all subjects, significant variability was observed in 5/18 contact pair comparisons, with hemispheres in 2 subjects exhibiting no difference ($p < 0.05$; using a Kolmogorov-Smirnov test to compare distributions [P1: (L) 1/3 (R) 1/3, P2: (L) 0/3 (R) 1/3, P3: (L) 0/3, (R) 2/3)]. Finally, we noted a significant asymmetry in the presence of beta power magnitude between the hemispheres ($F_{1,15478} = 1536$, $p < 0.001$). We found that *in vivo* assessment of a pathophysiological biomarker (peak beta power to determine the therapeutic contact) predicated on LFP information recorded from a DBS electrode (up to 5 years post-surgery) may inform therapeutic stimulation parameters.

Disclosures: S. Lewis: None. **L. Hirt:** None. **M. Case:** None. **C. Pulliam:** None. **J. Eubanks:** None. **S. Goetz:** None. **R.S. Raike:** None. **S. Ojemann:** None. **D. Kramer:** None. **D. Kern:** None. **J.A. Thompson:** None.

Poster

P943. Neurodegenerative Diseases: Methods, Tools, and Diagnostics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P943.07

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: Boettcher Collaboration Grant

Title: Automatic extraction of kinematic motifs captured with markerless tracking during deep brain stimulation implantation

Authors: *S. BAKER[1], S. OJEMANN[2], D. KRAMER[2], D. S. KERN[3], J. A. THOMPSON[3]; [1]Colorado Col., Colorado Springs, CO; [2]Dept. of Neurol., [3]Dept. of Neurol. & Neurosurg., Univ. of Colorado Sch. of Med., Aurora, CO

Abstract: Optimal placement of deep brain stimulation (DBS) electrodes for the treatment of movements disorders, including Parkinson's disease (PD), can involve intraoperative assessment of neural activity during upper limb movements. Conventional motor mapping procedures during DBS surgery have remained largely unchanged. Given advancements in machine learning, we pursued a novel approach to objective quantification of movement over time using "markerless tracking" during the surgery. Patients (n=9) underwent DBS implantation, targeted to the subthalamic nucleus (STN), as treatment for PD. Two cameras were oriented (one facing the patient at the end of the operating table and the second contralateral to the limb under investigation) to record kinesthetic testing evaluated by a movement disorders neurologist during various active and passive upper limb movements. After intraoperative video data collection, 21 labels were applied to patients' digits and palm on a subset of frames from each recording (700-912 frames per surgery, 1.52-5% of total frame count) and trained using the Python-based suite DeepLabCut. Training was realized with the *resnet-50* convolutional neural network (pretrained to human behavior) until a Huber loss plateau ($\mu = 0.022$), equating to a mean iteration count of 224,250. Refinement of additional parameters permitted acceptable accuracy in tracking, and suboptimal pose estimations were discarded. Post-hoc data analyses used Matlab to extract kinematic data, including Euclidean distance (ED), acceleration, and velocity. From these metrics, we defined an objective movement threshold, and used extracted ED epochs that represented specific patient movements such as hand clenches and raised arm postures to begin development of a movement categorization pipeline. We assessed exemplar sets of ED epochs from three behaviors to fit the polynomial $-a(x^2 - c) + b$. Each behavioral cluster's three coefficients were significantly different from one another, as suggested by independent-samples t-tests (all $p<0.001$, two-tailed) with Dunn's indices among pairwise coefficients ranging from DI: 0.059-0.3889. These preliminary results suggest accurate and precise capture of neuromotor behavior during surgical interventions for PD as well as development at objective behavioral categorization, which may be used improve surgical targeting, optimize DBS therapy, or develop novel DBS targets.

Disclosures: S. Baker: None. **S. Ojemann:** None. **D. Kramer:** None. **D.S. Kern:** None. **J.A. Thompson:** None.

Poster

P943. Neurodegenerative Diseases: Methods, Tools, and Diagnostics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P943.08

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: Microglial imaging with MRI diffusion radiomics

Authors: *A. P. SINGH, V. S. JAIN, J.-P. J. YU;
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Abstract: MR imaging with neurite orientation dispersion and density imaging (NODDI) has provided new opportunities to interrogate the composition of the axonal and extra-axonal compartments of the brain. Previous work from our group has shown the high sensitivity, but low specificity, of the extra-axonal NODDI parameter orientation dispersion index (ODI) to microglial density. We demonstrate the first application of radiomics in NODDI to track changes in microglial density using a well-characterized murine model of colony stimulating factor 1 receptor (CSF1R) inhibition resulting in microglial depletion throughout the brain. 12-week-old C57BL/6J male mice depleted of microglia with PLX5622 (a CSF1R small molecule inhibitor) were sacrificed 1- and 7-days following withdrawal of treatment (n=24; 12 control, 12 experimental). Following high-resolution, multi-shell *ex-vivo* diffusion imaging, all images underwent standard image preprocessing and spatial normalization. Images were then masked and segmented with a standard atlas. ODI was calculated for the right hippocampus (HC), amygdala (Am), and globus pallidus (GP). Regions of interest (ROI) were processed using the python library *PyRadiomics* (bin width=0.1) to generate original and wavelet-transformed first-order statistics and image texture matrices for a total of 792 calculated features per ROI. Student's *t*-tests were performed to compare the mean value of features across conditions and FDR-corrected to determine significantly different features. Statistical analyses were performed using python *scipy* and *statsmodels* libraries. Significant differences in radiomic features of ODI were found in all analyzed ROIs between day 1 treated (microglial depleted animals) and control animals (# of significant differences: HC = 102, Am = 160, GP = 21; FDR p-value < 0.05). No significant differences were found in any of the ROIs at the day 7 timepoint. These findings correlate with histologically confirmed changes in microglial density at the examined timepoints. Previous mean ODI intensity comparisons did not correspond with the known biology, with no significant difference in right HC or GP at day 1 (2/3 ROIs incorrect) and significant difference in right GP at day 7 (1/3 ROIs incorrect). Radiomic features generated from the NODDI model are sensitive to changes in microglial density. Texture matrices and other first-order statistics provide greater imaging specificity corresponding to underlying biological changes than mean intensity alone. Our results highlight the increased specificity and potential utility of radiomic analyses to capture important cellular changes in healthy and diseased brains.

Disclosures: A.P. Singh: None. **V.S. Jain:** None. **J.J. Yu:** None.

Poster

P943. Neurodegenerative Diseases: Methods, Tools, and Diagnostics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:00 AM - 10:00 AM

Program #/Poster #: P943.09

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Support: Craig H Neilsen Foundation 651037

Title: Evaluation of axonal injury with filtered diffusion weighted magnetic resonance imaging in cervical spondylotic myelopathy

Authors: *S. MURPHY, R. FURGER, S. SHABANI, S. KURPAD, M. BUDDE;
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Abstract: Diffusion tensor imaging (DTI) has gained popularity in studying microstructural changes in the spinal cord of patients with degenerative cervical spondylotic myelopathy (CSM). While DTI is sensitive to microscopic pathologies in the injured spinal cord, it lacks specificity because many different pathologies can cause DTI changes. CSM is characterized by a complex pathophysiology. Axonal injury is believed to be a main correlate to function, but it is accompanied by substantial edema or myelomalacia. DTI is unable to distinguish these features. Recently, filtered Diffusion Weighted Imaging (fDWI) has been demonstrated to have greater specificity for axonal injury by using one diffusion gradient pair to suppress or 'filter' non-axonal signals and a second to explore the axonal-specific diffusion changes. In this study, we evaluate the sensitivity of fDWI compared to DTI in studying axonal injury in patients with CSM. We hypothesis that fDWI will have greater sensitivity to axonal damage compared to DTI for patients with CSM. All MRI diffusion weighted images (DWI) were obtained on a GE 3T MRI for 22 CSM patients prior to surgery. Diffusion tensor imaging was obtained using 25 directions and a b-value of 800 s/mm^2. Filtered diffusion imaging employed two pairs of diffusion weighted pulses using 26 directions and a b-value of 2000 s/mm^2 tailored specifically for the spinal cord. The change in axial diffusion from above the site of greatest compression to the site of greatest compression was compared between fDWI ($fADC_{||}$) and DTI (AD) to investigate the impact of edema and other artifacts on each acquisition. $fADC_{||}$ showed a greater reduction (-38.13±24.82) at the level of maximal compression compared to AD (-6.67±14.47). In addition, fDWI showed a significantly greater decrease in the axial diffusion for patients with evidence of edema (-50.97±25.60%) compared to patients without edema (-29.18±21.02%) ($p = 0.044$). These findings were not observed using AD derived from DTI which showed no change with (-6.06±1.38%) and without (-7.10±16.89%) the presence of edema ($p = 0.889$). The results showed that DTI and fDWI exhibit divergent features in CSM patients. The decrease in $fADC_{||}$ at the site of maximal compression with and without edema is suggestive of axonal injury and highlights the ability of fDWI to capture more specific information regarding the status of the chronically compressed spinal cord. These trends illustrate the translational potential of using fDWI and may prove useful for improved prognostication in larger studies of CSM.

Disclosures: S. Murphy: None. **R. Furger:** None. **S. Shabani:** None. **S. Kurpad:** None. **M. Budde:** None.

Poster

P016. Stem Cell Differentiation and Reprogramming

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P016.01

Topic: A.03. Stem Cells and Reprogramming

Support: Craafordska Stiftelsen
The Royal Physiographic Society of Lund
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The Swedish Research Council

Title: The human bone marrow harbors a CD45−CD11B+ cell progenitor permitting rapid microglia-like cell derivative approaches

Authors: A. BRUZELIUS[1], I. HIDALGO[1], A. BOZA-SERRANO[1], A.-G. HJELMÉR[1], A. TISON[1], T. DEIERBORG[2], J. BENGZON[1], ***T. RAMOS-MORENO**[1];
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Abstract: Microglia, the immune sentinel of the central nervous system (CNS), are generated from yolk sac erythromyeloid progenitors that populate the developing CNS. Interestingly, a specific type of bone marrow-derived monocyte is able to express a yolk sac microglial signature and populate CNS in disease. Here we have examined human bone marrow (hBM) in an attempt to identify novel cell sources for generating microglia-like cells to use in cell-based therapies and in vitro modeling. We demonstrate that hBM stroma harbors a progenitor cell that we name stromal microglial progenitor (STR-MP). STR-MP single-cell gene analysis revealed the expression of the consensus genetic microglial signature and microglial-specific genes present in development and CNS pathologies. STR-MPs can be expanded and generate microglia-like cells in vitro, which we name stromal microglia (STRM). STR-M cells show phagocytic ability, classically activate, and survive and phagocyte in human brain tissue. Thus, our results reveal that hBM harbors a source of microglia-like precursors that can be used in patient-centered fast derivative approaches.

Disclosures: A. Bruzelius: None. **I. Hidalgo:** None. **A. Boza-Serrano:** None. **A. Hjelmér:** None. **A. Tison:** None. **T. Deierborg:** None. **J. Bengzon:** None. **T. Ramos-Moreno:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); patent holder.

Poster

P016. Stem Cell Differentiation and Reprogramming

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P016.02

Topic: A.03. Stem Cells and Reprogramming

Support: NIH R01 HL141326-03
NIH T32 CA009171-44

Title: Exploring transcriptional dynamics of neural differentiation in an induced pluripotent stem cell model.

Authors: *A. ZUCCO[1], Y. ZHANG[1], C. HILL[2], S. DELIARD[1], A. GARDINI[1];
[1]The Wistar Inst., Philadelphia, PA; [2]Univ. of Pennsylvania, Philadelphia, PA

Abstract: The formation of complex tissue and organ systems through development requires tightly regulated programs of coordinated gene expression. These patterns of gene expression are mediated by a network of different genomic elements, including enhancer regions and other regulatory elements distal to promoters and the gene body itself. Many of these distal elements are poorly characterized, and the role of different regulatory factors at these regions remains to be described. To better describe this epigenetic landscape during differentiation we utilize human induced pluripotent stem cells (hiPSCs) to best recapitulate neuronal stem cell development in-vitro. Coupled with next generation whole genome sequencing we have built snapshots of enhancer activation across the timeline of differentiation from pluripotency to hiPSC-derived neuronal cells. Here we show distinct shifts in both chromatin accessibility and activation as differentiation proceeds, and that these shifts predict activity at nearby genes in a checkpoint specific manner through differentiation. We further show that alterations in the transcriptional regulatory complex Integrator resulted in abnormal neuronal differentiation and decreased activation of enhancer elements, suggesting that Integrator may play a larger role in regulating differentiation than previously recognized. Our data reveal for the first time how subunits of the Integrator complex modulate activation of a neuronal-specific enhancer.

Disclosures: A. Zucco: None. **Y. Zhang:** None. **C. Hill:** None. **S. Deliard:** None. **A. Gardini:** None.

Poster

P016. Stem Cell Differentiation and Reprogramming

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P016.03

Topic: A.03. Stem Cells and Reprogramming

Title: Relationship between neural activity and differentiation in Hydra revealed through 3D functional imaging

Authors: *A. ESCOBAR[1], A. S. PRIMACK[2], C. JULIANO[2], J. T. ROBINSON[1];
[1]Rice Univ., Houston, TX; [2]Univ. of California, Davis, CA

Abstract: Hydra is an emergent model organism in neuroscience with key advantages to study regeneration, such as a simple body plan, limited number of cell types and the ability to regenerate large portions of the body. However, little is known about the relative timing of neural differentiation and recovery of neural circuit function during regeneration. Here we developed a dual-expression transgenic Hydra line that expresses a cell-type-specific red fluorescent protein (tdTomato) in ec5 neurons that are present in the foot of the animal, and a calcium indicator (GCaMP) in all neurons. Using SCAPE (Swept Confocally Aligned Planar Excitation) microscopy we were able to track single cell calcium dynamics in 3D during regeneration of the foot. We found that between 4 and 30 hours post amputation (hpa) Hydra shows unsynchronized spontaneous neural activity along with the absence of tdTomato-positive cells in the peduncle. Between 32 and 40 hpa we observe an emergence of tdTomato-positive cells with low level of synchrony followed by an increase in the number of tdTomato-positive cells and significant increase in synchronization. This synchronous activity is the hallmark of a fully regenerated neural circuit in the Hydra foot. These data suggest that during Hydra regeneration, certain neuronal cells make terminal fate decisions prior to re-establishing their functional role in the nervous system. Thus, in Hydra there are likely additional molecular mechanisms that connect molecular differentiation to functional neural activity after cells are committed to a specific fate. This ability to reproducibly study functional neural regeneration at specific times relative to molecular differentiation events makes Hydra a potentially powerful model system for understanding the key molecular and functional processes involved in neuroregeneration following injury.

Disclosures: A. Escobar: None. **A.S. Primack:** None. **C. Juliano:** None. **J.T. Robinson:** None.

Poster

P016. Stem Cell Differentiation and Reprogramming

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Program #/Poster #: P016.04

Topic: A.03. Stem Cells and Reprogramming

Support: Natalya Krutovska was supported by BP ENDURE at Hunter College/NYU NIH Grant R25 NS 80686
NINDS R01NS100897

Title: Understanding Hox gene regulation in response to Wnt signaling during motor neuron differentiation

Authors: *N. KRUTOVSKA[1], B. RAGIPANI[2], D. P. RAHE[2], E. O. MAZZONI[2];
[1]Hunter College, CUNY, New York, NY; [2]Dept. of Biol., New York Univ., New York, NY

Abstract: Abstract: Understanding patterning and neuronal subtype diversity in the developing neuraxis is at the crux of understanding neuronal diversity. The vertebrate Hox gene family assigns a positional identity to differentiating neurons along the spinal cord to differentiate into anatomically appropriate subtypes. Hox genes are organized in compact clusters surrounded by distal regulatory elements. During development, retinoic acid (RA) and Wnt signaling activate rostral and caudal Hox genes, respectively. Preliminary data demonstrate that regulatory elements within the Hox cluster are sufficient to induce Hox gene expression in response to RA signaling. However, it is not known if the Hox cluster without long-distance regulatory elements can respond to Wnt signaling as well. Differentiating mouse embryonic stem cells (mESCs) respond to RA by activating rostral Hox genes. Thus, we first aimed to establish a differentiation protocol that will rely on Wnt signaling to activate caudal Hox genes. The cells were treated with different concentrations of the Wnt signaling activator CHIR 99021, a GSK3β inhibitor. Additionally, we treated the cultures with a Sonic Hedgehog Agonist (SAG) to induce ventral spinal cord fate. This approach allowed us to develop a homogenous cell culture model and identify the proper dosage amount needed to activate the posterior hox region and grow neuronal cells. Next, we tested if a transposed minimal Hox cluster, devoid of any distal regulatory elements, would respond to Wnt signaling. With the dosages of CHIR 99021 and SAG needed to activate the endogenous posterior hox genes, we can also confirm that a minimal Hox cluster could activate caudal Hox gene expression responding to Wnt signaling. These results suggest the intrinsic ability for the HoxA cluster to decode positional information in the absence of enhancers or other long-range information outside the cluster. This study provides insights on the mechanisms of posterior Hox induction through Wnt activation in regulating transcription and patterning signals for neuronal differentiation.

Disclosures: N. Krutovska: None. **B. Ragipani:** None. **D.P. Rahe:** None. **E.O. Mazzoni:** None.

Poster

P016. Stem Cell Differentiation and Reprogramming

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Program #/Poster #: P016.06

Topic: A.03. Stem Cells and Reprogramming

Support: Kuwait University OMICS # SRUL02/13

Title: Ferulic Acid Enhance Neuronal Differentiation In Hippocampal Stem Cells Derived Neurospheres

Authors: *M. S. RAO, S. SHIVANANDAN;
Fac. of Medicine, Kuwait Univ., Jabriya, Kuwait

Abstract: ***Introduction and objectives:*** Asafoetida is an oleo-gum-resin obtained from the exudates of the roots of the plant *Ferula asafoetida*. Ferulic acid (FA, 4-hydroxy-3-methoxycinnamic acid) is the active component in asafetida. FA has been shown to be neuro stimulant, enhance neuronal stem cells proliferation in the hippocampus. Objective of the present experiment was to investigate the effects of FA on neuronal differentiation of progenitor cells in the hippocampal stem cells derived neurospheres. **Methods:** Stem cell culture was made from embryonic day18 hippocampal cells. Neurospheres were grown for one week and were divided into control (C), and three ferulic acid treatment groups (FA-5µM, FA-10µM and FA-15µM), Control neurospheres remained without any further treatment, FA-5µM, FA-10µM and FA-15µM groups were treated with 5µM,10µM and 15µM ferulic acid respectively for one week. Triplicate cultures were maintained for each group. After treatment, the neurospheres were grown in differentiation media for one week. Cultures were terminated, and immunostaied for TuJ-1, (Neuronal marker), GFAP (astrocytic marker) to label the neurons and astrocytes. Number of neurons and astrocytes differentiated in each neurosphere were quantified. Data were expressed as mean±SEM analyzed with one-way ANOVA. **Results:** Number of differentiated neurons from the neurosphere that are treated with 10µM and 15µM ferulic acid are found to be significantly (30-40%, $p<0.01$) more compared to control group. 5µM ferulic acid had no effect on neuronal differentiation. Number of differentiated astrocytes from the neurosphere that are treated with 10µM and 15µM ferulic acid are also found to be marginally increased (10-20%, $p<0.05$) compared to control group. 5µM ferulic acid had no effect on astrocyte differentiation. **Conclusion:** These results suggest neuron and astrocytes stimulatory role of ferulic acid in neurosphere. It facilitates the differentiation of neurons and astrocytes in the neurosphere.

Disclosures: M.S. Rao: None. **S. Shivanandan:** None.

Poster

P016. Stem Cell Differentiation and Reprogramming

Location: Virtual Only

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Program #/Poster #: P016.07

Topic: A.03. Stem Cells and Reprogramming

Support: "Scholarships grant program for the second round of postgraduate studies" from resources of OP "Human Resources Development (No. 2017-050-0504-9804) , Education and Lifelong Learning" 2014-2020
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Fondation Santé Biomedical Research grant
Operational Programme «Human Resources Development, Education and Lifelong Learning 2014- 2020» in the context of the project ‘’Long non-coding RNA – Transcription factor regulatory networks in mammalian neural stem cells’’ (MIS 5049258)

Title: Long non-coding RNA Lacuna regulates neuronal differentiation of neural stem cells

Authors: *E. NINOU, A. MICHAIL, P. K. POLITIS;
Biomed. Res. Foundation, Acad. of Athens, Athens, Greece

Abstract: Although long non-coding RNAs (lncRNAs) is one of the most abundant classes of RNAs encoded within the mammalian genome and are highly expressed in the adult brain, they remain poorly characterized and their roles in the brain development are not well understood. Here we identify the lncRNA *Lacuna* as a negative regulator of neuronal differentiation in the neural stem/progenitor cells (NSCs) during mouse brain development. In particular, we show that *Lacuna* is transcribed from a genomic locus near to the *Tbr2/Eomes* gene, a key player in the transition of intermediate progenitor cells towards the induction of neuronal differentiation. *Lacuna* RNA expression peaks at the developmental time window between E14.5 and E16.5, consistent with a role in neural differentiation. Overexpression experiments in ex vivo cultured NSCs from murine cortex suggest that *Lacuna* is sufficient to inhibit neuronal differentiation, induce the number of Nestin+ and Olig2+ cells, without affecting proliferation or apoptosis of NSCs. CRISPR/dCas9-KRAB mediated knockdown of *Lacuna* gene expression leads to the opposite phenotype by inducing neuronal differentiation and suppressing Nestin+ and Olig2+ cells, again without any effect on proliferation or apoptosis of NSCs. Interestingly, despite the negative action of *Lacuna* on neurogenesis, its knockdown inhibits T*br2/Eomes* transcription, implying a simultaneous, but opposite, role in facilitating the *Tbr2/Eomes* gene expression. Collectively, our observations indicate a critical function of *Lacuna* in the gene regulation networks that fine tune the rate of neuronal differentiation in the mammalian NSCs.

Disclosures: E. Ninou: None. **A. Michail:** None. **P.K. Politis:** None.

Poster

P064. ADHD, SLI, Dyslexia, and Other Specific Disorders of Neurobehavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P064.01

Topic: A.07. Developmental Disorders

Support: NIH - NIDCD Grant 1R15DC016438-01A1

Title: The effect of feedback timing on learning in children with atypical language development

Authors: *I. FITZPATRICK, Y. ARBEL, A. GUL;
MGH Inst. of Hlth. Professions, Charlestown, MA

Abstract: The study was designed to evaluate the effect of feedback timing on learning in school-age children with resolved and persistent developmental language disorder (DLD). Fifteen participants (mean age = 10.7) completed a declarative paired-associate learning task under two conditions. In both conditions, participants learned through trial and error guided by performance feedback. The timing of the feedback was manipulated in the two conditions with the immediate condition presenting feedback 1000ms after the participant's response and the delayed condition presenting feedback 7000ms after the participant's response. Previous research proposes that immediate feedback is linked to the implicit learning system, a system that is hypothesized to be impaired in children with DLD, while delayed feedback is linked to the declarative learning system. Participants' electrophysiological data were recorded during the learning tasks, and their accuracy was evaluated at the end of each task through a test that was free of feedback. Participants completed the Clinical Evaluation of Language Fundamentals (CELF-5), and their Core Language Score (CLS) was used as a measure of language skills. EEG data were time-locked to the presentation of feedback, and two event-related potentials (ERPs), the Feedback Related Negativity (FRN) and the N170, were evaluated. No difference in test accuracy between the two feedback conditions was found. Correlational analysis indicated that language scores positively correlated with test accuracy when feedback was delayed. The results of a mixed ANOVA of the FRN and N170 suggested that while there were no differences in the amplitude of the FRN across conditions, the N170 amplitude was larger in the delayed feedback condition. Correlational analysis indicated that smaller N170 was found associated with higher accuracy in the immediate feedback condition. Taken together, these findings suggest that feedback-based learning that relies on the declarative system (i.e., when feedback is delayed) depends on language skills, and that the activation of the declarative system, as reflected by the N170 when feedback is immediate, is associated with poorer test accuracy.

Disclosures: I. Fitzpatrick: None. **Y. Arbel:** None. **A. Gul:** None.

Poster

P064. ADHD, SLI, Dyslexia, and Other Specific Disorders of Neurobehavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P064.02

Topic: A.07. Developmental Disorders

Support: R25 NS09098502

Title: Relationship between neighborhood poverty and externalizing symptoms in children: mediation and moderation by environmental factors and brain structure

Authors: *M. MAXWELL[1], D. M. BARCH[2], R. TAYLOR[3];
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Abstract: Increased rates of internalizing and externalizing behaviors have been observed in children from more impoverished neighborhoods, and correlates of neighborhood poverty such as toxin levels, altered brain structure, or exposure to crime may contribute to this relationship. Additionally, receipt of socioemotional support as a protective factor can mitigate the impact of adversity on children's developmental outcomes. The goal of this project is to examine the extent to which neighborhood poverty relates to children's mental health outcomes independent of household socioeconomic status, and to determine whether toxin levels and brain volume serially mediate this relationship and whether socioemotional support moderates it. Data from the Adolescent Brain Cognitive Development study were obtained for 8,623 9-10 year old children; neighborhood poverty was measured using nine census tract variables from the Area Deprivation Index and mental health symptoms were assessed by the Child Behavior Checklist. Toxin levels included particulate matter, nitrogen dioxide, and lead risk measured by the NASA SEDAC while amygdala, dlPFC, and intracranial volumes were obtained by MRI. Socioemotional support consisted of parent, peer, and school sources assessed by various questionnaires. Generalized linear models and structural equation modeling tested for evidence of a mediation and moderation. Increased neighborhood poverty was significantly associated with increased externalizing symptoms, and reductions in intracranial volume mediated this relationship. Increased parental support as indexed by the Parental Monitoring Survey attenuated this relationship, but only for children lower in poverty. These results highlight the importance of identifying environmental and neurological markers that may increase risk for later psychopathology in order to better inform holistic interventions designed to reduce the burden of mental illness.

Disclosures: M. Maxwell: None. **D.M. Barch:** None. **R. Taylor:** None.

Poster

P064. ADHD, SLI, Dyslexia, and Other Specific Disorders of Neurobehavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P064.03

Topic: A.07. Developmental Disorders

Support: MRC Grant

Title: Reduced amygdala responsivity to fear in children with antisocial behaviour that persists after treatment

Authors: *S. O' BRIEN[1], A. SETHI[1], M. MEHTA[1], C. ECKER[1], N. BLACKWOOD[1], M. PETRINOVIC[1], S. SCOTT[1], M. CATANI[1], E. VIDING[2], J. BLAIR[3], D. MURPHY[1], M. CRAIG[1];
[1]King's Col. London, London, United Kingdom; [2]Univ. Col. London, London, United Kingdom; [3]Boys Town Natl. Res. Hosp., Boys Town, NE

Abstract: Reduced amygdala responsivity to fear in children with antisocial behaviour that persists after treatment.
Background: Parenting interventions reduce antisocial behaviour in some children with conduct problems (CP), but not others. Understanding the neural basis for this disparity is important because persistent antisocial behaviour is associated with lifelong morbidity and places a huge burden on our health, and criminal justice, systems. One of the most highly replicated neural correlates of antisocial behaviour is amygdala hypoactivity to another person's fear.
Objective: We aimed to assess whether amygdala hypoactivity to fear in CP children is remediated after successful treatment, and/or if it is a specific marker for persistent antisocial behaviour.
Design: We conducted a prospective, case-control, study of CP and typically developing (TD) boys. Both groups completed two MRI scanning sessions, separated by 18±5.8 weeks, with antisocial behaviour assessed at each visit. Parents of CP boys also engaged in a parenting intervention between sessions.
Setting: Parenting interventions referrals from Child and Adolescent Mental Health Services, Local Authorities, Charities, & Social Enterprises
Participants: Participants included parents, and their CP sons, following referral to a parenting intervention group, and TD boys recruited from the same schools and geographical regions
Exposure: Parenting interventions delivered for 12-14 weeks.
Main Outcomes and Measures: We used functional MRI to compare amygdala activity to fearful faces in TD boys, with CP boys whose antisocial behaviour improved or persisted following a parenting intervention.
Results: Final MRI data was available for 57 CP and 36 TD boys. Amygdala hypoactivity to fear was only observed in CP boys with antisocial behaviour that persisted following a parenting intervention, and was absent in CP boys with antisocial behaviour that improved.
Conclusions and Relevance: Our findings suggest amygdala hypoactivity to fear is a marker for antisocial behaviour that is resistant to change following a parenting intervention, and a putative target for future treatments.

Disclosures: S. O' Brien: None. **A. Sethi:** None. **M. Mehta:** None. **C. Ecker:** None. **N. Blackwood:** None. **M. Petrinovic:** None. **S. Scott:** None. **M. Catani:** None. **E. Viding:** None. **J. Blair:** None. **D. Murphy:** None. **M. Craig:** None.

Poster

P064. ADHD, SLI, Dyslexia, and Other Specific Disorders of Neurobehavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P064.04

Topic: A.07. Developmental Disorders

Support: JSPS Grant-in-Aid for Early-Career Scientists(20K14058)
Japan Health Research Promotion Bureau Research Fund for Young Investigators (JH2021-Y-01)

Title: A closer look at impairments of time perception function in individuals with attention-deficit/ hyperactivity disorder

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Abstract: Individuals with attention deficit/ hyperactivity disorder (ADHD) have impairments of time or timing perception. The studies in typically developing individuals (TD) have shown functional segregation that basal ganglia and cerebellum networks were involved in the sub-second timing tasks, whereas the supra-second timing tasks activated cortical structures, such as the supplementary motor area and prefrontal cortex. Individuals with ADHD show deficiencies in time perception for both sub- and supra-second range, which are reflected in performances of behavioral experiments, e.g., a standard deviation (SD) of trial-to-trial performance and duration discrimination accuracy. This study explored temporal cognitive dysfunction specific to ADHD in both sub- and supra-second range. Twenty-one children with ADHD (age: 9.0± 1.1) and 20 typically developing children (age: 10.3± 1.2) participated in this study. For sub-second time perception, the duration discrimination task (DD) was conducted. The participants detected a target tone among three tones, consisted of two pure tones with 1200 ms as standard and one target tone with 400, 700, 800, 900, 1000 or 1100 ms. For supra-second time perception, the duration estimation task (DE) was conducted. The participants were asked to keep pressing a button for reproducing the same duration of time as the preceding stimuli (5 seconds). This study protocol was approved by the Ethics Committee of National Center of Neurology and Psychiatry. As a result, in the DD, the discrimination rate of children with ADHD (63.8± 21.8%) was lower than that of TD (80.2± 5.6%). In the DE, children with ADHD had fewer successful attempts (15.3± 5.8/30) to reproduce the presentation time of the preceding stimuli than TD children (20.0± 4.3/30), though there is no significant difference in mean duration of the button press between groups (ADHD: 4819 ms, TD: 4710 ms). Furthermore, the SD of the button-press duration of the third session was smaller than that of the first session in both groups, but the SD

of children with ADHD was larger than that of TD in the latter half of each session. Our results coincide with previous evidence that ADHD has time perceptive deficits in sub- and supra-second range. While convergent response in the length of the button press suggests that children with ADHD have an ability to learn motor timing in supra-second range, the large SD and no mean difference of button-press duration in children with ADHD may reflect less tolerance in performing the task. This study suggests the importance of considering task characteristics for understanding temporal cognitive function in ADHD.

Disclosures: Y. Egashira: None. **S. Hayashi:** None. **M. Ukezono:** None. **S. Uono:** None. **Y. Kaga:** None. **Y. Kitamura:** None. **Y. Kita:** None. **Y. Yamashita:** None. **T. Okada:** None. **M. Inagaki:** None.

Poster

P064. ADHD, SLI, Dyslexia, and Other Specific Disorders of Neurobehavior

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Program #/Poster #: P064.05

Topic: A.07. Developmental Disorders

Support: NIH NICHD R15HD087937
Alan & Wendy Pesky Foundation Research Grant
Humboldt Research Fellowship

Title: Identifying dyslexia: Link between maze learning and dyslexia susceptibility gene, DCDC2, in young children

Authors: *L. A. GABEL[1], K. VOSS[2], E. JOHNSON[4], E. R. LINDSTRÖM[5], Y.-C. YU[3], D. T. TRUONG[6], E. M. MURRAY[2], K. CARIÑO[2], K. SHYNTASSOV[3], C. M. NIELSEN[5], D. CARROLL[3], S. PANIAGUA[7], J. R. GRUEN[8];
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Abstract: Dyslexia is a common learning disability that affects processing of written language despite adequate intelligence and educational background. If learning disabilities remain untreated, a child may experience long term social and emotional problems which influence future success in all aspects of their life. Dyslexia has a 60% heritability rate, and genetic studies have identified multiple dyslexia susceptibility genes (DSGs). DSGs, such as *DCDC2*, are consistently associated with the risk and severity of reading disability (RD). Altered neural connectivity within temporo-parietal regions of the brain are associated with specific variants of DSGs in individuals with RD. Genetically altering DSG expression in mice results in visual and

auditory processing deficits as well as neurophysiological and neuroanatomical disruptions. Previously, we demonstrated that learning deficits associated with RD can be translated across species using virtual environments. In this two-year longitudinal study, we demonstrate that performance on a virtual Hebb-Williams maze in pre-readers, is able to predict future reading impairment, and the genetic risk strengthens, but is not dependent on, this relationship. Using computational modeling of students' actions using maze locations and latency values, prediction results can be integrated into the test for real-time feedback of the performance in the form of at-risk percentages for reading impairment. A participant's performance in each trial was based on an accumulated score of the step-by-step distance between the participant's actual location and the nearest ground truth location in the maze throughout the trial. The better a participant performs in a trial, the lower the calculated score. Further examination of performance scores indicated that gender and age are two key factors in performance on this task. We are currently developing a computational model with deep learning capability to identify children at risk for reading impairment. Due to the lack of oral reporting and use of letters, this easy-to-use tool may be particularly valuable in a remote working environment, as well as working with vulnerable populations such as English language learners.

Disclosures: **L.A. Gabel:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); US patent pending (US patent Apl. No. 15/934,567). **K. Voss:** None. **E. Johnson:** None. **E.R. Lindström:** None. **Y. Yu:** None. **D.T. Truong:** None. **E.M. Murray:** None. **K. Cariño:** None. **K. Shyntassov:** None. **C.M. Nielsen:** None. **D. Carroll:** None. **S. Paniagua:** None. **J.R. Gruen:** None.

Poster

P064. ADHD, SLI, Dyslexia, and Other Specific Disorders of Neurobehavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P064.06

Topic: A.07. Developmental Disorders

Title: Analysis of brain electrical activity through qEEG in young adults at risk of ADHD

Authors: W. V. HERRERA-MORALES[1], D. ORTEGÓN-ABUD[1], L. RAMÍREZ-LUGO[2], J. V. REYES-LÓPEZ[3], ***L. NÚÑEZ-JARAMILLO**[1];
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Abstract: Attention deficit hyperactivity disorder (ADHD) is a neurodevelopmental disorder in which patients present inattention, hyperactivity and impulsivity. The condition has its onset during childhood, although some patients are diagnosed until adulthood. Moreover, more than 50

% of children diagnosed with ADHD continue with this condition as adults. Studies on the ADHD neurophysiological correlates address mainly pediatric patients, while comparatively fewer studies have been performed on adults. Some studies on adults report different results when compared with those performed on children, suggesting a variation due to age in the pathophysiology of this condition. Herein we analyzed brain electrical activity in young adults at risk of ADHD (n=46) who rated negative for alcohol use disorders, depression and suicide risk, comparing them versus controls (n=45). Brain activity was measured through quantitative electroencephalography. We analyzed absolute power (AP), relative power (RP) and mean frequency (MF) of delta, theta, alpha and beta frequency bands. We found that subjects at risk of ADHD presented decreased alpha AP at central region. While we found decreased alpha activity, as reported previously for ADHD children, we did not observe increased slow wave activity, as frequently reported for children and adolescents with ADHD. Our results support an age associated variation in the neurophysiological correlates of ADHD.

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Poster

P064. ADHD, SLI, Dyslexia, and Other Specific Disorders of Neurobehavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P064.07

Topic: A.07. Developmental Disorders

Title: Effect of the Neurologist's verbal behavior on the follow-up behavior of the ADHD patient

Authors: *G. OCHOA-ZENDEJAS, O. NÚÑEZ-GAYTÁN, R. GONZALEZ-CASTAÑEDA, A. GÁLVEZ-CONTRERAS, I. VARGAS-DE LA CRUZ;
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Abstract: Background: Lack of adherence to treatment is one of the most common problems in child neurologist care interventions (Sola, 2015). The number for non-compliance with ADHD treatment are worrying, ranging between 15% and 87% (Treuer et al., 2016). Adherence includes behaviors such as attendance at scheduled follow-up appointments, participation in health programs, and seeking care (Gil, Belda & Piñeiro, 1999). Although, there are a few research work about treatment factors related with adherence (Dilla, Valladares, Lizan and Sacristán 2009; WHO, 2003). Studies are still lacking in the identification of the doctor's verbal behavior related with a better patient's follow-up. We propose to incorporate a rigorous methodology that has been applied in the study of psychotherapy (Frojan et al., 2008). This methodology analyses the verbal behavior emitted along the treatment. **Purpose:** To identify the difference between the doctor's verbalization pattern in cases where there is a second appointment and in cases where

that is not the case. **Methods:** The methodology used was observational. The neurologist's verbal behavior was categorized using the SISC-INTER-CVT coding system (Ruiz-Sancho, 2011). The coding and analysis were carried out with the Behavioral Observation Research Interactive Software (BORIS) 7.4.11. SPSS software (22.0) was used for statistical analysis. The reliability of the categorization (inter and intra-judge) was calculated (percentage of agreement and the Kappa Cohen index). The participants were the neurologist and 40 children (7 girls and 33 boys), aged 4 to 9 years old. All the children had a diagnosis of Attention Deficit Hyperactivity Disorder assessed by the neurologist, considering the diagnostic criteria of the DSM-V. **Results:** The non-parametric Mann-Whitney U test was applied to analyze the statistically significant differences between the groups. In the non-attendance group cases there were more Punishment-type verbalizations (Z=-3.767, p= 0.000); while in the attendance group cases there were more informative-type verbalizations (Z=-2.281, p=0.023) and, although there were not statistically differences, there were more instruction-type verbalizations. **Conclusions:** Due to the characteristics of the study, the results can only point out some hypotheses to probe in future research. The verbalizations emitted by the neurologist can be an important factor to take into account in the patient's attendence or not-attendance to the medical appointments. The methodology used in the study seems a suitable research tool for the analysis of the doctor's verbal behavior and to clarify its role in the patient's adherence.

Disclosures: G. Ochoa-Zendejas: None. **O. Núñez-Gaytán:** None. **R. Gonzalez-Castañeda:** None. **A. Gálvez-Contreras:** None. **I. Vargas-de la Cruz:** None.

Poster

P064. ADHD, SLI, Dyslexia, and Other Specific Disorders of Neurobehavior

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Program #/Poster #: P064.08

Topic: A.07. Developmental Disorders

Support: National Science Foundation Division of Biological Infrastructure (Grant # 1659831).
MU School of Medicine Summer Medical Student Research Fellowship
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Title: The effects of stimulant medication on heart rate variability and relationship to creativity in adults with attention-deficit/hyperactivity disorder.

Authors: *C. APPLING[1], N. NURAINI[1], D. Q. BEVERSDORF[4], M. MCBRIDE[2], A. GONZALEZ[2], A. SCHAEFFER[2], A. ZAND[2], D. B. WANG[5], A. SAM[2], E. HART[2], A. TOSH[2], I. FONTCHA[2], B. FERGUSON[3];

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Abstract: Central nervous system stimulants are commonly prescribed for individuals with attention-deficit/hyperactivity disorder (ADHD). Studies suggest that stimulant medications such as norepinephrine-dopamine reuptake inhibitors used to treat ADHD are associated with enhanced performance on creativity tasks. Specifically, stimulants have been shown to enhance fluency, flexibility, and originality scores on the Torrance Test of Creative Thinking, a standardized measure of creativity, in ADHD. Stimulants have also been shown to enhance verbal fluency in adults with ADHD, with no effect on convergent thinking abilities. Recent findings led to questioning the specific roles of norepinephrine and dopamine on creative cognition in ADHD. Furthermore, baseline heart rate variability (HRV), a measure of autonomic nervous system functioning, may be related to creativity in ADHD. However, the full range of effects of stimulant medication on HRV pertaining to creativity in those with ADHD is not known. In the present study, seventeen adults with ADHD attended two sessions: one after taking their prescribed stimulant dose, and one after withholding the dose. Resting autonomic nervous system functioning was assessed using HRV at both visits. Participants completed convergent problem-solving and divergent generative tasks at both sessions in a counterbalanced fashion. We hypothesize 1) that an inverted u-shape relationship exists between HRV and creativity in adults with ADHD. We also expect 2) HRV will be lower on the day stimulant medication is taken compared to control. These results have the potential to reveal important autonomic nervous system relationships with creativity in ADHD.

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Poster

P064. ADHD, SLI, Dyslexia, and Other Specific Disorders of Neurobehavior

Location: Virtual Only

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Program #/Poster #: P064.09

Topic: A.07. Developmental Disorders

Support: 5 T32 MH 111360-4
NIH grant P41 EB015922

Title: Brain white matter microstructure and ADHD: A tensor distribution function analysis

Authors: *S. BETTS, J. VILLALON-REINA, L. SALMINEN, P. M. THOMPSON;
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Abstract: Introduction: Converging evidence suggests that abnormalities in fronto-striatal-cerebellar circuitry contribute to behavioral abnormalities in Attention Deficit Hyperactivity Disorder (ADHD). Diffusion magnetic resonance imaging (dMRI) allows detailed examination of cortico-subcortical white matter (WM) tracts, yielding indices of the network abnormalities thought to arise in ADHD. Altogether, dMRI studies of children and adolescents with ADHD show age-dependent patterns of altered fractional anisotropy (FA), indicative of a developmental delay in neurite density and/or fiber dispersion, in regions associated with executive function and attention. However, findings are inconsistent regarding the direction of effects, regions implicated, and their connection to specific functional deficits of ADHD. Although FA derived from the single-tensor model (FA^{DTI}) is the most widely used metric to characterize (WM) microarchitecture, it is possible that discrepancies in the ADHD literature are partly driven by known limitations in the FA measurement, which performs poorly in regions with crossing fibers. The tensor distribution function (TDF) is an alternative FA measurement that may be used to model multiple underlying fibers by representing the diffusion profile as a probabilistic mixture of tensors. We aimed to reconcile discrepancies in the literature by comparing the ability of FA^{DTI} and TDF-derived FA (FA^{TDF}) to predict ADHD symptom severity in a large sample of adolescents from the Adolescent Brain Cognitive Development (ABCD) Study. Methods: We analyzed brain and behavioral data from 745 children (ages 8.9-11 years, 341F/404M) from the ABCD Study. As ABCD consists of many groups of siblings, we selected one participant from each family to avoid confounding effects of relatedness. Raw scores from the ADHD DSM5 scale of the child behavior checklist (CBCL) were used to evaluate ADHD symptom severity as the primary outcome measure. Higher CBCL scores were predicted by higher FA^{TDF} of the body and genu of the corpus callosum, bilateral anterior corona radiata, bilateral superior corona radiata, bilateral posterior thalamic radiation, bilateral tapetum, and by an average of FA^{TDF} across all ROIs. No associations were found between FA^{DTI} and ADHD symptomatology. Collectively our results show that FA^{TDF} offers superior performance over the traditional single-tensor FA measurement, further emphasizing the utility of FA^{TDF} for assessing microstructural brain alterations in heterogeneous and complex conditions such as ADHD.

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Poster

P069. Development of Motor Systems

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P069.01

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Support: R01-NS105748
T32 GM007183
DGE-1746045
K01 NS102342
MGCB start-up funds
International Student Fellowship

Title: Linking neuronal stem cells to circuit wiring decisions in the Drosophila larval motor system

Authors: *J. L. MENG[1], Y. WANG[2], R. A. CARRILLO[2], E. S. HECKSCHER[3];
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Abstract: How circuit wiring is specified is a key question in developmental neurobiology. Previously, using the Drosophila motor system as a model, we found the classic temporal transcription factor Hunchback acts in NB7-1 neuronal stem cells to control the number of NB7-1 neuronal progeny form functional synapses on dorsal muscles (Meng et al., 2019). However, it is unknown to what extent control of motor neuron-to-muscle synaptic partnerships is a general feature of temporal transcription factors. Here, we perform additional temporal transcription factor manipulations—prolonging expression of Hunchback in NB3-1, as well as precociously expressing Pdm and Castor in NB7-1. We use confocal microscopy, calcium imaging, and electrophysiology to show that in every manipulation there are permanent alterations in neuromuscular synaptic partnerships. Our data show temporal transcription factors, as a group of molecules, are potent determinants of synaptic partner choice and therefore ultimately control circuit membership.

Disclosures: J.L. Meng: None. **Y. Wang:** None. **R.A. Carrillo:** None. **E.S. Heckscher:** None.

Poster

P069. Development of Motor Systems

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Program #/Poster #: P069.02

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

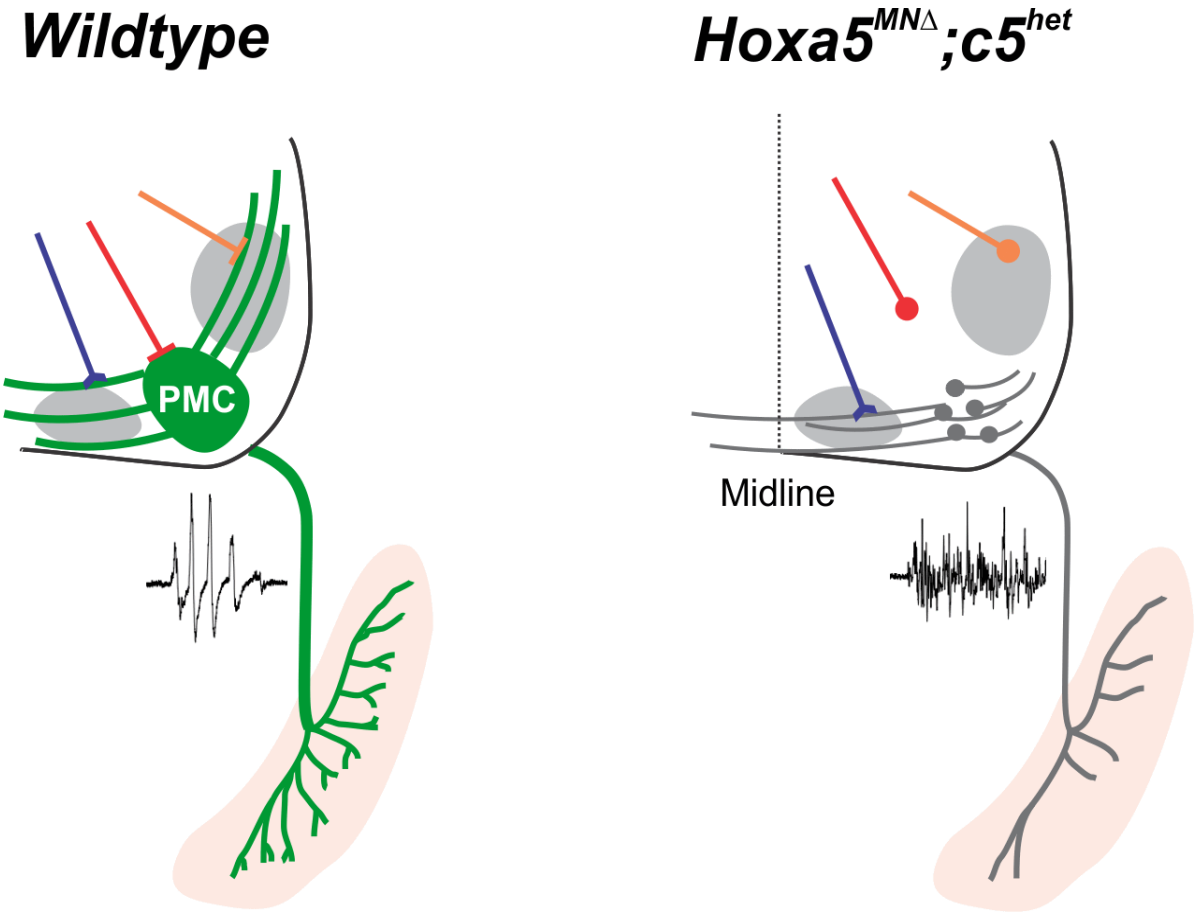
Support: F30HD096788
T32GM007250
R00NS085037
R01NS114510

Mt Sinai Health Foundation

Title: Phrenic-specific transcriptional programs shape respiratory motor output

Authors: *A. N. VAGNOZZI[1], K. GARG[1], C. DEWITZ[2], M. T. MOORE[1], J. M. CREGG[1], L. JEANNOTTE[3], N. ZAMPIERI[2], L. T. LANDMESSER[1], P. PHILIPPIDOU[1];
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Abstract: Breathing is the most essential motor behavior. The frequency and amplitude of breathing movements are controlled by neural networks residing in the brainstem and spinal cord. Degeneration of these networks leads to respiratory disorders, such as central sleep apneas, and, eventually, respiratory failure. In mammals, contraction of the diaphragm muscle is vital for driving airflow into the lungs during inspiration. Diaphragm contraction is controlled by a single motor input, phrenic motor neurons (MNs) in the cervical spinal cord, making this MN population a critical node in the respiratory network. Despite their essential role, the molecular determinants that control the patterns of phrenic MN activity and their integration into respiratory circuits are largely unknown. We show that Hox5 transcription factors are required for robust and efficient breathing. Hox5 loss renders mice vulnerable to respiratory dysfunction in the first two weeks of life, suggesting that Hox5 mutations may contribute to early life respiratory conditions, such as sudden infant death syndrome (SIDS). We show that Hox5 proteins establish phrenic MN clustering and topography through the regulation of a network of cell adhesion molecules. We find that a subset of cadherins are specifically expressed in phrenic MNs and that loss of cadherin function through conditional disruption of downstream β/γ-catenin signaling leads to phrenic MN cell body disorganization and dendrite displacement. Remarkably, after MN-specific deletion of Hox5 genes, phrenic MN firing becomes asynchronous and erratic due to the selective loss of inhibitory inputs to phrenic MNs. Our results demonstrate that Hox5 transcription factors determine phrenic MN topography and connectivity to generate robust breathing behaviors. Our findings support a model where MN-intrinsic transcriptional programs shape the pattern of motor output by orchestrating distinct aspects of MN connectivity.



Disclosures: A.N. Vagnozzi: None. **K. Garg:** None. **C. Dewitz:** None. **M.T. Moore:** None. **J.M. Cregg:** None. **L. Jeannotte:** None. **N. Zampieri:** None. **L.T. Landmesser:** None. **P. Philippidou:** None.

Poster

P069. Development of Motor Systems

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Program #/Poster #: P069.03

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Support: Rehabilitation Medical Research Foundation Research Grant

Title: Effects of Mechanical Force on Motor system Development in postnatal mouse with Sciatic Denervation

Authors: *Y. USAMI[1], Y. MINEGISHI[2], H. FUNAKOSHI[1], K. TAKAHATA[1], T. KOKUBUN[3];

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Abstract: INTRODUCTION : The musculoskeletal system is an efficient assembly of tissues that coordinate to enable movement by an order from the central nervous system. Musculoskeletal development needs mechanical stimulation, yet little is known that the electrical signals from a peripheral nerve to muscle affect tendon development. Here, we use a model of the Sciatic Denervation to investigate the impact of electrical signals from the peripheral nerve on musculoskeletal system development in postnatal mice.
METHODS : We used C57BL/6 mice aged postnatal day seven. The sciatic nerves of the lateral hind limbs were cut (Sciatic Denervation: DN). The Contralateral limb was sham-operated (Sham) as a control(n=6/group). We evaluated the muscle and tendon development factors at 3,7,14 post-operated days (POD). The muscle and tendon were examined to determine the expression of Scx is a bHLH transcription factor expressed in tendon cells from progenitor stages and through development, and TGF-b associate with growth factor. For histological evaluation, the limbs were sectioned focus on the Achilles Tendon at 12 μm thickness and stained with Alcian blue, Hematoxylin and eosin, and Picrosirius red to visualize collagen.
RESULTS : Our results show the tendon markers Scx were decreased in DN group tendons compared to sham group as time proceeds.On the other hand, expression of Scx in Sham group tendons increased from from POD3 to POD7 and decreased from POD7 to POD14. TGF-b expression in the tendon at POD3 was higher DN group than Sham group. Histological sections results for the DN group at POD14 suggested clear atrophy of skeletal muscle. The DN group at POD14 caused an immature collagen arrangement in the tendon tissue.
DISCUSSION : We focus on the role of electrical signals from peripheral nerves in motor system development using postnatal mice. Expression of Scx in DN group decreased from POD3 to POD14, so that means electrical signals from peripheral nerves promote Scx expression in postnatal tendon development. We show the functional role of the sciatic nerve in the soleus and gastrocnemius muscle, and these dysfunctions affect achilles tendon development. Signals by the peripheral nerve provide a contraction for the muscle, leading to tensile stress for the tendon. Peripheral nerve resection inhibited the postnatal growth of muscle and tendons. This study demonstrates that mechanical loading or electrical signals from the peripheral nerve affects the postnatal development of musculoskeltal systems.

Disclosures: Y. Usami: None. **Y. Minegishi:** None. **H. Funakoshi:** None. **K. Takahata:** None. **T. Kokubun:** None.

Poster

P069. Development of Motor Systems

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Program #/Poster #: P069.04

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Support: NIH Grant R37-HD081168

Title: Sensory coding of limb kinematics in motor cortex across a key developmental transition.

Authors: *R. M. GLANZ, J. C. DOOLEY, G. SOKOLOFF, M. S. BLUMBERG;
Univ. of Iowa, Iowa City, IA

Abstract: Primary motor cortex (M1) plays key roles in the production of movements and in motor learning in adults, but in early development it functions instead as a prototypical *sensory* structure. Throughout the first postnatal week in rats, M1 is strongly activated by self-generated limb movements—especially by the twitches that occur during active (or REM) sleep. Here, we quantify the kinematic features of forelimb movements to reveal receptive-field properties of individual units within the forelimb region of M1. At postnatal day (P) 8, nearly all units were strongly modulated by movement amplitude, especially during active sleep: Large twitches produced significantly more M1 activity than did small twitches. By P12, however, only a minority of units continued to exhibit amplitude-tuning, regardless of sleep-wake state. At both ages, movement direction also modulated M1 activity, though to a lesser extent. Finally, at P12, M1 population-level activity became sparse and decorrelated, accompanied by a substantial reduction in the frequency of "all-or-nothing" M1 responses to limb movements observed at P8. These changes in M1 activity were not due to the disappearance of spindle bursts (10-20 Hz thalamocortical oscillations) before P12. Currently, we are extending this work to identify the mechanisms responsible for the transition to decorrelated activity at P12. Our findings thus far reveal a transition toward a more complex and informationally rich representation of movement long before M1 develops its motor functionality.

Disclosures: R.M. Glanz: None. **J.C. Dooley:** None. **G. Sokoloff:** None. **M.S. Blumberg:** None.

Poster

P069. Development of Motor Systems

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P069.05

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Support: NIH Grant R37-HD081168
NIH Grant F32-NS101858

Title: Theta rhythmicity during REM sleep functionally integrates behavior with neural activity in primary motor cortex and red nucleus in preweanling rats

Authors: *J. C. DOOLEY, G. SOKOLOFF, M. S. BLUMBERG;
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Abstract: Myoclonic twitches are abundantly produced during REM sleep in skeletal muscles across the body. In infant rats, movements are produced by the red nucleus (RN), with the RN both sending motor commands and receiving sensory feedback from twitches. The RN's role in producing twitches contrasts with that of primary motor cortex (M1), which does not generate motor commands at early postnatal ages. Instead, M1 functions as a sensory structure, processing sensory feedback from self-generated movements, including twitches. By postnatal day (P) 12, the RN also begins to exhibit a continuous theta rhythm (~6 Hz) during REM sleep that promotes sensorimotor integration with other brain areas. At P12, however, REM-associated theta is not present in M1. Because we know that in adult rats, both the RN and M1 collaborate to control movement, we hypothesized that theta emerges in M1 to facilitate coordination of movement-related activity in both structures. To test whether theta promotes sensorimotor integration in the RN and M1, we recorded local field potentials and unit activity in the RN and the forelimb region of M1 in unanesthetized preweanling rats from P12 through P20. Rats were head-fixed but were able to locomote and cycle freely between sleep and wake. Neurons in both the RN and M1 continued to show twitch-related activity through P20. Further, as predicted, we observed the developmental emergence of REM-associated theta oscillations in M1 by P20 that were coherent with theta in the RN. Additionally, neural activity was phase-locked to theta; surprisingly, twitches were also phase-locked to theta, with twitches being more likely during the troughs of the oscillation. Finally, forelimb twitches—but not hindlimb, whisker, or tail twitches—were associated with phase-dependent nesting of fast gamma oscillations (80-100 Hz) within theta in M1. Taken together, these results show how theta during REM sleep promotes the developmental integration of behavior with activity in M1 and the RN. Because the nested fast gamma oscillations in forelimb M1 were specific to forelimb twitches, these results also implicate twitches and twitch-related activity in the development of somatotopically precise functional connectivity between the RN and M1.

Disclosures: J.C. Dooley: None. **G. Sokoloff:** None. **M.S. Blumberg:** None.

Poster

P069. Development of Motor Systems

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P069.06

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Title: Investigation of a human disease variant in GSX2 which alters DNA binding specificity and leads to basal ganglia agenesis

Authors: *L. TWEEDIE[1,2], J. SALOMONE[1,2], S. QIN[1], B. CAIN[1], B. GEBELEIN[1], K. CAMPBELL[1];
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Abstract: The striatum, a major component of the basal ganglia, is known to derive largely from neural progenitors in the lateral ganglionic eminence (LGE) during embryogenesis. Gsx2, a homeobox transcription factor, is necessary for the correct patterning of neural progenitors in the LGE and ultimately the specification of distinct neuronal subtypes, such as striatal projection neurons. Mice carrying germline *Gsx2* null alleles show severe agenesis of the striatum and do not survive after birth. A recent report in humans (De Mori et al., (2019) *Brain*, 142:2965-2978), identified a homozygous null allele and another homozygous mutation within the GSX2 homeodomain (HD) which changes the "Q" to "R" at amino acid 251 (i.e. $GSX2^{Q251R}$). Both mutations resulted in viable children with similar severe agenesis of the basal ganglia and neurological symptoms including spastic tetraparesis, dystonia, and intellectual impairment. Preliminary electrophoretic mobility shift assays (EMSAs) using mouse Gsx2 with an analogous mutation (i.e. $Gsx2^{Q252R}$) show that this mutant protein can bind a subset of DNA sequences normally bound by WT Gsx2, but with an altered sequence preference more closely analogous to K50 HD factors (e.g. Otx2) than the usual Q50 HD sites of Gsx2. We therefore hypothesized that the $Gsx2^{Q252R}$ mutation leads to aberrant DNA binding and thus a modified gene regulatory network (GRN) which alters LGE progenitor specification and differentiation. To determine the impact of this human variant on basal ganglia development, we generated a mouse model of the $Gsx2^{Q252R}$ HD variant using CRISPR-Cas9. Importantly, the $Gsx2^{Q252R}$ protein is nuclear localized and expressed in progenitors of the mutant LGE. Preliminary characterization of these mutant mice indicates an intermediate phenotype between *Gsx2* null and WT animals, both for gene expression changes and striatal size. These data support the idea that the $Gsx2^{Q252R}$protein maintains proper regulation of some target genes but does not regulate, or inappropriately regulates, other genes. To better understand the impact of this HD variant on the *Gsx2* GRN, we utilized CRISPR-Cas9 to introduce the human variant on a $^{2xFLAG}Gsx2$ background. Future studies will use these $^{2xFLAG}Gsx2^{Q252R}$ mice for CUT&RUN analysis to determine the genomic DNA binding profile of this variant and compare it with our recent findings for WT ^{2xFLAG}Gsx2 (Salomone et al., (2021) *Genes & Development*, 35:157-174). Finally, as $Gsx2^{Q252R/Q252R}$ mice are viable postnatally (unlike germline null *Gsx2* mice), these mice will provide a powerful model to investigate behavioral changes as a result of this mutation.

Disclosures: L. Tweedie: None. **J. Salomone:** None. **S. Qin:** None. **B. Cain:** None. **B. Gebelein:** None. **K. Campbell:** None.

Poster

P069. Development of Motor Systems

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Program #/Poster #: P069.07

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Support: R01 MH090740
F30 MH123056

Title: A pioneering role for striatonigral projections in the formation of the internal capsule and cerebral peduncle: Lessons from Isl1 conditional knockouts

Authors: *J. EHRMAN[1,3], P. MERCHAN-SALA[1], L. A. EHRMAN[2], R. R. WACLAW[2], K. J. CAMPBELL[1];
[1]Developmental Biol., [2]Exptl. Hematology and Cancer Biol., Cincinnati Children's Hosp. Med. Ctr., Cincinnati, OH; [3]Med. Scientist Training Program, Univ. of Cincinnati Col. of Med., Cincinnati, OH

Abstract: The basal ganglia are a group of subcortical nuclei with known roles in the coordination of voluntary motor programs, aspects of cognition, and the selection of appropriate social behaviors. The striatum is the principle input nucleus of the basal ganglia; disruptions in striatal connectivity are thus implicated in the motor, cognitive, and social dysfunction characterizing neurodevelopmental disorders including autism spectrum disorder and attention-deficit/hyperactivity disorder. Additionally, striatonigral (striatal direct pathway) projections form in close proximity to the ascending (thalamocortical) and descending (corticofugal) tracts of the internal capsule, suggesting that malformed striatal circuits may disrupt adjacent axon trajectories and thereby contribute to the pathology of these developmental disorders. Despite these notions, both the molecular mechanisms underlying striatal circuit assembly and the contributions of striatonigral axons to the establishment of other critical circuits traversing the internal capsule/cerebral peduncle remain understudied. Using *Sox8-EGFP* (striatal direct pathway) and *Fezf2-TdTomato* (corticofugal pathway) BAC transgenic reporter mice in combination with immunohistochemical markers, we show that striatonigral axons precede corticofugal and thalamocortical fiber entry into the internal capsule and forming cerebral peduncle. Thus, striatonigral axons are well positioned to guide the trajectories of descending cortical fibers. To investigate the impact of direct pathway malformation on the development of internal capsule axon tracts, we are using telencephalon- or ventral forebrain-specific *Isl1* conditional knockout (cKO) mice which exhibit selective, early death of striatonigral neurons and disrupted direct pathway axon outgrowth. Both types of *Isl1* cKO mutants show striking defects in descending cortical axons, despite the fact that Isl1 is not expressed in these projection neurons. To examine the molecular processes mediating these axonal trajectory defects, we generated an RNAseq dataset from the embryonic *Isl1* cKO striatum. We have identified and validated differentially expressed genes with roles in cell adhesion, growth cone dynamics, and extracellular matrix composition, focusing on those previously implicated in neurological disorders. Taken together, our findings support a novel role for the striatal direct pathway in pioneering the internal capsule/cerebral peduncle and suggest abnormal development of basal ganglia circuits may contribute to secondary defects in cortical trajectories adding to the pathology of common neurodevelopmental disorders.

Disclosures: J. Ehrman: None. **P. Merchan-Sala:** None. **L.A. Ehrman:** None. **R.R. Waclaw:** None. **K.J. Campbell:** None.

Poster

P105. Modulation of Synaptic Transmission: Molecular Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P105.01

Topic: B.06. Synaptic Transmission

Support: NIMH Grant 084989
NICHD Grant 007502

Title: Convergence of Drosophila cognitive dysfunction disease models: Corkscrew/SHP2 phosphatase activity in synaptic transmission

Authors: *S. N. LEAHY[1], D. J. VITA[1], C. SONG[1], K. BROADIE[1,2,3];
[1]Biol. Sci., [2]Vanderbilt Brain Inst., Vanderbilt Univ., Nashville, TN; [3]Kennedy Ctr. for Res. on Human Develop., Vanderbilt Univ. Med. Ctr., Nashville, TN

Abstract: Corkscrew (Csw; human SHP2) is a protein tyrosine phosphatase that acts downstream of receptor tyrosine kinase (RTK) function as a core regulator of MAPK/ERK signaling. Gain-of-function (GOF) and loss-of-function (LOF) mutants disrupt this key signaling to cause Noonan Syndrome (NS) and Noonan Syndrome with Multiple Lentigines (NSML), respectively. NS and NSML patients are known to manifest a wide variety of shared symptoms, including cognitive dysfunction. This GOF/LOF phenotype convergence is hypothesized to be caused by the increased MAPK/ERK signaling characterizing both diseases, although the exact cause of the cognitive dysfunction is unknown. To address hypothesized synaptic foundations of these cognitive deficits, we dissected Csw/SHP2 roles with disease-relevant mutations at the *Drosophila* neuromuscular junction (NMJ) model glutamatergic synapse. Using two-electrode voltage-clamp (TEVC), we find both GOF and LOF Csw/SHP2 mutations cause significantly increased glutamatergic neurotransmission as measured by excitatory junction current (EJC) recordings (LOF: n=>10 p=<0.0001; GOF: n=>10 p=0.02). We find the Csw/SHP2 requirement to be presynaptic based on evidence from miniature EJC (mEJC) analyses and targeted RNAi knockdown (mEJC: n=>10 p=0.008; RNAi: n=>10 p=0.001). We also find loss of Csw/SHP2 disrupts synaptic vesicle (SV) availability in fatigue and facilitation analyses, suggesting that Csw/SHP2 regulates transmission strength via control of SV availability (fatigue: n=6,7 p=<0.0001; facilitation: n=16,21 p=<0.0001). Additionally, we find the Csw/SHP2 functional role dependent on regulation of synaptic MAPK/ERK signaling. Importantly, we find Csw/SHP2 interacts with Fragile X Mental Retardation Protein (FMRP), an mRNA-binding protein that regulates activity-dependent translation of synaptic proteins, and whose epigenetic silencing causes Fragile X Syndrome (FXS), the most common heritable

intellectual disability and autism spectrum disorder. We find genetic evidence that Csw/SHP2 and FMRP interact to control synaptic function (n=>20 p=<0.0001). Our mEJC analyses indicate FMRP and Csw/SHP2 interact in presynaptic mechanisms of SV release (n=>15 p=0.0001). Importantly, synaptic pERK levels are elevated in both Csw/SHP2 and FMRP mutants. This work has revealed a previously unexplored role for Csw/SHP2 at the synapse, established disease relevant mutations disrupt synaptic function, and discovered an interaction between FMRP and Csw/SHP2, indicating a mechanistic connection between previously unlinked FXS and NS/NSML disorders.

Disclosures: S.N. Leahy: None. **D.J. Vita:** None. **C. Song:** None. **K. Broadie:** None.

Poster

P105. Modulation of Synaptic Transmission: Molecular Mechanisms

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Program #/Poster #: P105.02

Topic: B.06. Synaptic Transmission

Support: NIH Grant 1R01NS123648-01

Title: Hyperpolarization-activated cyclic nucleotide-gated channels regulate response probability in the terminals of parvalbumin positive basket cells

Authors: *T. BOCK[1], E. W. BUSS[1], O. M. LOFARO[1], B. SANTORO[2], S. A. SIEGELBAUM[3]; [1]Dept. of Neurosci., Columbia Univ., New York, NY; [2]Dept. of Neurosci., Columbia Univ. Press, New York, NY; [3]Dept of Neurosci., Columbia Univ. Coll P & S, New York, NY

Abstract: Hyperpolarization-activated, cyclic nucleotide-gated (HCN) channels generate the cationic current I_h, which has been found to regulate the excitability of neuronal networks. The function of HCN channels depends, in part, on their subcellular localization. HCN channels are abundantly expressed in the dendrites of pyramidal neurons in hippocampal area CA1 where they act as an inhibitory constraint on dendritic integration and synaptic plasticity. HCN1 is also strongly expressed in presynaptic terminals of parvalbumin-positive (PV+) inhibitory neuron basket cells (PVBC), which provide strong inhibitory control over hippocampal activity. Previous electrophysiological studies in our lab found that application of HCN channel blockers ZD7288 and Ivabradine strongly inhibit IPSCs in CA1 pyramidal neurons evoked by either electrical stimulation or PVBC-specific optogenetic stimulation (see Buss et al., SfN Abstract 2018). To directly investigate the impact of HCN channels in presynaptic terminals, we performed 2-photon imaging in acute brain slices of mice, expressing the fluorescent calcium reporter GCaMP6s specifically in the terminals of PVBCs within CA1. Fast excitatory transmission was blocked with D-APV and CNQX . Bath application of ZD7288 strongly reduced the amplitude of dF/F transients in PVBC terminals elicited by a train of 5 pulses of

electrical stimulation (30 Hz) delivered to the stratum pyramidale. To determine whether this effect was due to a reduction in the Ca^{2+} response to a presynaptic action potential or to a reduction in the probability that the stimulation would elicit an action potential, we examine responses to single electrical stimuli. We found that the Ca^{2+} response of individual terminals to single stimuli was probabilistic; some stimuli elicited a measurable response (successes) and some stimuli failed to elicit a response (failures). ZD7288 did not reduce the amplitude of the successful responses to single electrical stimuli, but rather decreased the probability with which single stimuli evoked a successful response. Thus we conclude that HCN channels in the axonal terminals of PVBCs upregulate the probability of terminal activation (and thus vesicle release) rather than the amplitude of the Ca^{2+} response to such activation.

Disclosures: T. Bock: None. **E.W. Buss:** None. **O.M. Lofaro:** None. **B. Santoro:** None. **S.A. Siegelbaum:** None.

Poster

P105. Modulation of Synaptic Transmission: Molecular Mechanisms

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Program #/Poster #: P105.03

Topic: B.06. Synaptic Transmission

Support: NIH Grant DA048946
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NIH Grant DA052109
NIH Grant CA77598

Title: Angiotensin-converting enzyme non-canonically constrains opioid-sensitive synaptic transmission in nucleus accumbens

Authors: *B. H. TRIEU, B. C. REMMERS, C. TODDES, D. D. BRANDNER, S. S. MORE, P. E. ROTHWELL;
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Abstract: Angiotensin-converting enzyme (ACE) is canonically known to regulate blood pressure by cleaving angiotensin I to form angiotensin II peptide primarily in the lungs. However, ACE is also highly expressed in the striatum and particularly enriched in medium spiny projection neurons expressing the Drd1 dopamine receptor (D1-MSNs) that form the striatonigral pathway. The physiological function of neuronally-expressing ACE is unknown though it has been previously reported to degrade neuropeptides. Using a combination of electrophysiology and targeted tandem mass spectrometry in female and male mice, we show

that ACE negatively regulates endogenous Met-enkephalin-Arg-Phe ("MERF"), an overlooked enkephalin opioid peptide that potently constrains excitatory synaptic transmission in a cell-type specific manner. Captopril, a clinically-approved ACE inhibitor, blocked this non-canonical mechanism to enhance mu opioid receptor activation and subsequently induced long-term depression of presynaptic glutamate release onto D1-MSNs. The physiological impact of ACE inhibition on striatal circuits is reflected in behavioral assays that demonstrated reduced conditioned place preference caused by fentanyl but increased reciprocal social interaction following captopril administration in female and male mice. These results provide preclinical evidence for a class of cardiovascular medications that could be repositioned and redesigned toward brain diseases characterized by aberrant striatal pathophysiology.

Disclosures: B.H. Trieu: None. **B.C. Remmers:** None. **C. Toddes:** None. **D.D. Brandner:** None. **S.S. More:** None. **P.E. Rothwell:** None.

Poster

P105. Modulation of Synaptic Transmission: Molecular Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P105.04

Topic: B.06. Synaptic Transmission

Support: NIH IRTA 77439

Title: Aging alters mitochondrial morphology in cholinergic terminals in the basolateral amygdala.

Authors: *D. FREEMAN[1], D. A. TALMAGE[2], L. ROLE[1];
[1]Natl. Inst. of Neurolog. Dis. and Stroke, Bethesda, MD; [2]Natl. Inst. of Mental Hlth., Bethesda, MD

Abstract: Cholinergic projection neurons innervate diverse regions of the brain in highly complex axonal arbors that require a steady energy supply provided primarily through mitochondrial respiration. Axonal mitochondria are morphologically distinct from those in the soma and their structure is specialized to support synaptic activity. Aged mitochondria lose their functional and structural integrity and mitochondrial dysfunction is an early molecular feature of several neurodegenerative diseases including Alzheimer's disease (AD). The effect of aging on the integrity of cholinergic mitochondria has not been explored. The degeneration of cholinergic terminal fields projecting from the basal forebrain is a hallmark of AD, and yet the dynamics of mitochondria in cholinergic axons is minimally understood. The goal of this study was to examine the effects of aging on axonal mitochondrial morphology and localization in cholinergic projections. We used old (30 weeks) and young (4 weeks) mice genetically labeled by the expression of cholinergic acetyltransferase (ChaT) promoter driven tau-GFP for easy

identification of cholinergic axons in fixed coronal sections of the basolateral amygdala (BLA). Mitochondria were labeled with a fluorescent antibody against the voltage-dependent anion channel (VDAC) located on the outer mitochondrial membrane. To compare cholinergic axons versus other axons, mitochondria will be examined in non-ChaT-tau GFP labeled axons stained with pan-axonal markers. We observed an increase in mitochondrial size in cholinergic axons compared to non-cholinergic axons. Cholinergic mitochondria also appeared less spherical than non-cholinergic across ages. Aging further increased mitochondrial size and altered the distribution of spherical and elongated axonal mitochondria. Since mitochondrial size and shape can determine motility and calcium uptake at the pre-synapse, it is hypothesized that changes in mitochondrial morphology alters synaptic activity and axonal branching. These data will provide insights into the dynamics of mitochondria within cholinergic terminal fields and will might reveal key aspects of mitochondrial dysfunction in neurodegeneration and Alzheimer's disease.

Disclosures: D. Freeman: None. **D.A. Talmage:** None. **L. Role:** None.

Poster

P105. Modulation of Synaptic Transmission: Molecular Mechanisms

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P105.05

Topic: B.06. Synaptic Transmission

Support: NIH/NINDS R21 NS111945-02
Research Acceleration Award (internal funding)

Title: The effect of bidirectional regulation of O-GlcNAcylation on neural circuitry

Authors: *S. PHILLIPS, L. MCMAHON;
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Abstract: Effective neuronal circuitry relies on the balance of excitation to inhibition (E/I). E/I imbalances are found in various neurodegenerative diseases like autism spectrum disorders and Alzheimer's disease. Targeting the mechanisms that contribute to and reestablish the E/I balance in the normal and diseased brain provide a fundamental understanding of neuronal circuit function. Our lab has begun to address this by examining the role of protein O-GlcNAcylation and its bidirectional effects on hippocampal excitatory and inhibitory transmission. O-GlcNAcylation is a post-translational modification that involves the O-linked attachment of β-N-acetylglucosamine to Ser/Thr residues on proteins. O-GlcNAc transferase (OGT) and O-GlcNAcase (OGA) add and remove the O-GlcNAc moiety on proteins respectively and is tightly regulated by UDP-N-acetylglucosamine (UDP-GlcNAc) availability via the Hexosamine Biosynthetic Pathway (HBP). Because OGT and OGA are highly abundant in the hippocampus, targeting both enzymes to modify O-GlcNAc plays a significant role in hippocampal synaptic

transmission and neuronal inhibition. Using brain slice electrophysiology, we previously reported that an acute increase in O-GlcNAc using glucosamine, a HBP substrate, or the OGA inhibitor Thiamet-G (TMG), induces O-GlcNAc modification of GluA2 AMPAR subunits triggering an NMDAR independent LTD and dampens pathological hyperexcitability at CA3-CA1 synapses (Taylor et al., 2014 and Stewart et al., 2017). Additionally, an acute increase in O-GlcNAc also induces a LTD of GABAAR mediated transmission and decreases spontaneous IPSC amplitude and frequency. However, the net effect is an overall decrease in the E/I ratio resulting in decreased excitability. While there are several highly selective inhibitors of OGA, identifying effective and specific OGT inhibitors presents its challenges. In this study, we are investigating whether pharmacological inhibition of OGT using OSMI-4 modifies the strength of basal excitatory and inhibitory transmission as well as prevent glucosamine induced LTD at excitatory and inhibitory synapses in rat hippocampal slices. Our preliminary results show that OSMI-4 partially blocks glucosamine-induced depression of sIPSC amplitude, indicating that bidirectional changes of O-GlcNAc regulates hippocampal synaptic transmission and solidify the therapeutic benefits of decreasing O-GlcNAc levels in disease states the exhibit increased O-GlcNAcylation.

Disclosures: S. Phillips: None. **L. McMahon:** None.

Poster

P105. Modulation of Synaptic Transmission: Molecular Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P105.06

Topic: B.06. Synaptic Transmission

Support: NIH Grant HL088052
NIH Grant NS150251

Title: Dehydration-induced glutamatergic excitation of the hypothalamic paraventricular nucleus (PVN) dynamically increases local inhibitory GABAergic transmission

Authors: *J. YAMAGUCHI, T. TRUONG, M. A. ANDRADE, G. M. TONEY;
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Abstract: Presympathetic neurons (PSNs) of the hypothalamic PVN mostly lack basal firing in unstressed animals due to the dominance of local GABAergic inhibition. We previously determined that synaptic inhibition of PVN PSNs by GABA is actively maintained by excitatory amino acid transporter 3 (EAAT3)-mediated uptake of L-glutamate (Glu) into synaptic terminals of GABA neurons in the surrounding peri-nuclear zone (PNZ). We termed this mechanism Glu-GABA strengthening (GGS). Here we tested the hypothesis that a physiological challenge that increases sympathetic nerve activity through lamina terminalis glutamatergic excitation of PVN

does so by disrupting the GGS mechanism. Male C57Bl/6 mice were euhydrated (EUH) or water deprived (WD) for 48 h. Contrary to our hypothesis, western blot analysis revealed increased, not decreased, PVN EAAT3 in WD (132 ± 1.2%) compared to EUH (100 ± 9.5 %) mice (P = 0.0210, n=8/group). This result, together with our previous report of reduced PVN expression of the glial Glu transporter GLT-1 (EAAT2) in WD rats, suggests that Glu clearance in PVN is shifted during dehydration from glial uptake by EAAT2 to neuronal uptake by EAAT3, which would be predicted to increase, not disrupt, PVN GGS. To quantify the potential impact of this, mice were instrumented with telemetry transmitters to record arterial pressure and were subsequently WD. As expected, mean arterial pressure (MAP) in WD mice increased from 109.2 ± 2.6 mmHg at baseline to 120.5 ± 2.6 mmHg after 48 h. Unexpectedly, MAP fell below baseline (103.4 ± 3.3 mmHg) upon rehydration, raising the possibility that this could reflect exaggerated PVN GGS that persists upon relative silencing of glutamatergic excitation. To investigate this possibility, we recorded evoked inhibitory post synaptic currents (eIPSCs) and generated a cumulative amplitude curve from 1000 stimuli delivered at 5 Hz to the GABAergic PNZ. The slope (-pA/stimulus) of this relation in WD mice was compared to that in mice lacking GGS due to CRISPR-Cas9 knockdown of EAAT3 expression in PNZ GABA neurons of vGAT-spCas9 mice. Early during the stimulus train (stimuli 1-100), slope values were similar for PSNs from WD (186 ± 0.9) and $PNZ^{EAAT3\text{-}KD}$ (180 ± 0.9) mice. By contrast, the late phase (stimuli 901-1000) slope value in the WD group (52 ± 0.2) was 3X greater than in the $PNZ^{EAAT3\text{-}KD}$ (17 ± 0.1) group. The latter indicates that eIPSCs in WD mice are resistant to rundown/depression and, contrary to our hypothesis, suggests that PVN GGS is enhanced, not diminished, by dehydration. We posit that enhanced PVN GGS may be critical for preserving synaptic homeostasis and preventing excessive PVN-driven sympathoexcitation during dehydration.

Disclosures: J. Yamaguchi: None. **T. Truong:** None. **M.A. Andrade:** None. **G.M. Toney:** None.

Poster

P105. Modulation of Synaptic Transmission: Molecular Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P105.07

Topic: B.05. Neurotransmitter Release

Support: AA020919
DA035958

Title: Role of a6-nicotinic receptors in alcohol-induced GABAergic synaptic plasticity in cholinergic interneurons in the nucleus accumbens

Authors: *H. A. WADSWORTH[1], E. Q. ANDERSON[1], J. K. MOEN[2], A. M. LEE[3], J. MCINTOSH[4], J. WU[5], J. T. YORGASON[1], S. C. STEFFENSEN[1];

[1]Brigham Young Univ., Provo, UT; [3]Pharmacol., [2]Univ. of Minnesota, Minneapolis, MN; [4]Univ. Utah, Salt Lake Cty, UT; [5]Barrow Neurolog Inst., Phoenix, AZ

Abstract: The prevailing view is that enhancement of dopamine (DA) transmission in the mesolimbic system, consisting of DA neurons in the ventral tegmental area (VTA) that project to the nucleus accumbens (NAc), underlies the reward properties of ethanol (EtOH) and nicotine (NIC). We have shown previously that EtOH modulation of DA release in the NAc is mediated by α6-containing nicotinic acetylcholine receptors (α6*-nAChRs), that α6*-nAChRs mediate low-dose EtOH effects on VTA GABA neurons and EtOH preference, and that α6*-nAChRs may be a molecular target for low-dose EtOH. Thus, the most sensitive target for reward-relevant EtOH modulation of mesolimbic DA transmission and the involvement of α6*-nAChRs in the mesolimbic DA reward system remains to be elucidated. The aim of this study was to evaluate EtOH effects on GABAergic modulation of VTA GABA neurons and VTA GABAergic input to cholinergic interneurons (CINs). Injecting DIO channel rhodopsin-2 (ChR2) viral constructs into the VTA of VGAT Cre mice, we found that VTA GABA neurons send an inhibitory projection to CINs, replicating what has been demonstrated by others. Low-dose EtOH enhanced GABAergic input to VTA GABA neurons via typical $GABA_A$ receptors that was blocked by knockdown of α6*-nAChRs by α6shRNA or superfusion of the α6-conotoxin MII[H9A;L15A]. VTA GABA neurons expressing α6*-nAChRs projected to NAc CINs and EtOH reduced GABAergic input to CINs via atypical $GABA_A$Rs. Ethanol enhanced CIN firing rate which was blocked by knockdown of α6*-nAChRs or MII. This study also investigated plasticity at this VTA-NAc GABAergic synapse. The baseline firing rate of CINs was markedly enhanced by knockdown of α6*-nAChRs, CINs were not activated by EtOH in EtOH-dependent mice, and low frequency stimulation (LFS; 1 Hz, 240 pulses) caused inhibitory long-term depression at this synapse (CIN-iLTD) which was blocked by knockdown of α6*-nAChRs, MII, atypical GABA receptor antagonists, and in EtOH-dependent mice. Taken together, these findings suggest that EtOH affects the VTA GABAergic projection to CINs via α6*-nAChRs and that atypical GABA receptors play a role in plasticity associated with chronic EtOH at this synapse.

Disclosures: H.A. Wadsworth: None. **E.Q. Anderson:** None. **J.K. Moen:** None. **A.M. Lee:** None. **J. McIntosh:** None. **J. Wu:** None. **J.T. Yorgason:** None. **S.C. Steffensen:** None.

Poster

P105. Modulation of Synaptic Transmission: Molecular Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P105.08

Topic: B.01. Neurotransmitters, Transporters, and Signaling Molecules

Support: 2R01GM058055-21

Title: Volatile anesthetics interfere with presynaptic cGMP signaling in mouse hippocampal neurons

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Abstract: Volatile anesthetics alter essential presynaptic functions including Ca^{2+} influx and neurotransmitter release, promoting unconsciousness and amnesia. Nitric oxide and cyclic guanosine monophosphate (NO/cGMP) signaling is implicated in several presynaptic mechanisms, and animal models of disrupted NO/cGMP signaling have altered sensitivity to volatile anesthetics. We investigated these pathways in relation to volatile anesthetic action in hippocampal neurons using pharmacological tools and genetically encoded biosensors for interrogating cGMP levels. Using the fluorescent biosensor cGull we found that activity-driven NMDAR-independent presynaptic cGMP transients were depressed by isoflurane or sevoflurane. Isoflurane and sevoflurane depression of action potential-evoked increases in presynaptic Ca2+ and SV exocytosis was blunted in neurons expressing the cGMP scavenger sponGEE, an effect recapitulated by inhibiting HCN1, a cGMP modulated effector that normally facilitates glutamate release. HCN1 had a distinct contribution to isoflurane depression of GCaMP6f-measured presynaptic Ca^{2+} in somatostatin^{+} and VIP^{+} interneurons, which differ in expression of cGMP signaling components. We propose that volatile anesthetics depress presynaptic cGMP signaling and downstream effectors like HCN1 which normally sustain presynaptic function and excitability, enabling volatile anesthetics to depress neurotransmission via second messenger signaling.

Disclosures: I.A. Speigel: None. **V. Osman:** None. **H.C. Hemmings Jr:** None.

Poster

P107. Short-Term Plasticity: Physiology and Modeling

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P107.01

Topic: B.07. Synaptic Plasticity

Support: R35 NS111573

Title: Synaptotagmin-7a potentiates facilitation of $Ca_v2.1$ channels

Authors: *A. DJILLANI, W. CATTERALL;
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Abstract: Voltage-gated calcium channel $Ca_v2.1$ undergoes Ca^{2+}-dependent facilitation and inactivation, which play key roles in short-term and long-term synaptic plasticity. In the

presynaptic terminal, $Ca_v2.1$ channels conduct P/Q-type Ca^{2+} currents and form large complexes with up to one hundred presynaptic proteins, including synaptotagmins. Synaptotagmin-7, a high Ca^{2+} affinity sensor, is essential to mediate synaptic plasticity in several types of synapses. In this study, we have investigated the role that synaptotagmin-7α, the main isoform in the brain, may play in regulation of $Ca_v2.1$ channel facilitation by calmodulin and neuronal Ca^{2+} sensor proteins. We found that synaptotagmin-7α co-immunoprecipitates with the α_{1A} subunit of $Ca_v2.1$ channels in lysates from mouse brain presynaptic membranes and from co-transfected tsA-201 cells through interaction with the synaptic protein interaction (synprint) site in the intracellular linker between domains II and III. Interaction of $Ca_v2.1$ with synaptotagmin-7α potentiates facilitation in several paired-pulse and repetitive-stimulation protocols and accelerates the onset of facilitation. In addition, synaptotagmin-7α induces a significant depolarizing shift in the voltage-dependence of activation of $Ca_v2.1$ and decreases the peak Ca^{2+} current amplitude due to enhanced Ca^{2+}-dependent inactivation. Our results taken together identify a new interaction between $Ca_v2.1$ channels and synaptotagmin-7α in vivo and in vitro, mediated through the synprint site, which potentiates $Ca_v2.1$ facilitation and modulates other aspects of $Ca_v2.1$ channel function in the brain. This unexpected protein interaction reveals a mechanism for convergent regulation of $Ca_v2.1$ function and synaptic facilitation by neural Ca^{2+} sensor proteins and synaptotagmin-7, providing
new insights into the role of $Ca_v2.1$ channel modulation in synaptic plasticity. Supported by NIH Research Grant R35 NS111573.

Disclosures: A. Djillani: None. **W. Catterall:** None.

Poster

P107. Short-Term Plasticity: Physiology and Modeling

Location: Virtual Only

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Program #/Poster #: P107.02

Topic: B.07. Synaptic Plasticity

Support: Wellcome Trust/DBT India Alliance Grant IA/I/12/1/500529
DST/INSPIRE Fellowship/2015/IF150919

Title: Cross-talk between active zones is essential for short-term plasticity in hippocampal mossy fiber

Authors: *N. SINGH, S. NADKARNI;
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Abstract: Granule cells in the dentate gyrus are an important junction for information processing and transfer in the hippocampus. These cells project onto CA3 pyramidal neurons via mossy fibers (MF). MFs have a complex ultrastructural organization: multiple active zones (AZ)

coupled to voltage-dependent calcium channels (VDCC) and an associated readily-releasable pool (RRP) of vesicles. This synapse shows characteristic forms of short-term plasticity (STP) essential for its role as a transmission line and a pattern separator. Upon stimulation it rapidly transitions from a low release probability (Pr) to a high Pr synapse. Loose coupling between active zones and VDCC, and large inter-AZ distance maintains low basal release probability—necessary for large facilitation—despite a large number of AZs. Large RRP size ensures that the vesicles do not deplete at high release probability. However, the implication of this elaborate synaptic design on STP is not well understood.We developed a physiologically realistic spatial model of the MF bouton to explore the role of the structural and biochemical properties that orchestrate synaptic signaling and plasticity. Our study quantitatively describes the synaptic basis for short-term plasticity at MF. MFs, unlike the CA3 presynaptic terminal, have several release sites, each seemingly associated with a distinct source of calcium. Accordingly, most studies consider MF as an extension of the CA3 terminal with multiple autonomous lines of signal transmission. In a direct contrast, our in-silico approach shows not only that calcium crosstalk across multiple release sites exists, but also that this communication is crucial for the observed plasticity. Apart from the coupling between the release sites, the biophysical properties specific to a family of calcium sensors for vesicle release, synaptotagmin-7 (Syt7), and local buffer saturation contribute to the observed facilitation. Furthermore, we show that this synapse can multiplex between the autonomous activity of AZs and coupled activity of AZs during a train stimulus that affects release transients and consequential information transmission.

Disclosures: N. Singh: None. **S. Nadkarni:** None.

Poster

P107. Short-Term Plasticity: Physiology and Modeling

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P107.03

Topic: B.07. Synaptic Plasticity

Title: Enhancement of input frequency dependent information integration in hippocampal granule cell.

Authors: *N. NAKAJIMA[1], T. KAMIJO[2], H. HAYAKAWA[1], T. AIHARA[1];
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Abstract: Hippocampal dentate granule cell (GC) was received spatial (place) and non-spatial (e.g., odor) information via entorhinal cortex (EC). Spatial information is input from medial EC (MEC) through the medial perforant pathway (MPP) to the medial dendrite (MD) of GC. On the other hand, non-spatial information is input from lateral EC (LEC) through the lateral perforant pathway (LPP) to the distal dendrite (DD) of GC. It was known that theta and gamma rhythmic

inputs from MEC and LEC are received MD and DD during associate learning, respectively. However, it is not clear how the GC integrate information for these inputs. First, to investigate the frequency-response characteristics of MD and DD, electrophysiological experiments were performed using rat hippocampal slices. A pulse train consisted of 10 pulses at 10-40 Hz was applied to DD and MD of GC through the LPP and MPP, respectively. D-AP5, NMDA receptor antagonist, was applied to block synaptic plasticity by high frequency stimulation. In addition, picrotoxin, GABAA receptor antagonist was applied. As the physiological experimental result, responses were transiently decreased at MD of GC, in the both cases, with and without inhibitory inputs. On the other hand, DD showed sustained responses to 10-30 Hz. However, when inhibitory inputs were blocked, DD did not show sustained response. Next, computer simulation was performed using a multi-compartment model of the GC by using NUERON simulator. The model was fixed by parameter fitting for the physiological experimental data. MD and DD were applied theta burst (8 Hz) and random pulse input (10-40 Hz, gamma), respectively. As the computational result, the temporal-pattern sensitivity for burst inputs was not clear when theta burst was applied to MD. When random pulse inputs were simultaneously applied to DD at 10 Hz, GC activation for theta burst was enhanced. Moreover, 20Hz-30Hz random pulse inputs were simultaneously applied to DD, GC activation was more facilitated so that temporal pattern sensitivity for theta burst was clearly observed. However, 40 Hz random pulse inputs were simultaneously applied to DD, temporal pattern sensitivity for theta burst was broad. Our results suggest that temporal-pattern processing of spatial information was enhanced by non-spatial information at 10-30 Hz in GC.

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Poster

P107. Short-Term Plasticity: Physiology and Modeling

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Program #/Poster #: P107.04

Topic: B.07. Synaptic Plasticity

Support: National Science Foundation Graduate Research Fellowship under Grant No.DGE-1839285
NIDA P50 award DA044118A

Title: The lateral perforant path as a low-pass filter to the dentate gyrus: substrates and LTP-related interactions

Authors: *J. QUINTANILLA[1], B. G. GUNN[2], J. C. LAUTERBORN[3], Y. JIA[1], C. D. COX[4], C. M. GALL[5], G. LYNCH[1];
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Abstract: Despite its critical role in episodic memory, little is known about how the lateral perforant path (LPP) filters and transforms incoming signals from cortex to hippocampus. The present studies used hippocampal slices from mouse (C57BL6) to investigate such transformations across the first node of the hippocampal circuit, the dentate gyrus (DG), using short trains of stimulation delivered to the LPP at frequencies corresponding to three prominent cortical rhythms. A ten-pulse train applied at 5 Hz (theta frequency) caused a modest enhancement of DG fEPSPs that fell to baseline levels (pulse 10: 97 ± 3% baseline, n=21), while gamma (50 Hz) stimulation quickly drove DG responses to below baseline levels (pulse 10: 57± 3%, n=22). Beta trains (20 Hz) produced intermediate effects (pulse 10: 93 ± 3%, n=22.). Outputs (population-spikes) of the target granule cells underwent parallel but larger within-train changes. By lowering the external calcium levels, we tested whether reducing the initial release probability (*p*) attenuated the within train suppression observed following LPP-stimulation at higher frequencies. Reducing extracellular calcium shifted the gamma frequency response profile from a marked depression to consistent facilitation (pulse 10: 148 ± 8%, n=13) with similar increases to theta (pulse 10: 127 ± 10%, n=10) and beta (pulse 10: 178 ± 10%, n=12) stimulation. Interestingly, treatment with the $GABA_AR$ antagonist, picrotoxin, had no significant influence on the depression seen at gamma stimulation. Within this system, long-term potentiation (LTP) is dependent upon stable presynaptic modifications that result in reductions in the paired pulse ratio (Wang et al. 2016) indicating an increase in *p*. Based on the results mentioned above, an increase in *p* would be expected to enhance train suppression, effectively altering the short-term plasticity within the synapses. Indeed, induction of LTP enhanced LPP responses but, as a result, also exaggerated presynaptic filtering to gamma stimulation (pulse 10: 45 ± 5%, n=7). Together, these findings describe unusual properties of LPP terminals that function as a low pass filter. The within-train suppression of responses to gamma stimulation was associated with factors in the presynaptic element as opposed to the recruitment of GABAergic inhibition, and evidence for enhancement of this suppression with LTP reveals novel consequences to memory-related synaptic plasticity to network function.

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Poster

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Topic: B.07. Synaptic Plasticity

Support: Incheon National University (International Cooperative) Research Grant
Research Grants Council of the Hong Kong Special Administrative Region (11102120)

Title: Dysfunctional longitudinal dentate gyrus network in hearing loss-induced anxiety

Authors: *S. PAK, S. YANG, D. CHO;
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Abstract: Anxiety is a characteristic comorbidity of noise-induced hearing loss (NIHL). NIHL causes physiological changes within the dentate gyrus (DG), a subfield of the hippocampus that is known to modulate anxiety. However, it remains unclear which DG circuit underlies hearing loss-induced anxiety. Here, we utilize an NIHL mouse model to investigate short- and long-term synaptic plasticity in DG networks. Animals with NIHL demonstrate increased anxiety-like behaviors which are similar to those observed in response to chronic restraint stress. This behavior is concurrent with enhanced synaptic responsiveness and the suppression of short- and long-term synaptic plasticity in the longitudinal DG-DG network, but not in the transverse DG-CA3 connection. These findings suggest that DG-related diseases can be typified by synaptic alterations of the longitudinal DG-DG network.

Disclosures: S. Pak: None. **S. Yang:** None. **D. Cho:** None.

Poster

P107. Short-Term Plasticity: Physiology and Modeling

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Program #/Poster #: P107.06

Topic: B.07. Synaptic Plasticity

Support: NSF Grant 1651396

Title: Predictable fluctuations in synaptic strength due to natural variation in presynaptic firing rate

Authors: *N. REN[1], A. GHANBARI[2], G. WEI[1], I. H. STEVENSON[1];
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Abstract: Many *in vitro* studies have shown how synaptic strength can vary with presynaptic spike timing due to short-term synaptic plasticity (STP). Synapses with short-term depression (STD), for instance, should be weaker when presynaptic firing rates are higher and stronger when rates are lower. However, it is still unclear how the natural fluctuations in presynaptic firing affect synaptic strength *in vivo*. Here we examine natural fluctuations in synaptic strength *in vivo* using large-scale spike recordings from the Allen Institute Neuropixels dataset. Using cross-correlations between neuron pairs, we detect >9,000 putative excitatory synaptic connections between ~4 million neuron pairs using a convolutional jitter method. We then use an extension of the Generalized Linear Model to describe each cross-correlogram and to quantify fluctuations in synaptic efficacy over time. For individual connections, we find that the synaptic

efficacy and presynaptic firing rate often vary substantially over time (both measured on a minute timescale): the mean coefficient of variation of the fluctuations in the presynaptic firing rate is 0.41 (sd = 0.25, n = 9800); the mean coefficient of variation of the fluctuations in the presynaptic firing rate is 0.72 (sd = 0.23, n = 9800). For many connections, the efficacy fluctuations mirror fluctuations in the presynaptic firing rate in patterns consistent with short-term synaptic facilitation or depression. In order to understand the potential mechanisms underlying the relationship between fluctuations in presynaptic firing rate and synaptic efficacy, we fit a statistical model of STP to the observed presynaptic and postsynaptic spike trains - a generalized bilinear model (GBLM). The GBLM describes the postsynaptic spiking with 1) a time-varying baseline rate that is fit using a point process adaptive smoothing, 2) a history term that describes the effect of the recent spiking history of the neuron itself, and 3) a time-varying coupling term that describes how the recent spiking of the presynaptic neuron influences the synaptic strength. We find that, for many connections, the GBLM reproduces the observed efficacy fluctuations, suggesting that these fluctuations can be predicted based on the time-varying presynaptic firing rate and can be, at least partly, explained by a cumulative effect of STP.

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Poster

P107. Short-Term Plasticity: Physiology and Modeling

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Program #/Poster #: P107.07

Topic: B.07. Synaptic Plasticity

Support: NSF XSEDE ACI-1548562

Title: Modeling facilitation of asynchronous neurotransmitter release using Monte Carlo simulation of the frog neuromuscular junction active zone

Authors: *R. POAGE[1], G. CHAVIS[1], R. LAGHAEI[2];
[1]Biol., UNC Pembroke, Pembroke, NC; [2]Pittsburgh Supercomputing Ctr., Pittsburgh Supercomputing Ctr., Pittsburgh, PA

Abstract: Computational models allow investigation of presynaptic function in unique ways. Here, we have used a spatially and temporally realistic model of the neuromuscular junction (NMJ) active zone (AZ) that reproduces many *in vivo* experimental findings to investigate molecular events underlying both synchronous and asynchronous fast exocytosis during a short train of action potentials. Some of the critical molecular events believed to underlie neurotransmitter released by an action potential were reproduced in MCell. The resulting model uses spatial and temporal parameters taken from experimental work to assign diffusion and

reaction rates in a Monte Carlo simulation of fast exocytosis at the AZ of the frog NMJ. In prior work by our collaborators, this model showed good agreement with experimental data of synchronous vesicle fusion events that produce endplate potentials at the NMJ. Further. it reproduced the facilitation of synchronous release events seen during and following a short train of physiological stimuli, showing an increased probability of vesicle fusion in the absence of increased calcium influx (Ma et al., 2014; Laghaei et al., 2018). However the ability of the model to recreate the profile of spontaneous (asynchronous) events, presumably those that generate miniature endplate potentials, or mEPPs, have not been fully characterized.Short trains of action potentials applied to the frog NMJ AZ cause the frequency of mEPPS to increase and then to decay to baseline levels with a time course that is best described by a dual exponential (τ_{F1}=47ms ,τ_{F2} =472ms, Zengel & Magleby, 1981). We report that short trains of action potentials applied to this model AZ generate an increase in asynchronous release events. While facilitation of asynchronous vesicle fusion events in response to a single action potential were described well by a single exponential decay, in response to a 100Hz 5-impulse train, the model shows an increase that is better described by a biphasic exponential decay (τ_1=0.44ms, τ_2= 2.3ms) than by any single exponential decay. This decay was more rapid than that observed experimentally.Multiple synaptotagmin 1 and synaptotagmin 7 energy-based rates for calcium ion binding and synaptic vesicle fusion were incorporated into the model, but these modifications of the energy assigned to calcium-binding to these molecules did not greatly improve the model's ability to reproduce asynchronous synaptic activity.

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Poster

P107. Short-Term Plasticity: Physiology and Modeling

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Topic: B.07. Synaptic Plasticity

Support: NIH R00MH092294
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The Whitehall Foudation

Title: Associative learning drives longitudinally-graded presynaptic plasticity of neurotransmitter release along axonal compartments

Authors: *A. STAHL[1], N. NOYES[1], T. BOTO[1], M. JING[2], J. ZENG[3,4,5], Y. LI[7,8,6,4], L. KING[1], R. DAVIS[1], S. TOMCHIK[1];
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Abstract: Anatomical and physiological compartmentalization of neurons is a mechanism to increase the computational capacity of a circuit, and a major question is what role axonal compartmentalization plays. Axonal compartmentalization may enable localized, presynaptic plasticity to alter neuronal output in a flexible, experience-dependent manner. Here we show that olfactory learning generates compartmentalized, bidirectional plasticity of acetylcholine release that varies across the longitudinal compartments of *Drosophila* mushroom body (MB) axons. The directionality of the learning-induced plasticity depends on the valence of the learning event (aversive vs. appetitive), varies linearly across proximal to distal compartments following appetitive conditioning, and correlates with learning-induced changes in downstream mushroom body output neurons (MBONs) that modulate behavioral action selection. Potentiation of acetylcholine release was dependent on the $Ca_V2.1$ calcium channel subunit *cacophony*. In addition, contrast between the positive conditioned stimulus and other odors required the inositol triphosphate receptor (IP_3R), which was required to maintain responsivity to odors in untrained conditions. These data demonstrate that learning drives valence-correlated, compartmentalized, bidirectional potentiation and depression of synaptic neurotransmitter release, which rely on distinct mechanisms and are distributed across axonal compartments in a learning circuit.

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Poster

P125. Synaptic Plasticity: Mechanisms and Behavioral Effects

Location: Virtual Only

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Program #/Poster #: P125.01

Topic: B.07. Synaptic Plasticity

Support: NIH R01 MH123212
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Kavli Neuroscience Discovery Institute

Title: Longitudinal imaging of millions of synapses in vivo reveals that synaptic plasticity in retrosplenial cortex is required for contextual fear learning

Authors: *A. R. GRAVES[1], G. COSTE[1], T. ZHU[1], J. SULAM[2], A. CHARLES[2], R. HUGANIR[1];
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Abstract: Synapses are the fundamental unit of the nervous system. They enable rapid communication between neurons and provide a substrate for plasticity that drives adaptive behavior. Synaptic plasticity, the bidirectional control of synapse strength via dynamic regulation of AMPA receptors, is the leading cellular mechanism underlying many cognitive functions, including learning and memory. However, the distribution of memory encoding, storage, and retrieval within vast networks of synapses remains almost completely unknown. Here, we employ longitudinal in vivo imaging of millions of fluorescently tagged AMPA receptors to directly observe the dynamics of endogenous synaptic networks in cortex throughout the entire process of contextual fear learning. We find that synaptic plasticity in retrosplenial cortex is strongly correlated with the strength of fear memory. Further, we show that blocking synaptic plasticity only in retrosplenial cortex—achieved using a conditional NR1 knockout mouse and targeted injections of AAV-Cre into bilateral retrosplenial cortex—significantly impairs fear memory, suggesting that synaptic plasticity in retrosplenial cortex plays an indispensable role in encoding fear learning and memory. Together, these results represent the most detailed spatiotemporal map of learning to date, uncovering the role of synaptic plasticity with brain-region-wide scale at single-synapse resolution in behaving animals.

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Poster

P125. Synaptic Plasticity: Mechanisms and Behavioral Effects

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P125.02

Topic: B.07. Synaptic Plasticity

Title: Serotonin-dependent mechanism of non-associative learning in earthworms

Authors: *Y. KITAMURA, T. NAKAHARA, H. TAKAHASHI;
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Abstract: We previously reported molecular mechanism of non-associative learning (habituation) in the earthworm. Extracellular recordings from the ventral nerve cord of the earthworm showed that number of action potentials decrease by repeated tactile stimulus to same region of the body wall. It is likely that this habituation is induced due to via serotonin (5-HT) and nitric oxide (NO) signaling, relatively high concentration of 5-HT or NO accelerated habituation. In this study, we further investigated effect of 5-HT receptor antagonists and 5-HT-

related reagent on establishment of habituation. From these results, it is revealed that both 5-HT_2 and 5-HT_3 antagonists inhibit and tryptophan hydroxylase inhibitor delay establishment of habituation. In conclusion, non-associative learning such as habituation in the earthworm by repeated tactile stimulus to the body wall is due to via 5-HT-NO signaling.

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Poster

P125. Synaptic Plasticity: Mechanisms and Behavioral Effects

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Program #/Poster #: P125.03

Topic: B.07. Synaptic Plasticity

Support: NIMHR01MH114888
1R01MH120156

Title: Circadian protein TIMELESS regulates cAMP in hippocampus controlling synaptic plasticity and cognitive memory

Authors: *E. BARRIO-ALONSO[1], M. NOTARAS[1], R. ALBERO[3], Y. BOUCHEKIOUA[2], D. P. CALDERON[2], D. COLAK[1];
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Abstract: Mutations in circadian-rhythm genes are linked to learning and memory deficits. Studies in multiple organisms have revealed that circadian rhythmicity is important for the formation, stability, and recall of memories. This involves the coordinated rhythmic expression of genes, the presence/absence of specific proteins and their active role shaping memory processing. However, the neuronal events underlying the circadian rhythm-mediated regulation of synaptic plasticity and cognitive function is not fully understood.The first set of genes controlling this rhythmicity was described in Drosophila, including TIMELESS, as part of the circadian clock. While mammalian TIMELESS seems to be dispensable in regulating circadian rhythms in the suprachiasmatic nucleus (SCN), TIMELESS does contain a DNA binding domain and has been implicated in DNA damage control. TIMELESS is also indispensable for embryonic development since its embryonic ablation is lethal. Mutations in *Timeless* are associated with numerous cognitive diseases like schizophrenia, autism or familial advanced sleep phase.In this project, we aim to determine, for the first time, the precise role of neuronal TIMELESS in synaptic plasticity and cognitive function in mammals by using conditional TIMELESS mice (*Timeless*$^{fl/fl}$; αCaMKII:CreER^{T2}). TIMELESS deletion led to a poorer performance in short-term spatial memory and contextual fear and tone memory together with significantly increased spine density in TIMELESS CKO hippocampal dendrites compared with

control dendrites *in vivo*. No differences in the sleep phenotype between TIMELESS CKO and control mice could explain these results. In search of the cellular mechanisms, we found that TIMELESS, which follows a circadian regulation in hippocampus, acts as a transcriptional factor that regulates phosphodiesterase 4B (PDE4B) expression. Through PDE4B regulation, we found that TIMELESS regulates cAMP/PKA signaling cascade, leading to a disruption in the AMPA receptor GluA1 phosphorylation activity and therefore affecting the functional properties of the neurons. The results from this study provide insight into the basic principles of how circadian protein TIMELESS participates in the regulation of learning and memory. These results increase our understanding of the etiology of multiple neuropsychiatric/neurodevelopmental diseases.

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Poster

P125. Synaptic Plasticity: Mechanisms and Behavioral Effects

Location: Virtual Only

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Program #/Poster #: P125.04

Topic: B.07. Synaptic Plasticity

Support: NSERC

Title: Immediate early gene induction by trace fear conditioning is preserved in NMDA receptor knockdown mouse hippocampus

Authors: *K. INTSON, R. ISLAM, C. A. MIELNIK, P. ARNOLD, V. MOHN, W. HORSFALL, A. SALAHPOUR, A. J. RAMSEY;
Univ. of Toronto, Toronto, ON, Canada

Abstract: Classical molecular depictions of contextual memory acquisition necessitate NMDA-receptor-mediated induction of immediate early gene (IEG) expression in the hippocampus. Pharmacological NMDA receptor blockade or interference with IEG induction, particularly Arc, greatly impair contextual fear conditioning. Both NMDAR activation and IEG induction are thought to be required for normal contextual fear learning. Here we demonstrate that IEG induction can occur despite dramatic impairments in NMDAR function, and that IEG induction does not correlate with fear learning or memory. Our studies were performed with $Grin1^{KD}$ mice, which have an intron insertion mutation in *Grin1* that causes ~90% knockdown of the gene encoding the obligatory GluN1 subunit for NMDA receptors. Thus, they are a unique model for studying contextual fear memory acquisition in mice with a global hypofunction of NMDA receptors. First, we confirmed using western blot that $Grin1^{KD}$ mice express only 7-15% normal wild-type (WT) protein in the hippocampus. Next, we trained $Grin1^{KD}$ and WT mice to associate a 95dB auditory tone with 0.5mA footshock in a novel context as per typical trace fear

conditioning protocols. Grin1^{KD} mice did not display normal WT freezing behaviour during tone-shock training or when placed in the fear conditioning context 24 hours later. These findings demonstrate expected, profound deficits in contextual fear learning and fear-associated contextual memory in Grin1^{KD} mice. Surprisingly, however, there were no differences in basal or induced IEG expression between WT and Grin1^{KD} mice, including Arc, Fos, Egr1, and Nr4a1. Basal and conditioning-induced CaMKIIa and Dusp1 gene expression also did not differ between WT and Grin1^{KD} mice. Contrary to previous findings, our study suggests that induction of IEG in the hippocampus is not sufficient for contextual memory-related synaptic plasticity, and can occur despite dramatic NMDAR hypofunction. Ongoing experiments are examining whether preservation of the IEG induction pathway in Grin1^{KD}s can be explained by conservation of a subpopulation of NMDARs within their hippocampus, and whether restoration of NMDA receptors to Grin1^{KD} mice in adulthood improves fear conditioning.

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Poster

P125. Synaptic Plasticity: Mechanisms and Behavioral Effects

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P125.05

Topic: B.07. Synaptic Plasticity

Support: MRC Grant MR/N02530X/1
MRC DTP Studentship
Gates Cambridge Stipend

Title: Electrophysiological Signature of the Functional Disengagement of the Basolateral Amygdala Over the Course of the Development of a Cocaine Seeking Habit

Authors: *S. STIEBAHL, S. JONES, D. BELIN;
Univ. of Cambridge, Cambridge, United Kingdom

Abstract: The use of cocaine and heroin among young Europeans and Americans has been steadily increasing over the past decade. As such it has never been more important to understand the mechanisms whereby some individuals switch from recreational controlled drug use to habitual and eventually compulsive drug seeking and taking, the hallmark of addiction. In humans, non-human primates, and rodents a shift occurs in the neural locus of control over behaviour from the ventral to the dorsolateral striatum (DLS) over the course of a history of drug use which sets the stage for the development of compulsivity and the tendency to relapse. We have recently shown that this functional shift is driven by a functional transition within the amygdala from the basolateral nucleus (BLA), which is involved in goal-directed behaviour, to

the central amygdala (CeN) which is involved in habits. In rats trained to seek cocaine under the influence of the conditioned reinforcing properties of a cocaine-paired cue, as measured under a second order schedule of reinforcement (SOR), cocaine seeking initially depends on the BLA - Acbc circuit, but when it becomes well-established and habitual, drug seeking relies on dopaminergic mechanisms in the anterior dorsolateral striatum (aDLS), the persistence of which depends on the CeN. The mechanisms underlying these intra-amygdala transitions have not been elucidated. In order to address this progressive shift from early to established cocaine seeking, male Sprague-Dawley rats were trained to seek cocaine under a SOR for 3 or 21 days (N=6 each), time points at which drug seeking is known to be independent of, or mediated by the CeN - aDLS habit system, respectively. Whole cell slice patch-clamp electrophysiology was used to record from pyramidal BLA (N=18) and CeN (N=20) neurons of both groups to determine their intrinsic excitability and spontaneous synaptic activity. While the activity and excitability of CeN neurons did not change between day 3 and day 21, that of BLA neurons was significantly decreased after long-term cue-controlled cocaine seeking. These results show that the switch in neural control over drug seeking when it becomes habitual is accompanied by a decrease in BLA activity which may enable control by the. These findings could indicate an amygdala mechanism by which to re-engage an otherwise downregulated A-O system in addiction.

Disclosures: S. Stiebahl: None. **S. Jones:** None. **D. Belin:** None.

Poster

P125. Synaptic Plasticity: Mechanisms and Behavioral Effects

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P125.06

Topic: B.07. Synaptic Plasticity

Support: Human Frontier Science Program (RGY0069)
European Research Council (Starting Grant FLEXNEURO 716643)
Leverhulme Trust (ECF-2020-352)
Issac Newton Trust

Title: Self-healing neural codes: Hebbian and homeostatic mechanisms can track evolving neural representations

Authors: *M. E. RULE, T. O'LEARY;
Engin., Univ. of Cambridge, Cambridge, United Kingdom

Abstract: Until recently, it was believed that homeostatic processes maintain single-neuron function, stabilizing long-term neural representations in the presence of molecular and synaptic turnover. However, recent experiments have revealed widespread instability not only in the structure of single neurons, but also in their computational function. This "representational drift[1]"

has been found throughout the brain, and occurs even in neural codes for habitual tasks. We examine a type of representational drift observed in the hippocampus[2] and parietal cortex[3]. These areas exhibit population codes with low-dimensional manifold structure that correlates with behavioral variables. However, the relationship between this low-dimensional structure and single-neuron firing rates is unstable, and the neural population code reconfigures over long timescales. Previous works found that such drift is compatible with stable network function in the presence of ongoing supervised[4] or unsupervised[5] learning, and that error correction could stabilize codes that employ discrete, binary neural assemblies[6,7]. We extend these ideas to the case of continuous, low-dimensional manifold representations. We show that drifting codes can be tracked readily via reactivation rather than ongoing relearning. We find that homeostasis and unsupervised Hebbian learning interact to allow neurons to error-correct and track unstable population codes, conferring long-term stability without relying on an external reference. These results suggest that long-term stable learned associations are compatible with continual, and complete, reconfiguration of neural population codes. This highlights a role for Hebbian plasticity in stability as well as learning, and the importance of homeostatic processes in maintaining distributed representations.
References: [1]Rule et al. (2019) Current opinion in neurobiology 58:141-147; [2]Ziv et al. (2013) Nat. Neuroscience 16.3:264-266; [3]Driscoll et al. (2017) Cell 170.5:986-999; [4]Rule et al. (2020) Elife 9:e51121; [5]Gillett et al. (2020) PNAS 117.47:29948-29958; [6]Kossio et al. (2020) bioRxiv; [7]Fauth & van Rossum (2019) ELife 8:e43717.

Disclosures: M.E. Rule: None. **T. O'Leary:** None.

Poster

P125. Synaptic Plasticity: Mechanisms and Behavioral Effects

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P125.07

Topic: B.07. Synaptic Plasticity

Support: NIH Grant R01NS095123

Title: Synaptic evidence for the cumulative effects of weak direct current stimulation in spaced learning in rat hippocampus

Authors: *M. SHARMA[1], F. FARAHANI[2], M. BIKSON[3], L. C. PARRA[2];
[2]Biomed. Engin., [3]Dept of Biomed. Engin., [1]City Col. of New York, New York, NY

Abstract: Electric fields generated during direct current stimulation (DCS) are known to modulate activity-dependent synaptic plasticity in-vitro. This provides a mechanistic explanation for the lasting behavioral effects observed with transcranial direct current stimulation (tDCS) in human learning experiments. However, previous in-vitro synaptic plasticity experiments show

relatively small effects despite using strong fields compared to what is expected with conventional tDCS in humans (20 V/m vs. 1 V/m). Leveraging the spaced learning paradigm, we found that DCS applied during repeated bouts of theta burst stimulation (TBS) resulted in an increase of LTP. Weaker TBS and the weakest electric fields of 2.5 V/m resulted in the strongest relative efficacies (12% boost in LTP per 1 V/m applied). These results bridge the gap in terms of efficacy between in-vitro DCS and human tDCS experiments.

Disclosures: M. Sharma: None. **F. Farahani:** None. **M. Bikson:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Soterix Medical LLC. **L.C. Parra:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Soterix Medical LLC.

Poster

P181. Brain Wellness and Aging: Dietary and Alternative Therapies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P181.01

Topic: C.01. Brain Wellness and Aging

Support: Health and Medical Research Fund (Ref: 15161201)
Central Research Fund (CRF, Ref: G-YBWJ & G-YBR0)

Title: Effects of electroacupuncture on post-operative cognitive dysfunction in mice

Authors: *J. Y.-S. HO[1], R. C. CHANG[2];
[1]Sch. of Nursing, The Hong Kong Polytechnic Univ., Hong Kong, China; [2]Lab. of Neurodegenerative Diseases, LKS Fac. of Medicine, Univ. of Hong Kong, Hong Kong, China

Abstract: Postoperative cognitive dysfunction (POCD) is a common complication after surgery which can have a profound impact on the patients. Clinical data suggest that acupuncture before and after surgery may be useful to reduce the cognitive decline. Limited data is available to explain the underlying mechanism. We hypothesize that electroacupuncture (EA) can attenuate inflammation to reduce cognitive dysfunction after surgery. Male C57BL/6N mice (12 weeks old) were divided into four groups: control, laparotomy only, laparotomy + EA, laparotomy + ibuprofen. The EA group mice received EA at Baihui (DU20) and Zusanli (ST36) for 20 minutes each day for 7 or 14 days after surgery. The other groups received physical restriction only. The ibuprofen group served as a positive control. We found that laparotomy caused impairment in recognition memory and associated memory as demonstrated by the novel object recognition test and the Y-maze test. The cognitive decline was attenuated both in the EA and ibuprofen-treated mice. Glial activation was also reduced in EA treated group. Western blot analysis demonstrated that EA could modulate the expression of some synaptic proteins and the phosphorylation of tau

protein. Taken together, our results suggested that EA given after surgery could reduce POCD and the suppression of neuroinflammation may play an important role in its action.

Disclosures: J.Y. Ho: None. **R.C. Chang:** None.

Poster

P181. Brain Wellness and Aging: Dietary and Alternative Therapies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P181.02

Topic: C.01. Brain Wellness and Aging

Title: Do we swallow the waste from our brain?

Authors: P. KULKARNI[1], J. LEASTON[1], C. A. GHARAGOUZLOO[2], J. QIAO[1], N. BENS[1], C. F. FERRIS[1];
[1]Ctr. for Translational Neuro-Imaging, Northeastern Univ., Boston, MA; [2]Imaginostics, Inc., Cambridge, MA

Abstract: Ferumoxytol an iron oxide nanoparticle was infused into the lateral cerebroventricle of awake rats to follow its movement and clearance from the brain using MRI. Within minutes the contrast agent could be observed accumulating on the soft palate at the back of the throat (Fig 1). In a subsequent study fluorescent quantum dots were infused into the brain of rats and within 15 min could be observed in the esophagus using microscopy (Fig 2). These imaging studies clearly show the gut is helping to clear waste from the brain.

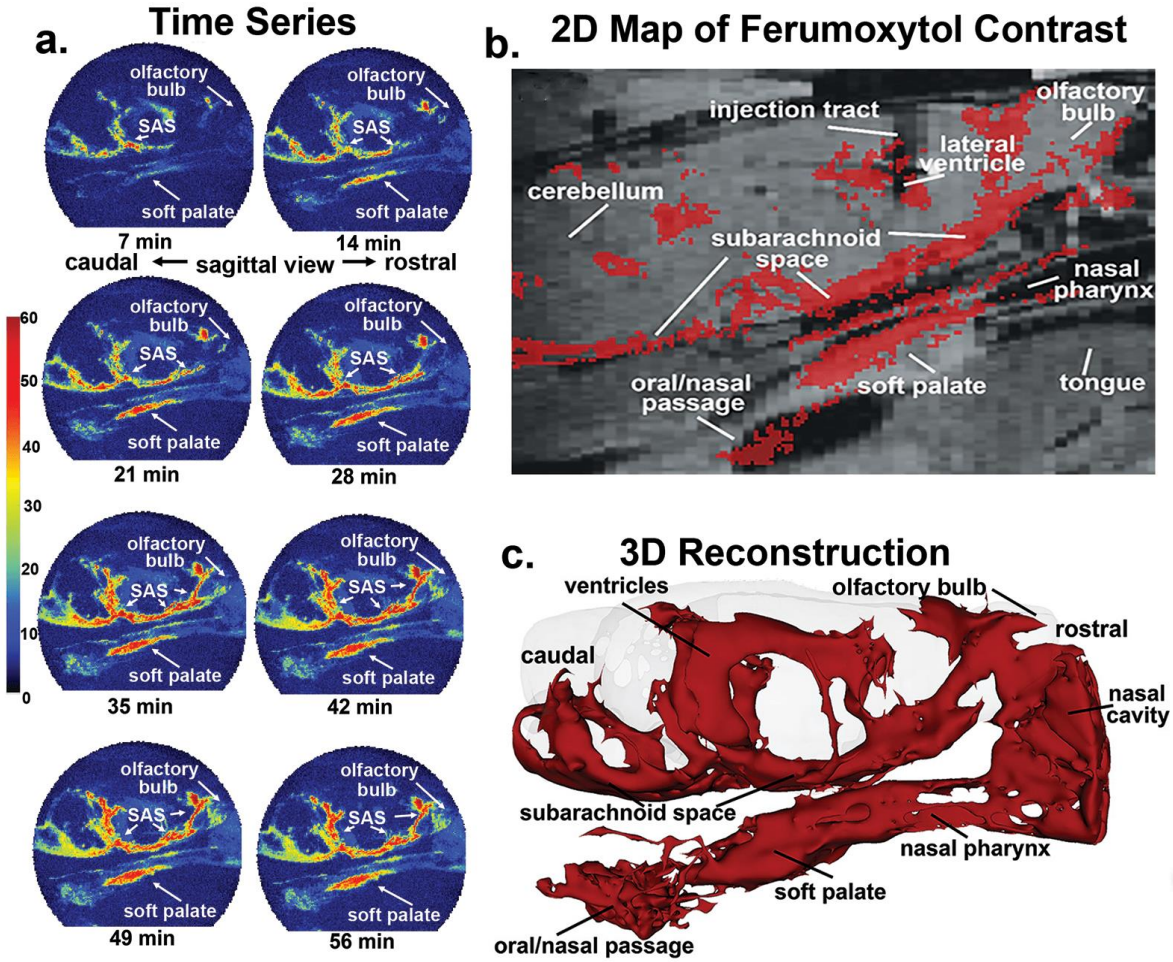


Fig 1. Sagittal reconstructions of ferumoxytol movement and accumulation. The time series (a.) are images from a single rat collected at 7 min intervals over 70 min showing accumulation of CA along the soft palate over time. A summary image (b.) generated as an average of CA signal from six rats is mapped onto the T2 weighted anatomy image of the brain. The 3D reconstruction of CA within and outside the brain was generated from a single rat, postmortem, at the end of the imaging study to eliminate the artifact of blood vessels.

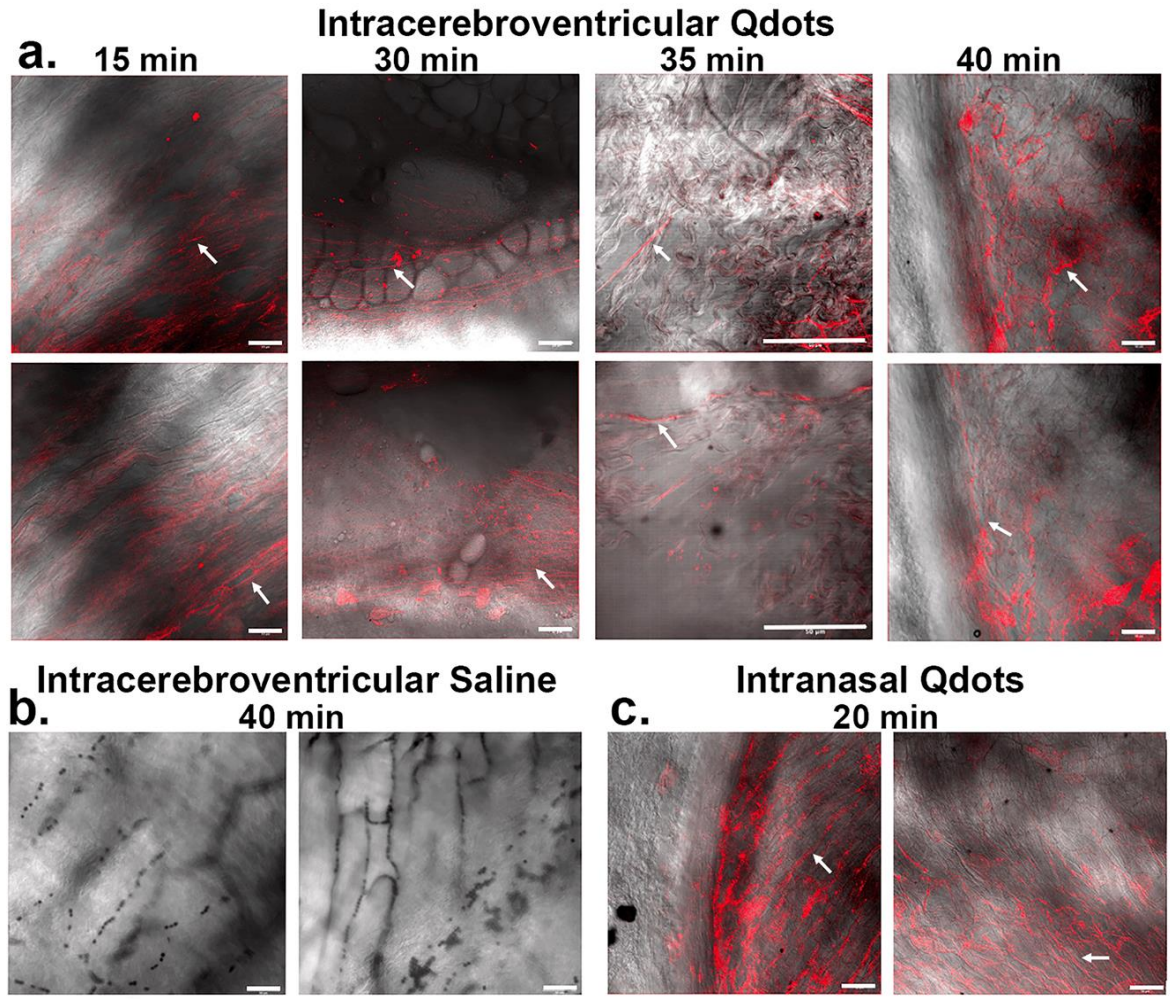


Fig 2. Fluorescent images of the lumen of the upper esophagus. Shown in the upper panel (a.) photomicrographs of the lining of the esophagus from ex-vivo tissue taken at different times following ICV infusion of Qdots. The lower left panel (b.) shows photomicrographs of esophagus from a male rat infused with 10 µl of 0.9% NaCl (saline). The lower right panel (c.) shows the presence of Qdots in the esophagus of a male rat following 1µl injections of Qdots into each nostril.

Disclosures: P. Kulkarni: None. **J. Leaston:** None. **C.A. Gharagouzloo:** A. Employment/Salary (full or part-time):; Imaginostics, Inc.. **J. Qiao:** None. **N. Bens:** None. **C.F. Ferris:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Animal Imaging Research.

Poster

P181. Brain Wellness and Aging: Dietary and Alternative Therapies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P181.03

Topic: C.01. Brain Wellness and Aging

Title: A resveratrol analog modulates TRH and TRH-like peptide expression throughout the brain and peripheral tissues of male rats

Authors: *A. PEKARY[1], A. SATTIN[2];
[2]Res., [1]VA Greater Los Angeles Healthcare Syst., Los Angeles, CA

Abstract: Resveratrol and related polyphenols have therapeutic effects ranging from treatment of depression, Alzheimer's and Parkinson's disease, obesity, neurodegeneration and aging. TRH and TRH-like peptides, with the structure pGlu-X-Pro-NH_2, where "X" can be any amino acid reside, have reproductive, caloric-restriction-like, anti-aging, pancreatic-β cell-enhancing, cardiovascular, and neuroprotective effects. The TRH/TRH-R1 receptor signaling pathway is an important mediator of brain-gut axis communication via the brain medulla. TRH and TRH-like peptides occur not only throughout the CNS but also peripheral tissues, with particularly high levels in rat and human prostate. Female rats were not studied because TRH and TRH-like peptide levels fluctuate 10- to 100-fold in response to the estrus cycle. For these reasons 16 young adult male Sprague-Dawley rats were divided into 4 groups. The control (CON) group remained on ad libitum chow and water for 10 days. The acute (AC) group received ad libitum chow and water for 9 days and then 0.9 g pterostilbene (PT)/250 g rat chow for 24 h. The chronic (CHR) animals received PT in chow for 10 days. The withdrawal (WD) rats received PT chow for 8 days and then normal chow for 2 days. TRH and TRH-like peptide levels were measured in medulla oblongata (MED), frontal cortex (FCX), hypothalamus (HY), amygdala (AY), hippocampus (HC), piriform cortex (PIR), nucleus accumbens (NA), entorhinal cortex (ENT), striatum (STR), cerebellum (CBL), anterior cingulate (ACNG), posterior cingulate (PCNG), prostate (PR), liver (L), testis (T), heart (H), pancreas (PAN), adrenals (AD) and epididymis (EP). Highly significant changes in the levels of TRH and TRH-like peptides occurred throughout the brain and peripheral tissues in response to PT treatment. In brain the number of these changes (in parentheses) declined in the order: PIR(16), HY(15), STR(14), PCNG(12), MED(11), ACNG (10), NA(10), AY(9). In peripheral tissues the corresponding order was: EP(17), PR(13), AD(8), T(4), H(0). We conclude the high responsiveness of PIR, CBL, HY, STR, PCNG, MED, FCX, NA, ACNG and AY in brain and EP and PR is consistent with TRH and TRH-like peptides participating in the therapeutic effects of PT.

Disclosures: A. Pekary: None. **A. Sattin:** None.

Poster

P181. Brain Wellness and Aging: Dietary and Alternative Therapies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P181.04

Topic: C.01. Brain Wellness and Aging

Title: Comparative effects of avocado (seed & pulp) on d-galactose induced brain aging in albino rats

Authors: *S. TABASSUM[1], P. TIKMANI[2], *M. SHEIKH[4], S. F. CHOHAN[4], S. AHMAD[3], S. HAIDER[5];
[1]SZABIST, Karachi, Pakistan; [3]BBS, [2]Agha Khan Univ., Karachi, Pakistan; [4]SZABIST, Karachi Campus, Karachi, Pakistan; [5]Biochem., Univ. of Karachi, Karachi, Pakistan

Abstract: COMPARATIVE EFFECTS OF AVOCADO (SEED & PULP) ON D-GALACTOSE INDUCED BRAIN AGING IN ALBINO RATS**Saiqa Tabassum[1*], Prashant Tikmani[1], Madiha Shaikh[1], Sameen Fatima Chohan[1], Saara Ahmad[2], Saida Haider[31]**Department of Biosciences, Faculty of Life Science, SZABIST, Karachi[2]Department of Biological and Biomedical Sciences, Agha Khan University, Karachi[3]Neurochemistry and Behavioral Neuropharmacology Research Unit, Department of Biochemistry, University of Karachi, Karachi**Abstract:**Aging is a well-known physiological process of getting older. It has a negative impact on functional abilities of an individual leading to detrimental deficits like cognitive dysfunction. Many factors contribute in brain aging particularly mitochondrial dysfunction characterized by a decrease in the activity of respiratory chain enzymes and ATP production leading to increased free radical generation, mitochondrial deoxyribonucleic acid (DNA) mutations, and impaired mitochondrial structures. D-galactose induced rodent brain aging model, used by previous researchers, is developed by progressive deterioration of brain cells via mitochondrial dysfunction increased oxidative stress, inflammation, and apoptosis contributing to cognitive decline. Persea americana, commonly known as Avocado, is a large berry-shaped fruit which is enriched with lutein and is helpful in improving brain functions. Lutein is a carotenoid which is commonly found in fruits and vegetables that accumulates in the blood, eye and brain. It may act as an antioxidant. So, it can be hypothesized that Persea americana might have neuroprotective properties and it might help in preventing brain aging. For this purpose, aging in rats was induced by D-galactose via oral administration for 6 weeks followed by supplementation of Avocado pulp and seed extract. After induction and treatment, neurobehavioral analysis was conducted followed by decapitation and brain dissection to collect hippocampus and cortex for neurochemical and histological analysis. Results showed that compared to avocado seed extract, avocado pulp is effective in improving spatial memory, muscle strength, anxiety, locomotion, depression and episodic memory and cognitive performance which is further validated by histopathological examination and alteration in levels of GABA and Glutamate. Thus, avocado pulp is found to be effective in preventing D-galactose-induced brain aging and can be suggested as a anti-aging supplement to improve neurological functioning.

Disclosures: S. Tabassum: None. **P. Tikmani:** None. **M. Sheikh:** None. **S.F. Chohan:** None. **S. Ahmad:** None. **S. Haider:** None.

Poster

P181. Brain Wellness and Aging: Dietary and Alternative Therapies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P181.05

Topic: C.01. Brain Wellness and Aging

Title: Novel adult brain processing method reveals age-, sex-, and species-dependent effects of pharmaceutical compounds on neuron survival and neuritogenesis.

Authors: *A. SEFIANI[1], C. G. GEOFFROY[2];
[1]Texas A&M Univ. Syst. Hlth. Sci. Ctr., College Station, TX; [2]Neurosci. and Exptl. Therapeut., Texas A&M, Bryan, TX

Abstract: Neurodegenerative diseases and neurotraumatic injuries are typically age-associated disorders that can reduce neuron survival, axon and neurite outgrowth, and synaptic plasticity leading to loss of cognitive capacity, executive function, and motor control. In pursuit of reducing the loss of said neurological functions, novel compounds must be identified that augment neuron survival, axon regeneration/neuritogenesis, and synaptic plasticity. However effective high-throughput in-vitro screenings are at finding compounds, most screens use iPSC derived, embryonic, or post-natal neurons. This is a major issue as they are likely to have different characteristics than the targeted neurons in clinical settings. Indeed, the majority of the patients suffering from neurodegenerative diseases and neurotrauma are of middle-age and older. This dichotomy in age between the neurons used in drug screens and patients certainly impedes chances of translational success. It has been historically challenging to culture adult neurons let alone conduct screenings; therefore, age-appropriate drug screenings have previously not been plausible. Thus, we have developed a novel adult neuron processing method and created the first high content morphology-based screening system using adult cortical neurons. This novel method allows for rapid and economical mass processing of large mammalian brains to maximize neuron survival for increased screening capacity. After conducting targeted screens utilizing cortical neurons from 2-year-old sheep (equating to 20-25 years old in humans) and mice of all ages, we have discovered age-, sex-, and species-dependent effects of compounds on neuritogenesis and neuron survival. Demographic-specific screens are expected to 1) prevent the premature dismissal of compounds with no effect in iPSC/embryonic screens yet would be advantageous to adults, 2) reduce the false positivity rate by efficiency vetting compounds with no benefit to adult neurons, and 3) provide quick multi-species validation to better predict success in clinical settings. Therefore, utilizing age-, sex-, and species-appropriate in-vitro models to find novel compounds increasing neuron survival and neurite outgrowth, made possible by our novel neuron processing method, will greatly increase the probability of translational success.

Disclosures: A. Sefiani: None. **C.G. Geoffroy:** None.

Poster

P181. Brain Wellness and Aging: Dietary and Alternative Therapies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P181.06

Topic: C.01. Brain Wellness and Aging

Support: University of North Carolina President Strategic Initiatives Awa
NIH grant 5R25GM077634

Title: Pharmacologic and non-pharmacologic enhancement avenues targeting the autophagy-lysosomal pathway increase cathepsin B activity, stimulate autophagic flux, and improve synaptic resilience

Authors: *M. F. ALMEIDA, K. M. RENTSCHLER, J. L. NGUYEN, I. GRIJALVA, M. A. DOS SANTOS, M. H. GIANG, K. L. G. FARIZATTO, U. S. IKONNE, B. A. BAHR; William C. Friday Laboratory, Biotech. Res. and Training Ctr., Univ. of North Carolina, Pembroke, Pembroke, NC

Abstract: Exercise and a healthy diet promote cognitive health, influencing the autophagy-lysosomal protein degradation pathway that is also manipulated by synaptic activity and important for memory (Kulkarni et al. 2021 *J Cell Biol* 220:e202002084). The growing incidence of proteinopathies warrants efforts to understand the crosstalk between protein clearance pathways and the targets for modulation (Farizatto et al. 2017 *PLoS ONE* 12:e0182895; Boland et al. 2018 *Nat Rev Drug Discov* 17:660). Interestingly, myokines irisin and cathepsin B (CatB) are elevated in human plasma by exercise of the type that increases BDNF expression and improves memory (Wrann 2015 *Brain Plast* 1:55; Moon et al. 2016 *Cell Metab* 24:332). Irisin was shown to rescue plasticity and memory functions in Alzheimer's disease (AD) mice (Lourenco et al. 2019 *Nature Med.*25:165) and CatB positive modulation increases protein clearance and cognitive measures in models of AD, α-synucleinopathy, and mild cognitive impairment (Mueller-Steiner et al. 2006*Neuron* 51:703; Butler et al. 2011 *PLoS ONE* 6:e20501; Wang et al. 2012 *JBC* 287:39834; Embury et al. 2017 *J Neuroimm Pharm* 12:340; Hwang et al. 2019 *Inter J Mol Sci* 20:4432). Here, we tested whether synapto-protective CatB modulation elicited by peptidyl and non-peptidyl modulators occurs via treatment with plant-derived flavanols, different ginseng extracts, and other plant extracts, to address how diet components influence the autophagy-lysosomal pathway for cognitive maintenance. A select group of the natural products enhanced the 30-kDa active form of the CatB protease (CatB-30) up to 300% in hippocampal explants. Derivatives like BW1002 ([3-(acetyl-benzyl-amino)-1-benzyl-2-hydroxy-propyl]-carbamic acid benzyl ester) typically produced 2-3-fold more CatB modulation than the natural agents. Flavonoid derivatives caused positive effects on the active CatB-30 and synaptic markers. Both pharmacological and non-pharmacological agents were

identified with strong correlations between increased CatB substrate degradation and synaptic resilience indicators. In addition, members of the two groups enhanced the LC3I to LC3II conversion which was confirmed to occur in the presence of the lysosomal inhibitor chloroquine. They also displayed close correspondences between increased levels of CatB-30 and beclin-1, an essential factor for initiation of autophagy, whereas reduced levels were found for p62, a selective autophagy substrate. Thus, the findings indicate that a diverse array of molecules can elicit beneficial effects on the autophagy-lysosomal pathway, perhaps in relation to the role diet plays in supporting brain health and cognition.

Disclosures: M.F. Almeida: None. **K.M. Rentschler:** None. **J.L. Nguyen:** None. **I. Grijalva:** None. **M.A. Dos Santos:** None. **M.H. Giang:** None. **K.L.G. Farizatto:** None. **U.S. Ikonne:** None. **B.A. Bahr:** None.

Poster

P181. Brain Wellness and Aging: Dietary and Alternative Therapies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P181.07

Topic: C.01. Brain Wellness and Aging

Support: USDA intramural funding
Tufts University
NASA

Title: Blueberry extract attenuates neuroinflammation and oxidative stress when administered before and/or after lipopolysaccharide in rat microglial cells

Authors: D. S. CAHOON, D. FISHER, *B. SHUKITT-HALE;
USDA-ARS Human Nutr. Res. Ctr. on Aging, Boston, MA

Abstract: Microglia, the primary immune cells of the central nervous system (CNS), are key regulators of inflammation and oxidative stress (OS) in the CNS. In response to stress, activated microglia produce inflammatory cytokines and oxidants to eliminate pathogens and initiate repair. However, overactivation of microglia can be neurotoxic, resulting in reactive microgliosis, a feed-forward amplification of inflammation and neuronal damage. Therefore, microglial activation can continue after removal of the initiating stimulus, leading to chronic inflammation, OS, and neurodegeneration. Blueberries (BB), rich in anti-inflammatory and antioxidant polyphenols, have been shown to reduce inflammation and OS when administered to microglia before stressors such as lipopolysaccharide (LPS), but the therapeutic value of BBs administered after microglial activation has not been examined. This study investigated the differential effects of pre-, post-, and pre-/post-BB on LPS-induced inflammation and OS in microglia.

HAPI rat microglia were pre-treated with freeze-dried BB diluted in media (0.5 mg/mL) or control media (C) for 24h, incubated overnight with LPS (0 or 200 ng/mL), and post-treated with BB or C for 24h. Griess methods, ELISA, and Western blot were used to measure biomarkers of inflammation (nitrite, tumor necrosis factor-α [TNFα], inducible nitric oxide synthase [iNOS], cyclooxygenase-2 [COX-2], phosphorylated IKBα [pIKBα]) and OS (NADPH oxidase [NOX2]). Data were analyzed using two-way ANOVAs with BB treatment period and LPS exposure as experimental factors. Post-hoc testing was performed using Tukey's test.
Results showed that LPS increased nitrite, TNFα, iNOS, COX2, pIKBα, and NOX2 compared to non-stressed control conditions ($P < 0.05$) and BB before and/or after LPS significantly reduced all markers compared to no BB ($P < 0.05$). BB treatment period did not produce significantly different attenuating effects on LPS-induced iNOS, pIKBα, and NOX2. However, pre-BB was more effective than post-BB at reducing nitrite, TNFα, and COX2 ($P < 0.05$). Pre-BB was also more effective than pre/post-BB at attenuating LPS-induced nitrite and TNFα ($P < 0.05$).
Results suggest that BBs can target the downstream effects of microglial activation in addition to preventing stressor-induced neuroinflammation and OS. Therefore, although potentially more effective, BBs may not need to be present prior to microglial activation for beneficial effects. These findings provide evidence to support that dietary interventions may be effective even after initiation of disease processes such as microglial-induced neuroinflammation and OS.

Disclosures: D.S. Cahoon: None. **D. Fisher:** None. **B. Shukitt-Hale:** None.

Poster

P210. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P210.01

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Phase-amplitude decoupling in an animal model of Alzheimer's disease

Authors: J. SU[1], S. YANG[1], H. WU[1], Z. LI[1], J. CAO[1], J. LI[1], Y. WANG[1], Z. ZHANG[1], C. ZHANG[1], Q. ZHU[1], P. ZHANG[1], H. LU[1], ***K. RUTTEN**[2], D. XU[1];
[1]Biol., WuXi AppTec, Shanghai, China; [2]Res. Service Div., WuXi AppTec, London, United Kingdom

Abstract: The clinical relevance of animal models for diseases, in terms of etiology, symptoms and diagnostic tests, is critical to preclinical efficacy studies. There are as many as ~200 animal models of Alzheimer's Disease (AD) with various symptomatic manifestations. However, the test methods are largely restricted to cognitive behaviors, histopathology and biomarkers, which are either pharmacologically indicative or experimentally terminal.
In an earlier clinical study, it was observed that the phase-amplitude coupling (PAC) of θ and γ oscillations (cross-frequency coupling of brain electrical activities) is impaired in AD patients,

and that the decoupling is progressively associated with dementia (Goodman et al., 2018). We followed the hippocampus local field potential in the 5x FAD mouse model of AD for 12 months, and observed abnormal power and modulation of different oscillations with the progression of disease. In particular, the onset of the aforementioned PAC decoupling was observed as early as in 4 month old model animals, and became almost completely diminished at 12 month. Although histopathological findings (plaque and gliosis) were seen as early as 4 month, cognitive deficiency became significant only at 9-12 month.
Together, our results suggest that PAC decoupling could be used in animal models to monitor the progression of AD. Since the onset of PAC decoupling occurs much earlier in comparison with cognitive behavioral deficiency, it provides a new method to predict the clinical outcome of AD for disease progression and therapeutic efficacy. The clinical relevance of the 5x FAD model and the PAC decoupling further suggests the predictive power of the latter not only in preclinical pharmacological tests but as well clinical tests.

Disclosures: J. Su: None. **S. Yang:** None. **H. Wu:** None. **Z. Li:** None. **J. Cao:** None. **J. Li:** None. **Y. Wang:** None. **Z. Zhang:** None. **C. Zhang:** None. **Q. Zhu:** None. **P. Zhang:** None. **H. Lu:** None. **K. Rutten:** None. **D. Xu:** None.

Poster

P210. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P210.02

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: R01AG042178
01AG47812
R01NS105473
AG060767
AG069333
AG066347
K99AG065645

Title: Novel microRNA-455-3p mouse models to study Alzheimer’s disease pathogenesis

Authors: *S. KUMAR[1], H. MORTAN[2], N. SAWANT[2], E. ORLOV[2], L. BUNQUIN[2], P. JANGAMPALLI[3], H. REDDY[2];
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Abstract: Novel microRNA-455-3p mouse models to study Alzheimer’s disease pathogenesis Subodh Kumar[1*], Hallie Mortan[1], Neha Sawant[1], Erika Orlov[1], Lloyd Bunquin[1],

Pradeep Adi Jangampalli[1] and P. Hemachandra Reddy[1-5]1. Internal Medicine Department, 2. Neuroscience & Pharmacology, 3. Neurology, 4. Public Health Department, 5. Speech, Language and Hearing Sciences, School Health Professions, Texas Tech University Health Sciences Center, Lubbock, TX, 79430 USA.***Subodh Kumar will be the presenting author**
Introduction: The purpose of our study is to determine the protective role of microRNA-455-3p in Alzheimer's disease (AD) progression. MicroRNA-455-3p is one of the highly conserved miRNAs involved in several human diseases but newly explored by our lab in AD. Our past studies unveiled the biomarker and therapeutic potentials of miR-455-3p in AD. Our *in vitro* study exhibited the protective roles of miR-455-3p against AD toxicities in terms of reducing full-length APP and amyloid-β (Aβ) protein levels, inducing defective mitochondrial biogenesis, impaired mitochondrial dynamics and synaptic deficiencies. **Methods:** We sought to determine the essential roles of miR-455-3p in AD using mouse models. Therefore, for the first time, we generated both miR-455-3p (+) transgenic (TG) and miR-455-3p (-) knockout (KO) mouse models. **Results:** We determined the positive and negative effects of miR-455-3p on mice cognitive function, AD proteins, mitochondrial biogenesis, mitochondrial dynamics, mitochondrial quality (number and length), dendritic spine density, synapse numbers and synaptic activity. Morris water maze test showed improved cognitive behavior, spatial learning and memory in miR-455-3p TG mice relative to KO and wild type (WT) mice. AD-related proteins were significantly altered in miR-455-3p TG mice relative to WT and KO mice. Further, mitochondrial biogenesis/dynamics and synaptic activities were enhanced in miR-455-3p TG mice, while these were reduced in KO mice. Overall, miR-455-3p TG mice displayed protective impacts and miR-455-3p KO mice exhibited deleterious effects in relation to AD pathogenesis. **Conclusions:** Both mouse models could be ideal research tools to understand the molecular mechanism of miR-455-3p in other human diseases including AD.
Keywords: Alzheimer's disease, MicroRNA-455-3p, Mouse models, Mitochondrial biogenesis, Synaptic activity

Disclosures: S. Kumar: None. **H. Mortan:** None. **N. Sawant:** None. **E. Orlov:** None. **L. Bunquin:** None. **P. Jangampalli:** None. **H. Reddy:** None.

Poster

P210. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P210.03

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: PAPIIT-DGAPA IN216821

Title: Effect of vanadium pentoxide (V_{2O5}) inhalation on memory and cytoskeleton alterations in brain structures related to Alzheimer disease

Authors: C. DORADO-MARTINEZ[1], E. MONTIEL-FLORES[2], J. ORDOÑEZ-LIBRADO[1], A. GUTIÉRREZ-VALDEZ[2], O. MEJÍA-GARCÍA[1], J. ESPINOSA-VILLANUEVA[2], L. REYNOSO-ERAZO[2], *M. AVILA-COSTA[2];
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Abstract: Alzheimer's disease (AD) is the most common neurodegenerative pathology worldwide; it has been reported that approximately 15 million people suffer from this disease. The incidence annually increases 0.5% in 65 and 8% in 85 years old. However, it was described that more than 100 years ago, and there is a lot of research being done about this pathology, it has been challenging to find an animal model that replicates all the characteristics of the neurodegenerative process of Alzheimer's. Previous experiments in our laboratory have shown that chronic exposure to vanadium pentoxide in rats causes morphological and behavioral changes similar to those seen in AD. To this end, 20 male Wistar rats were randomly divided into control and experimental group (n = 10) with an initial weight of 180-200 gr. The experimental group was exposed to 0.02M V2O5 1 h, 3 times a week, for 6 months. The two groups were trained in the T-maze test to determine behavioral changes that assess spatial behavior and an open-field test for 10 mins. Both groups were evaluated once a month for 6 months. After 6 months of inhalation, the hippocampal CA1 underwent argentic Bielschowsky impregnation and was analyzed to determine histological alterations. The T-maze show memory impairment since three months of V2O5 inhalation. During the open field test, differences were observed in the locomotion patterns of the experimental group. Motor activity decreased while freezing increased. Freezing behavior in control rats is nearly absent, this immobility behavior appears in rats exposed to V2O5 from the first month. Bielschowsky impregnation showed strong argyrophilic and collapsed somas and typical flame-shaped in all V-exposed rats hippocampus CA1 compared to controls. We also observe somatodendritic distortions. Axons and dendrites displayed thick dark bands replaced by noticeable thickening and nodosities and the cytoskeleton fibrillary proteins' linear traces. Our findings suggest that V_2O_5 inhalation induces Alzheimer-like cell death with evident cytoskeletal alterations.

Disclosures: C. Dorado-Martinez: None. **E. Montiel-Flores:** None. **J. Ordoñez-Librado:** None. **A. Gutiérrez-Valdez:** None. **O. Mejía-García:** None. **J. Espinosa-Villanueva:** None. **L. Reynoso-Erazo:** None. **M. Avila-Costa:** None.

Poster

P210. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models I

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Program #/Poster #: P210.04

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: CIHR FDN-148478
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HBHL
Alzheimer Canada
Canada Research Chair Tier 1

Title: Deep brain theta-burst optogenetic stimulation of the septohippocampal fibers in fornix improves memory performance in an Alzheimer's disease mouse model

Authors: E. CARMICHAEL, E. VICO VARELA, F. MANSEAU, G. ETTER, J. CHOI, ***S. WILLIAMS**;
McGill University, Douglas Res. Center,, Montreal, QC, Canada

Abstract: Electrical stimulation of the Fornix has been proposed to be an effective therapeutic approach to reverse memory loss in Alzheimer's disease. The Fornix is a white matter bundle containing axons connecting the hippocampus to the medial septum (cholinergic, GABAergic and glutamatergic fibers), the anterior thalamic nuclei and the mammillary bodies. While a large body of evidence shows that lesions of the Fornix lead to memory impairments in rodents and humans, there are only a few studies showing that acute or chronic electrical stimulation of the Fornix using theta-frequency related stimulations can improve memory. This study aimed to determine whether 5Hz optogenetic theta fornix stimulation of septohippocampal fibers was sufficient to rescue memory and what mechanisms may underlie the optogenetic improvements in AD mouse models. Using the hAPP-J20 AD mouse model aged 4-6 months, we show that septohippocampal fibers originated primarily from medial septum GABAergic neurons and that 5Hz optogenetic stimulation of the Fornix resulted in a substantial increase in power at that frequency during wake and REM sleep in the hippocampus. We found that a continuous 4 hours theta optogenetic stimulation between the encoding and recall phase of an object-location memory task rescued memory in the J20 mouse. Memory was also recovered in J20 after a 4Hr theta stimulation in the passive avoidance memory task. Interestingly, J20 mice, but not wild-type controls, showed a striking number of large-amplitude 6-9 ms long interictal spikes (IIS) occurring predominantly during REM and slow-wave sleep but not during wake. We observed that the 4hr optogenetic theta stimulation produced a significant reduction of IIs during REM and slow-wave sleep and that the extent of the decline was correlated with memory performance. We are now using calcium imaging to determine the effects of IIS during sleep. Our data suggest that fornix stimulation of septohippocampal GABAergic fibers is sufficient to improve memory in AD mouse models and that some of the underlying causes of the improvement may be related to the reduction of IIS during REM sleep.

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Poster

P210. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models I

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P210.05

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH U54AG062334 (GNN)

Title: Sex differences in age related modulation of synaptic mitochondrial function in the hippocampus and PFC of Wistar rats

Authors: *G. A. SHAW[1], M. M. HYER[1], A. J. WEGENER[1], S. K. DYER[1], G. N. NEIGH[2]; [1]Anat. and Neurobio., [2]Anat. & Neurobio., Virginia Commonwealth Univ., Richmond, VA

Abstract: Mitochondrial activity within the neuron, and specifically within the synapse, has been linked to disorders involving altered cognition, including Alzheimer's disease. These disorders are correlated with advanced age and mitochondrial dysfunction, yet the mechanism behind the increased prevalence of these neurocognitive disorders in women when compared to men has yet to be explored. Using male and female rats to assess neuro-mitochondrial activity across the lifespan, we determined the impact of sex and age on the function of synaptic mitochondria (SynMt) in the hippocampus (HPC) and prefrontal cortex (PFC) - regions susceptible to age-related changes in function. Brains were collected from 18 male and 18 female Wistar rats in either young (75 days), middle aged (6 months), or aged (1 year) groups. The HPC and PFC were dissected for isolation of synaptosomes via Percoll gradient. Oxygen consumption rate (OCR) of the SynMt was assessed using Agilent's Cell Mito Stress Test. Samples were examined for relative density of mitochondria within the synaptosomes using a ratio of Hexokinase I to SNAP25 through protein densitometry. OCR within the SynMt from the PFC and HPC was reduced in middle aged males when compared to young or aged males ($p < 0.0001$). Conversely, OCR within PFC SynMt was elevated in middle aged females compared to young ($p = 0.0323$) and aged females ($p < 0.0001$). OCR in HPC SynMt of aged females was also lower than that of young and middle aged females ($p < 0.0001$). A detailed analysis of mitochondrial dynamics reflected patterns seen in middle aged males, with reductions in basal respiration ($p < 0.0001$), maximal respiration ($p = 0.0006$), proton leak ($p = 0.0003$), ATP production ($p < 0.0001$), and spare capacity ($p = 0.0047$) than other male age groups. Middle aged females had higher basal respiration ($p < 0.0001$), maximal respiration ($p < 0.0001$), proton leak ($p < 0.0001$), ATP production ($p < 0.0001$), and spare capacity ($p < 0.0001$) in both the HPC and PFC than other female age groups. Densitometric analysis indicated young and middle aged females had a similar amount of mitochondria within each synaptosomal sample ($p > 0.05$), but aged female SynMt were reduced ($p = 0.0044$). In males, young and middle-aged animals were again parallel ($p > 0.05$) but aged males showed a considerably higher amount of SynMt within each sample ($p < 0.0001$). These findings shed light on a sex-specific mechanism that may contribute to the observed sex differences in age-related cognitive decline. SynMt show considerable changes in dynamics and respiration in the aging brain, suggesting a vulnerability where mitochondrial decline may contribute to the initiation of disease pathology with aging.

Disclosures: G.A. Shaw: None. **M.M. Hyer:** None. **A.J. Wegener:** None. **S.K. Dyer:** None. **G.N. Neigh:** None.

Poster

P210. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models I

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P210.06

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant 1R56AG056108-01

Title: Microglia-derived extracellular vesicles carrying Aβ induce progressive memory impairment and cortico-hippocampal network dysfunction.

Authors: *F. TOZZI[1], C. FALCICCHIA[2], M. GABRIELLI[3], O. ARANCIO[4], C. VERDERIO[3], N. ORIGLIA[2];
[1]Scuola Normale Superiore, Pisa, Italy; [2]CNR- Neurosci. Inst., PISA, Italy; [3]CNR- Neurosci. Inst., Milano, Italy; [4]Dept of Pathol, Columbia Univ., New York, NY

Abstract: Synaptic dysfunction is an early mechanism in Alzheimer's Disease (AD) that involves progressively larger areas of the brain over time. However, how it starts and propagates is unknown. We have previously demonstrated that amyloid-beta (Aβ) released by microglia in association with large extracellular vesicles (Aβ-EVs) alters dendritic spine morphology in vitro, and impairs synaptic plasticity both in vitro and in vivo in the entorhinal cortex-dentate gyrus (EC-DG) circuitry. In this work, we recorded EEG in freely-moving mice by means of chronic electrodes implanted both in the cortex (parietal cortex) and hippocampus (DG) and we investigated whether intracerebral Aβ-EVs injection (or control EVs derived from unstimulated microglia) was capable of altering network activity. In particular, we evaluated epileptic-like discharge looking at the clustered activity and the total number of spikes. Mice were recorded for 2h at 24h or 1, 2, 4 weeks post-injection. We observed that the intracerebral injection of Aβ-EVs can induce a significant increase in the number of spikes, and the total time spent in clustered activity, either at the level of the cortex or the hippocampus, when compared to mice injected with EVs alone, without Aβ (n= 6, 3 months old male C57bl/6j mice). Moreover, spectral analysis revealed that Aβ-EVs injection could induce alterations typical of transgenic models and preclinical AD patients, including spike-wave discharges (SWDs), a typical feature of absence epilepsy episodes. Interestingly, the spreading of synaptic dysfunction was also paralleled by progressive memory impairment. In fact, male mice (C57bl/6J, 3/4 months of age, n = 7) injected in the lateral EC (LEC) with Aβ-EVs and tested 1h, 24h, 2 weeks, 1 and 2 months following the injection, showed an early memory impairment (1h) exclusively confined to LEC-dependent memory tasks such as the novel Object-Place-Context Recognition Test (OPCRT) and then a progressive impairment (24h) also in hippocampal-dependent tasks as assessed by the novel

Object Recognition Test (ORT). This progressive spreading of memory impairment to the hippocampus was not observed in mice injected with Aβ alone or in mice injected with Aβ-EVs and Annexin V (inhibitor of EVs movement), suggesting that EVs contribute significantly to the propagation of memory dysfunction to the hippocampus. In conclusion, our results suggest that EVs carrying Aβ and spreading synaptic dysfunction along the Entorhinal-Hippocampal network, could contribute to memory impairment and propose a new neurobiological mechanism for the progression of AD neurodegeneration.

Disclosures: F. Tozzi: None. **C. Falcicchia:** None. **M. Gabrielli:** None. **O. Arancio:** None. **C. Verderio:** None. **N. Origlia:** None.

Poster

P210. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P210.07

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIA G1A62660

Title: Effects of age on sexually dimorphic food protection behavior associated with hippocampal cholinergic deafferentation.

Authors: J. R. OSTERLUND OLTMANNS[1], E. A. SCHAEFFER[1], A. A. BLACKWELL[1], S. A. PIETRUCHA[1], H. YANG[1], S. TSAI[2], G. L. KARTJE[2], ***D. WALLACE**[1];
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Abstract: Loss of hippocampal cholinergic projection originating from basal forebrain structures has been associated with the progression of Dementia of the Alzheimer’s Type. The role of these fibers in information processing deficits has been debated; however, spontaneous behaviors such as food protection have been observed to dissociate the contributions of hippocampal and cortical cholinergic function. Sexual dimorphism and age are critical factors in the progression of neurodegenerative disorders, yet these factors have not been evaluated in food protection behavior. The current study infused the immunotoxin 192-IgG-Saporin bilaterally into the medial septum to produce selective cholinergic deafferentation of the hippocampal formation. Female and male rats received infusion of the immunotoxin at either three or 18 months of age. Testing in the in the food protection paradigm began six weeks after the surgery. During the five days of testing, rats received two food protection sessions. Each of these sessions involved the rat (dodger) being placed in a transparent cylinder with a same sex conspecific (robber). The dodger was given a one-gram food item to consume, while the robber made multiple attempts to obtain the food item. The number, success rate, and type of food protection behaviors were recorded across all food protection sessions. Rats also received a third session each day in which the

latency to consume the food item was recorded in the absence of the conspecific. Preliminary results indicate that sex and age interact with cholinergic hippocampal deafferentation to influence the organization of food protection behaviors. These observations establish a foundation for future work investigating novel therapeutic interventions that target neuroplasticity within spared cholinergic systems.

Disclosures: J.R. Osterlund Oltmanns: None. **E.A. Schaeffer:** None. **A.A. Blackwell:** None. **S.A. Pietrucha:** None. **H. Yang:** None. **S. Tsai:** None. **G.L. Kartje:** None. **D. Wallace:** None.

Poster

P210. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P210.08

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R01 AG054654-01

Title: Involvement of CNS Luteinizing Hormone receptor in Alzheimer’s Disease pathogenesis

Authors: *M. MEY[1], G. CASADESUS SMITH[2];
[1]Kent State Univ., Kent, OH; [2]Pharmacol. and Therapeut., Univ. of Florida, Gainesville, FL

Abstract: CNS Luteinizing hormone and its receptor have been shown to modulate neuronal plasticity and cognition. Importantly, levels of brain LH are inversely related to levels in the periphery, thus low upon menopause-related upregulation of peripheral LH. Previously, we have shown that downregulation of peripheral LH improves function and plasticity in AD mouse models, improvements that are positively correlated with brain LH. However, whether these improvements are driven directly through the activation of central LHR signaling is not known. Therefore, the current study addresses whether central LHR activation was able to rescue cognitive and neuronal plasticity changes previously observed by peripheral LH downregulation. Female APP/PS1 transgenic and wild-type mice underwent ovariectomy or sham surgery and received ICV hCG or aCSF treatment for 8 weeks. Upon completion of cognitive testing, tissue was collected to determine changes in structural plasticity and Abeta pathology. cRISPR/cas9 methodology was used to study LHR in cell culture. Preliminary data demonstrates the ability of hCG treatment to differentially modulate cognition, pathology, and plasticity markers. This work supports the involvement of LHR in CNS function and begets its further study as a contributor linking menopause and increased AD risk.

Disclosures: M. Mey: None. **G. Casadesus Smith:** None.

Poster

P210. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P210.09

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant 1 R15 NS101608-01A1

Title: Characterizing the role of CD63-related tetraspanins, Tsp42Ee and Tsp42Eg, in Alzheimer's disease-associated synaptic dysfunction at the Drosophila neuromuscular junction

Authors: *E. L. HENDRICKS, F. L. W. LIEBL;
Southern Illinois Univ. Edwardsville, Edwardsville, IL

Abstract: The tetraspanin, CD63, is an established regulator of late endosomal trafficking and previous literature has positively correlated reduced CD63 blood serum levels to cognitive decline in Alzheimer’s disease (AD) patients. AD is characterized by the synaptic accumulation of amyloid beta (Abeta) plaques that result from the proteolytic processing of amyloid precursor protein (APP) by beta- (BACE) and gamma-secretases. CD63 physically interacts with the gamma-secretase complex and, thus, may regulate the cleavage of APP. However, CD63’s potential role in the pathogenesis of AD has not been well described. Therefore, we aim to identify a potential mechanism by which disruptions in the *Drosophila* CD63-related tetraspanins, *tsp42Ee* and *tsp42Eg*, lead to alterations in the endocytic pathway and processing of APP in control and AD-model animals. We find that pre- (*tsp42Ee*) and postsynaptic (*tsp42Eg*) knockdown of the CD63-related tetraspanins increases synaptic localization of two key endocytic proteins, Endophilin A and Dynamin. This increase in endocytic protein levels is coupled with a functional increase in synaptic vesicle internalization, indicating an upregulation of the endocytic pathway. Conversely, AD-model animals overexpressing APP and BACE (APP BACE OE) show a reduction in synaptic vesicle internalization. These opposing results suggest that either CD63-related tetraspanin expression, and subsequent endocytic upregulation, is independent of synaptic Abeta levels or APP BACE OE induces downregulation of these tetraspanins as a compensatory response to Abeta overload. These potential mechanisms align with findings that altered CD63 levels are not causative of AD, but rather a correlative indicator of advanced disease states in AD patients. Downregulation of CD63-related tetraspanins may promote endocytosis in states of compromised endocytic function in APP BACE OE animals and may also reduce CD63-directed cleavage of APP by reducing CD63 association with gamma-secretase. To explore these possibilities, we examine synaptic levels of another gamma-secretase interacting protein and endosomal trafficking regulator, Rab11. Rab11 also regulates the localization of BACE at the plasma membrane and, thus, may influence the production of Abeta. We find that *tsp42Ee* and *tsp42Eg* mutants phenocopy the reduced synaptic Rab11 levels and locomotor deficits observed in APP BACE OE animals, indicating that CD63-related tetraspanins may act upstream of Rab11 to regulate endocytosis and APP cleavage by BACE.

Disclosures: E.L. Hendricks: None. **F.L.W. Liebl:** None.

Poster

P210. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P210.10

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Institut Pasteur
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Inserm

Title: Nicotinic receptors mediate network dysfunction in early Alzheimer's disease

Authors: F. KOUKOULI[1,4], C. ZHANG[2], M. ROOY[5], I. LAZAREVICH[5], B. LAMOTTE D'INCAMPS[6], J. PEIXOTO[1], C. THIBERGE[2], S. PONS[2], J.-P. CHANGEUX[3], A. BACCI[4], B. S. GUTKIN[5], C. SCHMIDT-HIEBER[2], *U. MASKOS[2];
[1]Inst. Pasteur, Paris, France; [2]Inst. Pasteur, Paris Cedex 15, France; [3]Inst. Pasteur, Paris cedex 15, France; [4]Inst. du Cerveau et de la Moelle Epiniere (ICM), Paris, France; [5]Ecole Normale Supérieure, Paris, France; [6]SPPIN - St Pères Paris Inst. For Neurosciences, Paris, France

Abstract: Alzheimer’s disease (AD) is the most common form of dementia. Current treatments enhance acetylcholine (ACh) levels in the brain, but do not provide long-term amelioration. Patient and animal model data indicate a link between the cholinergic neuro-modulatory system and the amyloid beta (Aβ) peptide in the pathogenesis of AD. *In vivo* behavioral paradigms attempted to dissect the role of α7 nicotinic acetylcholine receptors (nAChRs) by using different AD mouse models crossed with knock-out mice for the α7 receptor (α7KO), but these studies came to opposite conclusions. In addition, previous work *in vitro* indicated either enhancement or blockade of nAChR function by Aβ of the α7 and β2* subtypes. Thus, comprehensive analysis of *in vivo* interactions between Aβ and the different nAChRs subtypes is essential. Here, we studied the early phase of AD pathology in a mouse model that we previously described, expressing soluble forms of the human Aβ peptide using an adeno-associated (AAV-hAPP) vector. We combined *in vivo* two-photon calcium imaging and *in vivo* patch-clamp electrophysiology in the prelimbic cortex (PrLC) of awake mice with synaptic slice electrophysiology and computational modeling. By analyzing the spontaneous activity of layers 2/3 in the mouse PrLC, we found pyramidal cell (PYR) hyperactivity early in the disease progression. However, Aβ peptide did not induce an increase in the PYR cell activity of α7KO mice. Targeted re-expression of α7 nAChRs in layers 2/3 in α7KO mice restored the activity of α7KO-APP mice to the levels of WT-APP, suggesting a key role of α7 subunits early in the disease. We further showed that PYR cell hyperactivity is due to an Aβ-mediated disruption of nAChRs signaling in parvalbumin-expressing interneurons that is accompanied by a differential

disruption of heteropentameric nAChR subtypes expressed by distinct types of GABAergic interneurons. Despite the massive investment in AD drug development, the majority of phase III clinical trials have failed in the past decade. We provide new experimental evidence to explain why clinical trials targeting α7 nAChR subunits with an agonist likely fail, as the drug target is no longer available. In contrast, heteropentameric nAChRs, are only partially inactivated throughout the disease progression, providing a novel drug target for intervention.

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Poster

P210. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models I

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P210.11

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: ZonMw 7330508160

Title: The contribution of glia to the onset of Alzheimer's Disease

Authors: *M. KATER, A. B. SMIT, M. H. G. VERHEIJEN;
Vrije Univ. Amsterdam, Amsterdam, Netherlands

Abstract: Alzheimer’s disease (AD) is a neurodegenerative disorder characterized by cognitive decline and the deposition of amyloid beta plaques and tau tangles. Activated glia cells, such as astrocytes and microglia, surround the amyloid plaques. This activation of glia cells, called gliosis, might be neuroprotective in removal of the plaques, while also detrimental by the release of cytokines by both types of glia damaging neuronal structures. The aim of this study is to determine whether gliosis contributes to early cognitive decline in AD, already before the presence of plaques. The commonly used AD mouse model APPswe/PS1dE9 was used to study the disease. Specific markers of astrocytes and microglia, earlier identified as altered during gliosis, were used to study the onset of gliosis. We found that gliosis is present in young AD-mice before the presence of plaques. These experiments will be followed by pharmacological interference with gliosis to study how this affects cognitive behaviour. Consequently, proteomics by mass spectrometry will be used to identify altered expression patterns of proteins that could identify the underlying molecular mechanisms. Taken together, this approach will provide information about early onset gliosis in AD and, ultimately, how early stage interference might be beneficial in treating AD.

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Poster

P210. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models I

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P210.12

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R01NS095799

Title: Extracellular neuronal activation triggers Tip60 HAT and HDAC2 subcellular dynamics in the brain: implications for cognition and Alzheimer's disease

Authors: *E. ARMOUR, G. GRECO, F. ELEFANT;
Biol., Drexel Univ., Philadelphia, PA

Abstract: Activity dependent gene control serves as the basis for long term memory formation and is driven by epigenetic mechanisms that regulate dynamic gene transcription in response to neuronal stimulation. Our lab has identified the histone acetyltransferase (HAT) Tip60 as a key regulator in the activity-dependent gene control of synaptic plasticity genes, co-regulating these genes with the histone deacetylase 2 (HDAC2). We recently demonstrated that stimulation of hippocampal neurons *in vitro* promotes Tip60 import from the cytoplasm into the nucleus with concomitant genomic reorganization and synaptic gene activation. Strikingly, we also found that Tip60 is largely excluded from the nucleus in hippocampal neurons from Alzheimer's disease (AD) patients. Nevertheless, whether Tip60/HDAC2 extracellular mediated subcellular dynamics functionally impacts activity dependent gene control and cognition *in vivo* and whether this process goes awry in AD remains to be elucidated. Here, we used immunohistochemistry (IHC) to compare the subcellular localization of Tip60 and HDAC2 in the *Drosophila* brain under normal conditions and in response to stimulation of fly brain neurons *in vivo* either by inducibly activating potassium channels using genetic manipulation or by exposure to natural environmental enrichment (EE) living conditions. Remarkably, we found that both methods of neural induction triggered changes in Tip60 and HDAC2 subcellular localization highlighted by a significant increase of both Tip60 and HDAC2 in the nucleus. These results support a model by which neuronal stimulation promotes import of Tip60 and HDAC2 chromatin modulators into the nucleus to activate neuroplasticity genes. To functionally test the impact of neural activation induced Tip60 subcellular changes, we are mutating Tip60's nuclear import and export signal sequences and assessing the consequences of disrupting Tip60 nucleocytoplasmic shuttling in the fly brain on neuroplasticity gene control, synaptic plasticity and cognition. We are also examining potential Tip60/HDAC2 subcellular disruption in the brain of flies modeling AD. Our studies will provide novel insights into the impact of neural induced Tip60 and HDAC2 nucleocytoplasmic shuttling on activity dependent gene control in cognition and AD.

Disclosures: E. Armour: None. **G. Greco:** None. **F. Elefant:** None.

Poster

P210. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models I

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CSC No. 201906100047

Title: Neural manifold analysis of hippocampal circuit dynamics during behavior in the 5xFAD mouse model of Alzheimer's Disease

Authors: *S. PRADO[1,2], M. GO[1], Y. LI[1], S. R. SCHULTZ[1];
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Abstract: In mouse models of Alzheimer's Disease (AD), amyloid or tau protein pathologies result in changes in cortical circuitry, which are particularly evident in the hippocampus. Although several treatment approaches that target amyloid-beta (AB) plaques have shown promising results in reducing their levels in the brain, it remains unclear whether reducing AB leads to cognitive improvement. One key issue is that the effect of amyloid pathologies on memory circuit dynamics at the network level is still not well understood, nor is the effect of these therapeutic strategies on the function of hippocampal-cortical circuits. In this study, we aim to characterize the effects of AB on the dynamics of neural circuits involved in spatial memory. We recorded two-photon calcium imaging data from mice running around a circular track floating on an air-pressurized table during a spatial memory task. Two- to ten-month old wildtype (WT) and transgenic 5xFAD mice were injected with hSyn1-GCaMP6f-mRuby in the hippocampal subfield CA1. To separate out memory encoding and recall processes, the mice were imaged after a week of behavioural training while exploring familiar and novel environments respectively. To track cells across multiple imaging sessions, we registered the motion-corrected images from different sessions using an image from a reference session as a template. ROI segmentation was performed on temporally concatenated videos from different sessions. We used Multidimensional Scaling (MDS) and Uniform Manifold Approximation and Projection (UMAP) as neural manifold learning algorithms to characterise the dynamics of CA1 neural population activity during formation, encoding, storage and retrieval of spatial memories. Both methods consistently revealed low-dimensional structures with datapoints clustered on a

manifold representing the spatial trajectory of the mouse and a cyclic pattern of transitions between network states over time across trials (loops). We compared the neural manifolds recovered from the different groups (5xFAD and WT mice of different ages) using the Fréchet distance between average manifold trajectories, finding a significant difference across groups. Neural manifold analysis methods provide a powerful tool for understanding how populations of neurons coordinate in representing task parameters, and here have allowed us to analyse how AB degrades the neural manifolds for spatial memory encoding and recall. By directly assaying the quality of the neural population representations of task-relevant information, they may also provide a sensitive test of whether AB clearance improves cognitive performance during therapeutic trials.

Disclosures: S. Prado: None. **M. Go:** None. **Y. Li:** None. **S.R. Schultz:** None.

Poster

P210. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P210.14

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R01 AG-055328

Title: Unanticipated stability of BDNF expression with aging in the hippocampal mossy fibers and resistance of dentate gyrus granule cells to Alzheimer’s disease neuropathology in a mouse model

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Abstract: It is well known that the neurotrophin brain-derived neurotrophic factor (BDNF) plays an important role in the developing CNS and in adults. In adults, BDNF contributes significantly to several normal functions, and deficits in BDNF have been implicated in disorders such as Alzheimer’s disease (AD). Most studies in mouse models of AD neuropathology show that hippocampal BDNF protein expression is reduced with age but there has been little chance to evaluate specific hippocampal areas because a specific antibody to BDNF has not been available recently. Therefore, we took advantage of a new antibody to develop a protocol for staining and then reexamined the levels of BDNF protein in specific areas of hippocampus at two ages and in a mouse model of AD neuropathology, the Tg2576 mouse.
For the first part of the study, we optimized an immunohistochemical protocol. For example, different fixation times were tested because previous experience suggested BDNF antibodies are sensitive to fixation. Then we compared Tg2576 mice with their littermates, including both

sexes, and focused on two different time points of the progressive neuropathology: 1) 3 months of age when oligomeric amyloid β (Aβ) is present but plaques are still absent, and 2) older than 11 months of age when plaques have started to accumulate.
Similar to past studies, BDNF protein expression in the mossy fiber axons (MF) of dentate gyrus granule cells (GCs) was strong in WT mice compared to other hippocampal layers. As previously shown in normal adult female rats, BDNF protein in female mice correlated with the stages of the estrous cycle. This result was true in both Tg2576 and WT mice. Surprisingly, BDNF protein expression was strong in Tg2576 mice at both young and older ages. Because BDNF expression increases with neuronal activity and Tg2576 mice show hyperexcitability, we also examined a marker of neuronal activity, ΔFosB. We found a correlation between the levels of MF BDNF protein expression and ΔFosB expression in the GCs. Despite the increase of activity in GCs of Tg2576 mice compared to WT, there was relatively little evidence of intracellular Aβ in GCs compared to pyramidal cells, using antibodies to Aβ (McSA1; 6E10). The results show that MF BDNF protein expression is remarkably stable in the Tg2576 mouse model. Furthermore, the reason that MF BDNF may remain stable could be due to the increased neuronal activity of GCs. Finally, we found evidence for a resistance of GCs to intracellular Aβ that is consistent with the resistance of GCs to insults and injury.

Disclosures: C. Criscuolo: None. **H.E. Scharfman:** None.

Poster

P210. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P210.15

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NSERC 171034-05
CIHR FDN - 159906
NSERC 40352
CIHR 390930
CIHR 390854
Weston Brain Institute

Title: Restoring the potassium chloride co-transporter KCC2 rescues functional deficits associated with Alzheimer’s disease-related mutations in mice

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Abstract: Growing evidence indicates that during early stages of Alzheimer's disease (AD) abnormal brain activity occurs due to disruption of $GABA_A$-mediated transmission. While disrupted $GABA_A$ signaling may result from several mechanisms, recent evidence points to deficits in the potassium-chloride cotransporter KCC2, responsible for maintaining low intracellular chloride in neurons to maintain robust inhibition. In this study, we validate whether KCC2 is downregulated in two transgenic mouse lines that develop AD-like amyloid-beta pathology and symptoms. Further, we examine whether by restoring KCC2 function we can alleviate deficits associated with AD. We found a decrease in the global and membrane protein levels of KCC2 in layer II/III of the prefrontal cortex in 4- and 6-months old 5xFAD mice and 6-months old APP^{NL-G-F} mice as compared to their non-transgenic littermates. Global and membrane KCC2 was also decreased in the hippocampus CA1 pyramidal layer of 6 months old 5xFAD mice. In addition, *ex vivo* chloride imaging revealed impaired Cl^- transport in the 5xFAD mice. Moreover, the power of hippocampal gamma oscillations was decreased in the APP^{NL-G-F} mice, as predicted from deficits in KCC2. Consistent with this prediction, treatment with CLP290, a KCC2 activity enhancer developed by our laboratory, restored the power of the higher band gamma oscillations. Finally, short-term administration of CLP290 in the 5xFAD mice improved spatial memory in the Morris Water Maze (MWM) test as compared to vehicle-treated mice. All together, these results indicate that KCC2 may be a viable target for reversing deficits in $GABA_A$-mediated inhibition in AD and attenuating several symptoms associated with AD pathology.

Disclosures: I. Keramidis: None. **B.B. McAllister:** None. **J. Mehla:** None. **J. Bourbonnais:** None. **D. Isabel:** None. **R. Sansonetti:** None. **P. Degagne:** None. **A.G. Godin:** None. **M.H. Mohajerani:** None. **Y. De Koninck:** None.

Poster

P220. Abeta as a Therapeutic Target

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P220.01

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant AG061708
NIH Grant AG063903

Title: NU-9, a novel inhibitor of amyloid beta oligomer accumulation as an Alzheimer's Disease neurotherapeutic

Authors: *E. A. JOHNSON[1], K. L. VIOLA[2], M. A. BICCA[2], W. ZHU[1], W. HUANG[2], R. B. SILVERMAN[1], W. L. KLEIN[2];
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Abstract: Objective and Rationale: Amyloid beta oligomers (AβOs) are a toxic species elevated in the brains of Alzheimer's disease patients. AβO administration in animals induces tau phosphorylation, synapse deterioration, and memory loss; antibodies selective for AβOs can prevent memory loss in Alzheimer's animal models. In this work, we identified a small molecule, NU-9, that prevents AβO buildup along dendrites of hippocampal neurons treated with amyloid beta (Aβ) monomer, which has the potential to be disease-modifying and neuroprotective in Alzheimer's disease, and characterized necessary components in the cell-dependent molecular mechanism of action. **Methods:** We performed immunofluorescence (15-30 images per condition, blind image analysis) and dot blot (3 bio-replicates per condition), using AβO-specific antibodies developed in-house, on primary E18 rat hippocampal neuron cultures at 14-25 days *in vitro*. All results were replicated in multiple cell cultures. For animal behavior studies, we treated 10-month old 5xFAD mice (4-5 per condition) with NU-9 by oral gavage. **Results and Conclusions:** Addition of NU-9 to neurons for 30 minutes prior to Aβ monomer administration significantly prevented the accumulation of AβOs on the dendrites of hippocampal neurons with approximately 60-80% reduction compared to vehicle. Several experiments demonstrate that the mechanism for this effect is cell-based. First, in media without cells, NU-9 did not reduce AβO formation, nor did conditioned media produced by NU-9 treated cells. Further, treatment of neurons with NU-9 prior to application of Aβ monomer did not reduce the number of unbound, extracellular AβOs, suggesting that NU-9 may act by altering the formation of a specific population of neuron-binding AβOs or by removing certain binding targets. Also, effectiveness of NU-9 was abolished in the presence of the lysosome acidification and fusion inhibitor Bafilomycin A, supporting a lysosome-dependent mechanism. The cysteine cathepsin inhibitor E64, additionally, mimicked the effect of NU-9, leading us to hypothesize that a cysteine cathepsin may be the target of NU-9. In a preliminary study with 5xFAD mice, oral administration of NU-9 also prevented memory loss in 75% of treated mice, allowing them to distinguish a novel object, while all vehicle controls failed. In summary, we have identified a small molecule inhibitor of dendritic AβO accumulation which functions via a lysosome-dependent mechanism, with potential to disrupt a key pathogenic process in Alzheimer's disease.

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Poster

P220. Abeta as a Therapeutic Target

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Program #/Poster #: P220.02

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH-R01 AG063903
NIH-R21 AG045637
NIH-R41AG054337

Title: An Amyloid β Oligomer-selective antibody rescues memory function in 5xFAD mice, potentially through prevention of AβO-induced reactive astrogliosis

Authors: *D. L. KRANZ[1], K. L. VIOLA[1], M. A. BICCA[1], A. BEBENEK[4], V. NANDWANA[2], E. A. WATERS[3], C. R. HANEY[3], T.-T. CHANG[5,6], A. PECK[5], V. P. DRAVID[2], G. KRAFFT[7], J. JERECIC[7], D. O'CONNELL[7], W. L. KLEIN[1];
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Abstract: The first FDA-approved drug to treat Alzheimer's disease (AD) in nearly two decades, Aduhelm, unfortunately has minimal efficacy and high dosage requirements in clinical trials, likely due to its preferential affinity for the less toxic, aggregated forms of amyloid beta (Aβ), rather than toxic soluble amyloid beta oligomers (AβOs). Here we report evidence further implicating AβOs as the species most germane to instigating AD, showing that a subpopulation of AβOs probed by a humanized AβO-selective monoclonal antibody induces memory dysfunction in the 5xFAD mice. We show that these AβOs first appear in the 5xFAD mice at 3 months of age, just prior to the onset of memory dysfunction, and that they build up with increasing memory dysfunction. In harmony with these results, intra-ventricular injection of AβOs into Wt mice induces hp-dependent memory dysfunction within 24 hours. Strikingly, intranasal inoculation of the AβO-selective antibody into 6 month-old 5xFAD mice rescued memory function 30 days post-inoculation. We also present evidence that probes based on AβO-selective antibodies are capable of distinguishing 5xFAD mice from Wt in MRI and PET imaging, respectively. These tools substantiate target engagement *in vivo* and hold significant diagnostic potential. A new aspect of the AβO mechanism was found in immunofluorescence experiments which showed 5xFAD mice of increasing age manifested concomitant buildup of AβOs and reactive astrocytes (GFAP). At high resolution, reactive astrocytes could be seen surrounding AβO+ cell bodies and micrometer-wide AβO+ puncta along astrocytic processes, strongly suggesting involvement of AβOs in the induction of reactive astrogliosis. These findings are particularly intriguing given recent studies indicating AD's dependence on astrocytes. One especially interesting study showed that when apolipoprotein E (ApoE), a protein which AβOs associate with at synapses, was knocked out in astrocyte-only populations of P301S mice, AD pathology markedly improved [Wang C, et. al, 2021]. Based on these new results, we propose that ApoE-dependent induction of reactive astrogliosis is triggered by the specific subset of AβOs which induces memory dysfunction in 5xFAD mice.

Disclosures: D.L. Kranz: None. **K.L. Viola:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Acumen Pharmaceuticals Inc.. E. Ownership

Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Acumen Pharmaceuticals Inc.. **M.A. Bicca:** None. **A. Bebenek:** None. **V. Nandwana:** None. **E.A. Waters:** None. **C.R. Haney:** None. **T. Chang:** None. **A. Peck:** None. **V.P. Dravid:** None. **G. Krafft:** None. **J. Jerecic:** None. **D. O'Connell:** None. **W.L. Klein:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Acumen Pharmaceuticals Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Acumen Pharmaceuticals Inc..

Poster

P220. Abeta as a Therapeutic Target

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Program #/Poster #: P220.03

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: FAPESP
CNPq
CAPES
FAEPA

Title: The conformational scFv antibody NUsc1 protects differentiated human neuroblastoma cells against Aß oligomers toxicity

Authors: *N. R. PINHEIRO, A. L. B. BITENCOURT, M. F. CARRARO, I. S. SANTOS, S. CHEDRAOUI-SILVA, A. SEBOLLELA;
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Abstract: BACKGROUND: Soluble oligomers of the Aß peptide (AßOs) are neurotoxins linked to synaptotoxicity and neurodegeneration in Alzheimer's disease (AD). AßOs are heterogeneous in conformation and toxicity and the extent to which different Aβ species contribute to the pathophysiology of AD remains uncertain. Antibodies sensitive to conformational differences between Aβ aggregates are powerful molecular tools for understanding the structure and function relationship of AßOs. In a previous work, we selected a scFv fragment antibody, named NUsc1, capable of distinguishing AßOs from both monomeric and fibrillar Aß. Different from other conformational anti-Aß antibodies, NUsc1 preferentially targets a subset of AßOs larger than 50kDa present in AD brain tissue. Here, we investigated the potential of NUsc1 in preventing the toxicity instigated by AßOs to differentiated human neuroblastoma cells. **MATERIAL AND METHODS:** SH-SY5Y neuroblastoma cells were differentiated into mature neurons and challenged with AßOs for 24h, either alone or in the presence of purified NUsc1. AßO binding to SH-SY5Y cells was assessed by immunofluorescence. Viability was evaluated by both MTT and Live/ Dead assays. In parallel,

the impact of NUsc1 on the conversion of AßOs into mature amyloid fibrils was evaluated by both Thioflavin T fluorescence emission and transmission electron microscopy. **RESULTS:** AßO binds to differentiated SH-SY5Y cells rendering a punctate binding pattern, as previously shown in primary neurons. Treatment with AßOs caused a decrease in cell viability followed using the MTT assay, although not leading to cell death. Importantly, the MTT reduction capacity was not affected in cells co-treated with AßOs and NUsc1. We have also compared the neuroprotection conferred by NUsc1 with the protection provided by the IgG monoclonal 6E10 (a pan-Aß antibody). Interestingly, AßO species neutralized by NUsc1 seem to be on-pathway to amyloid fibril formation, since a robust inhibition of AßO-to-mature fibril conversion by NUsc1 was observed. **CONCLUSIONS:** The scFv antibody NUsc1 neutralizes AßO-induced toxicity on differentiated SH-SY5Y cells. In the light of the recent advances in therapeutic strategies to AD based on antibodies selective for aggregated Aß, our data encourages a more in-depth investigation on the potential of NUsc1 to prevent AßO neurotoxicity.

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Poster

P220. Abeta as a Therapeutic Target

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P220.04

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: CityU11100318

Title: Chronic low dose ionizing radiation therapy for Alzheimer’s disease

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Abstract: Alzheimer's disease (AD) is characterized by Beta-Amyloid (Aβ) plaques, neurofibrillary tangles, and neuroinflammation leading to cognitive impairment. The primary role of Aβ peptide in the development of AD is well-known. It induces loss of synaptic connections and disrupts the neuronal circuit in the hippocampus, which contributes to memory deficits. Accumulating evidence suggests that reduction of Aβ accumulation could be a possible mechanism for the amelioration of AD symptoms. In our study, we used Streptozotocin (STZ) induced and transgenic (3xTg-AD) mouse of AD to imitate pathology and evaluate the therapeutic potential of low-dose ionizing radiation. Transgenic model (mutations of Swedish APP, MAPT P301L, and PSEN1 M146V) are linked to the early onset of Alzheimer's disease

and develop the extracellular Aβ deposition, hyperphosphorylated tau, gliosis, and cognitive impairment, as seen in AD patients. A sporadic AD mouse model was developed by intracerebroventricular (I.C.V) injection of Streptozotocin, which induces gliosis, neuroinflammation, and cognitive deficits in mice. The efficacy of low-dose ionizing radiation (LDIR) was evaluated in STZ and 3xTg-AD mouse model to assess the beneficial effect by improving learning and memory. Our results showed that the chronic treatment with LDIR improved the long-term and the short-term memory in both the 3xTg-AD mouse model and the STZ mouse model. We also observed a significant reduction of soluble as well as insoluble forms of Aβ in the hippocampus at early (AD mouse model at 9-month-old) and advanced (AD mouse model at 17-month-old 3xTg-AD mice) stages of disease progression. Also, the 17-month-old 3xTg-AD mice with LDIR treatment have shown a reduction of the hyperphosphorylated tau and the intensity of the microglia in the CA1 area of the hippocampus. Next, we used 3D reconstruction of extracellular Aβ plaques and the microglia in the CA1 area of the hippocampus and observed that LDIR treatment enhances the microglial phagocytotic clearance of fibrillar Aβ form. Furthermore, the western blotting results showed that low dose ionizing radiation could protect the neuron against Aβ by inhibiting the mitochondrial pathway of apoptosis in the hippocampus and the cortex of the 9-month-old and 17-month-old 3xTg-AD mice. In conclusion, chronic low dose ionizing radiation improved the memory in the mice model of AD by reducing the Aβ due to increased microglial phagocytotic activity and restored hippocampus neurons by inhibiting the apoptosis of the mitochondrial pathway.

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Poster

P220. Abeta as a Therapeutic Target

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Program #/Poster #: P220.05

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: FAPESP (Brazil)
CNPq (Brazil)
CAPES (Brazil)
FAEPA (University of Sao Paulo)

Title: Non-denaturing isolation of neurotoxic oligomers of the β-amyloid peptide using a conformation sensitive fragment antibody-based assay

Authors: *R. M. CAMPOS[1], A. L. B. BITENCOURT[1], E. N. CLINE[2], M. F. CARRARO[1], W. L. KLEIN[2], A. S. SEBOLLELA[1];

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Abstract: BACKGROUND: Soluble oligomers of the β-amyloid peptide (AβOs) are currently implicated in the pathogenesis of Alzheimer's disease (AD). Biochemical identity of those oligomers is debated, including their molecular mass and conformation. Importantly, neurotoxicity of AβOs has been shown to be neutralized by the scFv fragment antibody NUsc1, which preferentially targets a subpopulation of AβOs > 50 kDa. NUsc1 is thus a promising candidate to assist in the isolation and subsequent structure-function characterization of AD-relevant AβOs. In this work, we describe a method for the isolation of NUsc1-targeted AβOs under non-denaturing conditions from either *in vitro* prepared or tissue-extracted AβOs, followed by determination of their molecular mass. **MATERIAL AND METHODS**: NUsc1-functionalized magnetic beads were used in the isolation of AβOs. The complex NUsc1-AβOs was successfully released from beads using imidazole, as followed by Western Blot (WB) and liquid chromatography-mass spectrometry (LC-MS). NUsc1-targeted AβO species were stabilized by chemical crosslinking and molecular mass was determined by WB. Alternately, the NUsc1-AβOs complex was crosslinked and also analyzed by WB. In addition, molecular mass of isolated AβOs was investigated by analytical size-exclusion chromatography (SEC). **RESULTS:** The NUsc1-AβO complex was successfully isolated under non-denaturing conditions from a synthetic AβO preparation. Moreover, 5xFAD mouse brain-derived AβOs were efficiently pulled down by NUsc1-functionalized beads, as revealed by LC-MS. Interestingly, cross-linked AβO species targeted by NUsc1 were larger than 100 kDa, in agreement with results obtained with NUsc1-AβO cross-linked after complex isolation. Determination of molecular mass of NUsc1-reactive AβOs by SEC is ongoing, as well as investigations on the molecular shape of these species. **CONCLUSION:** In combination with other data from our group on NUsc1 neuroprotective actions, our findings indicate that NUsc1 preferentially targets AD-relevant AβO species larger than 100 kDa. This strategy may be applied to investigate mass and conformation of AβO assemblies from other complex samples under non-denaturing conditions, enabling the rationale development of more effective therapeutics to treat AD.

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Poster

P220. Abeta as a Therapeutic Target

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Program #/Poster #: P220.06

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH R01DA038635-S1

Title: Sri 22136, a novel delta opioid receptor (dor) antagonist for the treatment of alzheimer's disease

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Abstract: Alzheimer's disease (AD) is the most common form of dementia, which affects 47 million people worldwide. There are over five million AD patients over the age of 65 in the U.S. which is predicted to increase to 16 million by 2050. There are currently no approved disease-modifying therapies for AD and attempts to prevent or slow the progression of the formation of beta-amyloid plaques by targeting both the beta-secretase 1 (BACE1) and gamma-secretase enzymes have not yet achieved clinical success. Studies suggest that indirect modulation of the function of these enzymes via G-protein coupled receptors (GPCRs) may provide a novel strategy to reduce A-beta peptide production with potentially less side effects. Among GPCRs that influence amyloidogenesis, the delta opioid receptor (DOR), in particular, has been shown to play an important role in the trafficking and function of BACE1 and gamma-secretase and in the production of A-beta peptide. DOR activation increases BACE1 and gamma-secretase activity *in vitro* and in a mouse model of AD, and antagonism of DOR specifically blocks the amyloidogenic pathway and efficaciously prevents AD progression in mice. These effects were demonstrated using a known DOR antagonist, naltrindole. However, the potential of DOR antagonists as therapeutic agents for AD has yet to be explored.
The aim of this project is to create novel DOR antagonists via medicinal chemistry and identify the most promising lead compound based on binding, selectivity, and functional profile *in vitro*. Further, select a small set of the most promising compounds and evaluate the compound's ability to mitigate AD-like pathology *in vivo* using APP/PS double-transgenic mice. We report here the identification of several selective DOR antagonists with low nanomolar affinity/potency that have been shown to block BACE1 activity *in vitro*. Lead compound SRI-22136, showed systemic stability and blood brain barrier penetration using the *in vivo* tail flick assay in CD-1 mice. SRI-22136 also showed efficacy in Novel Object Recognition (NOR) behavioral studies at a dose of 1mg/kg twice daily for 90 days continuous treatment *in vivo* using the APP/PS AD model double-transgenic mice. We further report our progress in testing this compound's effect on histological and biochemical marker evaluation using the cortex and hippocampus regions of the APP/PS AD model double-transgenic mice.
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Poster

P220. Abeta as a Therapeutic Target

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Program #/Poster #: P220.07

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: CONACYT A1-S-16282

Title: Variants of amyloid beta peptide improve phenotypes related to amyloid toxicity in Caenorhabditis elegans

Authors: *D. P. QUIJANO GUERRERO[1], A. E. ESTRADA RODRÍGUEZ[1], M. GONZÁLEZ GONZÁLEZ[1], J. VALDÉS RODRÍGUEZ[2], R. VIDALTAMAYO[3], V. ZOMOSA SIGNORET[1]; [1]Univ. Autónoma De Nuevo León, Monterrey, Mexico; [2]Univ. Nacional Autónoma de México, Ciudad de México, Mexico; [3]Univ. of Monterrey, San Pedro, Mexico

Abstract: Amyloid beta peptide (Aβ) aggregation is one of the major hallmarks in Alzheimer's disease (AD) pathogenesis and therefore prevention of Aβ deposition is a relevant strategy in the treatment of AD. The development of aggregation inhibitors using Aβ as a starting point allows to create highly specific peptides as possible drugs. Using this approach, we generated three Aβ peptide variants of 42 amino acids (Aβ-42) by introducing the point mutations M35C, A30W or K28A which all lie in key regions that participate during the aggregation process. *In vitro* assays showed the mutant peptides disorganized and prevented *de novo* aggregation of the wild type peptide. For *in vivo* analysis we used the nematode *Caenorhabditis elegans* which is widely use for the screening of Aβ aggregation inhibitors and the evaluation of their associated effects on behavior. Assays done by feeding a small number of worms with the peptides showed some slightly improvement in N2 chemotaxis and CL2006 and CL4173 paralysis phenotype. In this work, to reduce the risk of peptide degradation and to assure its transportation into all body tissues, we standardized a transduction protocol based on the encapsulation of molecules within lipid vesicles. Using a commercial kit consisting of a lipid formulation (BioPORTER), the fluorescent marker FITC-IgG was delivered into worms of a wildtype strain to assess transduction efficiency. Following, peptide toxicity at three different concentrations (15, 25 and 40µM) was evaluated by treating N2 worms using the transduction protocol and tracking lifespan. Next, we used the muscular Aβ-expressing strain CL2006 to monitor the progression of a Aβ-dependent paralysis phenotype and to evaluate the number of amyloid deposits formed after treatment. Additionally, to relate Aβ toxicity with neurological functions, we treated the neuronal Aβ-expressing strain CL2355 and assayed its chemotaxis response to benzaldehyde. All assays were carried out with three groups of 30 synchronized worms for each treatment. Images obtained by fluorescent microscopy showed the transduction protocol allows the protein to reach all body tissues of the worm. Treatment with the mutant peptides caused a significant extension in N2 worm's lifespan, a significant delay in the onset of the paralysis in CL2006 worms, a slightly reduction in size and number of amyloid deposits and a recovery of the chemotactic index of CL2355 worms. Results suggest these mutant peptides could protect *C. elegans* against

Aβ-induced toxicity, probably due to a neuroprotective and antioxidant effect, and therefore could have a potential application in the development of treatments against AD.

Disclosures: D.P. Quijano Guerrero: None. **A.E. Estrada Rodríguez:** None. **M. González González:** None. **J. Valdés Rodríguez:** None. **R. Vidaltamayo:** None. **V. Zomosa Signoret:** None.

Poster

P220. Abeta as a Therapeutic Target

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P220.08

Topic: I.05. Biomarker, Drug Discovery, and Experimental Therapeutics

Title: A nasal formulation for the treatment of Alzheimer's disease

Authors: *T. ZIENELDIEN, Y. WANG, J. KIM, N. SHEN, X. LIN, M. MOMMER, C. CAO; Pharmaceut. Sci., Univ. of South Florida, Tampa, FL

Abstract: Due to the complexity of the causes of Alzheimer's disease (AD), combination therapy is considered the most effective approach. Furthermore, the adverse effects from long-term applications also represent a major obstacle in anti-AD drug development. The objective of this research was to develop a nasal formulation with nutraceutical molecules for AD prevention and treatment. The MCT oil-based nanoemulsion formulation was prepared with melatonin, insulin, and THC (MIT). In this case, 12-month-old APP/PS1 mice were used as the animal model and were treated with MIT daily via intranasal delivery for 3 months. Radial Arm Water Maze (RAWM) and plasma Aβ levels were used for grouping and treatment benefit evaluation. Post-treatment RAWM results demonstrated that MIT formulation can significantly decrease errors when compared to the no treatment transgenic mice group (N=9 per group). Blood samples were taken when treated for 1.5 months and at the end of treatment. Though plasma Aβ40 and 42 levels showed no difference among the groups at each time interval, both Aβ40 and 42 levels increased with treatment compared to the pre-treatment sample. Furthermore, brain soluble Aβ showed a tendency to increase during treatment, while the insoluble decreased, but there were no statistically significant differences ($P>0.05$). The flow cytometry analysis of the blood samples showed CD3+CD8a+ increased with MIT treatment ($P<0.05$), and this result implied that the MIT formulation not only targets Aβ but also can strengthen impaired immunity in aged mice. Overall, the MIT formulation appears to be a promising combination therapeutic for AD treatment since all the molecules used in this formula are safe and effective molecules for AD.

Disclosures: T. Zieneldien: None. **Y. Wang:** None. **J. Kim:** None. **N. Shen:** None. **X. Lin:** None. **M. Mommer:** None. **C. Cao:** None.

Poster

P220. Abeta as a Therapeutic Target

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Program #/Poster #: P220.09

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Brightfocus Foundation Postdoctoral Fellowship Award

Title: Irisin reduces Ab pathology by increasing neprilysin from astrocytes in 3D human neural cell culture models of Alzheimer's disease

Authors: *E. KIM[1], H. KIM[2], M. P. JEDRYCHOWSKI[2], G. BAKIASI[1], B. M. SPIEGELMAN[2], R. E. TANZI[1], S. CHOI[1];
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Abstract: Alzheimer's disease (AD) is the most common form of age-related dementia, characterized by progressive memory loss and cognitive disturbance. Two pathological hallmarks that are crucial in the disease are amyloid plaques and neurofibrillary tangles (NFTs). Physical exercise has been shown to counteract various aspects of AD pathology in animal models: it reduces amyloid-β (Aβ) levels and amyloid deposition in the brain, mitigates tau pathology, attenuates neuroinflammation, and ameliorates cognitive dysfunction. Irisin, a myokine that is cleaved from its precursor protein, fibronectin type III domain containing protein 5 (FNDC5), is a novel exercise-induced myokine which regulates energy metabolism in adipose and is recently considered to be a critical mediator for the beneficial effects of exercise in the brain. Using our three-dimensional (3D) human neural cell culture models of AD (3D-AD cultures) with Aβ-driven NFTs, we recently found that irisin significantly reduced Aβ pathology by increasing the activity/levels of the Aβ-degrading enzyme neprilysin (NEP) potentially secreted from astrocytes. We also identified that integrin αV/β5 acts as an irisin receptor on astrocytes, which was previously reported in bone and fat, and is required for the effects of irisin to enhance NEP-mediated Aβ clearance. Irisin reduced GFAP and S100β protein expression and inhibited STAT3 signaling via integrin αV/β5 in the 3D-AD cultures. Inhibition of IL-6/ERK-STAT3, a key modulator of astrocyte reactivity, was found to contribute to enhancement of NEP-induced Aβ clearance. Our findings suggest that irisin is a potential mechanism of exercise that acts on Aβ pathology which could emerge as a potential therapeutic target for AD.

Disclosures: E. Kim: None. **H. Kim:** None. **M.P. Jedrychowski:** None. **G. Bakiasi:** None. **B.M. Spiegelman:** None. **R.E. Tanzi:** None. **S. Choi:** None.

Poster

P228. Altered Energy Homeostasis in Alzheimer's Disease I

Location: Virtual Only

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Program #/Poster #: P228.01

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: RF1AG054052 (JK), R01AG044546 (CC), P01AG003991 (CC), RF1AG053303 (CC), RF1AG058501 (CC), U01AG058922 (CC), R01AG057777 (OH), A2020118F (JM)
The Mayo Clinic Alzheimer's Disease Genetic Studies, led by Dr. Nilufer Taner and Dr. Steven G. Younkin, Mayo Clinic, Jacksonville, FL using samples from the Mayo Clinic Study of Aging, the Mayo Clinic Alzheimer's Disease Research Center, and the May
P50 AG016574, R01 AG032990, U01 AG046139, R01 AG018023, U01 AG006576, U01 AG006786, R01 AG025711, R01 AG017216, R01 AG003949, NINDS grant R01 NS080820, CurePSP Foundation, and support from Mayo Foundation
Study data includes samples collected through the Sun Health Research Institute Brain and Body Donation Program of Sun City, Arizona. The Brain and Body Donation Program is supported by the National Institute of Neurological Disorders and Stroke (U24

Title: Alzheimer's disease alters oligodendrocytic glycolytic and ketolytic gene expression

Authors: *E. R. SAITO[1], J. B. MILLER[2], O. HARARI[3], C. CRUCHAGA[3,4,5,6], K. A. MIHINDUKULASURIYA[7], J. S. K. KAUWE[2], B. T. BIKMAN[1];
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Abstract: Sporadic Alzheimer's disease (AD) is the most common form of senescence-related dementia, characterized by neurodegeneration that progressively compromises memory and behavior. A growing body of evidence has demonstrated that AD is strongly correlated with impaired glucose metabolism and implicates metabolic dysfunction in disease onset and progression. However, the degree to which glia contribute to glucose hypometabolism and whether ketone metabolism is also impaired in AD is unknown. Because glucose metabolism is altered in AD, ketolysis has been suggested as an alternative pathway to fuel the brain. In the present study, RNA-seq data from postmortem AD brains were used to determine whether dysfunctional AD brain metabolism can be determined by impairments in glycolytic and ketolytic gene expression. Data were obtained from the Knight Alzheimer's Disease Research Center (62 cases; 13 controls), Mount Sinai Brain Bank (110 cases; 44 controls), and the Mayo Clinic Brain Bank (80 cases; 76 controls). These data were normalized to cell type – astrocytes,

microglia, neurons, oligodendrocytes – using cell-specific markers. In oligodendrocytes, both glycolytic and ketolytic pathways were significantly impaired in regions of the brain relevant to memory and cognition. However, ketolytic gene expression was not significantly altered in neurons, astrocytes, and microglia. Generally, these data indicate that oligodendrocytes may be larger contributors to brain hypometabolism and AD pathophysiology than previously thought. These demonstrate a potential link between AD hypometabolism and dysmyelination, and suggest that ketones may be therapeutic in AD due to their ability to fuel neurons despite widespread impairments in glycolytic brain metabolism.

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Poster

P228. Altered Energy Homeostasis in Alzheimer's Disease I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P228.02

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIA Grant R01AG061150

Title: Amyloid-beta initiates brain hypometabolism and network dysfunction via NOX2 activation: a potential onset mechanism of Alzheimer’s disease

Authors: A. MALKOV[1], A. I. IVANOV[2], I. POPOVA[1], S.-S. JANG[3], S. YOON[3], Y. HUANG[3], Y. ZILBERTER[4], **M. Y. ZILBERTER**[3];
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Abstract: A paramount driver of sporadic Alzheimer's disease (AD) is the synergy of oxidative stress and glucose hypometabolism in the brain. Oxidative stress damages macromolecules such as DNA, lipids and proteins, whereas glucose hypometabolism impairs cellular energy supply and antioxidant defense; Together, these cellular and functional alterations may be primary triggers of AD. However, the exact basis of AD-associated glucose hypometabolism as well as its causes and consequences has remained unknown. We utilized electrophysiology together with live imaging of NAD(P)H & FAD autofluorescence and metabolic sensors (glucose, lactate, oxygen) in activated hippocampal network. We report acute inhibition of brain glucose utilization by $A\beta_{1-42}$ and identify oxidative stress resulting from $A\beta_{1-42}$ -induced activation of NADPH oxidase 2 (NOX2) as the trigger of brain glucose hypometabolism with resulting network hyperactivity and behavioral disruptions. In hippocampal brain slices, $A\beta_{1-42}$ application

reduced network activity-driven glucose consumption and glycolysis by half, while NOX2 antagonism prevented this effect. *In vivo*, intracerebroventricular injection of $A\beta_{1-42}$ exerted a profound inhibitory effect on brain glucose consumption, resulting in long-lasting network hyperactivity and changes in animal behavioral profile. Critically, the novel bioavailable NOX2 antagonist GSK2795039 prevented all of the observed Aβ-related detrimental effects. These data provide the first experimental evidence behind AD-related brain glucose hypometabolism and its consequences, and suggest that targeting NOX2-induced oxidative stress is a promising approach to both the prevention and treatment of AD.

Disclosures: A. Malkov: None. **A.I. Ivanov:** None. **I. Popova:** None. **S. Jang:** None. **S. Yoon:** None. **Y. Huang:** None. **Y. Zilberter:** None. **M.Y. Zilberter:** None.

Poster

P228. Altered Energy Homeostasis in Alzheimer's Disease I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P228.03

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: 1R01AG062762-01A1
1R56AG062762-01

Title: Examining the effects of a blood glucose rescue on alzheimer's disease-related pathology in young vs. aged hyperglycemic mice

Authors: *A. A. ORTIZ[1], A. R. PLATT[2], A. M. OSSE-LEISGANG[1], J. W. WANG[1], K. C. ALCAZAR[1], E. MIRELES[1], A. M. SALAZAR[1], J. W. KINNEY[1];
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Abstract: Abstract
Alzheimer’s disease (AD) is an irreversible neurodegenerative disease that is characterized by progressive synaptic and neuronal loss, learning and memory deficits, and cognitive/behavioral decline. The pathological hallmarks of AD include senile plaques composed of amyloid-beta (Aβ); intracellular neurofibrillary tangles, composed of accumulated hyperphosphorylated tau protein (ptau); and chronic neuroinflammation that can promote and accelerate both Aβ and tau pathologies. AD affects approximately 6.2 million Americans and is the 6th leading cause of death in the US. AD is classified as familial (fAD) or late onset (LOAD). fAD (early onset) is associated with genetic mutations in several genes (i.e., amyloid precursor protein, presenilin 1, presenilin 2, etc.) and accounts for 3-5% of all AD cases. In contrast, LOAD accounts for 95-97% of all AD cases with no genetic etiology, however, several genetic and/or other comorbidities confer increased risk for LOAD.
A major risk factor for developing LOAD is diabetes mellitus (DM). DM confers up to a 4-fold

increased risk for developing AD, and approximately 80% of individuals with AD have type II diabetes (T2DM) or impaired fasting glucose levels. Furthermore, over 25% of the population over the age of 65 are diabetic. As age is also a risk factor for developing LOAD, DM and age may combine to impact AD onset and progression.
Hyperglycemia - abnormal elevated blood glucose level - is the primary characteristic of diabetes and is observed in T2DM. We have previously demonstrated that chronic hyperglycemia can initiate and promote neuroinflammation, resulting in significant increases in ptau and learning and memory deficits that are consistent with other AD models. To determine if hyperglycemia or the ensuing inflammatory response is the mechanism driving AD related changes, we evaluated if lowering blood glucose in our hyperglycemia model would rescue ptau and inflammation. We induced hyperglycemia via a staggered and low dose of streptozotocin (STZ) in young (7 months) and aged (18 months) wild-type male mice. Following 6 weeks of sustained hyperglycemia, we treated animals with Phloridzin (PZ) to lower blood glucose levels. Following PZ treatment, we evaluated if the AD relevant changes were rescued.
Briefly, STZ induced a sustained hyperglycemic state and PZ decreased blood glucose levels. However, in both the young and aged cohorts, PZ did not rescue the ptau or inflammatory changes consistent with AD. Our data demonstrate that even when hyperglycemia is controlled, AD relevant changes persist, and the underlying mechanism of how DM confers increased risk for AD is more than just a hyperglycemic state.

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Poster

P228. Altered Energy Homeostasis in Alzheimer's Disease I

Location: Virtual Only

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Program #/Poster #: P228.04

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R01AG057767
NIH Grant R01AG061937
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Illinois Health Improvement Association Grant
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Title: Metabolic and cognitive effects of altering core body temperature in APP/PS1 mice

Authors: S. A. MCFADDEN[1], M. F. COX[1], C. A. FINDLEY[1], M. R. PECK[1], L. A. ALEWELT[1], K. QUINN[1], Y. FANG[1], A. BARTKE[2], E. R. HASCUP[1], ***K. N. HASCUP**[1]; [1]Neurol. / CARE, [2]Intrnl. Med., Southern Illinois Univ. Sch. of Med., Springfield, IL

Abstract: Aging is the predominant risk factor for developing Alzheimer's disease (AD), so lifespan extending therapies may be effective treatments to ameliorate cognitive decline. Reducing core body temperature (Tc) promotes healthy aging through mechanisms associated with insulin signaling and glucose homeostasis. However, Tc declines with age, perturbs memory formation, and drives aggregation of both amyloid plaques and tau tangles. This paradox led us to hypothesize that increasing, rather than lowering Tc may improve cognitive performance in mouse models of Alzheimer's disease (AD). Starting at 6 months of age (mild amyloid pathology and cognitive impairment), male and female APP/PS1 and C57BL/6 (littermate controls) were chronically housed at either 16, 23, or 30°C for 6 months. At 12 months of age (advanced amyloid pathology and cognitive decline), their metabolic profile was assessed using an insulin and glucose tolerance test followed by cognitive performance on the Morris water maze (MWM) spatial navigation task. Plasma, hippocampal, and peripheral (adipose and hepatic) samples were procured postmortem and tissue-specific markers of amyloid accumulation and metabolism were assayed. Male and female APP/PS1 mice housed at 16°C for 6 months had similar insulin sensitivity, but significantly worse glucose tolerance and cognitive performance compared to temperature-matched littermate controls. Hepatic mRNA expression levels of INSR, PI3K, GCK, G6PC were significantly reduced in male, but unchanged in female APP/PS1 mice compared to temperature-matched littermate controls. In contrast, chronically housing mice at 30°C showed significant cognitive and metabolic improvements over genotype-matched mice housed at 23°C. Male C57BL/6 mice were more insulin sensitive while male APP/PS1 mice had improved glucose tolerance. Memory recall in male APP/PS1 mice chronically housed at 30°C was significantly improved compared to genotype-matched 23°C controls and were similar to littermate controls housed at 30°C. Chronically housing female mice at 30°C improved the insulin sensitivity and glucose tolerance in C57BL/6 and made APP/PS1 mice more insulin sensitive compared to 23°C genotype-matched controls. Despite an enhanced metabolic profile after housing at 30°C, this did not lead to improved cognitive performance in female APP/PS1 mice. Hepatic mRNA expression of GCK and G6PC was reduced in female, but not male APP/PS1 mice. Although reducing Tc is beneficial for successful aging, our data supports this exacerbates cognitive deficits in a model of AD. Rather, increasing Tc may provide therapeutic benefits in males experiencing dementia.

Disclosures: S.A. McFadden: None. **M.F. Cox:** None. **C.A. Findley:** None. **M.R. Peck:** None. **L.A. Alewelt:** None. **K. Quinn:** None. **Y. Fang:** None. **A. Bartke:** None. **E.R. Hascup:** None. **K.N. Hascup:** None.

Poster

P228. Altered Energy Homeostasis in Alzheimer's Disease I

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Program #/Poster #: P228.05

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: RGC-GRF 16100219
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RGC-CRF (C4033-19EF)
NSFC-EYSF (32022087)

Title: Loss of nuclear estrogen-related receptor alpha activity impairs metabolic plasticity and mitochondrial OXPHOS in neurons: Implications of sex differences observed in Late-onset Alzheimer's disease

Authors: *H.-M. CHOW[1], J. SUN[1], M. LAU[1], R. P. HART[2];
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Abstract: Late onset Alzheimer's disease (LOAD) is the most common form of age-related dementia, causing memory loss, cognitive deficits and behavioural changes. Currently, LOAD remains untreatable and the high failure rate of clinical trials over the past decade has increased the urgency to better understand the heterogeneity of the disease in order to facilitate more personalised therapies. Females are at the epicentre of the LOAD epidemic, and that better longevity found in women over men alone is insufficient to explain such phenomenon. Previous studies indicated that neurons located within LOAD-relevant brain regions are estrogen sensitive, and that they are also one of the extra-gonadal sites of estrogen synthesis. Here we report that loss of neuronal estrogen signalling transforms a sex-biased benefit to an increased risk of pathological changes associated with LOAD in older women. Supported by our unsupervised transcriptomic meta-analyses using multiple public datasets, various nuclear-encoded genes for oxidative phosphorylation (OXPHOS) components were consistently reduced specifically in neurons of LOAD brains. When the same analysis was further stratified based on the sex of the patients, such losses were observed only in female subjects. Mechanistically, systems analyses indicated that the loss in activity of estrogen-related receptor alpha (ERRa) is the most likely link behind such changes in gene expression observed in LOAD. ERRa is an orphan receptor; our integrated ChIP-seq analysis as well as in vitro mitochondrial bioenergetic studies revealed it is an under-appreciated trans-activator of nuclear-encoded respiratory and metabolic genes in neurons. Further analyses revealed that ERRa expression was not reduced in LOAD, but insufficient cholesterol ligand availability due to reduced cellular uptake mechanism may render ERRa inactive, particularly in female subjects. Taken together, our study suggested an unexpected link between the infidelity of estrogen-mediated cholesterol homeostasis and mitochondrial dysfunction in female neurons. This situation may be further exacerbated in the presence of apolipoprotein E4 allele—the greatest genetic risk factor of LOAD, offering a new explanation to the sex-dimorphic distribution occurred in the disease pathogenesis.

Disclosures: H. Chow: None. **J. Sun:** None. **M. Lau:** None. **R.P. Hart:** None.

Poster

P228. Altered Energy Homeostasis in Alzheimer's Disease I

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Program #/Poster #: P228.06

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: K01AG057815
1R01NS079965
5AZ06

Title: Mild deficiency of mitochondrial Complex III in a mouse model of Alzheimer's disease decreases amyloid beta plaque formation

Authors: *M. PINTO[1], N. NISSANKA[1], C. GUASTUCCI[2], P. ILLIANO[2], R. BRAMBILLA[2], C. T. MORAES[1];
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Abstract: For decades, mitochondrial dysfunctions and the generation of reactive oxygen species have been proposed to promote the development and progression of the amyloid pathology in Alzheimer's disease, but this association is still debated. In particular, it is still unclear if mitochondrial dysfunctions are a trigger or rather a consequence of the formation of amyloid aggregates, and in particular, the role of the different mitochondrial oxidative phosphorylation complexes in Alzheimer's patients' brain remains poorly understood. To study how mitochondrial Complex III defects affect amyloid beta pathology *in vivo,* we partially knocked out mitochondrial Complex III ($CIII^{KO}$) in mature forebrain neurons of an Alzheimer's mouse model that develops plaque pathology (APP/PS1). We found that Complex III dysfunction in adult neurons induced mild oxidative stress which did not correlate with increased amyloid beta accumulation. In fact, $CIII^{KO}$-AD mice showed decreased plaque number, decreased Ab-42 toxic fragment and altered amyloid precursor protein cleavage pathway. Our results support a model in which mitochondrial dysfunction is not the cause of amyloid oligomer accumulation but rather a consequence of amyloid beta toxicity.

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Poster

P228. Altered Energy Homeostasis in Alzheimer's Disease I

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Topic: C.02. Alzheimer's Disease and Other Dementias

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The Family Erling-Persson Foundation

Title: Apolipoprotein CIII may link metabolic diseases and brain disorders

Authors: *P. RECIO-LÓPEZ, I. VALLADOLID-ACEBES, P.-O. BERGGREN, L. JUNTTI-BERGGREN;
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Abstract: Apolipoprotein CIII (apoCIII) is a pro-inflammatory factor that increases in obesity and diabetes mellitus (DM). Extensive research has been done to link different apolipoproteins with neurodegenerative diseases. However, the presence of apoCIII in the central nervous system (CNS) and its relationship to metabolic disorders have never been investigated. We used leptin-deficient obese diabetic (*ob/ob*) mice and lean wild type controls, to investigate whether apoCIII is present in CNS and if it is an inducible factor that can be modulated by RNA-based therapy. Expression of apoCIII was analyzed in fresh frozen brains, microdissected *in loco* from five *ob/ob* mice and five controls, by quantitative Real-Time polymerase chain reaction. We found that apoCIII was up-regulated in the frontal cortex, hypothalamus, hippocampus, thalamus, substantia nigra, pons and cerebellar cortex in *ob/ob* compared to control mice. Thereafter *ob/ob* mice were injected intravenously with a mix of two apoCIII-specific siRNAs, or an inactive siRNA as control. There were five mice in each group and injection doses of siRNAs and inactive siRNA were 1mg/kg for three consecutive days. When *ob/ob* mice were given the apoCIII siRNA mix, apoCIII levels were reduced in hypothalamus and thalamus. Importantly, we have now demonstrated the presence of apoCIII in mouse brain. We provide evidences that apoCIII can be produced locally in the brain under conditions of obesity and DM and that its expression can be effectively modulated by siRNA. Hence, our findings not only suggest that brain apoCIII might have a role linking metabolic diseases with neuronal dysfunction but also a tentative way for therapeutic intervention.

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Poster

P228. Altered Energy Homeostasis in Alzheimer's Disease I

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Program #/Poster #: P228.08

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: R01AG057767
R01AG061937
Dale and Deborah Smith Center for Alzheimer's Research and trEatment
Kenneth Stark Endowment

Title: Effects of Visceral Fat Removal on Insulin Sensitivity and Cognition in $APP^{NL-F/NL-F}$ Mice

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Abstract: Background: Visceral fat (VF) contributes to the development of insulin resistance, type 2 diabetes, and dyslipidemia that are risk factors for developing Alzheimer's disease (AD). Previous work from our laboratories demonstrates that VF pad removal (VFR) improves insulin sensitivity and glucose tolerance in the growth hormone receptor knockout mouse model of successful aging. We hypothesized that VFR would improve metabolic signaling and slow disease progression in an AD mouse model. **Methods:** At 4-5 months of age, prior to cognitive decline, male and female $APP^{NL-F/NL-F}$ (knock-in) and C57BL/6 (genetic background controls) underwent either VFR or sham surgery. In the VFR group, perigonadal and perinephric adipose tissue was removed in both sexes. For the sham operation, similar incisions were made and the same adipose depots were mobilized but not removed. Insulin and glucose tolerance tests (ITT, GTT) were conducted at 18 months of age to examine metabolic function followed by cognitive assessment using novel object recognition (NOR). **Results:** Preliminary metabolic data support an increase in insulin sensitivity in the C57BL/6 male VFR group (n=5) when compared to their sham control mice (n=2). However, this trend was not seen between the male $APP^{NL-F/NL-F}$ VFR (n=7) and sham (n=13) groups. VFR in female $APP^{NL-F/NL-F}$ mice (n=7) significantly impaired insulin sensitivity compared to the sham group (n=5; $p<0.01$). This difference was not observed in the female C57BL/6 sham group. No change in glucose tolerance was observed in either sex in any of the genotypes. VFR negatively impacted NOR performance in female VFR mice ($p<0.03$) when compared to their sham littermates. A similar trend was seen in the male $APP^{NL-F/NL-F}$ VFR mice. **Conclusions:** Prodromal VFR significantly affected insulin signaling in male control mice, but this improvement was not seen in male $APP^{NL-F/NL-F}$. This, coupled with the absence of changes to glucose tolerance, support that VFR does not significantly alter metabolic signaling in male $APP^{NL-F/NL-F}$ mice. However, the significant reduction of insulin sensitivity observed in female $APP^{NL-F/NL-F}$ mice after VFR, but not C57BL/6 controls, points to sex-dependent VF function in insulin signaling in AD. Reduced cognitive performance in both sexes of $APP^{NL-F/NL-F}$ VFR mice demonstrates that changes in peripheral insulin signaling may not contribute to VFR-related cognitive decline. Future studies investigating VFR at 16 months of age could help elucidate the influence of VF on insulin signaling and cognition during later stages of disease progression in these mice.

Disclosures: S.A. McFadden: None. **C.A. Findley:** None. **A. Barke:** None. **K.N. Hascup:** None. **E.R. Hascup:** None.

Poster

P228. Altered Energy Homeostasis in Alzheimer's Disease I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P228.09

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: CNPq (Grant number: 310347/2018-1).
CAPES
Congresswoman Beatriz Cerqueira (PT-MG)
FAPEMIG

Title: Cholesterol diet promotes improvement of cognitive decline in mice models of Alzheimer's disease.

Authors: *S. KOERICH, G. L. MARTINS, M. L. N. BRANT, G. G. FREITAS, W. F. BRAGA, H. J. REIS, A. C. P. DE OLIVEIRA;
UFMG: Univ. Federal de Minas Gerais, Belo Horizonte, Brazil

Abstract: Introduction: Alzheimer's disease (AD) is the main cause of dementia in the world, in which behavioral changes, cognitive impairment and progressive memory loss are observed. Despite its high social impact, the pathophysiological mechanisms involved in the disease need further elucidation. It has recently been suggested that cholesterol homeostasis is involved in the pathophysiology of AD. In addition, recent studies demonstrated the influence of a cholesterol diet on cognition of mice models of AD. However, the effects of cholesterol diet on AD pathology are still controversial. To explore this issue, we investigated the effect of a cholesterol diet in a memory paradigm in different models of AD. Aim: The present study aimed to investigate the effects of cholesterol on cognitive impairment and hepatic lipid profile in models of neurotoxicity induced by Aβ and in the transgenic mouse model for AD. Methods: For this, male C57Bl/6 mice, aged 8 weeks, were treated with 1.25% of cholesterol or control diet for four months, then received intrahippocampal microinjection of Aβ (400 pmol) or saline. Additionally, we evaluated the effects of the cholesterol diet in the APP/PS1 transgenic model. For this purpose, male and female APP/PS1, aged 10 months, were treated for four months with a 1.25% cholesterol or control diet. For cognitive evaluation, we performed the novel object recognition test (NORT). In addition, hepatic lipids (triglycerides and cholesterol) were measured in the liver. All the experiments followed the principles of animal experimentation adopted by the Animal Use Ethics Committee of the Federal University of Minas Gerais (UFMG), approved under the number 374/2018. This is an exploratory research. Results: Even considering the higher hepatic cholesterol and triglycerides in the group that received cholesterol diet, in both

APP/PS1 and Aβ models, we observed that the cholesterol diet reversed cognitive deficit in NORT in male and female APP/PS1 mice and prevented cognitive deficit induced by Aβ 1-42. Conclusion: These results indicate that a cholesterol diet induces beneficial cognitive effects, despite appearing to induce systemic alterations, such as those observed in the liver. This data enhances the knowledge about the complexity of the pathogenesis of AD.Financial Support: FAPEMIG; CNPq (Process number: 310347/2018-1); CAPES; Congresswoman Beatriz Cerqueira (PT-MG) provide financial support directly from the House of Representatives.

Disclosures: S. Koerich: None. **G.L. Martins:** None. **M.L.N. Brant:** None. **G.G. Freitas:** None. **W.F. Braga:** None. **H.J. Reis:** None. **A.C.P. de Oliveira:** None.

Poster

P228. Altered Energy Homeostasis in Alzheimer's Disease I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P228.10

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: K99AG065445

Title: Neuronal loss of NCLX-dependent mitochondrial calcium efflux contributes to cognitive decline

Authors: *P. JADIYA[1], D. KOLMETZKY[2], D. TOMAR[2], J. ELROD[2];
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Abstract: Neuronal loss of NCLX-dependent mitochondrial calcium efflux contributes to cognitive decline
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Abstract:
Alzheimer's disease (AD) is characterized by memory loss, neuronal dysfunction, and the deposition of amyloid β plaques and tau neurofibrillary tangles. These AD-phenotypes are associated with mitochondrial dysfunction and impaired intracellular calcium homeostasis. We recently reported that mitochondrial calcium ($_{m}Ca^{2+}$) overload, due to remodeling of the mitochondrial calcium uniporter channel (mtCU) and by early neuronal loss of the mitochondrial Na^{+}/Ca^{2+} exchanger (NCLX), occurs before the appearance of AD-associated phenotypes. The loss or re-expression of NCLX in an AD mouse aggravates or reduces AD-associated memory decline and pathology, respectively. However, whether disrupted $_{m}Ca^{2+}$ signaling causally contributes to AD development or progression independent of amyloid-beta remains enigmatic. In the present study, we generated wild-type mice with neuronal-specific deletion (Camk2a-Cre)

of NCLX and evaluated age-associated changes in cognitive function and neurohistology. We found that neuronal-specific loss of $_{m}Ca^{2+}$ efflux in aged mice (12-15 months) impaired spatial and cued recall memory, elicited minor amyloidosis and tau hyperphosphorylation, and caused synaptic remodeling. Our findings suggest that loss of NCLX-dependent $_{m}Ca^{2+}$ efflux alone is sufficient to induce AD-like pathology in aged mice. These findings further advocate for modulating $_{m}Ca^{2+}$ exchange as a therapeutic strategy in AD.

Disclosures: P. Jadiya: None. **D. Kolmetzky:** None. **D. Tomar:** None. **J. Elrod:** None.

Poster

P252. Mitochondrial Mechanisms and Genetics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P252.01

Topic: C.03. Parkinson's Disease

Support: NIH Grant DK113652
Center for Compulsive Behavior & Addiction

Title: Assessment of endogenous synuclein in olfactory circuits and striatum in a progressive rat model of Parkinson's disease

Authors: *B. DESAI BRADARIC[1], I. D. CALMA[1], A. F. S. SHACK[2], J. G. NOEL-TORRES[3], G. K. MCCORMACK[4], A. E. KIRBY[1], A. L. PERSONS[1], T. C. NAPIER[1];
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Abstract: Methamphetamine (meth) abuse increases the risk to develop Parkinson's disease (PD) later in life. To decipher neurobiological links between meth abuse and PD, we use meth self-administering (SA) rats undergoing protracted abstinence to study prodromal and early stages of PD. Hyposmia is a prodromal symptom of PD. We hypothesized that olfactory brain circuits will exhibit pathogenic markers in rats with a history of meth SA. The mitochondrial toxin rotenone, which produces PD, synergizes with meth to dysregulate mitochondria and produce PD-like brain and behavior pathology (see Calma Persons Napier, this meeting). Pathological α-synuclein can interfere with mitochondrial ATP production. Accordingly, we conducted immunohistochemical evaluations of α-synuclein in the olfactory bulb (OB) and output structures, the anterior olfactory nucleus (AON) and piriform cortex (PIR) in meth SA (male, Sprague-Dawley) rats with and without subthreshold doses of rotenone. Meth was provided as 0.1mg/kg per 0.1mL infusion in daily 3h sessions for 2 weeks; controls were saline-yoked. Rotenone was delivered as 1mg/kg/day for 6 days *via* subcutaneous osmotic minipumps starting on day 14 of meth abstinence; controls received vehicle only. Brains were harvested 60 days after meth (n=5-9 per treatment). Meth SA rats exhibited progressive deficits in postural

instability, and there was an increase in synuclein-stained cell bodies in the nigra, which were both enhanced by rotenone, validating the relevance of this model to PD. Also as reported for PD, AON area was reduced by meth ($p<0.05$). α-Synuclein staining was not changed in the OB, AON or PIR layer 1 by meth or rotenone. In PIR layer 2, α-synuclein was decreased by 50% in meth SA rats and by 48% with rotenone ($p<0.05$); however, meth+rotenone was not different from either treatment alone. To ascertain if similar results were obtained in a nigral output, we measured striatal α-synuclein. Meth reduced striatal staining by 43% ($p<0.05$), but no changes were observed for rotenone alone or meth+rotenone. These outcomes illustrate regional differences in synuclein regulation. As α-synuclein was measured 60 days after meth exposure and 40 days after rotenone, results may reflect compensation to initial exposure, or incubation, a phenomenon well-described for meth-induced seeking behaviors. α-Synuclein is degraded by a ubiquitin pathway, and ubiquitin is increased in humans with fatal meth overdose (Quan *et al, Legal Med*, 2005). Increased metabolism would explain the observed meth-induced reductions in α-synuclein and mitochondrial dysregulation. Ongoing studies are determining if such mechanism link meth abuse to PD.

Disclosures: B. Desai Bradaric: None. **I.D. Calma:** None. **A.F.S. Shack:** None. **J.G. Noel-Torres:** None. **G.K. McCormack:** None. **A.E. Kirby:** None. **A.L. Persons:** None. **T.C. Napier:** None.

Poster

P252. Mitochondrial Mechanisms and Genetics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P252.02

Topic: C.03. Parkinson's Disease

Support: RTI2018-095311-B-100

Title: Differential mitochondrial dysfunction in striatum of HD depending on astrocyte population

Authors: *L. LOPEZ-MOLINA[1,2,3], A. PEREDA-VELARDE[1,2,3], S. GINES[1,2,3];
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Abstract: Huntington's Disease (HD) is a neurodegenerative disorder characterized by the selective loss of striatal medium spiny neurons. Although the molecular mechanisms of this striatal vulnerability remain unclear, compelling evidences point out that mitochondrial dysfunction could play a major role. However, much of what we know regarding the mechanisms underlying mitochondrial disturbances in HD has been gained from studies

conducted in neurons. Less is known about the role and regulation of mitochondrial dynamics in glia, particularly in astrocytes, the predominant type of glia cell in the brain. Indeed, relatively unexplored is whether mitochondrial dynamics in astrocytes may also contribute to neuron disturbances and therefore, to pathology in HD. Hence, we aimed to define the specific contribution of astrocytic mitochondria as well as the intercellular mitochondria communication between neuronal and astrocytes to HD pathology. In this work, we used the transgenic R6/1 mouse model of HD to either isolate adult astrocytes at different stages of the disease and to culture primary striatal astrocytes. Since astrocytes is a complex and heterogeneous population, we evaluated several astrocyte markers at the striatum of R6/1 mouse throughout the disease using biochemical and immunofluorescence assays. Then, we studied how mitochondria could be affected in HD striatal astrocytes depending on specific population. We could observe that mitochondrial abnormalities in HD astrocytes differed whether it occurs in GLAST+ or GFAP+ astrocytes. Finally, preliminary results showed that mitochondria could be a crosstalk mechanism between astrocytes and neurons and HD condition could hamper this cell-to-cell communication.

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Poster

P252. Mitochondrial Mechanisms and Genetics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P252.03

Topic: C.03. Parkinson's Disease

Support: NIH R21ES025920
the Center for Compulsive Behavior & Addiction

Title: Changes in Striatal Mitochondria in a Rat Model of Methamphetamine-Induced Increased Risk for Parkinson's disease

Authors: *I. CALMA[1], A. L. PERSONS[2], T. NAPIER[3];
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Abstract: Parkinson's disease (PD) is a progressive degeneration of nigrostriatal dopaminergic neurons resulting in motor disturbances and the emergence of α-synuclein rich protein aggregates in several brain regions, including the substantia nigra. Methamphetamine (meth) abuse increases the risk for developing PD. We revealed that meth self-administering (SA) rats have a force abstinence (FA) time-dependent loss of nigrostriatal tyrosine hydroxylase (TH), a marker for dopamine neurons (Kousik et al., JPET 351:432, 2014), and that this loss in TH is parallel by the emergence of PD-like postural instability (Persons et al., DAD 225:108746, 2021). Mitochondrial health is important for proper energy production and normal cell function, and mitochondrial dysfunction is implicated in meth toxicity and the pathogenesis of PD. We

hypothesized that meth-induced trajectory towards PD is associated with striatal mitochondrial dysfunction. Male Sprague Dawley rats were trained for meth SA (0.1mg/kg/0.1mL/infusion) 3h daily for 2 weeks or were saline-yoked. We administered subthreshold (behaviorally ineffective) doses of the mitochondrial toxin, rotenone, or vehicle *via* subcutaneous osmotic minipumps during FA days 14-20. Meth SA rats exhibited progressively worsening motor deficits and reductions in striatal TH that were exacerbated by rotenone ($p<0.05$). The rats were killed on FA day 62, and brain harvested. In the substantia nigra, the number of α-synuclein+ cells was increased by meth and the effect was enhanced by rotenone ($p<0.01$). Several markers of mitochondrial dysfunction were discovered in the striatum. Cytochrome c translocation to the cytosol was increased by meth and further enhanced by rotenone ($p<0.01$). Rotenone reduced levels of electron transport chain Complex I and II protein ($p<0.05$) and Complex I activity ($p<0.01$). Meth reduced Complex II protein levels ($p<0.05$), and meth-induced reductions in Complex I and Complex II activity were potentiated by rotenone ($p<0.01$). These findings reveal that mitochondrial dysfunction occurs with SA meth, these effects were observed months after terminating meth and were potentiated by subthreshold doses of rotenone, and the brain pathology parallels with motor deficits seen in early stages of PD. Thus, protracted dysregulation of mitochondria may underpin the link between meth-abuse and PD.

Disclosures: I. Calma: None. **A.L. Persons:** None. **T. Napier:** None.

Poster

P252. Mitochondrial Mechanisms and Genetics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P252.04

Topic: C.03. Parkinson's Disease

Support: Southern Research Change Campaign
R01NS101958
MJFF #16964

Title: Investigating the Neuroanatomical Distribution of Disease-implicated transcripts to explore cell-type-specific etiologies in Parkinson's Disease

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for Neurodegenerative Sci., Van Andel Inst., Grand Rapids, MI; [9]Psychiatry and Behavioral Neurobio., Univ. of Alabama, Birmingham, Birmingham, AL; [10]Van Andel Res. Inst., Grand Rapids, MI; [11]Biol. Sci., The Univ. of Alabama, Tuscaloosa, AL; [12]Psychiatry, UAB Med. Ctr., Birmingham, AL

Abstract: Parkinson's disease (PD) is a progressive movement disorder characterized by the loss of midbrain dopaminergic (DAergic) neurons and widespread accumulation of intracellular aggregates containing misfolded α-synuclein (Lewy bodies; LB). LB pathology is observed across multiple brain regions in PD, however, hallmark motor symptoms are attributed to the selective loss of DAergic terminals and neurons in the basal ganglia. Despite the identification of risk genes for inherited forms of PD, the etiological contributors to the vulnerability of DAergic neurons in idiopathic PD are not known. While genetic risk variants for PD provide starting points for exploring mechanistic contributors to disease etiology, it has been challenging to determine ways to use this information to uncover convergent mechanisms of disease. Here, single-cell transcriptomic data from mouse brain were leveraged to identify enrichment patterns of PD-implicated genes. Gene lists were derived from statistical clustering based on region- and cell-specific enrichment patterns, with empirical confirmation of top prototypical genes in mouse and human brain. Additionally, the top DAergic prototypical gene was modified in a *C. elegans* model of synucleinopathy; modification of this gene was shown to affect DAergic neuronal viability in this model. Single-cell data was further utilized to identify a subpopulation of midbrain DAergic neurons vulnerable to loss in a mouse model of synucleinopathy. Altogether, these results provide a strategy to prioritize and contextualize PD-linked risk genes for biological studies, highlighting the value of cell-type-specific resolution.

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Poster

P252. Mitochondrial Mechanisms and Genetics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P252.05

Topic: C.03. Parkinson's Disease

Support: Greehey Family Foundation Endowment for Alzheimer's Research
Bartell and Mollie Zachry Endowment for Alzheimer's Disease

Title: excitotoxic commitment to die in dopaminergic neurons is associated with changes in mrna expression consistent with failure of the mechanism of internalization of ampa receptors. p { margin-bottom: 0.1in; line-height: 115%; background: transparent }

Authors: O. MALDONADO[1], L. PADILLA[2], *G. A. DE ERAUSQUIN[1];
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Abstract: Mesencephalic dopaminergic neurons (DN) in primary culture are selectively susceptible to excitotoxic injury mediated by activation of the α-amino-3-hydroxy-5-methylisoxazole-4-propionic acid (AMPA) glutamate receptors, through a pathway requiring loss of intracellular calcium homeostasis, opening a mitochondrial permeability transition, nuclear translocation of nuclear factor kappa B (NFkB) and phosphorylation of the transcription factor p53 leading to programmed neuronal death. Glyceraldehyde-3-phosphate dehydrogenase (GAPDH) acts as a stimulator of programmed cell death, and forms protein complexes with glutamate receptor AMPA type-2 subunit (GluA2) leading to its internalizion during excitotoxicity in hippocampal neurons in culture. Further, nuclear accumulation of GluA2, GAPDH, and p53 were observed in the substantia nigra of Parkinson's disease post-mortem. We evaluated the expression of GAPDH and GluA2 mRNA in single dopaminergic neurons during an excitotoxicity challenge. Primary mesencephalic mouse cultures were exposed to conditioned medium alone, or containing 30 or 100 μM AMPA for 1, 3, or 6 h (the latter known to result in commitment to die in this model). DN were labeled with 5,7, dihydroxytrptamine (DHT) and manually picked live under fluorescent microscopy, and the dopaminergic phenotype was confirmed by expression of Tyrosine Hydroxylase (TH) and Paired-Like Homeodomain Transcription Factor 3 (pitx3) mRNA. Single DN were then processed for multiplex microfluidic PCR gene expression for genes included in the molecular pathway of AMPA induced excitototoxicity in DN. We found that commitment to die is correlated with dose-dependent significant increases in GluA2 expression, as well as decreses in GAPDH and p53. These results suggest a failure of the mechanism of internalization of AMPA receptors at the time of commitment to die in DN. p { margin-bottom: 0.1in; line-height: 115%; background: transparent }

Disclosures: O. Maldonado: None. **L. Padilla:** None. **G.A. de Erausquin:** None.

Poster

P252. Mitochondrial Mechanisms and Genetics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P252.06

Topic: C.03. Parkinson's Disease

Support: R01NS101958

MJFF #10758
F99AG068428

Title: Enhanced transcriptional drive through estrogen related receptor gamma (Esrrg) for mitochondrial and synaptic gene programs protects dopaminergic terminals from synuclein-mediated toxicity

Authors: *S. FOX[1], L. MCMEEKIN[2], C. SAVAGE[1], K. JOYCE[1], G. HAGGARD[1], S. BOAS[1], M. SIMMONS[2], J. PAUL[1], C. FARMER[1], J. AUWERX[3], S. SUDARSHAN[1], J. MA[4], K. GAMBLE[1], R. ROBERTS[1], D. CROSSMAN[1], A. KRALLI[5], R. COWELL[2];
[1]Univ. of Alabama At Birmingham, Birmingham, AL; [2]Southern Res., Birmingham, AL; [3]Swiss Federal Inst. of Technol. Lausanne, Lausanne, Switzerland; [4]Van Andel Inst., Grand Rapids, MI; [5]Johns Hopkins, Baltimore, MD

Abstract: Many studies implicate mitochondrial dysfunction as a key contributor to age-related neurodegenerative diseases, including Parkinson Disease (PD). Previously published gene expression analyses from laser captured dopaminergic (DAergic) neurons from the substantia nigra of patients with Lewy-body pathology revealed a deficiency in nuclear-encoded genes for mitochondrial respiration. The functional consequences of these changes and their role in disease progression and pathology are still unknown. The coordinated downregulation of a large number of nuclear-encoded mitochondrial genes suggests the disruption of a central regulator for mitochondrial gene expression. In fact, most affected genes are putative targets for the transcription factor estrogen-related receptor gamma (*Esrrg*/*ESRRG*), and several recent studies indicate that in addition to regulating mitochondrial genes, *Esrrg* can modulate dopaminergic phenotype and neuritogenesis. Here, we determine the dependence of DAergic neurons on *Esrrg* for gene expression, survival, and regulation of motor function and explore how cell type-specific manipulation of *Esrrg* expression influences DAergic neuron vulnerability in the pre-formed fibril model of synucleinopathy. We demonstrate that, while knockdown is well-tolerated in normal adult mice, it leads to a reduction in the number of mitochondria in DAergic neurons and renders DAergic terminals and neurons more susceptible to synuclein toxicity. Remarkably, overexpression of *Esrrg* provides neuroprotection from alpha-synuclein-induced toxicity at the terminals. Translating ribosome affinity purification-mediated transcriptional profiling of DAergic neurons revealed that *Esrrg* is required for the expression of genes related to synaptic vesicle cycling, vesicle-mediated transport, and tricarboxylic acid metabolic processes; however, overexpression was associated with a robust induction of non-*Esrrg*-dependent genes involved in mitochondrial respiration. In contrast to adulthood deletion, developmental deletion caused a reduction in classic transcriptional markers for DAergic neurons without causing overt cell loss. Interestingly, differences in behavioral sensitivity to amphetamine across the various mouse lines revealed a positive association between DA availability and eventual neuron loss. Altogether, these findings indicate that *Esrrg* is required for normal transcription and function of DAergic neurons and that overactivation of *Esrrg*-dependent transcriptional programs should be considered as a strategy for maintaining DAergic innervation in PD.

Disclosures: S. Fox: None. **L. McMeekin:** None. **C. Savage:** None. **K. Joyce:** None. **G. Haggard:** None. **S. Boas:** None. **M. Simmons:** None. **J. Paul:** None. **C. Farmer:** None. **J.**

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Poster

P252. Mitochondrial Mechanisms and Genetics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P252.07

Topic: C.03. Parkinson's Disease

Support: R01NS101958
Alabama Power Foundation

Title: Multiple roles for estrogen-related receptor-gamma in gene regulation in neurons

Authors: K. L. JOYCE[1], S. N. FOX[1], J. A. MOBLEY[1], L. PEREBOEVA[1], ***R. M. COWELL**[2]; [1]Univ. of Alabama at Birmingham, Birmingham, AL; [2]Dept. of Neurosci., Southern Res., Birmingham, AL

Abstract: The dysregulation of mitochondrial and synaptic genes occurs with neurodegeneration. Recently, members of the estrogen-related receptor (ERR) transcription factor family have been implicated in the regulation of mitochondrial and synaptic genes in the brain. Previous and ongoing studies indicate that ERRγ is involved in the regulation of cellular identity and neuritogenesis-related genes in dopaminergic neurons, yet little is known about its putative gene targets or interacting proteins in these cells. Here, we performed RNA-Seq and BioID proximity assays in an SH-SY5Y neuroblastoma line to identify putative ERRγ targets and interacting factors in dopaminergic neuron-like cells. In doing so, we discovered that 800 genes are upregulated and 3000 genes downregulated by ERRγ overexpression. Among these genes is ADARB2, an enzyme involved in RNA editing, and NEAT1_2, a long non-coding RNA which forms a scaffold for nuclear paraspeckles. Interestingly, BioID studies determined that overexpressed ERRγ resides in close proximity to proteins enriched in paraspeckles, such as SFPQ and FUS. Accordingly, ERRγ overexpression increases, and shRNA-mediated knockdown decreases, the abundance of paraspeckle foci in SH-SY5Y cells. Other putative ERRγ-interacting proteins of interest included proteins involved in RNA editing, splicing and trafficking, suggesting a more complex role for ERRγ in gene regulation beyond its role as a DNA-binding transcription factor. Furthermore, it is interesting to note that ERRγ was in close proximity to both transcriptional coactivators and repressors, suggesting that it can be recruited into both transcriptionally active and repressed transcriptional complexes, depending on the locus. Neuroanatomical localization of a subset of altered genes and co-factors to cell types in the mouse brain guided studies to test for ERRγ-responsive gene expression in mice lacking ERRγ in different cell types, demonstrating the utility of these studies for understanding ERRγ's roles *in*

vivo. Future experiments will explore whether alterations in ERRγ and its targets are associated with dopaminergic vulnerability in Parkinson's Disease.

Disclosures: K.L. Joyce: None. **S.N. Fox:** None. **J.A. Mobley:** None. **L. Pereboeva:** None. **R.M. Cowell:** None.

Poster

P268. Therapeutic Strategies: Preclinical Animal Models: Other Therapeutics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P268.01

Topic: C.03. Parkinson’s Disease

Support: European Research Council grant agreement no. REP-SCI-693038
Lundbeck Foundation (R276-2018-183)
Novo Nordisk Foundation Laureate Program (NNF15OC0016016)
The Swedish Research Council

Title: Targeted activation of midbrain neurons restores locomotor function in mouse models of parkinsonism

Authors: *D. MASINI, O. KIEHN;
Dept. of Neurosci., Copenhagen Univ., Copenhagen, Denmark

Abstract: The pedunculopontine nucleus (PPN) is a locomotor command area in the midbrain that contains glutamatergic neurons directly involved in the control of locomotor initiation and maintenance, especially slower explorative locomotion. Motor output is suppressed in Parkinson’s disease (PD), where dopaminergic neurodegeneration alters basal ganglia activity leading to strong inhibition of its targets. The PPN is a subregion of the MLR that is under direct inhibitory control from the basal ganglia. Activating the PPN to release it from the excessive basal ganglia inhibition in PD is, therefore, a logical step to ameliorate Parkinsonian locomotor deficit. However, when deep brain stimulation targeting the MLR is applied in the clinics, it exhibits variable and sometimes no locomotor improvement. In the present study we hypothesize that the lack of consistent clinical results is caused by the variable and/or nonspecific activation of the MLR. We therefore use cell-type-specific chemogenetic and optogenetic approaches to activate localized MLR subpopulations in two distinct mouse models of parkinsonism. To induce an acute parkinsonian-state we pharmacologically interfere with dopaminergic signaling by antagonizing D1- or D2-type dopamine receptors. We show that excitation of caudal glutamatergic PPN neurons can alone, normalize the otherwise severe locomotor deficit in PD, whereas targeting the GABAergic PPN population is not sufficient to support full locomotor recovery. Moreover, we compare the motor recovery driven by glutamatergic PPN stimulation to

that of the Cuneiform nucleus. Our observations pinpoint caudal glutamatergic PPN neurons as an optimal target for neuromodulatory restoration of locomotor function in Parkinson's disease.

Disclosures: D. Masini: None. **O. Kiehn:** None.

Poster

P268. Therapeutic Strategies: Preclinical Animal Models: Other Therapeutics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P268.02

Topic: C.03. Parkinson's Disease

Support: Aufzien Family Center for the Prevention and Treatment of Parkinson's Disease (APPD)
ERA-NET, ADNPinMED, Israel Ministry of Health
Elton Laboratory
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Selma Schottenstein Harris Lab for Research in Parkinson's
Gardner Family Center for Parkinson's Disease and Movement Disorders

Title: The microtubule-protecting drug candidate SKIP modifies Parkinson's disease deficits: rotenone and 6-hydroxydopamine neurotoxicity models

Authors: Y. IVASHKO-PACHIMA[1], K. B. SEROOGY[2], Y. SHARABI[3], ***I. GOZES**[1]; [1]Sch. Med/Tel Aviv Univ., Tel Aviv, Israel; [2]Dept Neurol, Univ. of Cincinnati, Cincinnati, OH; [3]Sheba & Sch. Med/Tel Aviv Univ., Tel Aviv, Israel

Abstract: Parkinson's disease (PD) poses a significant challenge to public health, requiring disease-modifying therapies. The Gozes laboratory discovered activity-dependent neuroprotective protein (ADNP), which is deregulated (decreased) in PD (Chu et al., J. Parkinsons Dis., 2016; Hadar et al., Mol. Psychiatry, 2021). ADNP contains a NAPVSIPQ (NAP) domain, including the SxIP motif, which interacts with microtubule (MT) end-binding proteins (EB1, EB3) enhancing/replacing ADNP-MT interactions, thereby regulating axonal transport and synapse formation. The laboratory further designed SKIP (based on the SxIP MT-interacting motif in NAP). SKIP interacts with NAP in ADNP to protect MTs, presenting a potential drug candidate for PD disease modification. Here, we used both cell culture and animal models to determine if SKIP provides in vitro protection against rotenone and in vivo attenuation of nigrostriatal 6-hydroxydopamine (6-OHDA) neurotoxicity. Rotenone, a well-known pesticide/insecticide, inhibits mitochondrial complex I and depolymerizes MTs, combining catecholamines and MTs in PD toxicity. Using live cell imaging and morphometrics at the microscopical level, we found significant protection of neuronal-like cells (neuroblastoma SH-

SY5Y and N1E-115 cells) by SKIP against rotenone challenge. Mechanistically, rotenone disrupted MT dynamics, which SKIP partially repaired through EB1/EB3, coupled with increasing neurite branch length. We chose the established rat model of nigrostriatal 6-OHDA neurotoxicity to initially evaluate SKIP in vivo in a pilot study. Using the intrastriatal 6-OHDA partial lesion model in adult Sprague Dawley rats, we found that supranigral SKIP pretreatment (2 weeks prior to 6-OHDA lesion) improved motor behavior in the forelimb asymmetry test and protected against nigral dopaminergic cell degeneration (as determined by tyrosine hydroxylase immunohistochemistry and stereological cell counts) at 4-weeks post-lesion (Ivashko-Pachima et al., J. Mol. Neurosci., 2021, in press). These findings are the first to demonstrate the neuroprotective potential of SKIP against neurotoxin-induced neuronal impairment and dopaminergic neurotoxicity. With PD afflicting millions of people globally, and with disease modification challenges, SKIP holds promise for future therapeutic development.

Disclosures: Y. Ivashko-Pachima: None. **K.B. Seroogy:** None. **Y. Sharabi:** None. **I. Gozes:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); SKIP is under patent protection, Ramot at Tel Aviv University.

Poster

P268. Therapeutic Strategies: Preclinical Animal Models: Other Therapeutics

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Program #/Poster #: P268.03

Topic: C.03. Parkinson's Disease

Support: NIH NINDS R01 NS107336
Mayo Clinic

Title: Effect of subthalamic high frequency stimulation on exploratory behaviors in a parkinsonian animal model

Authors: *S. G. HILLAN[1], C. B. MULDER[2], J. L. SILVERNAIL[3], W. D. LUJAN[3], S. L. BOSCHEN DE SOUZA[3], J. L. LUJAN[3];
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Abstract: A well-established treatment for the motor symptoms of Parkinson's disease (PD) is high frequency deep brain stimulation (DBS) of the subthalamic nucleus (STN). Despite the effectiveness of DBS in treating the motor symptoms of PD, a subset of patients experience side effects from stimulation, including psychiatric side effects like anxiety. Stimulation-evoked anxiety commonly manifests in these patients six months or more following the onset of stimulation, though anxiety has also been identified as a transient side effect immediately

following surgery. Given the proximity of the stimulation target to the limbic region of the STN, anxiety may be the result of stimulation spread. However, the mechanisms responsible for stimulation-induced psychiatric side-effects are not well understood. A complicating factor is that psychiatric disorders have a high comorbidity with PD, with psychiatric symptoms presenting earlier than motor symptoms. Interestingly, psychiatric disorders are an exclusion criterion for DBS for PD. Thus, there is a need to better understand how STN DBS induces anxiety and other psychiatric side-effects. The goal of this study is to better understand the effects of DBS on anxiety-like symptoms by investigating changes in exploratory behaviors in a pre-motor rodent model of PD undergoing chronic and acute DBS. This study uses a partial 6 hydroxydopamine lesion to preserve motor function and reduce movement deficits that can affect behavior, while still exhibiting psychiatric symptoms associated with PD. The small size of the rodent STN relative to the size of the simulating electrode (both in the order of 1 mm) prevents selective targeting of the motor region. As such, stimulation spread to the entire STN can be expected. However, we expect larger increases in anxiety-like behaviors in the chronically stimulated cohort due to long-term stimulation-induced changes in neuroplasticity. Next steps include characterizing changes in neural activity in brain regions associated with anxiety-like behaviors to better understand the pathways responsible for inducing anxiety in response to DBS.

Disclosures: S.G. Hillan: None. **C.B. Mulder:** None. **J.L. Silvernail:** None. **W.D. Lujan:** None. **S.L. Boschen De Souza:** None. **J.L. Lujan:** None.

Poster

P268. Therapeutic Strategies: Preclinical Animal Models: Other Therapeutics

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P268.04

Topic: C.03. Parkinson's Disease

Support: DOD Grant W81XWH-19-1-0776

Title: Improving the forced running wheel as a neuroprotective treatment in mouse models of Parkinson's disease

Authors: *H. SKELTON[1], D. GROGAN[2], J. RUBIANO[3], K. BERGLUND[4], C.-A. N. GUTEKUNST[4], R. E. GROSS[5];
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Abstract: Exercise is associated with lower risk for and slower progression of Parkinson's disease (PD) and is demonstrably neuroprotective in animal models. If the underlying mechanisms were better understood, they could provide an avenue towards disease-course

modifying treatment with neuromodulation. However, exploring that physiology requires an exercise modality that is repeatably neuroprotective and conducive to the relevant assays, particularly tethered electrophysiologic recording. We settled on forced wheel running as an ideal solution to this problem, but were not convinced that existing commercial wheels were suitable. We developed a new forced running wheel platform designed to be comfortable to run on and difficult to grab onto. It also allowed high quality video recording and can be used with tethered mice. We then tested whether exercise on this new platform could provide neuroprotection in mice lesioned with 6-hydroxydopamine (6-OHDA). For that study, exercise was conducted according to a fixed, 9-week schedule with 30-minutes of wheel running per day at up to 12 m/min. Eight-week old, male C57Bl/6J mice were trained to run for 5 weeks. After training, they received a unilateral 6-OHDA injection in the striatum, after which they were exercised for 4 weeks followed by tissue collection. Exercise consisted of daily 30-minute wheel running at up to 12 m/min. We ran mice in either a commercial wheel or the one we developed and compared their neurodegenerative outcome to that of a sedentary group of mice. To assess neurodegeneration, tyrosine hydroxylase immunoreactive (TH+) cell counts in the substantia nigra on the lesioned side were compared to the non-lesioned side using stereology. Though it was difficult to rigorously quantify running performance on the commercial wheel, it was quite poor, with most mice refusing to run when it got above 8 m/min. These mice had a mean TH+ cell deficit of 58%, which was not significantly different from the mean of the sedentary control group, 74% (p=.010, independent t-test). Mice ran much more consistently on the new wheel, which also allowed for quantification of running via precise markerless, video pose tracking. These mice had a mean 41% loss of TH+ cells, significantly less than the sedentary control group (p=0.0021). In conclusion, our new forced wheel platform provided an effective intervention and can be used to test hypotheses about the underlying neurophysiologic mechanisms of exercise-based neuroprotection. Future work should also examine the effect of different exercise parameters, such as speed and duration, on neuroprotection in PD.

Disclosures: H. Skelton: None. **D. Grogan:** None. **J. Rubiano:** None. **K. Berglund:** None. **C.N. Gutekunst:** None. **R.E. Gross:** None.

Poster

P268. Therapeutic Strategies: Preclinical Animal Models: Other Therapeutics

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Program #/Poster #: P268.05

Topic: C.03. Parkinson’s Disease

Support: NIH NS110398

Title: Impact of sex and advancing age on genetic silencing of striatal CaV1.3 for amelioration of levodopa-induced dyskinesias in a rat model of Parkinson's disease

Authors: M. E. CAULFIELD[1], F. P. MANFREDSSON[2], J. A. STANCATI[1], C. E. SORTWELL[1], S. A. BOEZWINKLE[3], M. J. VANDER WERP[1], I. M. SANDOVAL[2], M. DAVIDSSON[4], T. J. COLLIER[1], *K. STEECE-COLLIER[1];
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Abstract: The loss of striatal dopamine (DA) in Parkinson's disease (PD) results in dysregulation of CaV1.3 calcium channels leading to synaptic pathology that appears to underlie levodopa-induced dyskinesias (LID). We previously demonstrated* that silencing of striatal CaV1.3 with rAAV9-CaV1.3-shRNA in young (3 mo at lesioning), male parkinsonian rats can: 1) provide uniform and near complete protection against induction of LID even with sustained high dose levodopa, and 2) significantly reduce severity of already established LID. This gene therapy approach of CaV1.3 silencing provides some of the most profound antidyskinetic benefit reported to date. As a next step in understanding the utility of this novel therapeutic approach for LID in individuals with PD, acknowledging the strong link between aging and PD, we examined the ability of striatal CaV1.3 silencing to impact LID in middle-aged (15 mo at lesioning) male and female parkinsonian rats. To examine 'LID reversibility' in middle-age rats, subjects were unilaterally lesioned with 6OHDA and lesion status confirmed with amphetamine rotational asymmetry 2 wks later. Rats were then primed with low dose levodopa (3mg/kg) for 3 wks resulting in low level LID at which time they received intrastriatal CaV1.3-shRNA or scrambled (Scr) vector. LID remained low for 30 days post-vector in both groups with the 3mg/kg dose, which was escalated to 6 mg/kg and then to12 mg/kg; 2 wks/dose. At the higher doses, there was significant amelioration LID severity in aged male, but not female rats receiving rAAV-CaV1.3 compared to Scr-shRNA. Despite evidence that aged female rats may be more resistant to reversal of LID, a subpopulation (~40%) of subjects showed therapeutic efficacy comparable to aged male rats. To examine 'LID prevention' in middle-age rats, subjects received CaV1.3-shRNA or Scr-shRNA, one week later rats were unilaterally lesioned with 6OHDA and 3 weeks later began receiving escalating doses of levodopa (6, 9, 12, 18 mg/kg, 2 wks/dose; per *PMID:31002755). Unlike the 'LID reversibility' study, data from the 'LID prevention' study suggests that viral vectored silencing of striatal CaV1.3 channels maintains potency in preventing development of LID in both middle-aged male and female parkinsonian rats. However, in contrast to young subjects where all parkinsonian rats receiving CaV1.3-shRNA showed near complete amelioration of LID, middle-aged rats showed more variability, especially in female subjects. The source of the apparent sex difference in functional efficacy of CaV1.3 silencing to reverse LID and the therapeutic variability in preventing LID induction are currently under investigation.

Disclosures: M.E. Caulfield: None. **F.P. Manfredsson:** None. **J.A. Stancati:** None. **C.E. Sortwell:** None. **S.A. Boezwinkle:** None. **M.J. Vander Werp:** None. **I.M. Sandoval:** None. **M. Davidsson:** None. **T.J. Collier:** None. **K. Steece-Collier:** None.

Poster

P268. Therapeutic Strategies: Preclinical Animal Models: Other Therapeutics

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Program #/Poster #: P268.06

Topic: C.03. Parkinson's Disease

Support: Fundação de Amparo a Pesquisa do Estado de São Paulo - 2019/27643-5
Hospital Sírio-Libanês

Title: Subthalamic stimulation-induced analgesia and serotonergic modulationin a rat Parkinson's disease model

Authors: *G. MAURICIO[1], K. V. LIMA[2], A. CAMPOS[2], E. T. FONOFF[3], C. HAMANI[4], R. L. PAGANO[2];
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Abstract: Parkinson's disease (PD) is a progressive disease that affects dopaminergic and non-dopaminergic pathways which contributes to the development and maintenance of motor and non-motor symptoms in PD. Deep brain stimulation (DBS) of the subthalamic nucleus (STN) has been used to treat symptoms in advanced PD patients; however, the mechanisms involved in the analgesic response are yet to be determined. Considering the importance of serotonergic circuitry in the control of the nociceptive response, in this study we investigate the effect of STN-DBS treatment on the nociceptive response and the serotonergic system of hemiparkinsonian rats. Male Wistar rats were evaluated at the basal nociceptive threshold. After that, animals were injected with 6-hydroxydopamine (6-OHDA) neurotoxin or saline in the left striatum. In the same surgical procedure, the animals were implanted or not with stainless steel electrodes in the left STN. Seven days after the surgery, animals were evaluated in the apomorphine-induced rotation and re-evaluated in the paw pressure test. On the 8th day, the animals were divided into four groups: saline (control), 6-OHDA (not implanted), 6-OHDA + DBS OFF (implanted but not stimulated) and 6-OHDA + DBS ON. Six-OHDA + DBS ON animals were stimulated for five consecutive days (8th to 12th day after the surgical procedure) during 2 h (130 Hz, 60μs, 0,1 mA). In the next day, animals were re-evaluated in the nociceptive test, and euthanized by transcardial perfusion. Brains were collected and immunolabeled for tyrosine hydroxylase (TH) in the substantia nigra (SN), and for 5-hydroxytryptophan (5-HT) in the magnus raphe nucleus (MRN) and dorsal raphe nucleus (DRN). Six-OHDA animals presented increased contralateral rotation and decreased TH immunoreactivity (IR) in the SN, confirming the nigrostriatal lesion. The PD model decreased the nociceptive threshold in both paws of the lesioned animals compared to control animals and the STN electrode implantation per se did not interfere with this response. However, the STN-DBS treatment was able to bilaterally reverse the mechanical hyperalgesia induced by neurotoxin. Six-OHDA animals showed a decrease of 5-TH-IR in the MRN and DRN when compared with controls. The 6-OHDA + DBS ON animals showed a reversal of the 5-HT deficit in both raphe nuclei when compared to the saline and 6-OHDA

groups. Our data lead us to suggest that STN-DBS treatment inhibits nigrostriatal injury-induced hyperalgesia due, at least in part, to restoration of serotonergic circuitry in the raphe nuclei, which allowed the reorganization of the descending analgesic pathway and thus the inhibition of persistent pain transmission.

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Poster

P268. Therapeutic Strategies: Preclinical Animal Models: Other Therapeutics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P268.07

Topic: C.03. Parkinson's Disease

Title: Closed-loop spinal cord stimulation creates a pro-locomotion brain state in rats with Parkinson's disease

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Abstract: Enhanced oscillatory activity is a dynamic neuropathological hallmark of Parkinson's disease (PD), and excessive synchronization in the beta frequency band (10-25 Hz) has been linked to motor impairments seen in the disease. Previously, we've shown that epidural dorsal column stimulation (DCS)-an emerging semi-invasive technique improves locomotion in animal models of PD. Here, we incorporated a closed-loop DCS (CLDCS) paradigm and investigated the mechanisms by which CLDCS facilitates locomotion in PD animal models. Through an analysis of the spiking behavior of neurons in the motor cortex and striatum, we hypothesize that CLDCS promotes motor functioning by disrupting beta activity in the brain and restoring neural functioning to a more pro-locomotion state. We implanted six Parkinsonian rats with epidural electrodes in thoracic spinal cord and microelectrode arrays in the primary motor cortex and dorsolateral striatum. CLDCS was delivered in response to periods of elevated beta activity in the striatum. Spiking activity of single units was recorded during off-stimulation and CLDCS periods and analyzed offline. Spectral power for each neuron was computed during periods of on- and off-stimulation and averaged for each recording session. Across all rats our results indicated a significant reduction in beta activity during periods of CLDCS compared to periods of no stimulation. To understand how CLDCS facilitates locomotion we identified bouts of locomotion and studied the firing rate of each unit before and during the locomotion bout. Principal component analysis was performed to reduce dimensionality and to represent the population activity in a three-dimensional neural state-space that evolved over time. We were able to capture a representative view of ensemble activity as the behavior evolved from a state of

pre-locomotion akinesia to pro-locomotion movement. The neural trajectory represented in the 3D state-space during CLDCS was compared to that during off-stimulation and a control pre-disease condition. We calculated the mean distance of all points to the centroid as well as the three-dimensional path distance of the neural trajectory across conditions. We found that the spread and path distance of neural trajectory during CLDCS was more identical to the pre-disease condition as compared to the off-stimulation condition. This suggested that CLDCS facilitated the transition from a pre-locomotion to locomotion state. Overall, our results suggest that CLDCS mitigates motor impairments in PD animal models by reducing beta activity within individual units and restoring the neural state closer to a more healthy, pro-locomotion condition.

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Poster

P268. Therapeutic Strategies: Preclinical Animal Models: Other Therapeutics

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Topic: C.03. Parkinson's Disease

Support: Innovation and Technology Commission of the Hong Kong Special Administrative Region Government (ITS/168/19FP)

Title: A novel target of deep brain stimulation restored Parkinson's disease symptoms in mice

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Abstract: Parkinson's disease (PD) is common chronic neurodegenerative disorder with unknown etiology affecting more than 1% of the world population. Levodopa treatment is frequently used to control PD symptoms. However, it is effective only in the early stages of the disease and develops resistance in majority of patient at later stages as disease progresses. Deep brain stimulation (DBS) has become vital therapeutic option for the resistance PD. Stimulation of subthalamic nucleus (STN), globus pallidus pars interna (GPi) or ventral intermediate nucleus (VIM) of the thalamus has been used for two decades for PD treatment. It modulates either direct or indirect pathway of thalamo-cortical-striatal loop. The imbalance between direct and indirect pathways results in abnormal activation of output nuclei and over-inhibition of thalamus and motor cortex, leading to undesirable side effects such as cognition, depression and anxiety. The relatively small STN and GPi stimulation site is potential limiting factor in surgical implantation of DBS electrode that high surgical precision is required. These limitations could be overcome by validating a new target site proposed in our current study, which offers (a) larger size for easy electrode implantation and increases the area of stimulation; (b) stimulates both direct and

indirect pathways of locomotion to mimics the normal physiology state in healthy people that send normal output signals to motor cortex; (c) prevents direct stimulation of STN or GPi to reduce adverse side effects. Our novel target is located in strategic positions and acts as integrating centre for motor control by activation of both direct and indirect pathways. We explored the DBS efficacy of our proposed novel target in rotenone and 6-OHDA induced PD in mice. Mice received DBS stimulation at 130Hz frequency, 100μA pulse amplitude, 80μs pulse width for 1 hour on a daily basis for 7 days. Strikingly, novel target-DBS showed significant increase in recovery of general locomotion test, beam walking test, pole climbing test in rotenone model and decreased apomorphine induced rotation in 6-OHDA model as compared to GPi-DBS. We further examine the electrophysiological properties on day after the last stimulation. Consistent with the animal behavior test results, novel target-DBS significantly reduced abnormal neuronal firing and restored neuronal bursts and interspike interval to nearly baseline levels in PD mice as compared to GPi-DBS. In conclusion, our novel target-DBS showed better efficacy than GPi-DBS in improvement of PD symptoms by restoring abnormal firing pattern in PD mice.

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Poster

P268. Therapeutic Strategies: Preclinical Animal Models: Other Therapeutics

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Program #/Poster #: P268.09

Topic: C.03. Parkinson's Disease

Title: Burst DBS induces persistent motor rescue in parkinsonian mice

Authors: *S. NANIVADEKAR[1], T. A. SPIX[1], A. H. GITTIS[2];
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Abstract: Deep brain stimulation (DBS) is a highly effective, but invasive treatment for patients with medication-resistant Parkinson's disease (PD). The neural mechanisms underlying its effectiveness remain unknown. This has limited the ability to optimize the stimulation parameters to allow resolution of the underlying dysfunction. Issues related to continuous stimulation protocols like battery replacement, appropriate onset of therapy, and adverse clinical side effects have motivated a study of patterns of stimulation that can prolong the therapeutic duration beyond the time of active stimulation, an outcome currently unexplored in clinical practice. Our lab has demonstrated that optogenetic manipulation that differentially modulates cell types - namely excites the PV neurons while inhibiting the Lhx6 neurons in the GPe can generate long-lasting motor recovery in a parkinsonian mouse model. However, optogenetic stimulation is currently not feasible in humans. Our lab also identified in an in-vitro model that synaptic differences can be leveraged to differentially drive these cell types by using bursts of

electrical stimulation. In this study we test whether an electrical DBS protocol, tailored to drive population-specific neuromodulation could produce such persistent recovery in parkinsonian mice after stimulation has stopped. Using an in-vivo mouse model of electrical DBS in mice rendered parkinsonian by bilateral injections of the toxin 6-OHDA into the medial forebrain bundle, we compared the therapeutic efficacy of conventional continuous delivery of electrical stimulation vs. delivery of 200 ms/second bursts of electrical stimulation at a 175Hz frequency. Our DBS platform was modeled after Schor et al. and used six-lead DBS electrodes implanted in the entopeduncular nucleus, an area where fibers from the STN and D1-SPNs are coactivated. Both conventional and burst DBS were comparable in reducing immobility during stimulation measured as the percent time the animal did not move in the open-field. After stimulation, mice who received conventional DBS quickly returned to the immobile state (pre vs. 90' post-stim = 0.195), whereas those that received burst DBS continued to move around the arena for up to 2.5 hours after stimulation had stopped (pre vs. 90' post-stim =0.016*). On average, the therapeutic effects of DBS persisted about 4.5-fold longer than those of conventional DBS (p=0.008, n = 8 mice). These results demonstrate the ability to prolong the therapeutic benefits of DBS beyond conventional methods, by translating principles of basal ganglia cell-type specific neuromodulation to an in-vivo parkinsonian mouse model.

Disclosures: S. Nanivadekar: None. **T.A. Spix:** None. **A.H. Gittis:** None.

Poster

P268. Therapeutic Strategies: Preclinical Animal Models: Other Therapeutics

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Program #/Poster #: P268.10

Topic: C.03. Parkinson's Disease

Support: Spanish Ministry of Science and Innovation, Ref PID2020-120308RB-I00

Title: Biodistribution and comparative performance of AAV-TT vs. AAV9 in the basal ganglia of non-human primates

Authors: *J. L. LANCIEGO[1], E. RODA[1], E. HENCKAERTS[2], A. J. RICO[1];
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Abstract: The adeno-associated viral vector variant known as true type (AAV-TT) is a novel capsid engineered to improve CNS tropism and retrograde transport through brain circuits. This retrogradely-spreading AAV-TT capsid was designed for enabling neuronal transduction in multiple brain regions innervating the injected area (e.g. multiple targeting to be achieved with one single AAV-TT intraparenchymal delivery). Here we have performed a single injection of a GFP-coding AAV-TT (5 uL; 1 x 10E13 vg/mL) into the left post-commissural putamen of two

adult juvenile *Macaca fascicularis*, together with a similar strategy conducted with AAV9-GFP in two more animals for comparison purposes. All animals were sacrificed 4 weeks post-AAV deliveries. GFP+ neurons were consistently found throughout the layer V of the cerebral cortex (ipsi- and contralateral to the injected side), the caudal intralaminar nuclei and the substantia nigra pars compacta. Within the cerebral cortex, cortical areas showing the highest neuronal transduction were the superior frontal gyrus, the precentral gyrus, the cingulate cortex and the postcentral gyrus. Although both AAV-TT-GFP and AAV9-GFP showed a roughly similar pattern of retrograde transduction, AAV-TT capsid, in the present study, exhibited a slight enhanced transduction when compared to AAV9, as shown by its ability to transport and transduce the dorsal raphe nucleus. Moreover, uptake of viral vectors through fibers of passage was only observed for AAV9 and not for AAV-TT capsid (e.g. the latter lacking false positive results). These data represent a powerful ability of AAV-TT for transducing projection neurons in multiple brain areas located far away from the injection site, thus opening unprecedented possibilities when using retrogradely-spreading AAV-TT capsids for both disease modeling as well as for therapeutic approaches in the field of neurodegenerative disorders

Disclosures: J.L. Lanciego: E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Handl Therapeutics B.V. F. Consulting Fees (e.g., advisory boards); Handl Therapeutics B.V.. **E. Roda:** None. **E. Henckaerts:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Handl Therapeutics B.V. F. Consulting Fees (e.g., advisory boards); Handl Therapeutics B.V.. **A.J. Rico:** None.

Poster

P268. Therapeutic Strategies: Preclinical Animal Models: Other Therapeutics

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Program #/Poster #: P268.11

Topic: C.03. Parkinson's Disease

Support: Lundbeckfonden
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Title: Establishing a pre-clinical human glial chimeric rodent model for studies of direct neuronal reprogramming in vivo

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Abstract: Parkinson's Disease (PD) is a neurodegenerative disorder, which is associated with focal loss of dopaminergic neurons in the substantia nigra. The therapeutic prospective of cell replacement therapy for treating PD has been established through clinical trials transplanting human fetal tissue and stem cell therapies are under development. Direct reprogramming of resident glia cells *in situ* into therapeutic neurons represents a strategy based on cell replacement but circumvents the need for cell transplantation. Proof-of-concept experiments has been performed in rodent models using genetic overexpression of lineage-specifying transcription factors or downregulation of key barriers such as *Ptbp1*, but it remains an open question if human glia can be converted *in situ* into clinically relevant neurons with the ability to restore function in the damaged brain. For studies of direct neural reprogramming of human glia, we have established a stem cell-based *in vitro* conversion model (Nolbrant et al., 2020) as well as a pre-clinical humanized glial chimeric rodent model for *in vivo* conversion. In this model, human glia progenitor cells (GPCs) derived from induced pluripotent stem cells are transplanted into neonatal immunodeficient rats. The GPCs engraft and proliferate, and upon maturation the rodent brain contains large numbers of human GPCs and astrocytes. We have also engineered human GPCs to express CRE recombinase, which allows us to specifically target the human cells *in vivo* upon stereotactic injection of CRE-dependent neural conversion factors. Using these two models, we are currently exploring both transcription factor-mediated conversion and *PTBP1* knockdown-mediated conversion *in vitro* and *in vivo*, with the goal to select neural conversion factors that specify the reprogramming of dopaminergic neurons from human glia directly in the brain.

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Poster

P278. Ataxia and Dystonia: Human Studies

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Topic: C.04. Movement Disorders other than Parkinson's Disease

Support: Cerebral Palsy Alliance Research Foundation Inc. Grant PG02518

Title: Deep brain stimulation reveals high-precision temporal coding in basal ganglia and thalamus in pediatric patients with movement disorders

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Abstract: Deep brain stimulation (DBS) is a common treatment of movement disorders. While the mechanisms of DBS remain largely unexplained, recordings from implanted electrodes provide valuable insight into the brain's response to the electrical stimulations. We here present novel findings of complex and high-frequency evoked potentials (EPs) in basal ganglia and thalamus from DBS in 7 pediatric patients with movement disorders.
Externalized DBS leads in thalamic nuclei and basal ganglia were used to concurrently stimulate and record. Stimulations were performed through one bipolar pair of conventional contacts at a time, while recordings were acquired from all high-impedance contacts with a sampling frequency of 24 kHz, allowing us to detect high-frequency neural responses. Complex EPs were recorded in every subject, in regions including ventral oralis (VO), ventral intermediate (VIM), globus pallidus interna (GPi), and the subthalamic nucleus (STN). These responses consisted of multiple high-frequency oscillations up to and above 6 kHz, standing in stark contrast to the more commonly observed ~1-kHz single-oscillation DBS EP shape.
To ensure that these complex responses were not merely artifacts of the electrical stimulation, multiple analytical tests were run. First, phase reversal of each stimulation contact pair was performed, which successfully reversed the stimulus artifact, while leaving the EP intact (as expected for a neural response). Second, the response shape could be seen consistently in individual recordings, ruling out the possibility that the stimulation averaged EP shape was merely generated by noisy samples. Finally, stimulations in different contacts and at different frequencies generated similar but slightly varying responses, conforming to the inherent neural variability expected, while differing from the anticipated replicability of artificial electrical stimulus artifacts.
While further analysis is needed to determine the origin of these complex responses, it is possible that local resonant circuits or multiple axons firing with different delays are key mechanisms of this high-precision temporal coding. These recordings also emphasize that high-impedance contacts are vital to detect the complex, high-frequency neural activity that conventional low-impedance contacts would inherently filter out. Such precision may prove to be vital for successful applications of e.g. closed-loop DBS and brain-machine interfaces.
The observed high-frequency, high-precision EPs highlight the complexity of the brain's response to DBS, and will help shed light on the mechanisms of DBS as a treatment for movement disorders.

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Poster

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Topic: E.03. Basal Ganglia

Support: Cerebral Palsy Alliance Research Foundation Inc. (PG02518))

Title: Dynamic causal modelling predicts the cortico-basal ganglia-thalamo-cortical loop neurotransmission in children with secondary dystonia undergoing deep brain stimulation

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Abstract: Dystonia is a movement disorder in which excessive or abnormal deep brain activity causes involuntary muscle contractions and abnormal postures, however its neurophysiology remains unknown. An effective method to reduce its symptoms in children is deep brain stimulation (DBS). DBS target may differ in different patients, including ventralis oralis anterior/posterior (VoaVop) in thalamus, internal globus pallidus (GPi) and subthalamic nucleus (STN) in basal ganglia. Knowledge about the brain network and dynamics helps us better understand the mechanism of dystonia and DBS. Therefore, we aim to model the cortico-basal ganglia-thalamo-cortical loop in children with dystonia to explore the coupling and connection strength between deep brain structures, using dynamic causal modeling (DCM) from Statistical Parametric Mapping (SPM12). We recorded GPi, VoaVop, and STN oscillations through externalized DBS electrode implants in five dystonic children, executing a voluntary reaching with their upper limbs. The data was preprocessed and divided into “active” and “rest” segments, and their cross spectral densities were used to fit the models. Seven neuronal architectures were tested on five subjects’ data where the movement acts as an external effect on the connections between the brain deep structures. We assumed a Mean Field Neural Model (MFM) for each structure. Bayesian model inference provided strong evidence in favor of a model corresponding to the ‘standard architecture’; GPi receives excitatory input from STN, and its connection gain strengthens during movement ($p < .001$). Moreover, GPi inhibits VoaVop and this connection gain also increases during movement ($p < .001$), however the later gain is significantly smaller ($p < .001$) than the former, which shows the dominant effect of STN-GPi connection over GPi-Vo connection. Besides, VoaVop and STN excite each other bidirectionally. We suggest that it could be through cortex. Our results confirm current hypotheses of cortico-basal ganglia-thalamus connectivity and signal flow, and they show that DCM with MFM can potentially be used with intracranial recordings to test different hypotheses about brain networks. Even when the intracranial neural models are not specified in detail, the current models suffice to predict the direction and type of connections, and how they differ during movement. Furthermore, the effect of DBS can be better understood by implementing a detailed neural model into DCM models and making more accurate predictions about how connectivity parameters change with stimulation.

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Poster

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Title: Impact of genetic variations on the age at onset in Spinocerebellar Ataxia Type 3 / Machado-Joseph disease

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Abstract: Spinocerebellar ataxia type 3 (SCA3) or Machado-Joseph disease (MJD) is an autosomal-dominantly inherited, neurodegenerative disorder caused by the expansion of a CAG repeat within the *ATXN3* gene resulting in an expanded polyglutamine repeat in the encoded ataxin-3 protein. SCA3/MJD, therefore, belongs to the group of polyglutamine diseases comprised of nine neurodegenerative diseases including Huntington's disease. Statistically, a correlation between the number of CAG repeats and the age at onset of SCA3 patients exists and patients with more CAG repeats have an earlier onset of symptoms. However, this statistical correlation is not perfect and the number of CAG repeats contributes only about 55% to the age at onset. Therefore, the remaining 45% are influenced by other factors, which we aim to identify in this study. In order to identify modifiers of the disease progression, we genotyped in a combined European and South American approach more than 500 SCA3/MJD patients for promising polymorphisms in candidate genes. Candidate genes included ataxin-3 itself and known interaction partners of ataxin-3, functional modifiers identified in previous studies as well as genes with known relevance for the pathophysiology of SCA3/MJD including genes involved in the nucleocytoplasmic transport. We selected polymorphisms with a high likelihood of having a functional relevance i.e. polymorphisms in the promoter regions as well as polymorphisms leading to amino acid changes. While controlling for ethnic origin we assessed the contribution of the respective polymorphism to the age at onset in addition to the already known modifying factor, the length of the expanded CAG repeat within *ATXN3*. We indeed identified interesting

polymorphisms contributing to the age at onset including certain haplotypes within *ATXN3* itself. Subsequent functional characterizations will reveal the impact of these polymorphisms on pathogenic mechanisms in SCA3/MJD. We hope that our results will improve the prediction of clinical symptoms and contribute to the understanding of pathogenic processes in SCA3/MJD.

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Poster

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Topic: C.04. Movement Disorders other than Parkinson's Disease

Support: University of Iowa, Department of Psychiatry

Title: Aberrant structural and functional brain differences in spinocerebellar ataxia subtypes 3 and 6

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Abstract: Aberrant structural and functional brain differences in spinocerebellar ataxia subtypes 3 and 6
Spinocerebellar ataxias (SCA) are a heterogeneous group of over 40 autosomal dominant, neurogenerative disorders often caused by CAG trinucleotide repeats. Patients with SCA subtype 3 (SCA3) often present with ataxia, pyramidal, and extrapyramidal symptoms, while patients diagnosed with SCA subtype 6 (SCA6) frequently report ataxia, tremors, and dysarthria. Previous literature suggests cerebellar atrophy in SCA3 and SCA6 brains, though SCA3 atrophy continues through the brainstem and portions of the cortex. SCA3 and SCA6 functional connectivity (FC) studies are rare, though one study found within-network FC differences for SCA6. The current project examines the relationship between structural brain abnormalities and functional connectivity among SCA3 and SCA6 patients.
Participants completed MRI scans at University of Iowa Psychiatry Department. Healthy controls (CTRL; 18 total, 11 males, mean age: 53.07) were age- and sex-matched with SCA3 (9 total, 3 males, mean age: 46.40) and SCA6 patients (5 total, 4 males, mean age: 61.05). Structural T1w and resting-state fMRI (rs-fMRI) scans were preprocessed to obtain jacobian determinants (JD) images and Schaefer 7-network parcellation. JDs, which measure the difference in shape and volume of the cortex, cerebellum, and brainstem, were compared across participant groups. Regions of significantly different clusters were individually correlated to

Schaefer networks to assess how structurally abnormal regions functionally connect to major networks.
Initial linear mixed-effects (LME) models revealed that multiple JD clusters across the cortex, brainstem, and cerebellum differed significantly between groups. Further, LME models controlling for age, sex, and hemisphere successfully predicted FC between these JD clusters and Schaefer networks. Specifically, the control, default mode, dorsal attention, and limbic networks were abnormally connected to JD clusters across both SCA subtypes compared to CTRLs.
This study demonstrates that structural differences throughout the brain are related to abnormal functional connectivity among SCA patient groups. While previous literature has suggested limited atrophy for SCA6, this study demonstrates both SCA subtypes have varied atrophy across the cortex, brainstem, and cerebellum, as well as aberrant functional connectivity among multiple networks when comparing SCA patients and CTRLs. Because SCA subtypes affect patient brains differently, it will be important to consider these subtypes separately for potential behavioral and clinical phenotypes.

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Poster

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Topic: C.04. Movement Disorders other than Parkinson's Disease

Support: NIH NS094946
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Title: Open loop training in Spinocerebellar Ataxia 6 (SCA6)

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Abstract: Approximately 150,000 people are affected by hereditary ataxia in the US. Common features of ataxias are cerebellar degeneration and significant loss of function and mobility. Ataxia is notoriously difficult to treat, and rehabilitation, the mainstay of care, has had limited success. Individuals with cerebellar ataxia are impaired in open-loop control; yet there have been no efforts specifically targeting its rehabilitation. We have devised a training based on open-loop control and hypothesized that this form of training will have generalized effects and benefit functional movements. Here we examined the effects of this novel training intervention on endpoint control, ataxic gait severity, postural balance and BOLD activity in the cerebellum and motor cortex in individuals with Spinocerebellar Ataxia 6 (SCA6), an autosomal dominant

mostly pure cerebellar ataxia. Participants with SCA6 (n = 14) were randomly assigned into a SCA-training group and a SCA-control group. Both groups visited the lab twice 6 weeks apart to perform baseline (pre) and return (post) measures. Only the SCA-training group performed in-home error-reducing training with ballistic goal directed leg movements (4 times a week for 1 hr.). We found that the SCA-training group exhibited greater pre-post improvements in endpoint control of ankle ballistic goal-directed contractions relative to the SCA-control group (35% vs. 15%; $p = 0.001$). We found that ataxic gait (Scale for the Assessment and Rating of Ataxia; SARA gait) improved for the SCA-training ($p = 0.03$) but not for the SCA-control group. Similarly, we found significant training-specific improvements in biomechanical variables associated with balance and gait; including significant reduction in sway path-length during a postural balance test ($p = 0.03$) and significant increase in stride length, during 14 m overground walking ($p = 0.03$). Overall, these improvements suggest that our open-loop error-reducing intervention has the potential to successfully mitigate functional deficits in SCA6. Also, our results provide preliminary evidence of transfer (generalization) of open loop goal-directed training to balance and locomotion. This is further supported by our preliminary finding that activation (change in BOLD fMRI) of the cerebellum and the motor cortex increased for the training group only ($p < 0.001$).

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Poster

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Title: Spinocerebellar ataxia type 10: Memory and executive impairments and their relation with cortical thickness

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Abstract: Spinocerebellar Ataxia Type 10 (SCA10) is a neurodegenerative disorder caused by the expansion of an ATTCT pentanucleotide repeat. Its clinical features include ataxia and, in

some cases, epileptic seizures. To date, little is known about SCA10 cognitive impairments and there is no information yet about the neural bases underpinning them. The aim of this study was to explore SCA10 patients' performance in two cognitive domains typically affected in SCAs: memory and executive functions. Our second objective was to correlate the cognitive impairments of SCA10 patients with their clinical features and cortical thickness, as quantified by Magnetic Resonance Imaging (MRI). To this end, we administered memory and executive functions tests to 17 genetically confirmed SCA10 patients and 17 healthy matched volunteers. SCA10 patients showed deficits in the durability of visual memory traces, storage volume of verbal memory, error detection (monitoring), planning and cognitive flexibility. Patients with seizures (n=9) and without seizures (n=8) did not show significant differences in cognitive performance. Cognitive deficits in SCA10 patients were independent of the cerebellar motor impairments and the age at onset of disease. Based on MRI performed in 16 patients, we found associations between short-term visual memory impairments and cortical thickness in the bilateral posterior lobe of the cerebellum (bilateral lobule VI, IX and right X). Cognitive flexibility deficiencies correlated with cortical thickness in left dorsolateral prefrontal cortex, cingulate gyrus and middle temporal regions. Motor impairments were correlated with cortical thickness in several bilateral cerebellar regions. In conclusion, this study offers novel information about the SCA10 cognitive profile, showing significant memory and executive deficits that correlated with atrophy in specific cerebral/cerebellar regions.

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Poster

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Title: Single-nucleus transcriptomic analyses reveal microglial activation underlying cerebellar degeneration in Ataxia Telangiectasia

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Abstract: While *ATM* loss-of-function has long been identified as the genetic cause of Ataxia Telangiectasia (AT), how this genetic mutation leads to selective and progressive cerebellar degeneration of Purkinje and granule cells remains poorly understood. We performed single-nucleus RNA-sequencing of the human cerebellum and prefrontal cortex from individuals with AT and matched unaffected controls to identify AT-associated transcriptomic changes in a cell-type- and brain-region-specific manner. We provide the largest single-nucleus transcriptomic atlas of the adult human cerebellum in health and degeneration to-date (126,356 nuclei), annotate major cell types in the human cerebellum and prefrontal cortex, and identify cell type proportion changes between AT and controls. We demonstrate cell-type-specific expression of cerebellar ataxia-associated disease genes, with remarkably enriched expression of *ATM* in microglia from human cerebellum and prefrontal cortex, suggesting a particularly important role of *ATM* in microglia. We analyze the cell-type-specific molecular pathways perturbed in AT, revealing prominent and widespread inflammation of microglia in AT cerebellum with transcriptional signatures similar to aged and neurodegenerative microglia. Our analyses suggest that microglia activation precedes Purkinje and granule neuron death in disease progression, consistent with a cell-intrinsic mechanism of activation. The CGAS-STING pathway was upregulated specifically in AT microglia, implicating cytosolic DNA as a potential cell-intrinsic trigger of microglial activation. In addition, apoptotic and ER stress pathways were upregulated in AT Purkinje and granule neurons, while genes involved in calcium ion homeostasis were downregulated in AT Purkinje neurons, including calcium genes implicated in other hereditary ataxias. Overall, our data implicates a novel role of microglial activation underlying cerebellar degeneration and a convergence of disease pathophysiology underlying hereditary ataxias.

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Poster

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Topic: C.04. Movement Disorders other than Parkinson's Disease

Title: Hematological Parameters in Friedreich's Ataxia Patients: Evidence for Subtle Features of Iron Deficiency

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Abstract: Introduction: Friedreich's Ataxia (FRDA) is a neurodegenerative disorder affecting 1 in every 50-100000 individuals in the United States. The most common mutation in FRDA is an expansion of guanine-adenine-adenine (GAA) trinucleotide repeats within the gene encoding mitochondrial protein frataxin, leading to its genetic deficiency. Mitochondrial dysfunction resulting from frataxin deficiency is a component of the underlying pathophysiology of FRDA. Recently, a new isoform of frataxin was discovered in erythrocytes; its role and impact on extramitochondrial processes such as hematologic iron stores and hemoglobin production remain unknown at a clinical level. This study aimed to elucidate the features of erythrocytes and other hematological parameters in FRDA patients. **Methodology**: Specific clinical parameters associated with iron disposition and hemoglobin synthesis were examined in a cohort of 114 FRDA patients in order to ascertain the effect of deficiency of erythrocyte frataxin on such parameters. The effect of FRDA on ferritin, a systemic intracellular iron reservoir proposed to serve as a biomarker in FRDA, was also analyzed in an overlapping cohort of 221 FRDA patients. Linear regression and unpaired, two-tailed t-tests were performed with a significant p-value < 0.05. **Results**: This study demonstrated that in spite of the low levels of frataxin in erythrocytes, its absence leads to no major abnormalities in clinical parameters of hemoglobin synthesis or red blood cell parameters. Hemoglobin levels were normal in FRDA patients, however, a small decrease in mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and ferritin suggested that patients with FRDA manifest subtle features of iron deficiency. When hematological data were ranked by tertiles according to disease biomarkers, MCV values decreased significantly as a function of guanine-adenine-adenine (GAA) repeats but increased as a function of age of onset, while MCH values significantly increased as a function of age of onset. This study also demonstrated that ferritin levels are sex-dependent, correlate with hemoglobin levels, and are predicted by age. **Conclusion**: Collectively, the present results demonstrate a subtle level of hematological abnormalities in FRDA and suggest that ferritin may serve as a biomarker in FRDA. As MCV and MCH were more associated with age of onset and genetic severity than disease duration, such changes suggest that these differences are part of the FRDA phenotype. Further studies are required to understand whether other compensatory iron homeostasis mechanisms might increase in response to reduced frataxin levels in erythrocytes.

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Poster

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Title: Discrimination of idiopathic REM sleep behavior disorder patients by single-trial event-related potential during visuospatial attention based on deep neural networks

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Abstract: Introduction: Idiopathic REM sleep behavior disorder (iRBD) is a sleep disorder characterized by loss of muscle atonia during REM (rapid eye movement) sleep. An early detection of neurodegeneration in iRBD is essential to cure or slow down the progression of the disease since the majority of patients develop neurodegenerative diseases. We try to develop an efficient algorithm for the discrimination of iRBD patients by single-trial event-related potential (ERP) during visuospatial attention task based on deep neural networks.**Methods**: Nineteen healthy subjects (63.47 ± 7.37 years) and 16 drug-naive iRBD patients (64.94 ± 6.92 years) participated in this study. 60-channel electroencephalograms (EEGs) were recorded while performing Posner cueing task. Cortical source activities were reconstructed using weighted minimum norm estimation. The single-trial ERPs were averaged within every 100 ms time-interval from target onset to 800 ms for all subjects. The cortical activities were projected onto 2D image using Mollweide projection. Finally, the 120x120x12 three-dimensional volumes were constructed and used as input for the pattern classifiers based on a number of deep neural network architectures. The structure of the proposed classification algorithms is illustrated in Fig 1.**Results**: Four pattern classification algorithms were developed and successfully discriminated iRBD patients from normal controls (deep multilayer perceptron (MLP), long short-term memory (LSTM), two- and three-dimensional convolutional neural networks (2DCNN and 3DCNN)). The accuracies achieved by each algorithms were as follows: MLP: 92.19 ± 0.79 %, LSTM: 90.96 ± 0.69 %, 2DCNN: 97.66 ± 0.48 %, 3DCNN: 97.2 ± 2.15 %.**Discussion**: The iRBD patients could be successfully discriminated based on single-trial cortical activities with high accuracy based on deep neural network classifiers. The CNN-based architectures showed best test performance by 97 %. Our results suggest that the spatial characteristics of cortical activities are crucial to discriminate iRBD patients.

MLP		LSTM		2DCNN		3DCNN	
Layer (Filter/pool size)	Output dimension	Layer (Filter/pool size)	Output dimension	Layer (Filter/pool size)	Output dimension	Layer (Filter/pool size)	Output dimension
Dense	500	Flatten	12 x 14,400	2D convolution (3 x 3)	118 x 118 x 6	3D convolution (3 x 3 x 3)	118x118x10x6
Dense	100	biLSTM	200	Max pool (2 x 2)	59 x 59 x 6	Max pool (2 x 2 x 2)	59x59x5x6
Sigmoid	1	Dropout	200	2D convolution (3 x 3)	57 x 57 x 16	3D convolution (3 x 3 x 3)	57x57x3x16
		Dense	100	Max pool (2 x 2)	28 x 28 x 16	BatchNorm	57x57x3x16
		Sigmoid	1	2D convolution (3 x 3)	26 x 26 x 16	Max pool (2 x 2 x 2)	28x28x1x16
				Max pool (2 x 2)	13 x 13 x 16	Flatten	12,544
				Dropout	13 x 13 x 16	Dense	128
				Flatten	2,704	Sigmoid	1
				Dense	64		
				Sigmoid	1		

Disclosures: H. Kim: None. **P. Seo:** None. **M. Kim:** None. **J. Huh:** None. **K. Cha:** None. **J. Sunwoo:** None. **J. Byun:** None. **K. Jung:** None. **K. Kim:** None.

Poster

P329. Therapeutic Approaches for Ischemic Stroke II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P329.01

Topic: C.08. Ischemia

Support: NIH/NINDS Grants R01NS100947
NIH/NINDS Grants R21NS109687

Title: Selective inhibition of mPGES-1 mitigates cerebral ischemia-induced neuronal injuries and functional deficits

Authors: *L. LI, R. HOU, Y. YU, J. JIANG;
Dept. of Pharmaceut. Sciences, Col. of Pharm., The Univ. of Tennessee Hlth. Sci. Ctr., Memphis, TN

Abstract: Ischemic stroke is one of the leading causes of death and disability among adults. It involves microglia-mediated neuroinflammation in its pathophysiology. Microsomal prostaglandin E synthase-1 (mPGES-1)-derived prostaglandin E2 (PGE2) is a key mediator of inflammation. We herein investigate whether targeting mPGES-1 with a selective inhibitor MPO-0063 (N-phenyl-N'-(4-benzyloxyphenoxycarbonyl)-4-chlorophenylsulfonyl hydrazide) may improve functional outcomes after experimental cerebral ischemia. We first found that the

release of PGE2 and pro-inflammatory cytokines from mouse primary microglia was considerably increased upon lipopolysaccharide (LPS) stimulation. The pretreatment with mPGES-1 inhibitor MPO-0063 and its analogs significantly attenuated these microglial responses in a dose-dependent manner (n=4). Meanwhile, 11-week old C57BL/6 male mice were subjected to a 45-min episode of transient middle cerebral artery occlusion (tMCAO) and were intraperitoneally administrated with MPO-0063 over 24 hours after stroke onset (n=13-15). At the acute stage (over 72 hours), the inhibition of mPGES-1 was associated with a better survival, milder neurological deficits, transient improvement of weight loss, and a notable reduction in infarction. The ischemia induced an extensive upregulation of inflammation-associated genes, which was largely mitigated by treatment with MPO-0063. Moreover, a batteries of behavior tests were performed throughout the sub-acute stage and long-term recovery. In open field test, the maximal instantaneous locomotor velocity was notably compromised at day 7, whereas this deficit was undetectable in MPO-0063-treated cohort. The progressive anxiety of mice was indicated by the ever-increasing presence at corners, and MPO-0063 provided a significant alleviation in this regard. In light/dark box, MPO-0063 conferred a shorter presence in the dark and an extended latency under the light, further highlighting its anxiolytic potential. Among the vehicle cohort, the failure of recognizing novel objects was still ongoing at day 30, and the spontaneous cognitive recovery was not seen until day 60. By contrast, the cognitive restoration appeared as early as day 30 and was maintained at day 60 upon MPO-0063 administration. Our findings suggest that the mPGES-1 inhibition might represent a novel strategy to mitigate neurological and neuropsychological dysfunctions caused by ischemic stroke.

Disclosures: L. Li: None. **R. Hou:** None. **Y. Yu:** None. **J. Jiang:** None.

Poster

P329. Therapeutic Approaches for Ischemic Stroke II

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Program #/Poster #: P329.02

Topic: C.08. Ischemia

Support: Usher 1F Collaborative
NIDCD/NIH: R01 DC012564

Title: Adeno-associated virus mediated expression of CIB2 restores the mechanosensory transduction & auditory function in mouse model of hearing loss

Authors: *S. REHMAN[1,2], A. P. J. GIESE[1], Z. M. AHMED[1], S. RIAZUDDIN[1];
[1]Univ. of Maryland, Baltimore, MD; [2]Ctr. of Excellence in Mol. Biol., Univ. of the Punjab, Lahore, Pakistan

Abstract: Background: Hearing loss is the most common sensorineural deficit occurring in approximately 1 in 1000 newborns. Hearing loss can be classified into acquired and congenital forms. Out of all congenital forms of hearing loss 50% cases are due to genetic mutations. Variations in *CIB2* are reported globally and it accounts 8.6% of all known genetic causes of hearing loss in Pakistani population. CIB2 localizes at the tip of stereociliary bundle and is essential for mechanosensory transduction function. *Cib2*$^{tm1a/tm1a}$ mouse models exhibit profound hearing deficits and replicate the human phenotype. Therefore, they are good model to execute preclinical evaluation of potential therapeutic strategy to restore hearing function. **Approach:** Here we investigated, Adeno-associated virus (AAV) mediated *Cib2* gene delivery to inner ear hair cells and evaluated impact on hearing restoration. To execute this study, First *Cib2*$^{tm1a/tm1a}$ cochlear explant cultures were infected with AAV vector and mechanosensory transduction function was assessed through FM1-43 uptake. Secondly, AAV Capsid harboring full length *Cib2* was injected into posterior semicircular canal at P0. Auditory function was assessed through auditory brainstem response(ABR) measurements and expression of CIB2 was examined through Immunolabeling. **Results:** In vitro infection of AAV into HEK293T cells led to significant expression of *Cib2* mRNA and protein when examined through RT-qPCR and Western blot analysis respectively. Mechanosensory transduction in *Cib2* $^{tm1a/tm1a}$ cochlear explant cultures was restored due to infection of *Cib2* containing AAV vector. A single in vivo delivery of AAV vector into inner ear of *Cib2*$^{tm1a/tm1a}$ mice resulted in restoration of the Auditory brainstem responses (ABR) within two weeks. Further, CIB2 specific immunolabelling confirmed the localized expression of CIB2 at the tips of stereocilia of inner ear hair cells. **Conclusion:** In summary, this study suggests that AAV mediated gene transfer into inner ear is a potential biological strategy to treat deficits that affect sensory hair cells and auditory function. The success of this translational study can help in targeting the larger animal models as well as a better understanding of the therapeutic approach in humans.

Disclosures: S. Rehman: None. **A.P.J. Giese:** None. **Z.M. Ahmed:** None. **S. Riazuddin:** None.

Poster

P329. Therapeutic Approaches for Ischemic Stroke II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P329.03

Topic: C.08. Ischemia

Support: RFAG042189

Title: Evaluation of miR20a-3p in alleviating stroke-induced cognitive impairment in reproductively senescent female rats.

Authors: *D. SAMPATH[1], K. G. MARKOWSKY[2], T. BRANYAN[3], C. MYERS[4], F. SOHRABJI[5];

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Abstract: Stroke is the major cause of death and disability worldwide. Our previous work has shown that epigenetic modifications in astrocytes of middle-aged rats resulted in reduced expression of microRNA (miR)20a-3p after stroke.
Additionally, mir20a-3p mimics injected 4h after stroke improved acute stroke outcomes including infarct volume and sensory motor performance. We hypothesized that this treatment will also improve chronic stroke outcomes such as cognitive impairment.
Middle-aged acyclic female (Sprague-Dawley, 12-15 months of age) were injected stereotaxically with endothelin- 1 in the left MCA region to create an ischemic stroke. Mir20a-3p mimics or scrambled oligo were administered i.v. at 4, 24 hours, and at 70 days after the induction of MCAo. Animals were evaluated for sensory-motor function pre-stroke and 2d and 5d after MCAo.
Cognitive assessments were performed using the Novel Object recognition task at 30- and 100-days post stroke and cued fear memory retrieval at 30, 60 and 100 days after stroke. Fear conditioning was performed using the tone-shock trace fear conditioning protocol prior to the induction of the stroke.
MCAo resulted in significant deficits on the right-side forelimb placement, and adhesive removal test ($p<0.003$) in comparison to sham and miR 20 treated female groups ($p<0.01$), and ($p< 0.002$) among the male group.The middle-aged female group that received MCAo+ scrambled oligos were resistant to fear extinction to conditioned fear at 100 days after MCAo ($p<0.01$), displaying a sustained freezing response even after 3 days of fear extinction.
Additionally, the retrieval of fear memory was impaired in the scrambled treated stroke rats ($p<0.05$) but was retained by the miR20 treated group of rats even at 30-, 60- and 100-days post stroke (multi dose cohorts). The declarative memory, which is tested with the novel object recognition task, was also found to be significantly affected in the scrambled treated stroke rats compared to the miR20 treated group of rats at 100 days after stroke ($p<0.05$).
Overall, this data show that mir20a-3p treatment during the acute period improved short-term outcomes related to sensory motor performance, long-term improvement of fear, and declarative memory related to cognitive impairments. This data suggest that stroke therapies that improve acute stroke outcomes will also ameliorate chronic stroke outcomes.
Supported by RFAG042189 to FS.

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Poster

P329. Therapeutic Approaches for Ischemic Stroke II

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Program #/Poster #: P329.04

Topic: C.08. Ischemia

Support: Florida Department of Health #20K09

Title: Nicotine exposure alters lipid metabolism and exacerbates ischemic brain injury in female rats

Authors: *S. H. PATEL, J. SIEGAL, A. P. RAVAL;
Neurol., Univ. of Miami, Miami, FL

Abstract: While it is known that smoking-derived nicotine (N) and oral contraceptives (OC) synergistically exacerbate global and focal ischemia, the underlying mechanisms by which this occurs remains elusive. Our previously published study has demonstrated that OC exacerbation of N toxicity occurred via altered oxidative phosphorylation, in particular the dysfunction of mitochondrial complex IV in the electron transport chain [1, 2]. Due to the essential role mitochondria play in cellular respiration, we analyzed the metabolomic fingerprint in the brains of adolescent and adult female rats exposed to N +/- OC. In our study, 6-week-old adolescent and 12-week-old adult female Sprague-Dawley rats were randomly (n = 8/group) exposed to either saline or nicotine (4.5 mg/kg) +/- OC for 16-21 days. Following this exposure, the rats were sacrificed, and brain tissue was collected and Metabolon Inc performed unbiased metabolomic analysis. This metabolomic data was complemented with enzyme activity and western blot analysis. Pathway enrichment analysis revealed significant alterations in lipid metabolism. Adolescent female rats treated with N or N +/- OC showed significant increases in carnitine conjugated fatty acid metabolites when compared to saline treated rats. However, this difference was not observed in the adult female rats. Along with these changes, an increase in a subset of free fatty acids was also observed, implying elevated β-oxidation of fatty acids in the mitochondria in order to meet energy demands. Additionally, the presence of the ketone body 3-hydroxybutyrate (BHBA) was significantly lower in OC and N+OC treated adolescent rats, suggesting a total shunting of acetyl CoA for energy production via the TCA cycle. The concentration of BHBA increases in a direct relationship with the rate of lipid oxidation as acetyl CoA accumulates. Diminished BHBA levels may also point to OC and N+OC treatment initiated mitochondrial disfunction. These altered metabolomic profiles in conjunction with altered fatty acid metabolism suggests a general disfunction in energy metabolism specifically in young female animals treated with N. Furthermore, these alterations were heightened with N+OC treatment. Exploring the molecular mechanisms that dictate mitochondrial function at different ages, as well as the exact effects N +/- OC has on general brain metabolism, will open a new avenue for therapeutic interventions in the future. References: 1.Diaz, F. and A.P. Raval, J Cereb Blood Flow Metab, 2021. **41**(4): p. 793-804.2.d'Adesky, N.D., et al., Int J Mol Sci, 2018. **19**(5).

Disclosures: S.H. Patel: None. **J. Siegal:** None. **A.P. Raval:** None.

Poster

P329. Therapeutic Approaches for Ischemic Stroke II

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Program #/Poster #: P329.05

Topic: C.08. Ischemia

Support: Florida Department of Heath#7JK01 funds
Florida Department of Heath#20K09

Title: Post-stroke low frequency whole body vibration improves cognition in middle-aged rats of both sexes

Authors: N. KERR[1], J. SANCHEZ[1], W. MORENO[1], O. E. FURONES-ALONSO[1], W. DIETRICH[1], H. M. BRAMLETT[1], ***A. P. RAVAL**[2];
[1]Neurolog. Surgery, Univ. of Miami, MIAMI, FL; [2]Neurol., Univ. of Miami, Miami, FL

Abstract: Low frequency whole body vibration (WBV; 40 Hz), a low impact form of exercise, intervention for a month following mild transient middle-cerebral artery occlusion (tMCAO) reduces infarct volume and improves motor function in reproductively senescent, middle-aged female rats [1]. Since post-stroke cognitive decline remains a significant problem, the current study aims to investigate the efficacy of WBV in ameliorating post-tMCAO cognitive deficits and to determine underlying putative mechanism(s) conferring benefits of WBV in middle-aged rats. Middle-aged rats of both sexes were randomly assigned to tMCAO (90 min) or sham surgery followed by exposure to either WBV (twice a day for 15 min each for 5 days a week over a month) or no WBV treatment groups. Following the last WBV treatment, rats were tested for hippocampus-dependent learning and memory using a water maze followed harvesting brain and blood samples for histopathological and inflammatory marker analyses, respectively. Results show that post-tMCAO WBV significantly lessens cognitive deficits in rats of both sexes. Post-tMCAO WBV significantly decreased circulating pro-inflammatory cytokines and increased serum levels of irisin, a muscle-derived hormone that may play a role in brain metabolism and inflammation regulation, which suggests putative beneficial mechanisms of WBV. References:1. Raval, A.P., et al., *Whole Body Vibration Therapy after Ischemia Reduces Brain Damage in Reproductively Senescent Female Rats.* Int J Mol Sci, 2018. **19**(9).

Disclosures: N. Kerr: None. **J. Sanchez:** None. **W. Moreno:** None. **O.E. Furones-Alonso:** None. **W. Dietrich:** None. **H.M. Bramlett:** None. **A.P. Raval:** None.

Poster

P329. Therapeutic Approaches for Ischemic Stroke II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P329.06

Topic: C.08. Ischemia

Support: Florida Department of Health #20K09

Title: Effect of endogenous estrogen fluctuations on stroke outcomes in young rats

Authors: *V. REDDY, B. MANKALIYE, S. H. PATEL, A. P. RAVAL;
Neurol., Univ. of Miami, Miami, FL

Abstract: One out of five women suffer stroke after menopause in the United States. Menopause is characterized by decline in endogenous estradiol-17 beta (E2). Estrogen is neuroprotective against ischemia; however, concern regarding the safety of E2 therapy in menopausal women has inhibited translation of this treatment to the clinic. Despite these findings, women appear to be naturally protected against ischemic neuronal damage during pre-menopausal life, suggesting some estrogen-influenced neuroprotective mechanism. Thus, a better understanding of the cellular and molecular mechanisms by which endogenous estrogen fluctuations govern the female brain is required. In a published study, we demonstrated that increasing titers of E2 during the transition from the diestrus to proestrus stage of the estrous cycle protected neurons against global cerebral ischemia in young rats (Raval et al., 2009). The goal of the current study is to investigate the effects of endogenous E2 on stroke outcome and its impact on synaptic function. We hypothesized that higher levels of prevailing estrogen would lead to an increase in synaptic vesicle release. The proposed hypothesis was tested using young cyclic female (4-6 months) rats. Animals were randomly exposed to transient middle cerebral artery occlusion (tMCAO; 90 min) or sham surgery. Twenty-four hours after tMCAO, brains were collected to quantify infarct volume. The area of infarction was visualized using the TTC (triphenyltetrazolium chloride) technique and measured using ImageJ software. In a separate cohort of rats, brain tissue was collected for western blot analysis 24h after tMCAO/sham surgeries. The brain tissue was incubated with primary antibodies against synaptic proteins including syntaxin, synapsin, synaptotagmin, synaptophysin, Munc-18, SNAP 25, Complexin-1, and PSD-95. We observed a significant ($p<0.05$) reduction in infarct volume in young female rats, which underwent tMCAO during the estrus stage (higher levels endogenous E2) as compared to diestrus (lower levels of circulating E2). Results also showed that the brain of young rats in the estrus stage had an increased expression of syntaxin, synapsin, and synaptotagmin compared to diestrus stage. Our study demonstrates that higher levels of endogenous E2 regulate synaptic vesicle release after stroke thus reducing ischemic brain damage in young female rats.Reference: Raval, A. P., et al., 2009. Pretreatment with a single estradiol-17beta bolus activates cyclic-AMP response element binding protein and protects CA1 neurons against global cerebral ischemia. Neuroscience. 160**,** 307-18.

Disclosures: V. Reddy: None. **B. Mankaliye:** None. **S.H. Patel:** None. **A.P. Raval:** None.

Poster

P329. Therapeutic Approaches for Ischemic Stroke II

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Topic: C.08. Ischemia

Support: AHA 19CDA34660032
AHA 19TPA34850047
AHA CDA 856826
NIH 1R01NS096225-01A1

Title: Serum/glucocorticoid-regulated kinase 1 in cardiac arrest-induced brain injury

Authors: C. Y.-C. WU[1], G. CLEMONS[2], C. H. ACOSTA[2], W. C. CARR[1], C. T. CITADIN[2], H. LIN[1], ***R. H. LEE**[1];
[1]Neurol., [2]Cell. Biol. and Anat., LSU Hlth. Sci. Ctr., Shreveport, LA

Abstract: Cardiopulmonary arrest (CA) is a major cause of death/disability in the U.S. with poor prognosis and survival rates. The current CA therapeutic challenges are physiologically complex because they involved hypoperfusion [decreased cerebral blood flow, (CBF)], neuroinflammation, and mitochondrial dysfunction. Therefore, identify these complex regulatory elements that ultimately control neuronal viability can lead to novel therapies against CA. We previously discovered that novel serum/glucocorticoid-regulated kinase 1 (SGK1, a serine/threonine kinase) is highly expressed in brain neurons that are susceptible to ischemia (e.g., hippocampus and cortex). SGK1 plays a critical role for numerous cellular processes, including regulating homeostasis, inflammation, and apoptosis in various organs. However, the role of SGK1 in the brain is understudied. To explore the potential role of SGK1 in CA-induced brain injury, we inhibited SGK1 following CA using pharmacological (specific SGK1 inhibitor) and cell type (neuron)-specific genetic approaches (e.g., shRNA) in our well-established rodent models of CA (asphyxia- and potassium chloride-induced cardiac arrest). Intra-vital two-photon laser scanning microscopy and laser speckle contrast imaging revealed that pre-treatment with GSK 650394 (1.2 μg/kg, intracerebroventricular injection) or SGK1-shRNA ($1x10^{11}$ viral particles, retro-orbital injection) attenuated cortical hypoperfusion after CA. Interestingly, neuroinflammation (via astrogliosis and M1 microglia polarization) and mitochondrial dysfunction (via Seahorse respirometry) were reduced, while neuronal survival was enhanced in the CA1 region of the hippocampus after pre-treatment with GSK 650394 or SGK1-shRNA. Finally, rodents' functional learning and memory after CA were evaluated using Y-maze and novel object recognition test. Rodents pre-treated with GSK 650394 or SGK1-shRNA exhibited better neurological outcomes following CA as compared to untreated animals. In conclusion, SGK is one of the major contributors to CA-induced brain injury, while pre-treatment with GSK 650394 or SGK1-shRNA to diminish SGK1 expression provides neuroprotection against CA-induced hypoperfusion, neuroinflammation, mitochondrial dysfunction, neuronal cell death, and neurological deficits. Since the FDA has approved over 46 kinase-related drugs for the treatment of various diseases, our study will be promptly translated into human clinical trials for the patients suffering from CA.

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Poster

P329. Therapeutic Approaches for Ischemic Stroke II

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Program #/Poster #: P329.08

Topic: C.09.Stroke

Support: Anderson Family Research Fund
Applebaum Foundation Inc. Grant
Department of Neurology, Miller School of Medicine, University of Miami

Title: Safety and efficacy of intra-arterial mesenchymal stem cell therapy following ischemic stroke

Authors: *R. THAKKAR[1], M. WATANABE[2], P. BHATTACHARYA[3], S. CHAKRAVARTY[4], C. DONG[5], A. P. RAVAL[5], J. HARE[6], A. KHAN[4], T. RUNDEK[5], D. YAVAGAL[5];
[1]Neurol., Miller Sch. of Medicine, Univ. of Miami, Miami, FL; [2]Jikei Univ., Tokyo, Japan; [3]Natl. Inst. of Pharmacol. Educ. and Res., Ahmedabad, India; [5]Neurol., [6]Med., [4]Univ. of Miami, Miami, FL

Abstract: Introduction: Stroke is the leading cause of disability and the 5th leading cause of death in the USA. Despite recent advances in the standard of care for ischemic stroke, over 50% of stroke survivors are chronically disabled. To overcome this, novel treatment strategies need to be developed, particularly ones that help patients regain their functional abilities. Research in the past 2.5 decades has shown great promise in the use of cell-based therapies for ischemic stroke. Intra-arterial (IA) delivery directs the cells to the injured brain, is minimally invasive and widely available, making it attractive for clinical translation. Our group and others have shown the safety and efficacy of IA Mesenchymal Stem Cell (MSC) treatment in a rodent stroke model at 24 hours post-reperfusion. The optimal timing of a single dose of IA MSC is unknown. Also, whether an additional dose will lead to greater benefit and the optimal timing of the second dose need to be determined for successful translation of IA MSC to clinic. Here, we hypothesize that dual dose IA-MSC treatment leads to enhanced improvement after stroke as compared to a single dose and the controls. **Methods:** For this study, we used a reversible middle cerebral artery occlusion (rMCAO) model for stroke in rats. IA-MSCs (10^5 cells) or control PBS were administered in dual doses first at 1 day and then at 4, 6- or 15-days post-stroke. Behavioral analysis, stroke volume and neuronal counting were determined at 30 days after treatment. **Results:** Significant improvement in behavior, reduction in stroke volume, and increased neuronal survival with improved brain tissue integrity were reported in the 1-6D group.

Conclusion: The results of our study indicate that dual dose IA-MSCs at 1-6days after stroke is safe and leads to enhanced symptomatic recovery, reduced stroke lesion, and increased neuronal survival. In summary, our findings contribute to the understanding of the benefit of dual dose of IA MSC and aid effective translation to stroke patients.

Disclosures: **R. Thakkar:** A. Employment/Salary (full or part-time):; University of Miami. **M. Watanabe:** None. **P. Bhattacharya:** None. **S. Chakravarty:** None. **C. Dong:** A. Employment/Salary (full or part-time):; University of Miami. **A.P. Raval:** A. Employment/Salary (full or part-time):; University of Miami. **J. Hare:** None. **A. Khan:** None. **T. Rundek:** A. Employment/Salary (full or part-time):; University of Miami. **D. Yavagal:** A. Employment/Salary (full or part-time):; University of Miami.

Poster

P329. Therapeutic Approaches for Ischemic Stroke II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P329.09

Topic: H.05. Working Memory

Title: Uncovering sex-specific differentially regulated molecular changes in chronic hypoxia-induced learning and memory impairments using zebrafish model

Authors: *S. CHAKRAVARTY[1], **K. SOREN**[2];
[1]Applied Biol., Indian Inst. of Chem. Technol. (CSIR-IICT), Hyderabad, India; [2]Applied Biol., CSIR-Indian institute of Chem. Technol., Hyderabad, India

Abstract: Uncovering sex-specific differentially regulated molecular changes in chronic hypoxia-induced learning and memory impairments using zebrafish model Kalyani Soren[1,3], Tapatee Das[1,3], Kodi Gnaneshwari[1], Pankaj Malakar[1], Arvind Kumar[2], Srinivas Kantevari[3,4], Sumana Chakravarty*[1,3] [1]Applied Biology, CSIR- Indian Institute of Chemical Technology , Hyderabad. [2]CSIR-Centre for Cellular and Molecular Biology, Hyderabad [3]Academy of Scientific and Innovative Research (AcSIR), Ghaziabad-201002, U.P, India. [4]Fluoro & Agrochemicals Division, CSIR- Indian Institute of Chemical Technology, Hyderabad. *Email: sumanachak@iict.res.in Chronic hypoxic (CH) episodes often lead to problems in the brain, which in turn lead to cognitive impairments such as decline in memory & inability to learn, besides other behavioral deficits. Many of the neurological disorders have sex-specific characteristics. We have recently reported sex differences in systemic responses in zebrafish (zf) at the cellular & molecular level during the post-hypoxia (PH) recovery. However, studies on sex differences in CH induced cognitive impairment are not reported yet. To address this, we have used zf of 6-7 months old in this study. We have examined the effects of hypoxic stress (HS) on the behavior of zf using the novel tank test following their exposure to CH. Our results showed PH behavioral deficits in most of the tests, which are thought to be known as the hallmarks of

HS in both sexes. We evaluated cognition of zf, using T-Maze test after training them to recognize reward/punishment zone. We observed a PH cognitive impairment with an indication of sex difference in choosing the reward zone. Further study showed significant increase in the levels of inflammatory markers found in male zf brain compared to that in female. Keeping in mind, restoration between protective & degenerative neuroinflammation is of great interest in therapeutic approaches, we have used a novel inhibitor compound of inflammasomes molecule NLRP3 screened through in-vitro assays. There was significant decrease in inflammation associated with an increase in glial markers & cognitive improvements, irrespective of sexes; the compound was injected at the dose 5mg/kg body weight. A noticeable increase in neurogenesis in females as compared to their male was also observed suggesting the beginning of neural repair mechanism. Revealing the sex specificity in HS response, this study emphasizes not only the importance of sex difference in behavioral & pharmacological analyses but also in modeling sex differences in clinical responses to anti-inflammatory drugs in developing CNS therapeutics, using an alternate animal model.

Disclosures: S. Chakravarty: None. **K. Soren:** None.

Poster

P363. Spinal Cord Injury: Neuro-Immune Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P363.01

Topic: C.11. Spinal Cord Injury and Plasticity

Title: Age-related differences in inflammatory and behavioral responses following spinal cord contusion in rats

Authors: *B. T. DAVID, S. Y. OH, Q. SHEN, Y. LOPEZ-RAMIREZ, D. TUCKER, N. WROBEL, B. AVONTS, J. ALBERTS, J. MOSS, K. RAUE, R. G. FESSLER;
Neurosurg., Rush Univ. Med. Ctr., Chicago, IL

Abstract: The damage that results from spinal cord injury (SCI) is caused by both the initial mechanical trauma and the secondary injury that exacerbates the inflammatory reaction at the lesion site. As the average age at which an individual suffers an SCI has increased over the past decades, it has become increasingly relevant to determine what role age has on that inflammatory response. The aging process has been thought to be coincident with a state of nonresolving activation of inflammatory pathways, but many studies that have investigated the role of aging and SCI have found mixed results. Here, using the Infinite Horizon Impactor, we administered moderate contusive injuries to rats at a variety of ages between 3 and 12 months. We found that the older animals had a trend towards a worse recovery of both hindlimb locomotion and bladder function, despite having a lower microglia/macrophage population at the injury epicenter. Moreover, the reduced inflammatory response in older rats was not detected during the acute

phase. This inflammation data is contrary to what some have reported in significantly older animals (18 months), suggesting a possible shift in the inflammatory response with age, as opposed to a gradual increase in overall systemic inflammation.

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Poster

P363. Spinal Cord Injury: Neuro-Immune Mechanisms

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Program #/Poster #: P363.02

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NRF- 2020R1F1A1069875
NRF-2021R1I1A3058902

Title: Transplantation of tauroursodeoxycholic acid-inducing M2-phenotype macrophages promotes an anti-neuroinflammatory effect and functional recovery after spinal cord injury in rats

Authors: *S. SOHN, W.-K. KO, S. KIM, G. HAN, D. LEE, S. SHEEN;
CHA University, CHA Bundang Med. Ctr., Gyeonggi-do, Korea, Republic of

Abstract: In this study, we study the transplantation of tauroursodeoxycholic acid (TUDCA)-induced M2-phenotype (M2) macrophages and their ability to promote anti-neuroinflammatory effects and functional recovery in a spinal cord injury (SCI) model. To this end, compared to the granulocyte-macrophage colony-stimulating factor (GM-CSF), we evaluated whether TUDCA effectively differentiates bone marrow-derived macrophages (BMDMs) into M2 macrophages. The M2 expression markers in the TUDCA-treated BMDM group were increased more than those in the GM-CSF-treated BMDM group. After the SCI and transplantation steps, pro-inflammatory cytokine levels and the mitogen-activated protein kinase (MAPK) pathway were significantly decreased in the TUDCA-induced M2 group more than they were in the GM-CSF-induced M1 group and in the TUDCA group. Moreover, the TUDCA-induced M2 group showed significantly enhanced tissue volumes and improved motor functions compared to the GM-CSF-induced M1 group and the TUDCA group. In addition, biotinylated dextran amine (BDA)-labeled corticospinal tract (CST) axons and neuronal nuclei marker (NeuN) levels were increased in the TUDCA-induced M2 group more than those in the GM-CSF-induced M1 group and the TUDCA group. This study demonstrates that the transplantation of TUDCA-induced M2 macrophages promotes an anti-neuroinflammatory effect and motor function recovery in SCI.

Therefore, we suggest that the transplantation of TUDCA-induced M2 macrophages represents a possible alternative cell therapy for SCI.

Disclosures: S. Sohn: None. **W. Ko:** None. **S. Kim:** None. **G. Han:** None. **D. Lee:** None. **S. Sheen:** None.

Poster

P363. Spinal Cord Injury: Neuro-Immune Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P363.03

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH NINDS R01 NS097880
NIH HEAL Initiative Supplement # NS097880-S1 (MRD)

Title: No monocyte-derived macrophages in the anterior cingulate cortex of mice with SCI-induced affective pain

Authors: *J. H. RICHARDS, M. R. DETLOFF;
Neurobio. & Anat., Drexel Univ. Col. of Med., Philadelphia, PA

Abstract: Spinal cord injury (SCI) is a neuroimmune disorder that results in chronic pain development. The pain experience is multidimensional, involving sensory, cognitive, and affective components. While most basic science research focuses on sensory discriminative alterations in SCI pain, the prevalence of affective disorders, like major depressive disorder is high in the SCI community. Affective pain is defined as the negative emotion ascribed to the perception or anticipation of noxious sensations that motivates behavior. Importantly, both chronic pain and affective disorders have been correlated with aberrant inflammation generated by resident or infiltrating immune cells throughout the nervous system. The anterior cingulate cortex (ACC) contributes to both decision-making and emotional regulation and its activity is associated with both pain and affective disorders. Thus, we hypothesize that SCI-induced neuroinflammation in the ACC causes alterations in affective pain in the mouse model. We completed moderate unilateral C5 contusions on LysM-eGFP reporter mice in order to visualize macrophages that infiltrate the ACC after injury. Alterations in affective behavior, like anhedonia, learned-helplessness, and social withdrawal were measured with sucrose preference, forced swim, and social interaction tests. von Frey and Hargreaves' testing measured sensory discriminative aspects of pain. At 6 weeks following SCI, both male and female mice exhibit sensory and affective dysfunction compared to naïve, uninjured mice. Immunohistochemistry performed on ACC tissue did not reveal any evidence of infiltrating macrophages at 7- or 28-days post injury. However, we identified increased IBA-1+ staining in the ACC at 7-day post-SCI, which suggests that SCI causes activation of microglia. We are currently utilizing qPCR to

measure changes in gene expression of cytokines that may be associated with affective pain. These findings indicate that supraspinal inflammation may be a potential target for future interventions that reduce affective pain.

Disclosures: J.H. Richards: None. **M.R. Detloff:** None.

Poster

P363. Spinal Cord Injury: Neuro-Immune Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P363.04

Topic: C.11. Spinal Cord Injury and Plasticity

Support: Chan Zuckerberg Initiative Inflammation Program

Title: Perturbations of the immune response after spinal cord injury in aged mice revealed by single-cell profiling

Authors: *A. SALVADOR[1,2], T. DYKSTRA[2], J. RUSTENHOVEN[2], S. BLACKBURN[2], I. SMIRNOV[2], J. HERZ[2], J. KIPNIS[2];
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Abstract: Traumatic CNS injuries such as spinal cord injury (SCI) affect millions of people worldwide. More aging individuals experience SCI which lead to more devastating consequences. While treatment modalities are limited, one promising strategy is to harness the power of the immune system as previous studies have demonstrated its beneficial function after CNS injury. In this study, we investigated the temporal changes of the immune (CD45+) cell response in the injured mouse spinal cord using single-cell profiling. We observe dramatic changes in the immune cell composition of the spinal cord characterized by a massive infiltration of myeloid cells in the acute phases of injury (3 dpi) followed by the recruitment of lymphocytes in the more chronic phase of injury (14 dpi). These changes were also accompanied by shifts in the transcriptional state of the resident immune cells of the spinal cord, microglia. We observed the emergence of activated and interferon-responding microglia states, which may have diversified functions in the injured spinal cord. Peripheral myeloid cells recruited to the injury site were also investigated, and our analysis reveals populations that may contribute to distinct function and ontogeny. Putative ligand-receptor interactions were also analyzed to predict signaling pathways between immune cells that can perpetuate inflammation to worsen damage and those that can support repair. We also studied the temporal changes in the immune response of aged mice after spinal cord injury, which remain unexplored. The aged naive spinal cord already harbors a striking proportion of activated microglia and T cells representing heightened inflammation. Moreover, injury in aged mice leads to an upregulated expression of cytokine and

chemokine genes in macrophages compared to injury in young mice. These results may indicate that there is exacerbated inflammation in aging that worsens recovery. Overall, the cellular atlas we generated in this study serves as a rich, comprehensive resource that can shed light on our mechanistic understanding on the role of the immune system and of aging in CNS injury, which may help identify critical targets that enhance repair.

Disclosures: A. Salvador: None. **T. Dykstra:** None. **J. Rustenhoven:** None. **S. Blackburn:** None. **I. Smirnov:** None. **J. Herz:** None. **J. Kipnis:** F. Consulting Fees (e.g., advisory boards); Puretech Health.

Poster

P363. Spinal Cord Injury: Neuro-Immune Mechanisms

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Topic: C.11. Spinal Cord Injury and Plasticity

Support: NINDS R01NS081040
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The Miami Project to Cure Paralysis/The Buoniconti Fund
UMiami Department of Neurosurgery Black Lives Matter Fellowship

Title: Foamy macrophages induce fibrosis via IL1R1 after spinal cord injury

Authors: *K. MADDY[1], C. RYAN[1], L. MILICH[1], J. CHOI[1], J. K. LEE[2];
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Abstract: Background: Following spinal cord injury (SCI), inflammation and fibrosis at the lesion reduces the regenerative capacity of neural tissue. Myeloid cells play a key role in modulating inflammatory responses and thus are an important target in limiting tissue damage and maintaining tissue integrity at the injury site. A subset of these cells are lipid-droplet filled proinflammatory macrophages, termed “foamy macrophages.” In order to develop more effective therapeutic interventions, a better understanding of these myeloid cellular interactions is necessary. Our previous single-cell RNA sequencing (sc-RNAseq) study using of virtually all cell types comprising the mouse SCI site identified a novel interaction between IL1R1+ fibroblasts and IL1a+ macrophages. However, the functional role of this receptor-ligand signaling pathway in SCI pathobiology remains poorly understood.**Methods:** scRNA-seq data from mouse SCI tissue was used to assess expression of IL1a and IL1R1 at the injury site. Immunohistochemistry and in situ hybridization was used for validation studies. Using in vitro models to further probe the mechanism of fibrosis after SCI, primary fibroblasts from wild-type (WT) and IL1R1 knockout (KO) mice were cultured and treated with foamy macrophage

conditioned media as a source of IL1a, to measure the expression of extracellular matrix (ECM) molecule fibronectin. For in vivo analysis, IL1R1 KO mice received a mid-thoracic contusive SCI and the injury site was assessed histologically.**Results:** scRNA-seq data showed IL1a expression in macrophages and IL1R1 expression specifically in fibroblasts and endothelial cells, which was validated using immunohistochemistry and in situ hybridization.. Injury sites in IL1R1 KO mice showed reduced fibrosis after SCI.**Conclusion:** Our results suggest an important role played by IL1R1+ fibroblasts and their interactions with IL1α+ macrophages in inflammation and fibrosis after SCI. Genetic deletion of IL1R1 results in reduced fibrotic scar formation after SCI. Understanding these receptor-ligand interactions and their role in SCI pathogenesis at the injury site can present new opportunities for targeted therapeutic interventions.

Disclosures: K. Maddy: None. **C. Ryan:** None. **L. Milich:** None. **J. Choi:** None. **J.K. Lee:** None.

Poster

P363. Spinal Cord Injury: Neuro-Immune Mechanisms

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Program #/Poster #: P363.06

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NINDS R01NS081040
NINDS F31NS115225
The Miami Project to Cure Paralysis/The Buoniconti Fund

Title: Pi3k/mtor inhibition reduces foamy macrophages after spinal cord injury

Authors: *C. B. RYAN, J. S. CHOI, H. A. ALI, J. K. LEE;
Dept. of Neurosurgery, The Miami Project to Cure Paralysis, Univ. of Miami Miller Sch. of Med., Miami, FL

Abstract: After spinal cord injury (SCI), macrophages infiltrate the injury site and phagocytose lipid debris, forming lipid-laden “foamy” macrophages. Foamy macrophages contribute to the propagation of inflammation at the injury site and remain in the lesion chronically. To better understand the mechanisms of foamy macrophage formation, we developed a novel in vitro foamy macrophage assay. The assay uses primary bone-marrow-derived macrophages isolated from wildtype 6-8 week old male mice treated with wildtype spinal cord homogenate to represent all lipid sources at the spinal cord injury site. Using this assay, we found increased expression of inflammatory cytokines and mitochondrial dysfunction in foamy macrophages, accompanied by increased production of ROS. Pharmacological inhibition of PI3K/mTOR resulted in decreased lipid content in foamy macrophages, which was associated with reduction

in ROS production and inflammatory cytokine production. RNA-sequencing of foamy macrophages following PI3K/mTOR inhibition identified induction of lipid transporters as a potential mechanism. Macrophage-specific inhibition of PI3K/mTOR using liposomes showed significant decrease in foamy macrophages at the injury site after a mid-thoracic contusive SCI in 8-10 weeks old female mice (7 dpi: n=9 PBS, n=9 Inhibitor; 28 dpi: n=8 PBS, n=8 Inhibitor). Together, our data suggests that foamy macrophages are important contributors to inflammation after injury and identify PI3K/mTOR pathway in foamy macrophages as a potential therapeutic target for SCI.

Disclosures: C.B. Ryan: None. **J.S. Choi:** None. **H.A. Ali:** None. **J.K. Lee:** None.

Poster

P363. Spinal Cord Injury: Neuro-Immune Mechanisms

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Program #/Poster #: P363.07

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH Grant T32NS096050
Craig H. Neilson Foundation Grant 642928

Title: Microbiome alterations lead to impaired enteric neuronal status following spinal cord injury

Authors: *A. M. HAMILTON, J. CHANG, S. M. GARRAWAY, S. SRINIVASAN, T. R. SAMPSON;
Emory Univ., Atlanta, GA

Abstract: Research Objective & Rationale: Neurogenic bowel dysfunction is one of the most cited detriments to quality of life in persons with spinal cord injury (SCI). In addition to the loss of supraspinal control over spinal defecation centers, SCI results in significant alterations to enteric neuromusculature, including enteric neuron loss. The gut microbiome changes significantly post-SCI and directly affects injury outcomes and recovery, likely through metabolic and inflammatory pathways. Interactions between host and microbiome modulate overlapping functions including enteric neurogenesis, endocrine signaling, and inflammation, all of which impact intestinal motility. We therefore sought to determine if post-SCI dysbiosis directly contributes to changes in enteric neuromusculature and gastrointestinal (GI) motility and if targeting the post-injury microbiome could improve GI outcomes associated with SCI.
Methods: Adult Mice were subjected to a midthoracic (T9) spinal cord injury or a midthoracic laminectomy (sham-surgery controls). GI motility was assessed, and molecular analysis of enteric neuronal status and colonic inflammation performed. Fecal microbiomes derived from mice with SCI or from sham controls were used to colonize germ-free mice to assess the impact

of the injury-induced microbiome on GI physiology in mice naive to injury. Following colonization, GI motility was assessed, colonic tissues collected, and molecular markers of neuronal and inflammatory status analyzed. **Results:** We observed that mice with SCI had significantly impaired intestinal transit, as compared to sham controls. Colonic tissues revealed decreased neuronal markers in injured mice, as well as evidence of increased inflammation. We noted alterations to serum serotonin metabolism, indicative of dysfunction present in intestinal endocrine cells. Colonization of injury naïve mice with microbiomes derived from injured donors recapitulated colonic dysmotility of their donors. Intriguingly, molecular analysis of mice colonized with SCI-derived microbiomes revealed increased intestinal inflammation and significant alterations to the status of the ENS and circulating intestinal neuropeptide hormones. **Conclusions:** Our data demonstrate a direct effect of the gut microbiome on post-SCI GI dysmotility and pathophysiology, suggesting that dysbiosis appearing after injury contributes to reduced enteric neurons, GI inflammation, and an altered serum metabolic status. These novel findings provide a clear rationale for the advancement of targeted dietary or probiotic therapies to ameliorate GI dysfunction in persons with SCI.

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Poster

P363. Spinal Cord Injury: Neuro-Immune Mechanisms

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Program #/Poster #: P363.08

Topic: C.11. Spinal Cord Injury and Plasticity

Support: VA 1I01BX004039-01A2

Title: Il-12 contributes to secondary damage and functional impairment after spinal cord injury.

Authors: *J. ROSAS[1], N. PELISCH[1], B. APERI[1], K. STEHLIK[2], M. D. BUDDE[2], K. SWARTZ[1], **A. KRONER-MILSCH**[2];
[2]Neurosurg., [1]Med. Col. of Wisconsin, Milwaukee, WI

Abstract: Traumatic spinal cord injury (SCI) is a relatively frequent event that imposes a massive burden on the health, quality of life and socioeconomic situation of affected persons. Tissue damage after SCI occurs in two phases: primary and secondary damage. Primary damage refers to the initial mechanical trauma causing cell death and tissue disruption while secondary damage includes multiple processes that can promote further damage. One critical secondary process is inflammation or the recruitment and activation of immune cells. A more complete understanding of individual contributors to inflammatory damage is necessary to specifically target and modify detrimental factors. Inflammation after SCI is exacerbated, with activated

microglia and monocyte-derived macrophages being the main immune cell populations in the injured tissue. Pro-inflammatory cytokines IL-12 and IL-23 share the p40 subunit which is strongly upregulated after phagocytosis of red blood cells. IL-12 and IL-23 can be expressed by a multitude of cell types and mediate multiple functions on different target cells. Here, we assess the functional effect of IL-12 and IL-23 via IL-12p40 on SCI, using a low thoracic (T11) moderate contusion injury (50 kDyne) in mice. First, we quantified protein expression using Western blot to find IL-12 factors upregulated compared to laminectomy controls. Furthermore, we observed a concurrent upregulation of IL-12p70 with hemoglobin, a biomarker for magnetic resonance imaging (MRI) spinal cord hemorrhage. We have identified IL-12p40 to be expressed primarily by astrocytes early after injury and by microglia by 28 days after injury. Both receptors are robustly expressed in astrocytes. To test the functional impact of IL-12 and IL-23 on SCI, we utilized IL-12p40 knockout mice and wildtype controls to quantify locomotor recovery after SCI using the Basso Mouse Scale (BMS). We observed improved recovery in IL-12p40 deficient mice compared to wildtype mice. IL-12p40 KO mice also demonstrate significantly more spared tissue 28 days after injury. In summary, these results suggest that the absence of IL-12p40, but not IL-23p19, mediates a neuroprotective effect after contusion SCI, serving as a new potential therapeutic target to ameliorate secondary damage and promote better functional recovery.

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Poster

P363. Spinal Cord Injury: Neuro-Immune Mechanisms

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Program #/Poster #: P363.09

Topic: C.11. Spinal Cord Injury and Plasticity

Support: TIRR Foundation
Craig H. Neilsen Foundation Grant 648714

Title: Neutrophil Extracellular Traps in Spinal Cord Injury

Authors: *S. K. REID[1,2], M. E. LEAL[2], M. A. KIRCHHOFF[2], S. J. LEE[2], S. W. MIN[2], M. E. PLATT[2], D. A. MCCREEDY[1,2];
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Abstract: Spinal cord injury (SCI) is a severely debilitating event that results in extensive tissue loss and long-term neurological deficits. Neutrophils are the first peripheral immune cell to infiltrate the spinal cord in large numbers following injury; however, their role in secondary tissue damage is not well characterized. Neutrophils employ numerous cytotoxic effector functions in response to injury that are important for eliminating potential pathogens, but may

result in collateral tissue damage. Neutrophil extracellular traps (NETs) are a recently identified effector function wherein decondensed chromatin decorated with granule proteins is expelled from the cell body as a mechanism to trap and destroy bacteria and other pathogens. Beyond their bactericidal role, NETs have been implicated in a variety of sterile pathologies as damaging to the host. The contribution of NETs towards secondary tissue damage in the context of SCI, however, remains unexplored. To determine if NET formation occurs acutely after SCI, we performed an ELISA for citrullinated histone 3 (CitH3) and DNA complexes in peripheral blood and spinal cord samples at 0, 4, 8, 12, 18, and 24 hrs post-SCI. We found that CitH3/DNA levels, which are indicative of NETs, markedly increased over the first 12 hours post-SCI and remained elevated through 48 hours post-SCI. Similar trends were observed in blood samples, albeit at lower levels. To confirm NET formation in neutrophils, we also performed flow cytometry to assess the presence of CitH3 in neutrophils from in blood and spinal cord samples at 24 hrs post-SCI. We observed a nearly 7-fold increase in CitH3+/Ly6G+ cells in the acutely injured spinal cord relative to the blood, further indicating that NET formation is occurring in response to SCI. Collectively, our data demonstrate the first evidence of NETs in the injured murine spinal cord, peaking by 24 hrs following injury. Our results suggest that pharmacological treatment to eliminate NETs, such as with the DNase I, may represent a therapeutic strategy to limit tissue loss and improve functional recovery after SCI.

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Poster

P363. Spinal Cord Injury: Neuro-Immune Mechanisms

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2019R1I1A1A01059432

Title: Oncomodulin derived from pro-regenerative macrophages following preconditioning injury promotes sensory axon regeneration

Authors: *M. J. KWON[1], Y. SEO[2], H. CHO[3], H. C. KANG[3], B. G. KIM[1];
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Abstract: We have previously reported the neuron-macrophage interaction mediating perineuronal activation of pro-regenerative macrophages in dorsal root ganglia (DRGs) following preconditioning peripheral nerve injury. Oncomodulin (ONCM) is a macrophage-derived factor previously identified to exert pro-regenerative activities for retinal ganglion axons. The current study found that ONCM expression was markedly increased in DRGs at 3 and 7 days following preconditioning injury. Preconditioning-induced enhancement of neurite outgrowth activities was abolished in ONCM-deficient mice. We determined whether the ONCM is involved in our neuron-macrophage interaction model, in which conditioned medium (CM) obtained from the neuron-macrophage cocultures treated with cAMP exhibits robust neurite outgrowth activities for DRG sensory neurons. The neurite-outgrowth activities were abolished when ONCM-deficient macrophages, but not ONCM-deficient neurons, were used for the cocultures, indicating that macrophage-derived ONCM play a critical role. Our previous study demonstrated that sensory neuron-derived chemokine CCL2 drives activation of pro-regenerative macrophage phenotype. Preconditioning injury in CCR2-deficient mice increased ONCM level to a much smaller extent. CCL2 overexpression in DRG neurons exhibited a dramatic increase in the extent of neurite outgrowth mimicking conditioning effects, while it failed to promote neurite outgrowth in ONCM-deficient mice. We further demonstrated that certain regenerative-associated genes, such as Neuropeptide Y, Sprr1a and Vip were upregulated in ONCM treated DRGs sensory neuron. We sought to examine whether ONCM can be therapeutically used to enhance intrinsic regenerative capacity for DRG sensory axons following spinal cord injury. When ONCM was injected into DRGs, sensory neurons showed very little, if any, improvement in neurite outgrowth. However, injection of ONCM conjugated to a reducible epsilon-poly(L-lysine)-nanogel (REPL-NG) dramatically increased neurite outgrowth. Furthermore, intraganglionic injection of REPL-NG/ONCM complex increased the extent of axon regeneration beyond the dorsal hemisection injury site. Thus, our study demonstrated that ONCM is a critical proregenerative factor derived from macrophages to enhance intrinsic regenerative capacity of sensory axons. We also suggest a possibility that the nanogel-mediated ONCM delivery can be exploited as a therapeutic agent for promoting sensory axon regeneration following spinal cord injury.

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Poster

P363. Spinal Cord Injury: Neuro-Immune Mechanisms

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Program #/Poster #: P363.11

Topic: C.11. Spinal Cord Injury and Plasticity

Support: DOD W81XWH-16-1-0387

Title: Association of metabolic markers and microbiota populations in a rat model of thoracic spinal contusion

Authors: *A. M. SMITH[1], B. A. WELCH[2], K. K. HARRIS[2], M. GARRETT[3], B. E. GRAYSON[4];
[1]Neurobio., Univ. Of Mississippi Med. Ctr., Jackson, MS; [2]Neurobio., [3]Pharmacol., [4]Dept of Neurobio. and Anatom. Sci., Univ. of Mississippi Med. Ctr., Jackson, MS

Abstract: Spinal cord injury (SCI) results in chronic changes to multiple body systems, including the digestive system. The gut microbiota exist symbiotically within the organism and are readily influenced by diet, health status, and physical activity. The relationship between metabolic markers and the microbial populations following SCI remains unknown. We performed thoracic level 10 contusions or sham injuries on male Long-Evans rats. After 4 weeks of recovery, the rats were exposed to either low-fat (9%, LF) or high-fat (40%, HF) diet for 12 weeks. Following euthanasia at 16 weeks, caecal samples were harvested. Microbial DNA was isolated and sequenced using the Illumina Iseq100 platform, and data processed using Microbiome Analyst software. Overall, sham rats weighed significantly more than the SCI rats ($p < 0.01$), but HF-fed rats were heavier than LF-fed rats ($p < 0.05$). HF rats also had elevated body fat composition ($p < 0.05$), with no difference in lean body mass in comparison to LF. Leptin levels were significantly reduced in SCI animals compared to sham ($p <0.05$). Leptin levels were also higher in the HF group compared to LF, independent of injury ($p < 0.05$). Triglyceride levels were significantly increased in HF compared to LF. With respect to the microbiome analysis, significantly increased richness in read diversity existed in SCI animals when compared to sham ($p < 0.01$). Beta diversity analyses showed significant differences in the four groups; LF-fed rats were found to have more diversity overall. There were abundant changes at all taxonomic levels when comparing LF vs. HF, independent of injury ($p < 0.05$). Simple linear regression analyses were performed in sham animals alone to determine the association between body fat; strong association existed at multiple taxonomic levels, including *Lautropia, Rhodobacteraceae,* and *Bacilli.* This relationship was abolished in SCI animals, who only demonstrated a strong relationship in four taxa, (e.g., *Olivibacter* and *Roseburia),* which were alternately not significantly correlated in sham animals. Similarly, analyses in sham animals to determine the relationship with leptin levels showed that there were strong associations with certain bacteria (i.e., *Firmicutes, Clostridia,* and *Enterobacteriales).* Again, this relationship was eliminated in SCI animals, whose leptin levels only had an association with the genus *Olivibacter.* These data suggest that the unique relationship that exists between markers of adiposity and microbiota in healthy, able-bodied rats are mitigated following SCI. More studies are required to understand the reason behind this altered relationship.

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Poster

P363. Spinal Cord Injury: Neuro-Immune Mechanisms

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Program #/Poster #: P363.12

Topic: C.11. Spinal Cord Injury and Plasticity

Support: Craig Neilsen Foundation 543659

Title: Role of Hemopexin in secondary damage following spinal cord injury

Authors: N. PELISCH[1,3], J. ROSAS ALMANZA[4,2], K. STEHLIK[3,2], B. APERI[4,2], ***A. KRONER**[4,2,5];
[1]Neurosurg., [2]Clement J Zablocki VA Med. Ctr., Milwaukee, WI; [3]Neurosurg., [5]Microbiology and Immunol., [4]Med. Col. of Wisconsin, Milwaukee, WI

Abstract: Tissue damage in spinal cord injury (SCI) occurs in two phases, with the primary damage being caused by the mechanical insult and the secondary damage mediated by a multifactorial cascade of events. Inflammation and hemorrhage from sheared blood vessels significantly contribute to secondary damage after SCI. Blood breakdown products such as hemoglobin, heme and iron are pro-inflammatory and highly cytotoxic through lipid peroxidation, DNA oxidation and iron deposition. In the vasculature, breakdown products are sequestered by designated capture proteins: Haptoglobin (Hp) and Hemopexin (Hx) bind free hemoglobin and heme, respectively, $Hx^{-/-}$ mice until the complexes are taken up by macrophages, thereby preventing its toxic effects. Despite low expression levels of Hp and Hx within the CNS, limiting the potential to reduce tissue damage and enhance functional recovery following hemorrhagic insults and trauma, the absence of Hp and Hx under such conditions resulted in further functional impairment. Here, we investigate the role of Hx on injury progression, axonal damage and functional outcome in male and female mice following thoracic contusion SCI. After SCI, Hx is expressed predominantly in astrocytes, while its receptor LRP is expressed mainly by macrophages. At the gene expression level, Hx was upregulated at day 1 post SCI, while LRP expression was increased at day 7 post SCI compared to the laminectomy control group. $Hx^{-/-}$ mice showed reduced locomotor recovery compared to wildtype mice, combined with an increase of axonal damage and reduced neuronal survival distal to lesion. These results indicate a detrimental effect of the lack of Hx and that boosting Hx availability could result in improved functional recovery and reduced secondary damage after SCI.

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Poster

P469. Visual System: Response Modulation and Adaptation

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P469.01

Topic: D.07. Vision

Support: NIH NS109978
NS107968

Title: Distinct effects of arousal and movement on visual spatial selectivity across network, cellular and subthreshold levels in mouse visual cortex

Authors: S. COLETTA[1], J. DEL ROSARIO[2], A. DEL CASTILLO VALERIO[1], B. HAIDER[3]; [1]Georgia Tech., Atlanta, Georgia; [2]Georgia Inst. of Technology/Emory Univ., Atlanta, AL; [3]Biomed. Engin., Georgia Tech. and Emory Univ., Atlanta, GA

Abstract: Sensory processing depends strongly upon factors such as arousal, movement, and task engagement. Several studies in primary visual cortex (V1) of head-fixed mice have revealed that locomotion increases arousal and this leads to membrane potential (Vm) depolarization, desynchronization, and elevated firing rates in excitatory neurons (Neske et al., 2019; Vinck et al., 2015). There remain open questions regarding the influences of arousal and motor action upon vision. First, most studies examine response amplitude without assessing changes to spatial selectivity. Second, most studies rely on one signal modality in isolation from others, providing an incomplete picture of potential effects of arousal and movement on spatial selectivity in V1 across multiple scales. Here, we performed multisite silicon probe and whole-cell patch-clamp recordings in awake, head-fixed, stationary mice (n = 23) while monitoring pupil and periorbital facial movements with high-speed video. We computed pupil size and facial motion energy to explore the impact of arousal versus movement on V1 receptive field (RF) properties. We showed small (9° wide) full contrast vertical bars throughout the visual field (>150°) to assess changes to spatial selectivity. In some probe recordings, we identified parvalbumin (PV) and somatostatin (SOM) interneurons via optogenetic stimulation, and identified putative excitatory and inhibitory neurons based on regular spiking (RS) or fast spiking (FS) waveforms. Even in stationary mice, we found that arousal and movement exerted clear and non-overlapping effects on visual responses and spatial selectivity. With increasing arousal, RS neurons (n = 1150) and local field potential (LFP) responses showed modest and potentially non-monotonic changes in RF amplitude. These effects of increased arousal were mirrored in Vm of excitatory neurons (n=35), where subthreshold response amplitude and spikes decreased while spatial selectivity increased; these trends were distinct from effects of movement, which generally evoked greater depolarization and firing. The effects were clearer and distinct among different inhibitory cell-types: in PV (n=25) and fast-spiking (FS) neurons (n=290), responses increased with arousal and especially movement, but both factors suppressed SOM neurons (n=20). We are examining timing between movement and pupil, and if inhibitory neuron responses explain those in excitatory neurons. These preliminary results suggest a complex interplay of arousal and

movement even in non-locomoting mice, with distinct contributions from cortical cell-types to visual spatial selectivity.

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Poster

P469. Visual System: Response Modulation and Adaptation

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Topic: D.07. Vision

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Title: Local field potential signatures of context-specific processing in the mouse cortical column during visual oddball

Authors: *C. G. GALLIMORE, J. P. HAMM;
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Abstract: Attentional demands shift on a moment-to-moment basis. Top-down attentional control, the ability to maintain focus on the task at-hand by filtering out irrelevant information, is in constant competition with bottom-up attentional capture, the propensity of non-goal related, but otherwise potentially salient, stimuli to evoke an orienting response in cortex. The oddball paradigm is a classic psychological assay to measure this interplay in cortical circuits, which manifests in the form of event-related potentials (ERPs) collected from the scalp. Human studies using this paradigm have shown that people in neuropsychiatric disease-states, such as schizophrenia, display reductions in prediction-error signaling to “oddball” stimuli. However, the cortical circuitry supporting this computation needs further characterization. Here, we used linear microelectrode arrays to measure intracranial local field potentials and current source density profiles across laminae 1-5b of the primary visual cortex (V1) of mice (n=7, male and female) while they viewed full-field oriented square-wave grating stimuli in visual oddball sequences and a “many-standards” control sequence. This allowed us to compare cortical responses to the same visual grating stimuli when they were predictable (i.e. redundant in the oddball run, p=.875), rare and unexpected (i.e. deviants, p=.125), and rare but not-deviant (i.e. in the control run, p=.125). We performed dimensionality reduction techniques on trial-wise time-frequency spectral to identify relevant frequency bands for analysis. We identified a superficial (layer 1) and deep (layer 4, ≈400 μm deep) increase in low-frequency (2-10 Hz) oscillatory power to contextually deviant stimuli, suggesting anatomical and frequency-specific signatures of sensory prediction error. Further analysis of current source density (an estimate of local synaptic current flux)

demonstrated deviant stimulus-specific sources and sinks in supragranular regions of V1, consistent with previous *in vivo* two-photon microscopy recordings from our lab which showed excitatory cell firing selective to contextually deviant stimuli in L2/3, but to negligible degrees in deeper layers. We also identified increased alpha (10-15Hz) desynchronization to deviant stimuli across layers 2-5 at the immediate onset of deviant stimuli, but decreased alpha desynchronization in the late stimulus period (300-500ms). These results suggest that bottom-up processing of contextually unexpected stimuli engages low-frequency oscillations and alpha-band dynamics in a layer-specific pattern in V1.

Disclosures: C.G. Gallimore: None. **J.P. Hamm:** None.

Poster

P469. Visual System: Response Modulation and Adaptation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P469.03

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Title: Rapid visual adaptation of LGN neurons in the awake macaque monkey

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Abstract: The nervous system needs to dynamically adapt to changing environments. A well-known form of visual adaptation is the reduction of neuronal spiking to prolonged exposure of sensory stimuli. This adaptation had long been thought to be not a feature in LGN neurons but one for cortical neurons. However, this view has been challenged by evidence of visual adaptation in the lateral geniculate nucleus of the thalamus (LGN) (Solomon et al., 2004). LGN adaptation in that study was limited to a specific class of neurons in anesthetized animals stimulated over prolonged periods of time (> 10 secs). However, under normal viewing

conditions, eye movements limit uninterrupted stimulation to durations of a few hundred milliseconds. This difference in time scales raises the question whether LGN adaptation occurs on time scales relevant to typical visual function. Here, we show that adaptation in the LGN occurs for short stimulus presentations in awake macaque monkeys. We recorded the visual responses of LGN neurons to one second presentations of 4Hz drifting sinusoidal luminance gratings. We then examined the relative change of each neuron's response across successive grating cycles. This analysis revealed significant decreases in the responsiveness of neurons from all cell classes (parvocellular, magnocellular, and koniocellular). However, only magnocellular neurons showed significant adaptation on the population level. Adaptation also increased response variability between neurons while the Fano factor of individual neurons remained largely constant. Lastly, we examined whether adaptation differed between binocular and monocular stimulation. We found that response reduction was equally pronounced when stimuli were shown to one or both eyes, suggesting a monocular (possibly retinal) origin. Taken together, these results demonstrate significant pre-cortical adaptation during active vision.

Disclosures: L.J. Daumail: None. **B.M. Carlson:** None. **B. Mitchell:** None. **M.A. Cox:** None. **J.A. Westerberg:** None. **C. Johnson:** None. **P.R. Martin:** None. **F. Tong:** None. **A.V. Maier:** None. **K. Dougherty:** None.

Poster

P469. Visual System: Response Modulation and Adaptation

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Topic: D.07. Vision

Support: NEI R01-EY025872
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Title: Temporal context in visual encoding and readout across species

Authors: *T. SHEEHAN[1], J. SERENCES[2];
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Abstract: How does stimulus history shape the encoding of incoming visual information and eventual perception? These questions have been explored in the context of adaptation studies which typically present inducing stimuli for unnaturally long periods, and examine the relative influence on neural coding (Patterson, et al., 2013) or perceptual reports (He & MacLeod, 2001). Recently, we found that changes in sensory coding do not directly shape perception but require a readout process that accounts for changes at encoding due to adaptation (Sheehan & Serences, 2021). We further noted that sensory effects were observable up to three stimuli (and over 50s) following the inducing item (presented for only 500ms), suggesting history effects may be robust

even under natural stimulus conditions.
To shed light on this further, we analyzed data from mice and non-human primates passively viewing stimuli and examined how sensory history shapes neural coding (Pachitariu & Stringer, 2019). We found that repulsive biases were pervasive across paradigms, recording technique, and model organisms with significant repulsive biases observable from items as many as 9 stimulus presentations back (12s). This is notable because at such short presentation times perceptual reports are not found to be repelled and are often attracted to past stimuli. As stimuli are naturally correlated across time (Felson et al., 2005), we propose that sensory readout 'expects' a certain amount of adaptation in the encoding population and adjusts its interpretation of incoming sensory signals accordingly. Such an account allows both perceptual attraction and repulsion from past stimuli depending on the strength and duration of stimulus presentations. We test this model on human observers while examining sensory representations in visual areas with BOLD fMRI decoding.
Citations
Sheehan, T.C. and Serences, J.T., 2021. Sensory readout accounts for adaptation. bioRxiv.
Patterson CA, Wissig SC, Kohn A. 2013. Distinct effects of brief and prolonged adaptation on orientation tuning in primary visual cortex. J Neurosci.
He, S. and MacLeod, D.I., 2001. Orientation-selective adaptation and tilt after-effect from invisible patterns. Nature.
Pachitariu, Marius; Michaelos, Michalis; Stringer, Carsen (2019): Recordings of ~20,000 neurons from V1 in response to oriented stimuli. Janelia Research Campus. Dataset. https://doi.org/10.25378/janelia.8279387.v3
Felsen, G., Touryan, J. and Dan, Y., 2005. Contextual modulation of orientation tuning contributes to efficient processing of natural stimuli. Network: Computation in Neural Systems

Disclosures: T. Sheehan: None. **J. Serences:** None.

Poster

P469. Visual System: Response Modulation and Adaptation

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P469.05

Topic: D.07. Vision

Title: Unaltered divisive normalization in V1 of a mouse model with altered short-term plasticity in thalamocortical synapses

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Abstract: Divisive normalization is a model that describes how much the response of a neuron to a set of combined stimuli is lower than would be expected by a linear summation of the responses to the individual stimuli. This type of normalization describes the interaction of stimuli on responses in V1 very well and has been suggested to serve the maximizing of sensitivity and reduction of redundancy (Carandini and Heeger, 2011). The source of this normalization is still unclear. One hypothesis is that divisive normalization in the early visual system is caused by short-term depression of afferent excitatory synapses (Carandini et al., 2002). In order to test this hypothesis, we used a mouse model with altered short-term plasticity in the thalamocortical synapse. This was achieved by knocking out one allele of the Stxbp1 gene in thalamic relay neurons. Heterozygous knockout of Stxbp1 (a.k.a. Munc18-1) has been shown to cause increased short-term depression (Toonen et al., 2006). We recorded visual responses from V1 of the anesthetized mice. Indeed, these mice show a decreased half-life of visual responses. However, contrast tuning and cross orientation suppression, in which the effects of divisive normalization can be shown, were unaffected. This suggests that divisive normalization is not caused by short-term depression of thalamocortical synapses. Based on our data, a cortical source of divisive normalization seems more likely.

Disclosures: J.L. Cazemier: None. **L.N. Cornelisse:** None. **J.A. Heimel:** None.

Poster

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Topic: D.07. Vision

Support: National Institute of Mental Health (R00MH115082)
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Fellowship (CNCD-2CI)

Title: Schizophrenia-relevant mutation in Kalrn yields neurooscillatory aberrations in mouse sensory cortices

Authors: *A. D. FERRELL[1], A. M. RADER[1,2], Z. NISSEAU-BEY[1], R. A. SWEET[3,4], M. J. GRUBISHA[3,4], J. P. HAMM[1,2];
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Abstract: Many neuropsychiatric disorders involve disruptions in basic sensory processing. Using standardized sensory stimulation paradigms, such as the auditory steady-state (aSSR) and oddball sequences, such disruptions can be reliably quantified with scalp electrophysiology (i.e. EEG) in human patient samples. From a translational standpoint, these passive stimulation

sequences present a strategic inroad for studying the underlying neurobiological basis of sensory pathology utilizing animal models. To this end, we have recently developed a mouse model of a naturally occurring mutation in Kalrn, initially described in a re-sequencing analysis of a schizophrenia cohort. These mice (Kalrn-PT) exhibit reductions in auditory cortical dendritic length, branching, and spine density and impaired silent gap detection in auditory testing that develops across adolescence. In order to gain insight into disease-relevant pathology at the circuit level, we recorded local field potentials (LFPs) in awake mice, employing two sensory stimulation paradigms used in human clinical research: aSSR and a visual oddball sequence. In the aSSR, we presented 1-second long pure-tone auditory stimuli (70 dB), from 2kHz to 21kHz, where sinusoidally modulated at 40-Hz (gamma-band), and stimulus induced power at 40-Hz was quantified in the LFP. In the visual oddball, full field moving grating stimuli were presented at 1-Hz (.5 sec ISI), with one orientation (e.g. 45-deg) serving as the redundant (p=.875) and the opposite orientation as the oddball stimulus (p=.125). Adult male and female mice (P84-154) were used with the following genotypes: homo-(n=6) or heterozygous (n=3) Kalrn-PT mutants; littermate wildtype controls (WT; n=8). Bipolar electrodes were stereotaxically implanted ≈300 µm deep into primary sensory cortex (auditory-A1 or visual-V1). For mice in the visual stimulus paradigm, bipolar electrodes were also inserted into ACa, a region of prefrontal cortex (PFC) which projects to V1. In A1, mutants displayed increased background noise across all frequencies. 40-Hz entrainment was altered as a function of A1 tonotopy; mutants showed decreased power to locally preferred pitches, but increased power to side-bands, relative to WT. In the dual recordings from V1 and PFC, the Kalrn-PT mice exhibited diminished fronto-visual synchrony across all frequencies. Collectively, these results suggest that Kalrn-PT mutations likely affect inter -regional/-columnar circuitry. This is consistent with known deficits in apical dendrites in Kalrn-PT mutants and suggests a deficiency in dendrite-targeted inhibition, potentially mediated by somatostatin-positive interneurons.

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Poster

P469. Visual System: Response Modulation and Adaptation

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Program #/Poster #: P469.07

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Bertarelli Foundation (LP)

Title: Task-dependent routing of visual information

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Abstract: Current computational models of visual processing focus on the bottom-up cascade of sequential operations along the visual cortex. However, feedback in the brain is abundant and its role is crucial, especially for goal-directed behaviors. In this exploratory research, we investigated how information is routed in a task-dependent manner by recording intracranial field potentials during two tasks in 21 epilepsy subjects implanted with electrodes. We propose a computational model whereby top-down signals modulate activations in a convolutional neural network. In experiment 1, subjects indicated the presence of a given object category (Bansal et al, 2014). Out of 776 electrodes, 101 electrodes (13 %) showed target-dependent response modulation. These electrodes were mainly located in the inferior and middle temporal gyri and the fusiform gyrus. Experiment 2 consisted of synthetic face images generated by the software FaceGen that varied along the male/female dimension or the young/old dimension and subjects classified images according to either dimension. The instructions about whether to discriminate according to one or the other dimension were provided either before stimulus presentation (*pre* condition) or after stimulus presentation (*post* condition). Focusing on the *pre* condition, out of 1201 electrodes, 25 electrodes (2 %) showed modulation by either one of the two tasks. These electrodes were mainly in the middle occipital gyrus, the lingual gyrus, and the middle temporal gyrus. We divided the temporal dynamics of the responses into two phases: an object recognition phase [0-t_i] ms after stimulus onset and a top-down modulation phase [t_i-t_f] ms after stimulus onset, where t_i and t_f represent the first and the last time point of significant divergence (t-test, $p < 0.01$) between the two conditions compared, respectively. To understand the mechanisms of target modulation we propose a computational model that starts with a deep convolutional neural network (VGG19) pre-trained on ImageNet. A top-down modulation signal is implemented in the top two layers of the model (from the classification layer to the topmost fully connected layer and from the topmost fully connected layer to the prior fully connected layer). This top-down signal was dependent on the inputs and on the current task. The same model was used for both experiments. The proposed model qualitatively approximated the task-dependent modulation effect observed in the two experiments. These results provide a proof-of-principle account for how task-dependent goals can be incorporated by the cortex to route information and achieve different goals with the same sensory inputs.

Disclosures: L. Pollina: None. **K. Bendtz:** None. **G. Kreiman:** None.

Poster

P469. Visual System: Response Modulation and Adaptation

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Program #/Poster #: P469.08

Topic: D.07. Vision

Support: NSF Career Award 1652617
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Title: Stimulus invariant population statistics in response to natural movies

Authors: *B. HOSHAL[1], C. M. HOLMES[3], J. M. SALISBURY[2], M. J. BERRY, II[4], O. MARRE[5], S. E. PALMER[2];
[1]Committee on Computat. Neurosci., [2]Organismal Biol. and Anat., Univ. of Chicago, Chicago, IL; [3]Dept. of Physics, [4]Princeton Univ., Princeton, NJ; [5]Inst. de la Vision - UPMC, Paris, France

Abstract: Over the course of the lifetime of an organism, its sensory organs must encode stimuli described by different statistics; we expect that neural mechanisms exist to support stimulus dependent adaptation. In the absence of adaptation, cells doing the same computation should have different statistics in response to different stimuli. Stationarity in the signal can therefore be a signature of adaptation. To investigate this, we simultaneously recorded the responses of 93 tiger salamander retina ganglion cells presented with multiple natural scene movies. A variety of maximum entropy-based analysis techniques revealed a consistent, stimulus invariant structure to the population codes. We believe that this invariance may be useful for downstream decoding, and may be an indicator of adaptation in response to natural scenes.

Disclosures: B. Hoshal: None. **C.M. Holmes:** None. **J.M. Salisbury:** None. **M.J. Berry:** None. **O. Marre:** None. **S.E. Palmer:** None.

Poster

P469. Visual System: Response Modulation and Adaptation

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Topic: D.07. Vision

Support: EY028626

Title: Adaptation to synthesized naturalistic textures in macaque V1 and V2

Authors: *A. DAVILA, A. KOHN;
Neurosci., Albert Einstein Col. of Med., Bronx, NY

Abstract: Adaptation alters neuronal responsivity and tuning, in a manner that depends on the cortical area and the stimulus used. However, previous adaptation work has largely used stimuli tailored to each targeted area. This approach has left unclear how adaptation with a given image affects neuronal encoding distributed across different stages of the visual hierarchy. To

understand the hierarchical effects of adaptation, we measured responses to synthetic, naturalistic textures in primary visual cortex (V1) and area V2 of anesthetized macaque monkeys using Neuropixel probes. V1 and V2 neurons have been shown to encode different texture statistics: V1 is sensitive to the spectral content of textures, whereas V2 is sensitive additionally to higher-order texture statistics. We presented 5 different textures and their spectrally-matched noise images, either under control conditions (no adaptation) or after exposure to an adapter (600-ms duration). Adapters were synthesized samples of one of the test textures or its corresponding noise image. Both V1 and V2 showed robust adaptation to textures and noise images. V1 responsivity was reduced more strongly by adaptation than V2 responsivity, for both texture and noise adapters. The reduction in responsivity was broadly shared across test textures and noise images, in both areas. In V1, responses to test stimuli matched to the adapter were not reduced more strongly than responses to novel textures or noise images. In V2, there was only weak evidence of stimulus specificity: responses to textures matched to the adapter were reduced slightly more than responses to novel textures. We also evaluated neuronal population information by training a classifier to discriminate among texture families, based on responses measured before and after adaptation. Adaptation either had little effect on discriminability, or led to slightly worse performance, consistent with its broad tuning. We conclude that adaptation with naturalistic textures induces broadly-tuned response suppression across early stages of the visual system.

Disclosures: A. Davila: None. **A. Kohn:** None.

Poster

P469. Visual System: Response Modulation and Adaptation

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Program #/Poster #: P469.10

Topic: D.07. Vision

Title: Perceptual adaptation to continuous versus intermittent spatial distortions

Authors: *I. R. MCLEAN[1], T. S. MANNING[1], E. A. COOPER[1,2];
[1]Sch. of Optometry, [2]Helen Wills Neurosci. Inst., Univ. of California, Berkeley, Berkeley, CA

Abstract: The visual system adapts to changing environmental demands. Adaptation during continuous exposure to a novel stimulus has been well studied and involves a range of neural mechanisms. However, little is known about how repeated exposure to a stimulus might change the dynamics and mechanisms of adaptation. In a psychophysical experiment, we compared perceptual adaptation in humans who were either continuously or intermittently exposed to an adapting stimulus for the same cumulative amount of time. We measured adaptation to spatial distortions caused by small changes in the retinal image size in one eye. These distortions change the apparent shape and orientation of objects due to altered binocular disparities. Two groups of

participants wore glasses with a 3.8% monocular horizontal magnifier for five hours in one day. Upon initially putting the glasses on, participants experienced distortions in the perceived shape and 3D slant of objects. The continuous group (n = 14, 10 females, mean age = 25 years) wore the glasses continuously for five hours, while the intermittent group (n = 14, 10 females, mean age = 26 years) wore the glasses for five one-hour periods with 20-30 minute breaks in between. To measure adaptation to the shape distortion, participants adjusted two corners of a quadrilateral until it appeared square. To measure adaptation to the slant distortion and changes in the weight given to binocular disparity, participants adjusted the slant of stereoscopic stimuli until they appeared frontoparallel. Stimuli isolating binocular disparity were randomly interleaved with stimuli that contained multiple slant cues. Measurements were taken immediately before and after the glasses were put on, and before and after the glasses were taken off at the end of the day. Adaptation was quantified by the difference in the judgments from the beginning to the end of the day. The magnitude of shape and slant adaptation in the two groups were descriptively similar, suggesting that intermittent exposure did not substantially alter perceptual adaptation. However, our results suggest a difference in the adaptation mechanisms between groups. Specifically, in the slant judgment task, the continuous group showed an increase in the weight given to binocular disparity and the intermittent group did not. These findings suggest that adaptation is present after one day of exposure to spatial distortions and that the number of hours exposed to the distortion may be more important than the continuity. Finally, the visual system may employ different adaptation mechanisms, such as cue weighting strategies, depending on whether an adapting stimulus is present continuously or intermittently.

Disclosures: I.R. McLean: None. **T.S. Manning:** None. **E.A. Cooper:** None.

Poster

P469. Visual System: Response Modulation and Adaptation

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Topic: D.07. Vision

Support: NIH RO1 EY024912
NIH P50 MH103204

Title: Contribution of individual features to repetition suppression in macaque inferotemporal cortex

Authors: *N. P. WILLIAMS, C. R. OLSON;
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Abstract: When an image is presented twice in succession, first as adapter and then as test, neurons in area TE of macaque inferotemporal cortex exhibit repetition suppression, responding

less strongly to the second presentation than to the first. Suppression is known to occur if the adapter and the test image are subtly different from each other. However, it is not known whether cross-suppression occurs between images that are radically different from each other but that share a subset of features. To explore this issue, we measured repetition suppression using colored shapes as stimuli. On interleaved trials, the test image might be identical to the adapter, might share its shape alone, might share its color alone or might differ from it totally. When an image was repeated precisely, the population response was strongly suppressed and also was altered in dynamic, taking the form of a trough during which suppression was especially deep followed by a rebound. This represented an accentuation of the tendency previously described in our laboratory for visual responses in TE to take the form of a damped 5 Hz oscillation. Suppression was of intermediate depth when the images shared only one attribute and was minimal when they shared neither attribute. At the level of the individual neuron, the degree of suppression depended not only on the properties of the two images but also on the preferences of the neuron. Suppression was deeper when the repeated color or shape was preferred by the neuron than when it was not. This effect could arise either from feature-specific adaptation or from adapter-induced fatigue. We favor an interpretation based on feature-specific suppression because neither we nor others have been able to obtain independent evidence for neuronal fatigue. The fact that response suppression occurs even when the test image differs from the adapter in shape or color tends to undercut the notion that suppression is a useful cue for recognition of the test image as recent or familiar. Further undercutting this notion, we have demonstrated that TE neurons, as a population, are unable to distinguish between an image that elicits a relatively weak response because it is intrinsically ineffective and one that does so because it is repeated. On these grounds, we argue that repetition suppression is more akin to classical adaptation rather than to a memory signal.

Disclosures: N.P. Williams: None. **C.R. Olson:** None.

Poster

P469. Visual System: Response Modulation and Adaptation

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Whitehall Foundation 2019-05-44
GSU CBN Next Generation New Scholar

Title: Characterizing the development of sensory prediction error in cortical circuits

Authors: *J. M. ROSS, J. P. HAMM;
Neurosci. Inst., Georgia State Univ., Atlanta, GA

Abstract: In naturally complex environments, organisms are inundated with sensory information. The ability to dynamically process this information to perceive salient changes is crucial to survival. One theoretical framework for explaining this computationally complex task is predictive coding, in which brain networks continuously generate and test predictive models about future sensory information, based on past experience. With simple "oddball" sequences and concurrent recordings in primary sensory cortices, past work shows that neurons exhibit reduced responses to repetitive stimuli (i.e. redundants) known as "stimulus specific adaptation" (SSA); rare stimuli (i.e. deviants) evoke augmented neuronal responses, referred to as "deviance detection" (DD), a putative type of sensory prediction error. While DD is robust in adult mammalian brains, whether and how such prediction error signals emerge postnatally, across adolescence, is unknown. This could hold clues to the underlying neurobiology of predictive coding in cortical networks, as well as its significance for cognitive development.
To assess how DD develops across adolescent development, we employed two-photon calcium imaging of excitatory pyramidal cells (PYRs) in L2/3 of primary visual cortex (V1). We presented visual stimuli (full-field oriented squarewave gratings) to awake mice while imaging neural responses during three different postnatal periods relevant to postnatal brain development: early adolescence (P28), late adolescence (P42), and adulthood (P84). We assessed the stimulus evoked activity of PYRs during two classic visual oddball sequences and a many standards control sequence. This paradigm allows the direct comparison of responses to the same stimuli (e.g. 45° grating) in different contexts - when the stimulus is expected (redundant), when it deviates from expectation (deviant), and when it is neither expected nor deviates from expectations (control). In general, we find that adapted responses to redundant stimuli are present in both sexes (SSA) well before adulthood, but that DD develops and changes over adolescence in V1. Further, this trajectory differs between males and females. Males do not demonstrate DD in V1 until adulthood, whereas females appear to generate the largest DD signals in early adolescence.
Ongoing work is aimed at further characterizing the underlying mechanisms that contribute to differential DD development between males and females, as well as its relevance for learning and behavior. Altogether, this work may inform our understanding of how postnatal brain development and biological sex contribute to aspects of predictive coding, such as SSA and DD.

Disclosures: J.M. Ross: None. **J.P. Hamm:** None.

Poster

P469. Visual System: Response Modulation and Adaptation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P469.13

Topic: D.07. Vision

Title: Divisive normalization is an efficient code for multivariate Pareto-distributed environments

Authors: *S. F. BUCHER[1,2], A. M. BRANDENBURGER[3];
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Abstract: Divisive normalization (DN) is a canonical computation (Carandini & Heeger 2012) reputed to solve the brain's problem of representing potentially unbounded stimuli via bounded firing rates. Originally discovered in the primary visual cortex, DN has since been observed in many other sensory domains as well as in decision-making. It is often considered to be an implementation of the ***efficient coding principle***, as it has been shown empirically to reduce redundancy in natural stimulus statistics (Schwartz & Simoncelli 2001). To date, however, the sense in which divisive normalization is efficient has not been made analytically precise. We close this gap with a theoretical result that states the conditions under which divisive normalization is an optimal encoding system, and specify a class of input distributions that DN encodes efficiently. Our approach differs from previous ones in that we ask what class of input distributions DN represents efficiently, rather than analyzing the properties of the DN computation for a given empirical input distribution. We show that in a low-noise environment DN maximizes the mutual information between an *n*-dimensional stimulus and its representation if and only if the stimulus' prevalence in the environment is described by a multivariate Pareto distribution. Divisive normalization transforms (only) this particular distribution into an entropy-maximizing distribution, which amounts to a multivariate analog of histogram equalization. We extend this result to allow for an arbitrary metabolic cost of the representation, and show how this impacts the optimally encoded distributions. Our result suggests that divisive normalization may have evolved to efficiently represent stimuli with Pareto distributions, consistent with empirical observations on naturalistic stimulus distributions such as the conditional variance dependence of filter responses to natural images. Beyond formalizing the sense in which this important computation adheres to the efficient coding principle, our theoretical result also yields empirically testable predictions across sensory domains on how the divisive normalization parameters should be tuned to features of the input distribution.

Disclosures: S.F. Bucher: None. **A.M. Brandenburger:** None.

Poster

P500. Cross-Modal Processing I

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Program #/Poster #: P500.01

Topic: D.09. Multisensory Integration

Support: NSERC, Individual Discovery Grant

Title: Haptic cue combination for size discrimination

Authors: *K. ALLEN, D. GOLDREICH;
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Abstract: Our senses provide noisy inputs (cues) which we interpret to perceive the world. Many studies have investigated multisensory cue combination: the integration of cues that arise through different senses. Fewer studies have investigated cue combination within a single modality. The haptic modality has been particularly under-investigated in this regard. Here, we investigated whether haptic cutaneous and hand configuration cues are combined optimally to discriminate between coin-sized discs of different sizes. When the hand is open such that the thumb and index fingers span the diameter of a disc to contact its perimeter, cutaneous cues occur from the indentation of the skin caused by the curvature of the disc (smaller discs cause greater indentation). Simultaneously, the hand configuration cue (proprioceptive), provides an additional cue to size. We considered three hypotheses for how humans may use these cues: humans rely solely on the least noisy cue (Winner-Take-All Model), humans combine cues based on a simple average (Average Model), or humans combine cues via an optimal weighted average (Optimal Model). Each experiment tested participants using a two-interval forced-choice (2IFC) paradigm with 3D printed disc stimuli. On each trial, under occluded vision, participants felt two discs sequentially and responded which felt larger. Participants were tested under four counterbalanced conditions to test performance with each finger's cutaneous cue alone, the hand configuration cue without the cutaneous cue, and all cues together. We applied Bayesian parameter estimation to fit cumulative normal psychometric functions to these data and Bayesian model comparison to determine a winning model for each participant. Results from Experiment 1 (12 participants) were ambiguous due to the poor reliability of the cutaneous cues. A new set of discs was 3D printed to enhance the cutaneous cues and used for Experiment 2 (11 participants). Results indicated optimal cue combination for 9 of the 11 participants. We conclude that humans may combine haptic cutaneous and hand configuration cues optimally to judge the size of held objects.

Disclosures: K. Allen: None. **D. Goldreich:** None.

Poster

P500. Cross-Modal Processing I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P500.02

Topic: D.09. Multisensory Integration

Support: Swiss National Science Foundation (project grant 310030B-170269)

European Research Council (ERC Advanced Grant BRAINCOMPATH, project 670757)

Title: Multisensory representation of object location in mouse barrel cortex and posterior parietal cortex

Authors: *A. HOFFMANN[1,2], F. HELMCHEN[1,2];
[1]Brain Res. Institute, Univ. of Zurich, Zurich, Switzerland; [2]Neurosci. Ctr. Zurich, Zurich, Switzerland

Abstract: For a stable and coherent perception of the world, the brain combines sensory information from different modalities using distinct reference frames. In the mouse brain, tactile input is organized somatotopically in the primary whisker somatosensory cortex (wS1) while the primary visual cortex (V1) contains a retinotopic map of the visual field. For nearby objects, these two sensory streams likely converge in the rostro-lateral (RL) area of the posterior parietal cortex (PPC). However, how the integration of multisensory information happens for naturalistic stimuli on a single-cell level remains unclear. Here, we investigated how neurons across mouse wS1, V1, and RL integrate visuotactile information about a pole that is in reach of the whiskers and can be seen and/or felt. Using two-photon calcium imaging, we recorded several 10,000 excitatory neurons expressing GCaMP6f in L2/3 of head-fixed mice (n=5, male triple transgenic Rasgrf2-2A-dCre;CamK2a-tTA;TITL-GCaMP6f mice) that were free to run on a disk. We presented a pole that was moved to different locations in reach of the whiskers, either under nearly dark conditions or during illumination of the pole. We recorded the interactions of the whiskers (only C-row) with the pole with a high-speed camera and tracked their kinematics offline with the deep learning pose estimation algorithm DeepLabCut. For reproducible and flexible data analysis, we organized the analysis workflow as a DataJoint pipeline and created interactive user interfaces to explore the raw and processed data. We find that around one third of neurons across wS1, V1 and RL are significantly more active when the pole is in reach of the whiskers, whereas around 15% of neurons reduce their activity. A higher fraction of neurons in V1 and RL were more modulated by light (significantly more active) compared to wS1 (~20% vs. ~5%). By comparing average activities at different pole locations, we find that subsets of neurons in RL and wS1 show selectivity for specific locations in the near space based on tactile and/or visual input. To test whether location selectivity was a result of altered whisker kinematics, we predicted neural activity using Generalized Linear Models (GLMs) based on different sets of regressors. We find that object locations predict neural activity in RL better than whisker kinematics, whereas the opposite holds for wS1. Together, these findings corroborate the notion of RL representing object locations based on visual and tactile information, potentially in a more modality-invariant manner.

Disclosures: A. Hoffmann: None. **F. Helmchen:** None.

Poster

P500. Cross-Modal Processing I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P500.03

Topic: D.09. Multisensory Integration

Title: Neural computations underlying tactile texture constancy

Authors: *A. F. TORTOLANI, E. OKOROKOVA, R. NGACHOKO, S. J. BENSMAIA; Univ. of Chicago, Chicago, IL

Abstract: A major function of sensory processing is to achieve neural representations of objects that are stable across changes in context and perspective. Small changes in exploratory behavior can lead to large changes in the neural signals at the sensory periphery, thus resulting in ambiguous neural representations of objects. Overcoming this ambiguity is a hallmark of human object recognition across sensory modalities. Our ability to achieve a stable perception of texture regardless of the way we explore a surface constitutes an instance of this process. Indeed, the responses of tactile nerve fibers to textured surfaces depend critically on the speed at which the surface moves across the skin. In contrast, texture responses of neurons in somatosensory cortex are less susceptible to changes in scanning speed and the resulting representation of texture is far more stable across changes in speed. The resulting texture percept is highly independent of scanning speed. The nature of the transformation from periphery to cortex that gives rise to texture constancy is not entirely understood. One hypothesis is that two computations -- spatial and temporal differentiation --, which have been shown to be implemented along the somatosensory neuraxis, are sufficient to give rise to a speed-invariant texture representation. To test this hypothesis, we simulated the responses of all the nerve fibers that innervate the glabrous skin of the hand to a large set of natural and artificial textures scanned across the fingertip using exploratory trajectories measured in human subjects. First, we found that the (stimulated) population response in the nerve provides ambiguous information about texture. Next, we simulated cortical responses by applying a set of temporal and spatial filters -- inspired by the known response properties of cortical neurons -- to the simulated afferent input. We then assessed the degree to which the simulated cortical responses yielded a texture representation that was more robust to changes in scanning speed than was its peripheral counterpart. We found that filters improved texture classification performance when textures were scanned with variable speed and force, demonstrating that temporal and spatial differentiation of peripheral input is sufficient to give rise to an invariant representation of texture.

Disclosures: A.F. Tortolani: None. **E. Okorokova:** None. **R. Ngachoko:** None. **S.J. Bensmaia:** None.

Poster

P500. Cross-Modal Processing I

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Program #/Poster #: P500.04

Topic: D.09. Multisensory Integration

Support: Research Training Group 2416 MultiSenses – MultiScales, RWTH Aachen University

Title: Neocortical activity of mice performing a visuotactile accumulation of evidence task

Authors: *G. NABBEFELD[1,2], A. OSTENRATH[1], S. GRAFF[1,2,3], S. MUSALL[1,2,3], B. KAMPA[1,2,4];
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Abstract: Much effort has been focused on understanding how the brain processes the information from individual senses. Yet, the neural mechanisms that allow the effortless integration of unisensory inputs into multisensory objects are less well understood. To study how neural circuits integrate visual and tactile information, we developed a multisensory discrimination task for head-fixed mice. Here, two sequences of visual, tactile or visuotactile stimuli are presented on both sides of the mouse which has to respond to the higher-rate target side to obtain a water reward. To ensure integration of sensory information over the entire stimulus period, a short delay was added before the response. Mice achieved high accuracy in all conditions, with improved performance in the multisensory condition. In line with the principle of inverse effectiveness, improvements were highest in the most difficult trials. We then used widefield imaging to measure cortexwide activity in transgenic mice, expressing the Ca^{2+}-indicator GCaMP6s in all excitatory neurons. Multisensory stimuli evoked higher neuronal activity compared to unisensory stimulation. This was most evident in the rostrolateral association area RL and parts of medial frontal cortex (mFC), which reliably responded to both visual and tactile stimulation. These responses, as well as activity in the primary visual (V1) and somatosensory barrel cortex (S1), evolved over the course of learning and increased in magnitude and the duration of sensory responses. To better isolate multi- from unisensory responses we then used a linear encoding model. Including a multisensory interaction term explained significantly more information as with unisensory regressors alone, especially in parietal and frontal areas. This suggests that cortical multisensory responses are not easily explained as a linear summation of unisensory responses.To causally test the relation between cortical activity and perceptual decisions, we then used the inhibitory opsin stGtACR2 to silence excitatory neurons in either V1, S1, RL and mFC at different times during the task. Inhibiting V1 or S1 strongly impaired task performance during the stimulus while inactivating mFC had the strongest effect during the delay period. Surprisingly, inactivating RL had a minor impact on performance, despite its robust multisensory activation.Our results demonstrate that multisensory stimulation leads to improved task performance and widespread cortical activation in mice. However, only sensory and frontal cortices were causally required, raising the question of the function of multisensory activity in expendable areas such as area RL.

Disclosures: G. Nabbefeld: None. **A. Ostenrath:** None. **S. Graff:** None. **S. Musall:** None. **B. Kampa:** None.

Poster

P500. Cross-Modal Processing I

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Program #/Poster #: P500.05

Topic: D.09. Multisensory Integration

Support: H2020, Grant/Award Number: 945539

Title: Sensory modality modulates neural coding ofresponse side:comparison of visual and somatosensory cortices, perirhinal cortex andhippocampus

Authors: *J. FIORILLI, P. MARCHESI, T. R. RUIKES, G. HUIS IN ‘T VELD, M. QUINTERO, R. BUCKTON, C. M. PENNARTZ;
Univ. of Amsterdam, Amsterdam, Netherlands

Abstract: Studies on rodent hippocampus present evidence for an increasing level of invariance in object and spatial representations along the sensory-cortical to hippocampal hierarchy. However, neural coding of object recognition and motor variation effects has not been compared directly between cortical and hippocampal areas. We conducted simultaneous ensemble recordings in primary somatosensory cortex (S1BF, N units= 119), secondary visual cortex (V2L, N units = 366), perirhinal cortex (PRH, N units = 210), and the hippocampal subfields (CA1, CA3, DG, N units = 221) from 4 rats that performed an object discrimination task using vision, touch, or both senses combined. This enabled us to quantify both the sensory evoked (tactile and visual) responses during active sampling, as well as illumination-invariant coding during object sampling and self-motion for individual cells.
We first explored the relative prominence of object- and motor-related neural coding when ascending the cortico-hippocampal hierarchy. All four areas showed a predominant coding of the response side, with surprisingly little evidence for coding of object-specific information. Whereas V2L, PRH and CA1 contained information on the response side starting from self-motion initiation, PRH choice-side encoding peaked around the nose-poke ports. Secondly, we asked how the neural coding of the response side is modulated by illumination, and if neural representations become more independent to photic changes when ascending the hierarchy. We found light-dark modulation of response side in all four areas, but with a stronger independence in area CA1 as compared to V2L and S1BF. Surprisingly, despite the illumination invariant coding of the response side in CA1, a significant proportion of cells in this area was modulated by the light onset. Different from the three other areas, PRH neurons revealed a sparse but highly selective coding of reward information that was specific to the combination of object and response side.

Altogether, under naturalistic movement conditions we demonstrate a dominant neural coding of spatial motor choice in sensory cortical and hippocampal areas, with increasingly light independent response side coding when ascending the hierarchy. The perirhinal cortex forms a rather exceptional entity, selectively and sparsely coding information on reward contingencies.

Disclosures: J. Fiorilli: None. **P. Marchesi:** None. **T.R. Ruikes:** None. **G. Huis in 't Veld:** None. **M. Quintero:** None. **R. Buckton:** None. **C.M. Pennartz:** None.

Poster

P500. Cross-Modal Processing I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P500.06

Topic: D.09. Multisensory Integration

Title: The impacts of audiovisual spatial congruency on pupil dilation

Authors: *Z. PARK, H. LEE, C.-Y. KIM;
Psychology, Korea Univ., Seoul, Korea, Republic of

Abstract: Pupillometry is a reliable measurement for investigating bisensory integration. Recent studies have reported faster and greater pupil dilation for audiovisual conditions compared to auditory and visual conditions (Rigato et al., 2016; Wang et al., 2017). However, little is known about the impacts of bimodal stimulus congruency on pupil dilation. As spatio-temporal congruency is a substantial factor in modulating the extent of bisensory integration, we investigated whether spatial congruency of audiovisual stimuli influences pupil dilation by adopting the ventriloquist illusion paradigm. Participants maintained fixation on a small dot at the center of a computer monitor with their heads fixed on a chin rest. On each trial, an auditory or visual stimulus was presented alone as A or V conditions, or a pair of auditory and visual stimuli were presented simultaneously (AV condition). A simple beep sound and a small dot were respectively used for the A and V stimuli, presented for 100ms following 1-1.5s of fixation. We manipulated spatial congruency of audiovisual stimuli by presenting the aforementioned auditory stimulus from the left, right, or center of each participant accompanied by a visual stimulus on the left or right. Here, we defined an AV condition as "spatially incongruent" (SI) if the visual and auditory stimuli were presented on different sides, while "spatially congruent" (SC) AV conditions featured collocated stimuli. Participants' eye responses were recorded using an eye tracker (Eyelink-1000, SR Research) for 2000ms following the onset of the stimuli. Afterward, participants performed a sound localization task by clicking their perceived location of the auditory stimulus. Behavioral results exhibited the ventriloquist effect (VE), which is the degree of bias towards a visual stimulus, for the SI condition. For the pupillometry results, transient pupil dilation was evoked for conditions featuring stimuli compared to those that did not, and greater pupil dilation was observed for the AV condition rather than the A and V

conditions, which replicates previous studies. In addition, we found that pupil dilation for the SI condition was greater than that for the SC condition: this effect remained persistent throughout the entire 2000ms, but was statistically significant for 950ms to 1200ms after the onset of the stimuli. These results indicate that spatial congruency modulates pupil dilation reflecting bisensory integration. We are now attempting to extend our current findings into another important eye measurement, microsaccades.

Disclosures: Z. Park: None. **H. Lee:** None. **C. Kim:** None.

Poster

P500. Cross-Modal Processing I

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Program #/Poster #: P500.07

Topic: D.09. Multisensory Integration

Support: DFG 233886668 / GRK1960
Studienstiftung des deutschen Volkes

Title: Multimodal Proprioceptive Integration in the Sensorimotor Network of an Insect Leg

Authors: *C. GEBEHART, A. BÜSCHGES;
Univ. of Cologne, Cologne, Germany

Abstract: To generate the internal representation of the limbs that is an essential aspect of stable motor control, an animal's nervous system integrates proprioceptive sensory input from multiple sense organs. Distinct proprioceptive modalities are monitored by separate sense organs. In insects, campaniform sensilla (CS) and the femoral chordotonal organ (fCO) provide information about leg loading and joint movement. Both sensory modalities are able to alter motor output, and must therefore converge along the sensorimotor pathways. Understanding the network computations and architecture underlying proprioceptive signal convergence thus requires knowledge about spatial and temporal summation of multimodal signals and their interaction. We combined the mechanical stimulation of trochanterofemoral and tibial CS (tr/fCS, tiCS) and the fCO with intra- and extracellular electrophysiological recordings of sensory afferents, premotor nonspiking interneurons (NSI), and motor neurons (MN) to analyze proprioceptive signal integration in the stick insect leg. Spatial summation - Load signals from tr/fCS and tiCS and movement feedback from the fCO was distributed into one common network of premotor NSIs. Signal processing for both sensory modalities was antagonistic, i.e. each individual NSI's responses could support or oppose the ensuing motor output. Temporal summation - Movement signal transmission along sensorimotor pathways was faster than load signaling, resulting in temporally shifted effects on motor output and muscle force. Using signal timing to establish network connectivity, we demonstrate that functional connectivity between NSIs was determined

by temporal aspects, i.e. to which NSI the sensory signal was transmitted first. Signal integration - During combined load and movement stimuli, fCO afferents were presynaptically inhibited by tiCS afferents. In postsynaptic NSIs, the interaction between both modalities led to nonlinear summation of sensory signals. The gain of NSIs opposing the resistance reflex of extensor tibiae (ExtTi) MNs increased. Consistently, the gain of the slow ExtTi MN to movement stimuli decreased in the presence of load, an effect that was abolished when presynaptic inhibition was blocked. In summary, load and movement were spatially summed by premotor NSIs, while being separated in time. The temporal delay of load signals predestines them for a role in tuning ongoing movement processing, which we verified by their effect on movement signal gain. Taken together, these results provide a framework for multimodal proprioceptive integration in which load feedback contextualizes movement signal processing.

Disclosures: C. Gebehart: None. **A. Büschges:** None.

Poster

P500. Cross-Modal Processing I

Location: Virtual Only

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Program #/Poster #: P500.08

Topic: D.09. Multisensory Integration

Support: DFG Grant SPP 2205

Title: Neural representations in larval zebrafish navigating different sensory-modality gradients

Authors: *V. PALIERI[1], E. PAOLI[1], I. C. GRUNWALD KADOW[2], R. PORTUGUES[1]; [1]TUM Sch. of Med., Technische Univ. München, Munich, Germany; [2]Sch. of Life Sci., Tech. Univ. of Munich, Freising, Germany

Abstract: Animal survival depends on the ability to find environments where chances of survival are maximized. Achieving this goal often requires navigating spatial gradients successfully. For example, animals can use odor gradients to locate food and they can exploit temperature gradients for thermoregulation. It has been shown, in larval zebrafish, *C. elegans* and *D. melanogaster*, that a temporal sampling strategy is used to probe the stimulus landscape and to detect the rate of change of stimulus intensity. As a consequence, animals increase their turning as they either move away from a favorable condition or come closer to a negative one. The majority of existing studies on gradient navigation focus on single sensory modalities to show how sensory neurons detect stimulus characteristics or how the behavioral output is modulated. However, an understanding of how sensory and motor components are linked in the brain and whether the representation of the rate of change of different sensory modalities converges onto the same circuit is still missing. In the present study, we took advantage of the optical accessibility and the well-defined behavioral repertoire of larval zebrafish to investigate

how and where the temporal derivative of stimulus intensity across different modalities is processed in the fish brain. We first developed an open-loop virtual gradient navigation assay in head-restrained larvae used in combination with lightsheet microscopy for whole-brain calcium imaging. Then, we used three different aversive stimuli while recording fish behavior to screen for areas representing the change of stimulus irrespective of its identity. These representations may convey information about improving or worsening conditions and could be a hub for the collection of relevant stimulus information used by the animal to successfully perform gradient navigation.

Disclosures: V. Palieri: None. **E. Paoli:** None. **I.C. Grunwald Kadow:** None. **R. Portugues:** None.

Poster

P502. Cross-Modal Processing III

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Program #/Poster #: P502.01

Topic: D.09. Multisensory Integration

Support: EY031532
NS073553

Title: The Behavioral Benefits of Multisensory Integration Require Multisensory Experience

Authors: *N. BEAN, S. SMYRE, B. STEIN, B. ROWLAND;
Neurobio. and Anat., Wake Forest Sch. of Med., Winston Salem, NC

Abstract: Concordant visual-auditory stimuli enhance the responses of superior colliculus (SC) neurons and the detection and localization functions they mediate. However, this capacity for multisensory integration is not an innate capability. Animals reared in omnidirectional noise ("noise-rearing") lack the needed visual-auditory experience required to instantiate this physiological capability. Here we tested the presumption that the multisensory enhancements in detection and localization in such animals would also be compromised. A cohort of cats reared in omnidirectional noise (n=3) and a cohort (n=2) of normally-reared animals were trained to detect, localize, and approach brief visual cues (50 ms) presented randomly at any of 8 locations (±60◦, 15◦ increments, excluding 0◦). They were also trained to remain at the start location ("No-Go") for stimulus absent ("catch") trials. After training, stimulus intensities were lowered so that performance did not exceed 25-40% correct approaches to the visual targets. Target stimuli were presented at random locations and consisted of either a visual stimulus, a clearly audible (but novel) auditory stimulus, or their spatiotemporally combination. The performance of normally-reared animals was enhanced by the cross-modal stimulus combination. They approached it more reliably (63±1.4%) than the visual target alone (38±1.2%). In contrast, noise-reared animals

showed no greater ability to respond to the visual-auditory stimulus (35±1.3%) than the visual stimulus alone (33±1.0%). Yet, they had no severe hearing deficits, and a second series of tests thereafter confirmed that even when trained that both the visual and auditory stimuli were targets, they failed to show enhanced performance to their combination. Normally-reared animals performance on multisensory trials far exceeded that to the best unisensory performance selected in a statistical facilitation model (80±1.1% vs 59.±4.6%); whereas the multisensory performance of noise-reared animals were impaired relative to statistical facilitation 47±1.3 vs 59±3.9%. Error analysis revealed defects in both detection and localization in the noise-reared cohort in both conditions. The data suggest that simultaneous activation of visual and auditory neural ensembles in noise-reared animals does not translate into better performance in these simple behavioral tests. Their behavioral defects resemble those noted physiologically in SC neurons and are consistent with a model in which these neurons are primary decoders for cross-modal information. Supported by NIH grants EY031532 and NS073553

Disclosures: N. Bean: None. **S. Smyre:** None. **B. Stein:** None. **B. Rowland:** None.

Poster

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Program #/Poster #: P502.02

Topic: D.09. Multisensory Integration

Support: NIH/REI RO1 EY031532
T32 NS73553

Title: Visual and auditory predictions independently affect superior colliculus responses

Authors: *S. A. SMYRE, N. L. BEAN, B. E. STEIN, B. A. ROWLAND;
Neurobio. & Anat., Wake Forest Baptist Hlth., Winston-Salem, NC

Abstract: The brain possesses multiple senses that transduce different energies, a feature that increases the amount of information available for decision-making and enhances adaptability in a greater variety of environments. Multisensory circuits like the superior colliculus (SC) simultaneously process signals derived from multiple senses, and cross-modal (e.g., visual-auditory) signals aligned in space and time are integrated into novel neural products. SC neurons use sensory context to enhance their efficiency: repeated stimulus presentations generate predictive signals that can either potentiate or attenuate (habituate) future responses depending on the presentation rate and sensitivities of individual neurons. The present study sought to better understand the origin of these predictive signals. The first objective was to determine whether they were modality-specific or cross-modal. SC neurons (n=50) of anesthetized cats were tested with stimulus trains containing three repetitions of either a visual (50ms flash) or auditory (50ms

broadband burst) stimulus followed by a fourth stimulus of either the same or the opposite modality presented at the same location at the same temporal interval. A presentation rate (2 Hz) was selected in which almost all (92%) of the neurons showed robust, significant attenuation in the auditory modality, but either potentiation or attenuation (46%/34% respectively, 20% no change) in the visual modality. We found that the predictive signals for almost all neurons (90%) failed to cross sensory modalities, suggesting that they were modality-specific. The second objective was to determine if the predictions were generated and applied to each modality independent of the other modalities in the immediate context, or if they would be different in multisensory vs. unisensory contexts. The same neurons were tested with stimulus trains containing three repetitions of a modality-specific (visual or auditory) or cross-modal (visual-auditory) stimulus followed by a fourth stimulus of the same or the opposite configuration. The cross-modal stimulus configuration reliably elicited enhance neural responses. The influence of the predictions crossed between modality-specific and cross-modal configurations, suggesting the predictions for each modality were generated and applied independently. Collectively, these findings are consistent with a model in which predictive modality-specific signals independently alter the modality-specific inputs to the SC, which are rapidly integrated by multisensory neurons into enhanced neural products. Supported by NIH/REI RO1 EY031532 and T32 NS73553.

Disclosures: S.A. Smyre: None. **N.L. Bean:** None. **B.E. Stein:** None. **B.A. Rowland:** None.

Poster

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Topic: D.09. Multisensory Integration

Support: NIH Grant R01NS081071
OHSU LaCroute Fellowship

Title: Input and output specific microcircuits of the insular cortex

Authors: *Y. CHEN, B. C. JONGBLOETS, M. A. MUNIAK, I. GINGERICH, T. MAO; Vollum Inst., Oregon Hlth. & Sci. Univ., Portland, OR

Abstract: The insular cortex (IC) is an information-integration hub that mediates diverse functions such as taste perception, decision making, and aversive state processing. While great progress has been made in the field using in vivo optogenetics, pharmacology, and calcium imaging to establish diverse functional roles of the anterior and posterior subregions of the IC, their underlying circuit mechanisms remain elusive. To better understand how local signal processing within the IC contributes to its diverse functions, our study aimed to decipher IC microcircuits that are associated with specific input sources and output targets. To

comprehensively identify local inputs to IC output neurons, we utilized laser scanning photostimulation (LSPS) of caged glutamate, coupled with patch-clamp recordings of neurons labeled with retrograde bead injections in either the basolateral or central amygdala (BLA, CEA), mediodorsal (MD) or perifascicular (PF) thalamus, or the contralateral IC, five of the known major projection targets of the IC. We found that IC output neurons exhibit stereotypical distribution patterns across cortical depths. Also, their local input patterns—often differing from the canonical microcircuits observed in primary cortices—are unique based on their projection target. Further, by recording postsynaptic responses to optogenetically stimulated long-range axons, we showed that long-range inputs to the IC that originate from different thalamic nuclei preferentially target distinct subsets of IC neurons, and that local circuits are required to connect these input-receiving neurons with output-projection neurons both across cortical layers and along the anterior-posterior axis of the IC. Notably, we uncovered an information pathway from the posterior to anterior IC. Although the two subregions have been reported to mediate distinct functions, our results suggest that they may be synergistically recruited during certain behaviors, thereby providing a new perspective on how functional diversity within the IC can be achieved. Together, our findings show that local circuits unique to the insular cortex are specifically tailored to integrate unique neuronal signals for different output pathways. Our work also elucidates the neuronal substrates responsible for information-integration within the IC, and sets the foundation for future studies on input- and output-specific insular functions.

Disclosures: Y. Chen: None. **B.C. Jongbloets:** None. **M.A. Muniak:** None. **I. Gingerich:** None. **T. Mao:** None.

Poster

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Topic: D.09. Multisensory Integration

Support: R01 NS081071

Title: Origin-specific dendritic integration of long-range cortical inputs to agranular insular pyramidal neurons

Authors: *I. GINGERICH, M. A. MUNIAK, Y. CHEN, B. C. JONGBLOETS, T. MAO; Vollum Institute/Oregon Hlth. & Sci. Univ., Portland, OR

Abstract: Origin-specific dendritic integration of long-range cortical inputs to agranular insular pyramidal neurons

Ian K Gingerich, Michael A Muniak, Yang Chen, Bart C Jongbloets, Tianyi Mao Vollum Institute, Oregon Health & Science University, Portland, OR 97239 USA

The insular cortex receives inputs from many cortical and subcortical structures and integrates these inputs to mediate diverse functions. These include proprioception, visceral sensation, taste perception, and aversive state processing. The agranular insula (AI) receives inputs from cortical areas associated with disparate sensory processes, including regions associated with olfactory, taste, and proprioceptive processing, as well as higher order cortical functions including memory and navigation. However, how these inputs integrate in the AI and facilitate AI function remain elusive. Here, we used subcellular ChannelRhodopsin-2 Assisted Circuit Mapping (sCRACM) to investigate the functional connectivity and the subcellular organization of these input patterns to the AI. sCRACM allows us to map the spatial distribution of synaptic inputs to the dendritic arbors of recorded neurons by expressing ChR2 in presynaptic neurons. Presynaptic axon terminals are excited via laser patterns, triggering local neurotransmitter release which, in combination with extracellular application of Na^+- and K^+- channel blockers, results in robust excitatory postsynaptic currents ($EPSC_{sCRACM}$). This technique is well suited to the investigation of long-range inputs, in which the presynaptic somata may reside in a different brain region than that of the recorded neuron.
To investigate cortical input organization, we systematically injected ChR2 in the known cortical regions that send projections to the AI. We performed sCRACM experiments by recording from pyramidal cells located in all layers of the AI. Using cluster analysis and pattern recognition algorithms, we identified insular region-specific, layer-specific, and dendritic-compartment-specific input patterns. In conjunction with morphologic analysis of the recorded pyramidal neurons, we conclude that the unique patterns associated with each input source is one of the ways by which the insular cortex may mediate its diverse functions within the same anatomical region.

Disclosures: I. Gingerich: None. **M.A. Muniak:** None. **Y. Chen:** None. **B.C. Jongbloets:** None. **T. Mao:** None.

Poster

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Topic: D.09. Multisensory Integration

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CAS Pioneer Hundreds of Talents Program
Shanghai Municipal Science
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Shanghai Key Basic Research Project 16JC14202001

Title: Neural implementation of causal inference during multisensory processing in the Macaque brain

Authors: *G. QI, W. FANG, S. LI, J. LI, L. WANG;
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Abstract: Natural perception relies inherently on inferring causal structure in the environment. However, the neural mechanisms and functional circuits that are essential for representing and updating the hidden causal structure during multisensory processing are unknown. To address this, monkeys were trained to infer the probability of a potential common source from visual and proprioceptive signals on the basis of their spatial disparity in a virtual reality system. The proprioceptive drift reported by monkeys demonstrated that they combined historical information and current multisensory signals to estimate the hidden common source and subsequently updated both the causal structure and sensory representation. Single-unit recordings in premotor and parietal cortices revealed that neural activity in premotor cortex represents the core computation of causal inference, characterizing the estimation and update of the likelihood of integrating multiple sensory inputs at a trial-by-trial level. In response to signals from premotor cortex, neural activity in parietal cortex also represents the causal structure and further dynamically updates the sensory representation to maintain consistency with the causal inference structure. Thus, our results indicate how premotor cortex integrates historical information and sensory inputs to infer hidden variables and selectively updates sensory representations in parietal cortex to support behavior. This dynamic loop of frontal-parietal interactions in the causal inference framework may provide the neural mechanism to answer long-standing questions regarding how neural circuits represent hidden structures for body-awareness and agency.

Disclosures: G. Qi: None. **W. Fang:** None. **S. Li:** None. **J. Li:** None. **L. Wang:** None.

Poster

P502. Cross-Modal Processing III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P502.06

Topic: D.09. Multisensory Integration

Support: European Research Council (ERC) under the European Union's Horizon 2020 research and innovation program (FeelAgain grant agreement No. 759998)

Title: Population analysis approach reveals interference of sensory modalities in spinal cord

Authors: *N. KATIC[1], J. M. BALAGUER[2], S. CONTI[3], O. GORSKY[4], N. PAVLOVA[4], P. MUSIENKO[4], S. RASPOPOVIC[5], M. CAPOGROSSO[2];
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Abstract: Restoration of somatic sensations from the missing limb of amputees through sensory neuroprosthetics improves the functionality and embodiment of prosthetic devices. Yet, while it is possible to generate touch percepts, proprioception restoration remains elusive. Theoretically, large-diameter proprioceptive afferents should have the lowest threshold for electrical stimulation of the peripheral nerves and should thus be the easiest sensory afferents to recruit with neural interfaces. Why, after decades of research in independent clinical trials, reports of controlled proprioceptive sensations have been so scarce? We hypothesized that sensory regulation mechanisms, such as presynaptic inhibition, may play a major role during non-selective recruitment of afferents classes. To unveil these mechanisms, we recorded signals in the gray matter of the spinal cord, while simultaneously stimulating muscle and cutaneous branches of the radial nerve. Multi-channel spinal neural signals are projected into a low-dimensional neural space. Proprioceptive inputs elicited robust trajectories and showed that processed proprioceptive information was reduced when the cutaneous nerve was concurrently stimulated. Our results suggest that interference of sensory modalities may pose an electrophysiological limit to sensory restoration and set a base for overcoming the current and defining the future neurotechnology challenges.

Disclosures: N. Katic: None. **J.M. Balaguer:** None. **S. Conti:** None. **O. Gorsky:** None. **N. Pavlova:** None. **P. Musienko:** None. **S. Raspopovic:** None. **M. Capogrosso:** None.

Poster

P502. Cross-Modal Processing III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P502.07

Topic: D.09. Multisensory Integration

Support: R35

Title: Neuronal responses of different types of neurons are modulated by arousal state in mouse sensory cortices

Authors: *S. JO[1], D. MCCORMICK[2];
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Abstract: Identifying meaningful sensory stimuli is required to survive, and the ability to detect sensory stimuli is significantly affected by ongoing brain activities. Recent works have shown that spontaneous brain activities were various in different brain areas. However, these studies used 1-photon widefield calcium imaging to observe brain-wide activities, or small parts of the brain were examined in 2-photon calcium imaging or electrophysiology. Therefore, it is important to understand how different types of neurons in different sensory cortices are related to arousal state. For a better understanding of neural network dynamics by arousal state, we performed calcium imaging of excitatory neurons (EXC), as well as vasoactive intestinal peptide (VIP), somatostatin (SST), and parvalbumin (PV) interneurons in three sensory cortices, auditory, visual, and somatosensory cortex, using a 2-photon mesoscope. We crossed VIP, SST, and PV mouse CRE lines with reporter line Ai162 which is expressing GCamp6s. To define each sensory cortex, we performed 1-photon widefield calcium imaging while tone clouds, visual stimuli, or whisker deflections were delivered to a mouse. To measure arousal state, running speed, whisker motion energy, and pupil size were recorded. We also delivered tone clouds as auditory stimuli in a subset of VIP and SST imaging sessions. Different types of neurons showed various response directionality to arousal state in different sensory cortices. In the visual cortex, all types of neurons, VIP, SST, PV, and EXC were activated with whisking and running. However, in the auditory cortex, the majority of VIP neurons were inactivated and SST neurons were activated, and PV and EXC neurons showed more heterogeneous responses to the arousal states. In the somatosensory cortex, VIP neurons showed heterogeneity with whisking, but were inactivated with running. SST, PV, and EXC neuronal activities were activated with both whisking and running. VIP was inactivated, but SST was activated by tone clouds during stillness, whisking, and running. In a subset of auditory stimuli sessions of VIP mouse, narrower band tone clouds were presented, and VIP neurons were activated, which is the opposite to the responses to the broader band tone clouds. Our results show that different types of neurons in different areas show different relationships to arousal, and their responses to sensory stimuli could be different to the characteristics of the stimuli. Our research contributes to a better understanding of neural network dynamics by revealing characteristics of four different types of neurons in three sensory cortices.

Disclosures: S. Jo: None. **D. McCormick:** None.

Poster

P502. Cross-Modal Processing III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P502.08

Topic: D.09. Multisensory Integration

Support: NIH F31

Title: Effects of experience on multisensory processing in primary somatosensory cortex

Authors: *D. KATO[1], R. M. BRUNO[2];
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Abstract: Several studies over the past few years have found that activity even in primary sensory cortical areas is modulated by multiple sensory modalities. However, the functional and computational roles of these responses remain unclear. To test whether these cross-sensory effects encode stimulus-specific information, we measured responses to different auditory stimuli in primary somatosensory cortex (S1) of C57 mice using in vivo 2-photon calcium imaging. We found that very few cells were preferentially driven by one auditory stimulus over the other, and a linear decoder could not distinguish between population responses to different auditory stimuli once the effect of whisking had been taken into account. Moreover, we found very few cells whose responses to tactile stimuli were specifically modulated by concurrent presentation of one tone versus another, and a linear decoder was unable to distinguish between different conjunctive audio-tactile stimuli. Repeatedly pairing one auditory stimulus but not the other with a tactile stimulus did not seem to have any effect on decoder performance, nor did pairing reward with the presentation of conjunctive audio-tactile stimuli. However, we also found attenuated auditory brainstem responses in these mice, suggesting that these results may be at least in part due to a general hearing deficit. We are currently repeating these experiments in CBA mice, which do not exhibit the same hearing deficits.

Disclosures: D. Kato: None. **R.M. Bruno:** None.

Poster

P502. Cross-Modal Processing III

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Program #/Poster #: P502.09

Topic: D.09. Multisensory Integration

Support: NIH BRAIN Initiative Diversity Supplement Fellowship 3R01NS110060-03S1

Title: Multisensory integration and social behavior in Drosophila

Authors: *E. NORMAND, D. DEUTSCH, T. D. PEREIRA, N. RANGARAJAN, M. WANG, M. MURTHY;
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Abstract: Social behaviors rely on multisensory integration. How multisensory responses are generated from unimodal inputs and how they are read out to inform behaviors remain open questions. Here, we dissect the cellular mechanisms underlying multisensory integration in the

fruit fly Drosophila, focusing on neurons involved in mated female rejection behavior (ovipositor extrusion) during courtship. We utilize this female behavior because it is driven by a specific cell type (DNp13 (Wang et al. Current Biology, 2020)). Using two available Drosophila female brain connectomic resources, FlyWire (Dorkenwald et al. bioRxiv, 2020) and the hemibrain (Scheffer et al. eLife, 2020), we have mapped sites of audiovisual integration onto DNp13 as well as its presynaptic partners. Auditory neurons in this circuit are selective for male courtship song, while visual neurons are uncharacterized. Through high resolution behavioral tracking technology (SLEAP; Pereira et al. bioRxiv, 2020) we trained a machine learning model to track male and female pose, including ovipositor extrusion. We tracked both pose and male song throughout courtship in over 45 pairs of virgin males with previously mated females. These data are used to build a model of audiovisual integration that predicts female rejection behavior. Via neuronal silencing and calcium imaging experiments, we are testing the role of specific neurons in this circuit in audiovisual integration. Taken together, this study expands our understanding of how cells and circuits process multisensory signals and transforms them into dynamic motor responses.

Disclosures: E. Normand: None. **D. Deutsch:** None. **T.D. Pereira:** None. **N. Rangarajan:** None. **M. Wang:** None. **M. Murthy:** None.

Poster

P504. Cross-Modal Processing In Humans I

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P504.01

Topic: D.09. Multisensory Integration

Support: NRF-2020R1F1A1076336

Title: Lack of sensorimotor contingency deters visuo-haptic interaction of realistic stimuli

Authors: *H. SHIN, C.-Y. KIM;
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Abstract: Sensory information from modalities other than vision is known to alleviate visual ambiguity. In a previous experiment, we explored the influence of visuo-haptic shape congruence on perceptual selection during binocular rivalry using 3D shape objects (Shin et al., VSS 2021). Spiky, round, and a neutral shape neither spiky nor round was created using Superformula (Gielis, 2003). Rotating 3D rendered spiky and round shape objects were presented dichoptically and participants simultaneously explored with their hand either spiky, round, or neutral haptic object by rotating it in a direction congruent or incongruent with the presented visual target. Results revealed that shape congruence between visual and haptic stimuli influenced visual selection by boosting the dominance duration of the shape congruent visual

target for both spiky and round shapes. The present experiment was conducted using ecologically valid stimuli to examine the same shape congruence effect in a setting with tighter visuo-haptic interactions. By changing the visual targets to a pre-recorded video of a hand exploring a spiky or round 3D shape object in a rotating motion, the influence of sensorimotor contingencies involved in dynamic visuo-haptic interactions was expected to be observed. Participants dichoptically viewed videos exploring a round or spiky shape and reported the consciously perceived visual target via keyboard key press using their left hand while exploring the given haptic object with their right hand. However, the pattern of results was largely different from the previous experiment - previously the interaction between visual and haptic shapes was significant, with no main effects of either visual or haptic shape. In the present experiment, visuo-haptic shape interactions were not statistically significant, and there existed a main effect of visual shape. We deduce that even with shape congruence between visual and haptic stimuli, the reality of the visual stimuli highly resembling (but not identical to) participants' hand movements bolstered the significance of sensorimotor contingency between what was being seen and what was being haptically explored. This suggests that visuo-haptic interactions underlying shape congruence effects in perceptual selection is dependent upon sensorimotor contingency, especially when the stimuli closely resemble perceptual experience in real life.

Disclosures: H. Shin: None. **C. Kim:** None.

Poster

P504. Cross-Modal Processing In Humans I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P504.02

Topic: D.09. Multisensory Integration

Title: Tactile Motion Coordinate Transforms in the Somatosensory Cortex

Authors: *H. AHUJA[1], S. SHIVKUMAR[1], C. FEISTRITZER[2], R. M. HAEFNER[1], G. C. DEANGELIS[1], M. GOMEZ-RAMIREZ[1];
[1]Brain and Cognitive Sci., Univ. of Rochester, Rochester, NY; [2]Neurosci., Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Our ability to perceive motion information on the skin is key to manipulating dynamic objects in the environment. Previous studies show that the brain derives tactile motion representations by integrating cues of the object that impinge on the skin (e.g., speed, intensity, direction), a mechanism known as the Full Vector Average model. This model was derived from studies that placed the hand in the same posture. Yet, object perception and manipulation with the hand (i.e., haptics) is a highly dynamic function. Thus, it is key to study whether motion inputs on the skin are transformed by hand position, and whether these transformations depend on the reference frame of the motion discrimination judgement. Here, we asked human

participants to discriminate motion stimuli on the index finger in a hand-centric or sternum-centric reference frame, while the stimulated hand was placed in different postures. Humans sat on a chair wearing a virtual reality headset, wherein a visual cue instructed participants to judge whether stimuli moved to the left- or right-side (relative to their sternum) or towards vs. away from their thumb (i.e., hand-centric task). The data revealed that humans flexibly represent tactile motion information according to the instructed reference frame, with motion judgements relative to the sternum perceived with higher sensitivity. We also developed a Bayesian generative model that accounts for the motion percepts in different reference frames and postural configurations. These data show that perceptual representations of tactile motion are generated by neural circuits that integrate cutaneous and proprioceptive inputs from the hand, and are under the control of goal-directed cortical signals.

Disclosures: H. Ahuja: None. **S. Shivkumar:** None. **C. Feistritzer:** None. **R.M. Haefner:** None. **G.C. DeAngelis:** None. **M. Gomez-Ramirez:** None.

Poster

P504. Cross-Modal Processing In Humans I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P504.03

Topic: D.09. Multisensory Integration

Support: BCM Seed Funds

Title: Compensatory changes follow somatosensory feedback loss in a dexterous manual task

Authors: *K. UNG[1], P. F. NORDMARK[2], J. YAU[1];
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Abstract: The role of sensory feedback is well established in current models of motor control, evidenced by severe deficits in movement coordination resulting from impaired sensory function. Goal-oriented motor behaviors involve extracting and integrating somatosensory and visual information about the action goal before the movement, and outcomes during and after the movement. How these sensory cues interact to contribute to dexterous motor behavior and how these interactions change following acute sensory loss have not been well studied. Here, we collect a large multimodal dataset from subjects performing a dexterous unimanual behavior (pegboard task) in order to address how behavior is impacted by loss of somatosensory feedback. Subjects retrieved, transported, and placed pegs under normal sensory conditions. They repeated the task with local anesthesia that removed somatosensory feedback at the fingers. During the performance of the task, we performed eye and hand tracking while also recording forces applied to the pegboard during the retrieval and placement of pegs. We hypothesized that impaired

tactile feedback disrupts the ability to finely tune dexterous motor behavior, leading to exaggerated movements and a greater reliance on visual feedback.
We found that loss of tactile feedback impairs performance of our manual dexterous motor task, leading to increased time to complete the task. Successful peg retrieval and placement required more time. Without tactile feedback, subjects also applied more force on the pegboard, implying impaired fine motor control. In some subjects somatosensory loss was accompanied by a shift in visual behavior marked by a closer alignment between eye and hand positions during all task stages. Together, these data support the important roles of sensory feedback on force control in service of dexterous object manipulation. Uncovering novel functional roles for sensory integration in dexterous motor tasks may not only better inform us of how the brain supports multimodal sensory integration, but may also help develop novel therapeutic approaches towards sensory feedback-based recovery of motor control.

Disclosures: K. Ung: None. **P.F. Nordmark:** None. **J. Yau:** None.

Poster

P504. Cross-Modal Processing In Humans I

Location: Virtual Only

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Program #/Poster #: P504.04

Topic: D.09. Multisensory Integration

Support: H2020-MSCA-ITN-2019 Grant 860114

Title: Multisensory integration of dynamic stimuli and the role of active touch

Authors: *M. CASADO PALACIOS[1,2], C. CAMPUS[1], *A. TONELLI[1], M. GORI[1];
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Abstract: Multisensory integration of dynamic stimuli and the role of active touch. M. Casado, C. Campus, A.Tonelli, M. Gori.
Multisensory integration is a topic that has been extensively studied. However, the mechanism underlying visual and audio integration with touch and especially the impact that active touch, defined as voluntary movements, might have in this integration has been neglected. Some studies have found the specific effect of the active movement at the behavioral level, such as better recognition of objects (Morton, 1984), or a more accurate perceived duration of tactile stimuli after adaptation (Tomassini et al., 2012). We wanted to shed some light into the role of active touch on the multisensory integration process with this work. In the framework of the project MCSA MULTITOUCH, we designed two different experiments that compare the velocity discrimination threshold using tactile and audio information during active and passive tactile perception. 15 healthy participants have performed the experiments.The participant had to perceive a sequence of two stimuli with different speeds and discriminate the faster one. Two

standard speeds were used as the first stimulus (3 and 7 cm/s), while the second was determined by the adaptive QUEST algorithm (Watson and Pelli 1983). Three experimental conditions were considered: unimodal tactile condition, in which the participant had to feel the movement of a wheel embedded on a tactile frequency greeting with the index finger; Unimodal audio condition, in which the information was produced by a device attached to the wheel; and a bimodal audio-tactile condition, where audio and tactile stimulation were provided. In experiment one, the finger was fixed in a certain position, while in experiment two, the participant had to move the finger in the opposite direction of the wheel's movement. Our results show that for the 3cm/sec standard velocity, the multisensory information is integrated into a Bayesian optimal fashion when tactile information is perceived during passive touch but not during dynamic touch. Interestingly, for the 7cm/sec standard velocity, the reverse is true. The audio and tactile information is integrated into a Bayesian fashion when active movements are involved but not for the passive touch.These results might indicate two different mechanisms of multisensory integration for active and passive multisensory speed perception. The passive touch could be more adapted to integrate multisensory signals that are slower, while the active touch could be more adapted to integrate multisensory signals faster.

Disclosures: M. Casado Palacios: None. **C. Campus:** None. **A. Tonelli:** None. **M. Gori:** None.

Poster

P504. Cross-Modal Processing In Humans I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P504.05

Topic: D.09. Multisensory Integration

Support: NIH Grant T32-EY007125

Title: Multisensory interaction of visual and proprioceptive cues in tactile motion perception

Authors: *C. FEISTRITZER[1,2], H. AHUJA[3], A. FULLER[3], M. GOMEZ-RAMIREZ[3]; [1]Neurosci., Univ. of Pittsburgh, Pittsburgh, PA; [2]Ctr. for Visual Sci., [3]Brain and Cognitive Sci., Univ. of Rochester, Rochester, NY

Abstract: The ability to perceive motion on the skin is crucial for manipulating objects with our hands. Recent work from our lab shows that tactile motion representations are derived by integrating motion cues impinging on the skin with the proprioceptive signals from the arm. We further showed that these representations are under control of goal-directed cognitive signals encoding the reference frame of the motion judgement. However, previous reach-to-grasp studies show that visual cues can modulate perceptual representations of the body's position. Here, we investigated whether visual signals relaying the proprioceptive state of the arm bias perceptual representations of tactile motion on the hand, and whether these effects depend on the reference

frame used to make the motion judgement. Human participants were asked to discriminate the motion direction presented on their index finger while the arm was placed in different postures. Participants wore a virtual reality headset that provided a visual representation of the arm's location. On certain trials we modulated the disparity between the visually-displayed arm and the physical position of the arm. Participants were asked to make motion judgements left vs. right relative to their sternum, or towards the tip vs. the base of the finger. Preliminary data showed that visual inputs modulated motion percepts according to a causal inference mechanism, wherein perceptual biases increased when visual and proprioceptive cues were closely aligned, and then decreased (and eventually disappeared) as visual and proprioceptive as disparity increased. These causal inference effects were observed in both reference frames. These data indicate that the brain integrates visual cues conveying proprioceptive state to derive tactile motion representations regardless of the reference frame used to make the motion judgements.

Disclosures: C. Feistritzer: None. **H. Ahuja:** None. **A. Fuller:** None. **M. Gomez-Ramirez:** None.

Poster

P504. Cross-Modal Processing In Humans I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P504.06

Topic: D.09. Multisensory Integration

Support: The Miami Project to Cure Paralysis
Craig H. Neilsen Foundation

Title: Multisensory Integration following Spinal Cord Injury

Authors: *R. VASTANO[1], M. COSTANTINI[2], W. H. ALEXANDER[3], E. WIDERSTROM-NOGA[1];
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Abstract: Multisensory integration is the process by which we integrate stimuli from different sensory modalities which generates a facilitatory effect on perception and action, namely *multisensory enhancement effect*. This requires absence of unisensory imbalance and appropriate exposure to cross-modal cues. Following spinal cord injury (SCI), multisensory integration may be altered by sensorimotor deficits, altered GABAergic inhibition and reorganization of multisensory areas, causing unisensory imbalance and altered responsiveness to cross-modal cues. The aim of this study was to determine if SCI causes deficits in multisensory integration, and where in the processing stream this would occur. We used a detection task in which

redundant unimodal (visual, auditory, and tactile) and bimodal (audio-visual, visuo-tactile, and audio-tactile) targets were delivered. Participants (17 controls and 15 individuals with SCI) were instructed to rapidly detect any of the stimuli and respond vocally by saying “yes”. We recorded vocal reaction times. To quantify the multisensory enhancement effect, we used the independent race model that dissociates multisensory integration from bimodal facilitation lead by redundant stimuli. The multisensory enhancement effect is expressed as a “violation of the race model”, in which bimodal stimuli are faster than the summed unimodal probabilities. We hypothesized a reduced multisensory enhancement effect in SCI participants compared with control subjects. We found evidence of reduced race model violations in SCI participants. This result involved all tested sensory modalities.In addition, we investigated whether independent processing or competition mechanisms would occur at the stimulus and/or perceptual level in the SCI group by using a computational model approach. We tested two model parameters (i.e., stimulus strength and multisensory competition). We hypothesized that multisensory integration deficits could either involve one of the model parameters in a specific processing stream (stimulus or perceptual level), or involve more than one parameter, suggesting that after an SCI both independent processing and competition mechanism may arise at multiple perceptual stages. The computational model results indicate that SCI participants exhibit a higher perceptual multisensory competition at the neural representation level.Our results indicate that SCI reduces multisensory integration due to competition mechanisms occurring at the neural representation level. This suggests that a reorganization of multisensory areas after an SCI may play a role in multisensory integration deficits.

Disclosures: R. Vastano: None. **M. Costantini:** None. **W.H. Alexander:** None. **E. Widerstrom-Noga:** None.

Poster

P504. Cross-Modal Processing In Humans I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P504.07

Topic: D.09. Multisensory Integration

Support: NIH NINDS U01 NS108922
Clinician Investigator Program, Schulich Medicine, Western University

Title: Vision and artificial touch through intracortical microstimulation integrate optimally in humans

Authors: *V. KARAPETYAN[1], M. L. BONINGER[2], J. L. COLLINGER[2], A. P. BATISTA[1], P. LOUGHLIN[1], R. A. GAUNT[2];
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Abstract: Sensory feedback rarely operates in isolation. For instance, skillful object manipulation relies on the interplay between somatosensation and vision. Brain computer interface (BCI) based prostheses can implement intracortical microstimulation (ICMS) of the somatosensory cortex to augment vision with localized sensory feedback. However, it remains to be determined whether, or to what extent such artificial somatosensation can integrate with natural vision. We used a two-alternative forced choice task to study the interaction of ICMS with vision in a human participant with tetraplegia and compared our results with a well-established Bayes' optimal integration model.
ICMS was delivered using microelectrode arrays implanted in Area 1 of the somatosensory cortex. It consisted of a constant frequency pulse train with a sinusoidally modulated amplitude to simulate the tactile sensation of running a finger across a corrugated surface. The visual stimulus consisted of random dots coalescing into vertical light and dark patches, matching the sinusoidal ICMS signal. The amount of dot coherence permitted control over noise in the visual signal. These modalities were presented together over two intervals, each interval with a different sinusoidal frequency, and our participant reported which had the faster oscillation. By delivering vision and ICMS incongruently (at different frequencies) we were able to determine the extent to which one modality was preferred over the other at different degrees of visual noise. Single modality performance was used to generate predictions under a Bayes' optimality model, and multi-modal observations were compared with these predictions.
We found that ICMS and vision do not operate in isolation but indeed do integrate in a manner consistent with minimal-variance optimality. As visual feedback noise decreased, participant preference became significantly biased towards the visual modality, in accordance with model predictions. This effect was replicated on two electrodes, projecting to different somatotopic locations and having different sensory profiles. Our results show that direct cortical stimulation can engage similar multisensory integration mechanisms as visuo-haptic exploration in able-bodied individuals. These results help interpret the manner in which somatosensory ICMS feedback operates in conjunction with other sensory modalities and guide its use in neuroprostheses.

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Poster

P504. Cross-Modal Processing In Humans I

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P504.08

Topic: D.09. Multisensory Integration

Title: Mirror Neuron Therapy for Central Pain Syndrome

Authors: *E. ALTSCHULER;
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Abstract: A 63 year old female presented with six years of a severe central pain syndrome from presumed medication toxicity. The pain started in the left arm, leg and face and then spread to the right leg as well. Even after eventually tapering the medication thought to have initiated the pain syndrome, severe pain remained. Pain was exacerbated by warm temperature, vibration and even wind against the limbs. The patient found icing the limbs to be useful, but still had to have the limbs covered at most times to avoid wind or other light touch sensations setting off a pain exacerbation. The topic of mirror neurons--neurons that respond not only when one moves, but when one watches another move--was discussed with the patient as a possible therapeutic avenue: The patient is a yoga instructor and she noticed that when doing yoga wearing socks and leggings, he pain was lessened when she watched her student performing yoga in bare feet without experiencing any pain. The patient then started for limited times working without socks while still watching her students perform yoga. Gradually, the patient was able to go an entire teaching session without socks. Concomitant with this improvement the patient's pain at other times improved as well. Over the course of three months the patient's pain decreased to only a minimal level. The patient is now able to well tolerate vibration, e.g., long drives, without significant pain, and the patient does not need to cover up to avoid a pain exacerbation caused by wind. This case suggests that a mirror neuron inspired approach may be a useful treatment for central pain syndromes. Controlled studies of this approach may be warranted.

Disclosures: E. Altschuler: None.

Poster

P504. Cross-Modal Processing In Humans I

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Program #/Poster #: P504.09

Topic: D.09. Multisensory Integration

Support: NIH Brain Initiative Grant UH3 NS109556-02
DARPA grant DARPA-BAA-14-09

Title: Perception and Chronic Pain: Anterior Cingulate and Orbitofrontal Cortex Activity during Musical Hallucinations

Authors: *A. SCHMITGEN[1], J. SAAL[2], N. SANKARAN[3], M. DESAI[6], I. JOSEPH[1], P. STARR[4], E. F. CHANG[4], P. SHIRVALKAR[5];
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Abstract: The anterior cingulate cortex (ACC) and orbitofrontal cortex (OFC) have been extensively implicated in the functional brain network underlying chronic pain. Electrical stimulation of the ACC and OFC have been proposed as therapies for refractory chronic pain, although, mechanisms of therapeutic action are still unclear. While the OFC is commonly associated with the valuation and cognitive manipulation of pain, the ACC plays a varied role in sensory and emotional integration of pain, as well as modulating internally generated perceptual states. In the context of perception, ACC activation has been commonly associated with audition, including detection of music, while the OFC has been described in the perception of taste, vision, and olfaction. In this case series, we report the emergence of subjective musical hallucinations (MH) after electrical stimulation of the ACC in two patients with refractory chronic pain. In an N-of-1 analysis from one patient, we identified neural activity (local field potentials, LFP) that distinguish MH from both the non-MH condition and during a task involving music listening. In contrast to an absence of notable LFP changes in the OFC, activity in the ACC (particularly in gamma and alpha bands), was associated with musical hallucinations and musical listening, respectively, extending prior results that internally-generated perceptual phenomena are supported by circuits in the ACC. We discuss these findings in the context of phantom perceptual phenomena and posit a framework whereby chronic pain may be interpreted as a persistent internally-generated percept.

Disclosures: A. Schmitgen: None. **J. Saal:** None. **N. Sankaran:** None. **M. Desai:** None. **I. Joseph:** None. **P. Starr:** None. **E.F. Chang:** None. **P. Shirvalkar:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Medtronic Inc. provided research devices for use in this study and technical support through a research agreement with UCSF, but no financial support..

Poster

P504. Cross-Modal Processing In Humans I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P504.10

Topic: D.09. Multisensory Integration

Support: NIH Grant F31-NS118832
NIH Grant T32-EB009406
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Title: Chemotherapy induces proprioceptive deficits in the proximal limbs of cancer survivors

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Abstract: The neurotoxic side effects of oxaliplatin (OX) chemotherapy contribute to movement deficits such as dexterity loss, imbalance, and falls in cancer survivors. It is often thought that OX-induced sensory nerve degeneration in the distal limbs, commonly termed sensory neuropathy, causes these movement deficits. However, recent animal studies have identified dysfunction in the muscle proprioceptors as an alternative mechanism. Muscle proprioceptors encode the muscle and limb motions and forces needed for accurate interactions with the physical world. Unlike distal sensory neuropathy, proprioceptive dysfunction could lead to more extensive motor deficits, including in the proximal limbs, where neuropathies are typically not reported. It is unclear whether chemotherapy leads to proprioceptive dysfunction independent of sensory nerve degeneration in humans. This study fills this gap by evaluating the functional use of proprioception in the shoulder and elbow joints of cancer survivors treated with OX. Thirteen cancer survivors and thirteen control subjects were tested. We implemented three sensorimotor tasks that assessed the use of proprioception in the horizontal plane: isometric force matching, unloaded reaching, and postural stability while countering external loads. Subjects were attached to a HapticMaster robot that measured endpoint displacements and forces and created the virtual environment required for each task. All subjects completed the tasks with and without visual feedback to separate the role of proprioception and vision. Cancer survivors had the greatest deficits in force matching. They were 62% less accurate ($p = 0.003$) and 37% less precise ($p = 0.03$) than controls when completing the task without vision. The group differences were small in the postural stability task (accuracy: 35%, $p = 0.05$) and minimal in the reaching task (accuracy: 22%, $p = 0.11$), suggesting that tasks involving force sensing and generation might be more affected in cancer survivors than those related to kinesthesia. The severity of the observed force deficits was correlated with cancer survivors perceived motor deficits, assessed using the clinical QLQ-CIPN20 questionnaire ($p < 0.001$). The most severely impaired cancer survivors had the poorest force-matching in directions that required significant use of shoulder muscles, indicating proximal involvement. While distal symptoms are routinely assessed in the clinical setting, our results suggest proximal assessments are also needed. Force tasks like those tested could be a starting point towards a more thorough and objective assessment of movement function in patients treated with chemotherapy.

Disclosures: A.B. Wang: None. **S.N. Housley:** None. **A.M. Flores:** None. **T.C. Cope:** None. **E.J. Perreault:** None.

Poster

P509. Eye Movements II

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P509.01

Topic: E.01. Eye Movements

Support: NIH Intramural Funding

Title: Multiple groups of neurons in superior colliculus control gaze and saccade based on object values

Authors: *A. GOPAL, O. HIKOSAKA;
NIH, Bethesda, MD

Abstract: Specialized networks in the basal ganglia (BG) assess the value of objects in the visual field. The neural activity in the caudate nucleus and substantia nigra reticulata distinguishes high-valued good objects from low-valued bad objects. This value signal from BG is then transmitted to the superior colliculus (SC), which contains at least three subtypes of neurons based on their functional role in generating saccades. This value signal must interface with these neuronal subtypes to modulate behavior, but the detailed connectivity is not well understood. To test this, we studied the response properties of different subtypes of SC neurons when monkeys directed saccades to objects of high and low value presented in their contralateral visual field. We identified four subtypes of SC neurons using a linear array that recorded cells across the different layers. Consistent with the previous findings, we found the four classes of SC neurons have distinct response profiles. Despite this, we found some common features in three groups: visual, visuomotor, and motor neurons. The early transient activity (0-80ms after target onset) did not differentiate between the value of objects. In the time interval 80-160ms, there was significantly higher activation in these neurons in response to good objects in the receptive field than bad objects. This suggests that the descending value inputs from the BG connect with these three classes of neurons. In addition, we found that the activity in the same interval has a systematic relationship with the latency of the upcoming saccade. The highest activation resulted in the fastest reaction time (RT), and the average activity progressively reduced for slower RTs. This systematic relationship between firing rate and RT was most prevalent in the visuomotor and motor neurons and to a lesser extent in the visual neurons. We also found a novel class of neurons (i.e., Inhibited) that are tonically active but decreased their activity in response to object onset in the receptive field. When the decrease was larger, the saccade tended to be faster, suggesting that these neurons may control saccades by their inhibitory connections to the other three groups: tonic inhibition (before object) and disinhibition (after object). However, these neurons are not sensitive to the value of objects as a population, hence unlikely to receive BG inputs. These data suggest that control of gaze/saccade based on object value involves the interplay of several mechanisms 1) Inputs from BG to visual, visuomotor, and motor neurons, 2) Interactions between visual, visuomotor, and motor neurons, 3) Inputs from Inhibited neurons to visual, visuomotor, and motor neurons.

Disclosures: A. Gopal: None. **O. Hikosaka:** None.

Poster

P509. Eye Movements II

Location: Virtual Only

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Program #/Poster #: P509.02

Topic: E.01. Eye Movements

Support: PAPIIT-DGAPA No. IA200920
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CONACYT A-1-S-10669
Laboratorio Nacional de Resonancia Magnética LANIREM

Title: Cerebellar role for planning motor behavior identified by Pro/Antisaccade Task and concurrent Functional Magnetic Resonance Imaging

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[4]Facultad de Medicina, Univ. Nacional Autónoma de México, Ciudad de México, Mexico

Abstract: Motor control can be divided into automatic and voluntary processes. These processes require planning and preparation in order to deciding in advance to perform one action and not another when a particular sensory event occurs. It is known that the primary motor, the premotor, and the dorsolateral prefrontal cortices are linked to motor preparation. The cerebellum is recognized as an important structure in motor control, however, we do not know its precise role in voluntary motor preparation. To delve into this problem, 18 healthy right-handed volunteers, 9 females and mean age 23.8 years ± 3.44 performed the Pro/Anti-saccade Task while undergoing a fMRI scan with concurrent aye tracking. A randomly interleaved design was used including full prosaccade (PS), antisaccade (AS); and preparatory-only (PO) trials aimed at examined both the preparatory an execution components of saccade. Participants fixated on a central white circle for 1000 ms (neutral fixation stimulus) to start each trial. The neutral fixation stimulus changed to an instructional cue indicating that a PS (green circle) (look toward the target location) or an AS (red circle) (look away from the target in the opposite direction) was required. After 1300 ms presentation of the instructional cue, a 200 ms black screen gap period was introduced. On saccade trials, a peripheral target was flashed for 100 ms to the left or right of the neutral fixation to signal a saccade. Participants had 1400 ms to execute the appropriate PS or AS. Once a trial was finished, the neutral fixation stimulus reappeared at the center of the screen for 500 ms. On PO trials, the instructional cue was briefly presented, but the peripheral target never appeared to signal the saccade. Participants did not know whether or not the peripheral

target would appear on any given trial; thus the instruction cue would always elicit preparation for PS or AS. There were statistical differences between correct PS Saccade Reaction Time (SRT) and AS-SRT (p=0.003), and differences between correct and error PS-SRT cumulative distributions (p=0.01). Our fMRI analysis identified the main oculomotor network previously reported for PS and AS. More importantly, on PP trials we showed significant activation in the cerebellar lobules VI, VIII A and B; and Cruz I – II (p=0.05). These findings suggest that the cerebellum is important not only in the execution but also in the motor preparation. This activity pattern can be explained by the functional link between cerebellar and frontal-motor areas controlling the preparation and inhibition of the motor commands, which involves attentional processes.

Disclosures: I. Vaca-Palomares: None. **R.O. San Pedro-Caligua:** None. **E.H. Pasaye-Alcaraz:** None. **J. Fernandez-Ruiz:** None.

Poster

P509. Eye Movements II

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Program #/Poster #: P509.03

Topic: E.01. Eye Movements

Support: MHRD fellowship through IISc
DBT IISc Partnership Programme grant

Title: Peripheral signatures of sequential saccade planning

Authors: *D. BASU[1], N. SENDHILNATHAN[2], A. MURTHY[1];
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Abstract: The conversion of neural motor plans into muscle activity at the periphery underlie all targeted behavior. Planning of single saccades in oculomotor centers like the frontal eye field (FEF) have been shown to rapidly engage neck muscle motor units involved in head movements, suggesting that a tight link exists between the eye movement and head movement machinery. However, it is not known if a similar motor unit recruitment pattern occurs during sequential saccade planning, or if the downstream leakage of sequential signals is blocked by inhibitory control centers like the basal ganglia. To determine the relationship between neural and peripheral firing patterns during sequential movement planning, we undertook simultaneous single-unit recordings from the FEF (neural activity) and the dorsal neck muscle (electromyographic or EMG activity) of two macaque monkeys performing a sequential saccade task. In our study, the monkeys were head-restrained and only execution of saccadic eye movements was required in the task.We have previously shown that when a sequence of two saccades is concurrently planned, a processing cost is imposed on both the motor plans and FEF

activity ramping up towards threshold slows down. Neural signatures of processing bottlenecks seen in the FEF were preserved in neck motor unit activity: the shorter the gap between two saccade plans, the higher is the processing cost, and slower is the rate of growth in both neural (FEF) and peripheral activity (neck muscle). Mimicking neural activity patterns, evidence of parallel programming of saccades was also found at the level of the neck muscle: motor unit activity for the second plan was initiated before the first plan reached completion, although the onsets of neck muscle activity was delayed when compared to the onsets of FEF activity. Thus, the correspondence of the results between the FEF and the muscle periphery suggests that a common gaze command originates in the FEF and is relayed down to the neck muscle for executing eye-head gaze sequences. Our results indicate that inhibitory control centers like the basal ganglia, which has been shown to limit the processing of concurrent motor plans, restricts the downstream flow of signals encoding the second saccade in the FEF-neck muscle circuit only to a limited extent. We posit that sequential oculomotor plans in the FEF are indexed by the motor units in the dorsal neck muscle to bring about coordinated sequential gaze shifts.

Disclosures: D. Basu: None. **N. Sendhilnathan:** None. **A. Murthy:** None.

Poster

P509. Eye Movements II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P509.04

Topic: E.01. Eye Movements

Support: Smith-Kettlewell Eye Research Institute

Title: A new model of binocular eye movement control

Authors: *S. HEINEN[1], A. CHANDNA[1], D. SINGH[1], S. WATAMANIUK[1,2];
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Abstract: Oculomotor physiologists usually record one eye, assuming that the other rotates identically (or oppositely). This methodology is reflected in the structure of virtually all oculomotor models, since these models describe pure conjugate or pure vergence eye movements. However, many eye movements acquire targets that lie off of the horopter or midline, requiring asymmetric eye rotations. How are asymmetric eye movements controlled? According to Hering's (1868) theories, a unitary vergence command is simply added to a unitary conjugate command to drive gaze shifts to any location, and most models assume this occurs. Our recent results during monocular viewing show asynchronous rotation of an occluded eye making it unlikely that a unitary vergence command drives gaze shifts to objects in depth (Chandna et al., 2021). Theoretical independent control of each eye could substitute for vergence

and solve the problem, but pure independent control does not account for sluggish midline vergence or inappropriate conjugate rotations during gaze shifts to targets aligned on an eye (Müller paradigm). Here we introduce a new theoretical model of binocular control. The core of the model is a rapid conjugate system that outputs a unitary command, and it is modulated by slow independent controllers. Like other oculomotor models, ours uses feedback and generates eye movement profiles with realistic temporal dynamics. Our model generates rapid saccades, as well as asymmetric eye movements, slow midline vergence and inappropriate conjugate rotations in the Müller paradigm. An additional endogenous component that interprets target motion when sensory evidence is lacking explains our covered eye results.

Disclosures: S. Heinen: None. **A. Chandna:** None. **D. Singh:** None. **S. Watamaniuk:** None.

Poster

P509. Eye Movements II

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Program #/Poster #: P509.05

Topic: E.01. Eye Movements

Support: Helen Bader Age & Community Scholarship
UW - Milwaukee Graduate Student Research Award

Title: Saccade amplitude, among other age-related differences in eye movements, is related to Archimedes spiral tracing performance in older adults

Authors: *B. HEINTZ WALTERS[1], W. E. HUDDLESTON[2], K. G. KEENAN[3];
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Abstract: Introduction. Archimedes spiral tracing is a common, clinical assessment sensitive to age-associated impairments in hand function. When performed on a touchscreen, tracing with the relatively larger diameter index finger versus stylus preferentially impaired performance in older adults, suggesting that decreased visual information of the spiral may contribute to age-associated spiral tracing impairments. However, no comprehensive analysis to date has evaluated the effect of eye movements on spiral tracing performance. The purpose of this study was to determine the association between eye movements and Archimedes spiral tracing performance in older adults. **Methods.** Eye movements were recorded from 21 young (age 20 - 38 yrs; 12 f, 9 m) and 20 older (age 65 - 85 yrs; 10 f, 10 m) adults during a spiral tracing task. Participants traced an Archimedes spiral template displayed on a touchscreen as accurately as possible using their index finger and a stylus. Live feedback of the participant's trace was superimposed onto the template. RMSE was calculated to quantify proximity of the participant's trace to the template. **Results.** RMSE was approximately 30% greater in older versus young adults when tracing with

the finger ($p < .001$) though there was no difference in RMSE when tracing with the stylus ($p = .130$). All participants used a series of saccades and fixations while tracing the template. Older adults made fewer saccades than young adults ($p = .029$). Saccade amplitude ($p = .029$) and saccade amplitude variability ($p = .011$) were greater in older versus young adults. Increased saccade amplitude was associated to increased RMSE in older adults ($r = .741$). Due to the nature of the task, the hand may have obstructed more visual information of the spiral depending on the part being traced. Participants made more saccades when tracing parts of the spiral where vision of the upcoming spiral template was likely less obstructed by the hand ($p < .001$) though there were no differences in RMSE ($p = .065$). **Conclusions**. Because saccades are used to orient gaze with areas of interest in the visual field, a greater number of saccades likely reflects a visual strategy that would allow for more up to date visual information of the spiral; however, this had no effect on spiral tracing performance. Results provide evidence of age-related changes in eye movements during the spiral tracing task and suggest that decreased visual information may not be the main contributing factor associated with impaired spiral tracing performance when tracing with the finger in older adults. Future work could continue to investigate other factors associated with the ability to perform the touchscreen spiral tracing task.

Disclosures: B. Heintz Walters: None. **W.E. Huddleston:** None. **K.G. Keenan:** None.

Poster

P509. Eye Movements II

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P509.06

Topic: E.01. Eye Movements

Support: E,C-H is a ConTex Postdoctoral fellow

Title: Neuronal computations associated with saccades in a widely distributed prefrontal cortex

Authors: *E. CONTRERAS-HERNANDEZ, G. M. JEANPIERRE, S. S. KOUKUNTLA, H. M. STEALEY, S. R. SANTACRUZ;
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Abstract: The role of different cortical prefrontal areas in the preparation of voluntary visual- and memory- guided saccades is well studied. In particular, the anterior cingulate cortex (ACC) and dorsolateral prefrontal cortex (dlPFC) play important roles in the control of intentional saccades and visuospatial working-memory, respectively. Studies that have analyzed neuronal computations associated with saccade movements at the level of single neurons have been primarily conducted in individual prefrontal regions. We analyzed the tuning of single neurons simultaneously recorded from the ACC, dlPFC and orbitofrontal cortex (OFC) while non-human primates (macaca mulatta) performed a center-out saccade task. After a center-fixation period,

visual cues appeared simultaneously in four quadrants, and then the subjects made a saccade at a free chosen direction. The employed data in this analysis were obtained by other authors and made openly available (CNCRS.org-PFC7). By using a linear discrimination-based neural classifier we find that the firing rate of the neurons recorded from each of these three areas are tuned at different extents to the direction of the saccades. Interestingly, we observe that the accuracy of the prediction of the saccades direction is proportional to the number of analyzed neurons regardless if they were recording simultaneously from the three regions (ACC, dlPFC and OFC) or from subgroups of regions (e.g. ACC and dlPFC). We also show that the firing rate of a modest fraction of single neurons are tuned with the amplitude, speed and onset time of the ocular movement. Additionally, all the correlates between neuronal activity and the features of the saccades appear after the presentation of the visual cues, and before, during and after of the saccades. These observations suggest that the dlPFC, OFC, and ACC form part of an integrative prefrontal network involved on the preparation and execution of voluntary saccades. Additionally, it supports the inclusion of the OFC in such network in non-human primates, whose participation has remained on debate.

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Poster

P509. Eye Movements II

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P509.07

Topic: E.01. Eye Movements

Support: R00EY027846

Title: Head tilt influences saccade directions during free viewing

Authors: *S. M. REEVES[1], E. A. COOPER[1,2], R. RODRIGUEZ[1], J. OTERO-MILLAN[1,3]; [1]Sch. of Optometry & Vision Sci. Program, Univ. of California Berkeley, Berkeley, CA; [2]Helen Wills Neurosci. Institute, Univ. of California Berkeley, Berkeley, CA; [3]Dept. of Neurology, Johns Hopkins Univ., Baltimore, MD

Abstract: When humans freely explore visual scenes they can make saccades in any direction, but it turns out that not all directions are equally likely. A bias toward horizontal and vertical saccade directions is well documented, though the reason for the bias is unknown. In addition to biases in oculomotor anatomy, scene structure, or past experience, it is possible that vestibular signals indicating head orientation play a role in the saccade direction biases. Thus, in the present study, we asked whether roll tilting the head influenced saccade direction distributions during free viewing. Study participants (n = 14) viewed natural scenes (outdoor and indoor) and abstract

scenes (radially symmetric patterns) using the FOVE virtual reality headset. We recorded eye movements with the integrated FOVE eye tracking system (120Hz, binocular). Participants sat in a chair with their heads stabilized and tilted at -30, 0, or 30 degrees while viewing scenes that could also be tilted by -30, 0, and 30 degrees relative to the head. All participants were exposed to all head tilt and scene tilt conditions. Head tilt angle was blocked, and scene type and tilt angle were randomized. To determine if the biases in saccade direction changed with head tilt (i.e., if the directions with the most saccades rotated) we calculated polar histograms of saccade directions in head-referenced coordinates for both natural scenes and abstract scenes for each head tilt block, and then cross correlated the distributions for -30 and 30 degree head tilts to find the angular displacement that resulted in the maximum correlation. A single sample t-test revealed that saccade distributions for abstract scenes were significantly rotated by a mean angle of 5.29 degrees ($SD = 5.41$; $t(13) = 3.65$, $p = 0.003$). Saccade distributions for natural scenes were also significantly rotated by a mean angle of 2.2 degrees ($SD = 2.36$; $t(13) = 3.51$, $p = 0.004$). These results suggest that the saccade direction biases can be affected by head tilt, and that non-visual information about head orientation, such as information from vestibular signals, likely plays a role in saccade generation.

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Poster

P509. Eye Movements II

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Program #/Poster #: P509.08

Topic: E.01. Eye Movements

Support: EU MSCA 734227

Title: Translational and rotational eye movements during blinks measured using real-time MRI

Authors: J. KIRCHNER[1], T. WATSON[2], *M. LAPPE[1];
[1]Inst. for Psychology, Univ. of Muenster, Muenster, Germany; [2]Western Sydney Univ., Penrith, Australia

Abstract: Blinks occur frequently in normal life and have increasingly been linked to perceptual and cognitive effects. However, some aspects of the oculomotor mechanics of blink-related eye movements remain unclear. Blink-related retraction of the eyeball has been observed when forcibly holding the eyelid open, but the full extent of the blink-related eye movement has not been measured so far due to technical limitations. Here we use real-time Magnetic Resonance Imaging (MRI) to image the eye in motion during blinks and developed a fully-automated segmentation method allowing to analyse eye movements in terms of both translation and

rotation. For our MR-based Eye Tracking (MREyeTrack) procedure we used a balanced steady-state free precession (bSSFP) MR sequence with a temporal resolution of 35 ms in the axial or 38 ms in the sagittal plane. Motion estimation was optimised by obtaining precise knowledge of the geometric shape of the eyeball from a 3D anatomical scan before undertaking kinematic estimation using the high temporal resolution single-slice data. For comparison, a video-based eye tracker (Eyelink 1000) simultaneously recorded gaze direction and pupil size of the right eye of the participants. We recorded various horizontal eye movements, blinks and longer periods of eye closure. Linear regression analysis of the MREyeTrack data and the horizontal gaze data from the video-based eye tracker yielded highly significant agreement between both data sources. We were able to measured the full extent of the blink-related eye movement, further demonstrating that the eye approaches a holding position during lid closure moving by as much as 35° in rotation and 2 mm in translation. Apart from the previously reported retraction, we additionally observed that the eyeball lifts during blinks. We also investigated the coordination of gaze shifts with blinks. We found that the gaze shift is tightly coupled in time to the translational blink movement and that blinks can induce significant temporal shifts of the gaze trajectory between left and right eye. We have demonstrated the application of real-time MR sequences to eye tracking and established MREyeTrack, an automated analysis method based directly on anatomical eye orientation. It is capable of establishing translational eye movements and those that occur when the eyelid is closed. We used this method to show novel aspects of the blink-related eye movement and think that these may provide useful insights into the biomechanics of the oculomotor system.

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Poster

P509. Eye Movements II

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Topic: E.01. Eye Movements

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MOST Grant 108-2410-H-038-002-MY3
MOST Grant 106-2410-H-038-004-MY2
MOST Grant 109-2410-H-038-010

Title: Role of the frontal eye field in human pupil and saccade orienting responses

Authors: *C.-A. WANG[1], T.-Y. HSU[2];
[1]Inst. of Cognitive Neurosci., Natl. Central Univ., Taoyuan City, Taiwan; [2]Grad. Inst. of Mind, Brain and Consciousnes, Taipei Med. Univ., Taipei City, Taiwan

Abstract: The appearance of a salient stimulus evokes a series of orienting responses including saccades and pupil size to prepare the body for appropriate action. The midbrain superior colliculus (SC) that receives critical control signals from the frontal eye field (FEF) is hypothesized to coordinate all components of orienting. It has shown that the FEF, together with the SC, is also importantly involved in the control of pupil size, in addition to its well-documented role in eye movements. Although the role of the FEF in pupil size is demonstrated in monkeys, its role in human pupil responses and the coordination between pupil size and saccades remains to be established. Through applying continuous theta-burst stimulation (cTBS) over the right FEF and vertex using magnetic resonance imaging-guided TMS in twenty-eight human participants, we investigated the role of the FEF in human pupil and saccade responses evoked by a salient stimulus, and the coordination between pupil size and saccades. Each cTBS session delivered a 40 seconds train of uninterrupted biphasic theta-bursts pulses. This consisted of 3 pulses, at 50 Hz, given in 200 ms intervals, comprising a total of 600 pulses for 40 seconds at 80% intensity stimulation for active motor threshold. The active motor threshold was defined as the lowest stimulator output in percentage that elicited 5 out of 10 twitches of more than 200 μV peak-to-peak amplitude in the contralateral hand, while the participant maintained 20% of a finger-thumb contraction, with the mean being 41.03% ± 6.15 (mean ± standard deviation) of maximum stimulator output. The participants came in twice (one week apart) for the same experiment with a different stimulation (the sequence of stimulation sites was counterbalanced across participants). All participants underwent the identical experimental procedure and conducted the behavioral task 1 minute after the completion of cTBS. Our results showed that neither saccade reaction times (SRT) nor pupil responses evoked by salient stimuli were modulated by FEF stimulation. Contrarily, the correlation between pupil size and SRTs in the contralateral stimulus condition (stimulus appeared in the contralateral field of stimulation) was diminished with FEF stimulation, but intact with vertex stimulation. Moreover, FEF stimulation effects between saccade and pupil responses associated to salient stimuli correlated across participants. This is the first TMS study on the pupil orienting response, and our findings suggest that human FEF was involved in coordinating pupil size and saccades, but not involved in the control of pupil orienting responses.

Disclosures: C. Wang: None. **T. Hsu:** None.

Poster

P509. Eye Movements II

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Program #/Poster #: P509.10

Topic: E.01. Eye Movements

Title: Hpa axis and cognitive function: circadian cortisol pattern and oculomotor task performance

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Abstract: The Hypothalamic-Pituitary-Adrenal (HPA) axis is a basic neuroendocrine system that regulates Circadian Cortisol Rhythm. Findings indicate that cortisol affects multiple aspects of cognitive function. In this study we attempted to explore the relationship between the diurnal variation of cortisol, measured by saliva samples collected five times a day, and cognitive function, by employing four experimental tasks that assess eye movements and inhibition, in two distinctive experimental paradigms. We compared the performance of 21 healthy young adults, aged between 18-33, in the Prosaccade (STP), GAP, Antisaccade (ANS) and in two task conditions of the Countermanding (CMN) task (CMN 40 and CMN 100), in three different time points (Morning, Afternoon, Evening) during one day. Participants presented a general improvement in their performance in the evening condition compared to the morning one, as measured by reaction time (main effect of time for: STP & GAP: $F_{2,40} = 8.16$, $p<.001$, ANS: $F_{2,40}= 4.88$, $p=.013$, CMN 40 & 100: $F_{2,38} =7.17$, $p =.002$), reaction variability (ANS: $F_{2,40}= 5.68$, $p =.007$, CMN 40 & 100: $F_{2,38} = 16.44$, $p <.001$) and percentage of correct responses (ANS: $F_{1.25, 25.01}= 7.88$, $p=.006$, CMN 40 & 100: $F_{2,38} =12.26$, $p <.001$). However, the diurnal variation of all oculomotor tasks' performance was not correlated to the expected and observed diurnal variation of cortisol. Results suggest that normal variability of cortisol levels in healthy young adults does not affect oculomotor performance and inhibition. We, also, compared the performance of 9 healthy young adults in the aforementioned oculomotor tasks during the morning of two consecutive days. Participants presented an improvement in their performance on the second day as measured by reaction time (main effect of day for: STP & GAP: $F_{1,8} = 17.84$, $p = .003$, ANS: $F_{1,7} = 39.26$, $p <.001$, CMN 40 & 100: $F_{1,8} = 5.62$, $p = .045$), reaction variability (STP & GAP: $F_{1,8} = 39.42$, $p <.001$, ANS: $F_{1,7} = 29.7$, $p <.001$) and percentage of correct responses (ANS: $F_{1,7} = 10.96$, $p = .013$, CMN 40 & 100: $F_{1,8} = 11.45$, $p = .009$) suggesting that the improvement observed in the first experimental paradigm, as well as in the second one, is due to learning effects.

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Poster

P509. Eye Movements II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P509.11

Topic: E.01. Eye Movements

Support: NSERC CREATE Brain-in-Action Program
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Title: Cortical contributions to transsaccadic discrimination of object orientation vs. shape changes: An fMRI study

Authors: *B. R. BALTARETU, W. D. STEVENS, E. FREUD, J. CRAWFORD;
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Abstract: Recently, Dunkley et al. (2016) showed modulations in parietal cortex (i.e., supramarginal gyrus, SMG) for transsaccadic changes in object orientation. However, this result did not generalize to other features, such as spatial frequency (Baltaretu et al., 2021). We hypothesized that the fundamental difference between these results was the detection of transsaccadic changes in object orientation (inferior parietal regions) vs. shape (medial occipital regions). Moreover, we anticipated that transsaccadic perception would occur through a unique functional network comprising saccade and object perception regions. Here, we used a double-dissociation fMRI task to determine the cortical correlates for transsaccadic change detection of multiple object features. 21 participants fixated a cross ±15.4° of centre, where an object was subsequently presented (rectangle, barrel, or hourglass) oriented at ±45° from vertical. The fixation cross remained in the same position (Fixation condition) or shifted (Saccade condition), followed by the same object re-presented at the orthogonal orientation (Orientation change) or another object at the initial orientation (Shape change). Change in object orientation or shape in each trial was indicated via button press. After excluding 4 participants, a region-of-interest analysis on two regions (supramarginal gyrus and cuneus) showed that only cuneus presented significant eye movement and feature sensitivity. A similar whole-brain univariate analysis was used to localize the optimal seed region for network analysis, which showed connections between *left medial occipital cuneus* and early-to-mid-level visual (e.g., lingual gyrus, superior occipital gyrus), object-relevant (e.g., medial occipitotemporal sulcus, transverse occipital sulcus), and oculo/sensorimotor (i.e., superior parieto-occipital cortex) regions. These results implicate medial occipital cortex in the transsaccadic memory and discrimination of multiple object features, with additional recruitment of regions in parietal and temporal cortex.

Disclosures: B.R. Baltaretu: None. **W.D. Stevens:** None. **E. Freud:** None. **J. Crawford:** None.

Poster

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Program #/Poster #: P509.12

Topic: E.01. Eye Movements

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Title: Dynamic visual processing coverage predicts observed differences in natural oculomotor behavior across mammals

Authors: *J. SAMONDS[1], M. SZINTE[2], C. BARR[1], A. MONTAGNINI[2], G. MASSON[2], N. J. PRIEBE[1];
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Abstract: Most vertebrates use saccadic eye movements to quickly change gaze and sample different portions of the environment. Several hypotheses have been proposed to explain saccadic dynamics, from low-level automatic image analysis (e.g. decorrelating receptive field responses) to cognitive scanpaths (e.g. directing gaze towards new objects). The adaptive properties of visual neurons require that saccades should be large enough to sample local information that are uncorrelated with the previous fixation in order to maximize responsiveness. Using this rationale, we previously were able to predict average saccade sizes from the combination of receptive field sizes and natural scene statistics. This prediction applies not only to small fixational saccades, but to all saccades when free viewing natural scenes for both foveate and afoveate mammals. Adaptation recovery times can also explain saccade rate distributions in these different animal species. Here, we examine what general principles would drive the differences for both saccade sizes and rates across three species (mice, marmosets, humans) scanning the same set of natural images. We report that in order to achieve similar coverage over time, animals with larger receptive field sizes require slower rates. For all three animals, saccade rates and sizes decrease with smaller images and over time. Differences in the number of photoreceptors and V1 cells available to process the sampled visual information also appear to play a substantial role in determining saccade rate. We conclude that a diverse set of mammals share a common strategy of maintaining sufficient sampling coverage of their environment over time, under the constraint of conserving energy.

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Poster

P509. Eye Movements II

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P509.13

Topic: E.01. Eye Movements

Title: Dopamine and control of memory-guided saccades in Parkinson's disease

Authors: *L. JIANG, A. KHIBOVSKA, H.-C. LEUNG;
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Abstract: Voluntary eye movement control has long been associated with dopaminergic modulation in the cortico-basal ganglia (BG) circuitry, yet its underlying neurocognitive mechanisms remain unclear. Using Parkinson's disease (PD) as a model, we examined how cortico-BG dopamine deficiency may influence selective memory- and visually-guided saccades (MGS and VGS). A group of early-stage PD patients (N = 12, both *on* and *off* dopaminergic medication) and age-matched healthy controls (HC, N = 14) completed an MGS and VGS task requiring selective processing. During the VGS task, participants were instructed to shift their gaze to one of two displayed items and ignore the other item. The paradigm was similar for the MGS task, except there was a delay period (1.7 or 4.3 s) between stimulus presentation and saccade response. We compared a wide range of saccade metrics between the two subject groups (PD vs. HC) and PD medication status (*on* vs. *off*), including saccade latency, gain, and peak velocity, number of corrective saccades, as well as systematic and unsystematic errors of the saccade endpoint with and without statistical modeling. Performance accuracy was comparable between PD and HC (MGS: 80.5% vs. 85.8%; VGS: 92.9% vs 93.6%) and between *on* and *off* (MGS: 77.1% vs. 83.8%; VGS: 92.5% vs 92.9%). As expected, individuals with PD showed reduced saccade velocity, prolonged latency, and more corrective saccades during the tasks, indicating impaired voluntary saccade generation in the early stage of the disease. In addition, individuals with PD showed significantly greater unsystematic errors, not systematic errors or mean saccade gain, in saccade endpoints of both tasks. These findings suggest an increased response variability, not a shift in the mean endpoint responses in PD relative to HC in the face of distraction. However, the dopaminergic medication did not show clear behavioral benefits. Compared to off medication, the same individuals with PD seemed to show prolonged saccade latency and were less accurate when they are on medication. These preliminary data suggest that dopaminergic medication impairs the ability to select the target or ignore the distractor in MGS and VGS in early-stage PD, potentially due to excessive upregulation of the indirect basal ganglia pathway dopamine or downregulation of the prefrontal dopamine. Taken together, these results revealed deficits in both visually and memory-guided saccadic responses in PD while dopaminergic medication did not seem to restore but may exacerbate these deficits, suggesting a complex role of cortico-BG dopamine in regulating voluntary control of eye movements during cognitive tasks.

Disclosures: L. Jiang: None. **A. Khibovska:** None. **H. Leung:** None.

Poster

P537. Cortical Planning and Execution: Premotor and Motor Cortex: Neurophysiology: Animal II

Location: Virtual Only

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Program #/Poster #: P537.02

Topic: E.04. Voluntary Movements

Support: NIH Grant 1R01NS114651-01
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Title: Effects of ischemic injury on the organization of complex forelimb movements in mouse motor cortex

Authors: *C. C. WOLSH, A. R. BROWN, S. MITRA, R. M. BROWN, M. M. LOPEZ, J. A. BOYCHUK;
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Abstract: Stroke is a predominant source of disability and there is a need to understand the neuronal processes underlying its pathophysiology and recovery. In this study, motor cortex was tested (using electric brain stimulation) as this structure is often damaged by stroke and motor impairments of the upper extremities are common post-stroke sequelae. Stroke was modeled by permanent ischemic injury using photothrombotic occlusion of the distal middle cerebral artery (MCAo-injury group). Control animals received sham-injury except one animal was non-surgerized (Uninjured group). Skilled motor behavior was assessed pre- and post-injury using the single pellet reaching task. Following completion of behavioral testing, long duration intracortical microstimulation (LD-ICMS; 500ms pulse trains) was applied to motor cortex to determine the location and proportion of complex forelimb movements of this structure. LD-ICMS was used here to study the resulting complex (multi-joint) movements since these responses may represent fundamental components of active motor behaviors. We detected four distinct complex forelimb movement regions of motor cortex in mice by determining the most consistent and robust movement at individual test sites during LD-ICMS; ‘advance’ (anterior forelimb displacement with flexion of the shoulder, and wrist extension), ‘elevate’ (dorsal forelimb displacement involving elbow flexion and wrist extension), ‘retract’ (posterior forelimb displacement involving flexion of the elbow and extension of the wrist) and ‘Dig/Push’ (posteroventral forelimb displacement involving extension of the shoulder and elbow). Comparison between the MCAo-injury group and Uninjured group detected a significant overall effect on reaching accuracy on the single pellet reaching task, with the MCAo-injury group being 56% less successful across the last 4 sessions. MCAo-injury also reduced the overall area of motor cortex capable of producing complex forelimb movements whereas the cortical area capable of producing simple movements (during LD-ICMS) was not reduced. The regions of most complex movement types (advance, retract, dig/push) were significantly decreased by MCAo (relative to the Uninjured group). Surprisingly, the cortical region of Elevate was not decreased by MCAo (relative to the Uninjured group). Kinematic testing of evoked LD-ICMS movements is ongoing. Future experiments will test for mechanisms that account for this

difference between Elevate and the other zones of complex forelimb movement in order to test parameters of intervention to improve motor outcomes after stroke injury.

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Poster

P537. Cortical Planning and Execution: Premotor and Motor Cortex: Neurophysiology: Animal II

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Topic: E.04. Voluntary Movements

Support: NIH 1R01NS092894-01
NSF IIS-1527747

Title: Physical Effort and Reward Value Representation in Sensorimotor (PMd, M1 And S1) Cortical Local Field Potentials

Authors: *T. YADAV, O. MAGAÑA-TELLEZ, J. T. FRANCIS;
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Abstract: Reward expectation in humans and animals is known to modulate behavior and its valuation is discounted by the physical effort associated with the movement. Our previous work has shown reward encoding within the macaque premotor (PMd), motor (M1), and somatosensory (S1) cortices. However, the involvement of these regions in concurrent processing of reward and effort information is not clearly understood. To address this gap, we asked how the reward representation in local field potentials (LFPs), recorded from the PMd, M1, and S1 are influenced by physical effort requirements. We trained a non-human primate (female, Macaca Mulatta) to perform three variations of a planar center-out reaching task: reward-only, effort-only, and reward-effort tasks. A visual cue (color and shape) presented at the beginning of each trial indicated the expected reward (3 levels of juice amount) and effort (4 levels of hand velocity dependent resistive force field) respectively. Cued amount of juice reward was delivered only for successful trials. While no viscous field was active for the reward-only task, a fixed reward (1 drop) was delivered in the effort-only task, whereas both reward and effort levels were varied in the reward-effort task. We assessed binned LFP during the post-cue period, before the target information was known, using generalized linear model (predictors: reward level, time bin for reward-only task; effort level, time bin for effort-only task; and reward level, effort level, and time bin for reward-effort task). In the reward-only task, we have previously shown that the post-cue amplitude of alpha and beta band LFPs encode reward value in PMd, M1 and S1. The effort-only task showed no significant effect of effort level or effort-bin

interaction, indicating negligible modulation in LFP amplitude with effort level (F-test, $p>0.05$). Interestingly, the reward-effort task showed a significant effect of reward-bin interaction, as well as reward-effort interaction (F-test, $p<0.05$), indicating effort-dependent reward representation in the alpha and beta oscillations. Taken together, our preliminary findings suggest subjective influence of effort information on the reward value encoded by the LFP amplitude from PMd, M1 and S1 regions. In addition to highlighting the neural processing of reward and effort information, results from this study could be critical to develop autonomous brain computer interfaces (BCIs) for clinical applications.

Disclosures: T. Yadav: None. **O. Magaña-Tellez:** None. **J.T. Francis:** None.

Poster

P537. Cortical Planning and Execution: Premotor and Motor Cortex: Neurophysiology: Animal II

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Program #/Poster #: P537.04

Topic: E.04. Voluntary Movements

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The University of Chicago
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Title: Mouse sensorimotor cortex reflects complex kinematic details during reaching and grasping

Authors: *H. A. GRIER[1], M. T. KAUFMAN[2];
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Abstract: Neural recordings in non-human primates have shown that activity in sensorimotor cortex correlates with the kinematic and postural details of reach to grasp movements. We characterized this relationship in the mouse. To obtain a diverse set of forelimb postures and movements, 5 head-fixed mice performed unimanual reaches to grasp water rewards from two spout targets cued by sound (based on Galinanes et al. 2018). We collected high speed camera data and tracked 15 points on the proximal and distal limb using DeepLabCut and DeepGraphPose. 3D triangulation allowed extraction of 25 angular degrees of freedom (DoFs) from skeletal relationships between markers. These variables showed that our task evoked complex kinematics: cross-validated dimensionality reduction (PCA) performed on single-trial joint angle time series revealed dimensionalities in the range of 6-11 across mice. To relate these variables to neural activity, we recorded from layer 2/3 excitatory cells of contralateral forelimb

S1 (S1-fl) and M1 (Caudal Forelimb Area, CFA) using 31 Hz two-photon calcium imaging in GCaMP6f transgenic mice (337-635 cells/dataset). Deconvolved calcium events showed that a substantial fraction of cells in both areas were significantly modulated over time or across spout targets: 61-66% in S1-fl and 38-80% in CFA. To determine how these responses reflected forelimb kinematics, we used cross-validated ridge regression encoding and decoding models. We found that 40-47% of significantly modulated neurons in S1-fl and 50-55% in CFA encoded 3D marker position, velocity or acceleration, and 40-44% / 51-54% encoded joint angles, velocities or accelerations. Position information was more accurately decoded than velocity or acceleration from both S1-fl and CFA, with a median of 73% variance explained (VE) across all markers and sessions. Multiple angular DoFs could be decoded from both areas, with wrist rotation, elbow flexion, and shoulder rotation decoded best (71-73%, 60-65%, 68-78%, VE). These results suggest that detailed cortical representations of reaching and grasping in the mouse are highly distributed across sensory and motor areas, and that activity strongly reflects the forelimb and digit positions across many angular DoFs.

Disclosures: H.A. Grier: None. **M.T. Kaufman:** None.

Poster

P537. Cortical Planning and Execution: Premotor and Motor Cortex: Neurophysiology: Animal II

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Topic: E.04. Voluntary Movements

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Title: Effect of Intracortical Microstimulation on Motor Learning in Macaques Manual Interception

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[1]Lab. of Neural Mechanism of Motor Control, Ctr. for Excellence in Brain Sci. and Intelligence Technol. (Institute of Neuroscience), CAS, Shanghai, China; [2]Univ. of Chinese Acad. of Sci., Beijing, China

Abstract: Recent progress in intracortical microstimulation (ICMS) has shown the promise of bi-directional brain-machine interface (BMI) for restoring sensorimotor function. However, the

artificial sensations evoked by ICMS in primary somatosensory cortex (S1) are still far from naturalistic. Despite that motor cortex is mainly responsible for motor generation, it has been recently shown that subthreshold ICMS in the motor cortex can evoke distinguishable sensations to guide specific movements. In contrast, a previous study claimed that subthreshold ICMS disrupted the activities in motor cortex and delayed the movements. To examine the effect of subthreshold ICMS in motor cortex in a stricter behavioral context, we delivered ICMS to monkey when doing the delayed manual interception task. One monkey (*Macaca mulatta*) was trained to manually intercept a target that moved circularly around a center on a vertical screen in his front. Subthreshold ICMS (100 ms of 0.2-ms biphasic pulses at 300 Hz) were delivered via single electrodes inserted into the primary motor cortex (M1) or dorsal premotor cortex (PMd) acutely. ICMS was delivered randomly in 30% trials, with a random onset time during the delay period. Arm kinematics was recorded using optical motion capture systems (vicon). Although subthreshold ICMS in M1 delayed the movements with endpoint error unaltered in the first eight sessions, reaction times (RTs) were gradually accelerated later in both non-stimulated (NS) and stimulated (ST) trials, while RT reduced more in ST trials without increasing endpoint error. The subsequent PMd ICMS no longer evoked RT increase, instead manifested a similar effect to M1 after the transformation of RT: shortened RT + movement time (MT), reduced RT/(RT+MT), and elevated peak hand velocity. Moreover, M1 ICMS made hand trajectory more curved without changing the error, whereas PMd ICMS induced an insignificant but modest increase in error and a straighter hand trajectory. Furthermore, we gradually dimmed the GO cue and found that the monkey learned to used subthreshold ICMS in PMd, M1, or S1 in place of visual GO cue to conduct delay-intercept task. Our study reveals that subthreshold ICMS in motor cortex induced a motor adaption with kinematic change in manual interception. Short-train subthreshold ICMS in motor cortex probably mainly evokes sensation rather than motor perturbation, which can be utilized by the monkey to improve action execution. In summary, in addition to S1, motor cortex is a candidate for feeding information into the brain which may help the development of more integrated bi-directional BMIs in the future.

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Poster

P537. Cortical Planning and Execution: Premotor and Motor Cortex: Neurophysiology: Animal II

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Topic: E.04. Voluntary Movements

Support: ERC Grant 678307
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Title: Largely shared neural codes for biological and nonbiological observed movements but not for executed actions in monkey premotor areas

Authors: *D. ALBERTINI[1], M. LANZILOTTO[2], M. MARANESI[1], L. BONINI[1];
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Abstract: The neural processing of others' observed actions recruits a large network of brain regions (the action observation network, AON), in which frontal motor areas are thought to play a crucial role. Since the discovery of mirror neurons (MNs) in the ventral premotor cortex, it has been assumed that their activation was conditional upon the presentation of biological rather than nonbiological motion stimuli, supporting a form of direct visuomotor matching. Nonetheless, nonbiological observed movements have rarely been used as control stimuli to evaluate visual specificity, thereby leaving the issue of representational similarity among executed actions and biological or nonbiological observed movements unresolved. Here, we addressed this issue by recording from two nodes of the AON that are attracting increasing interest, namely the dorsal premotor area F2 and the mesial pre-supplementary motor area F6 of macaques while they 1) executed a reaching-grasping task, 2) observed an experimenter performing the task, and 3) observed a nonbiological effector moving in the same context. Our findings revealed stronger neuronal responses to the observation of biological than nonbiological movement, but biological and nonbiological visual stimuli produced highly similar neural dynamics and relied on largely shared neural codes, which in turn remarkably differed from those associated with executed actions. These results indicate that, in highly familiar contexts, visuo-motor remapping processes in premotor areas hosting MNs are more complex and flexible than predicted by a direct visuomotor matching hypothesis.

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Poster

P537. Cortical Planning and Execution: Premotor and Motor Cortex: Neurophysiology: Animal II

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P537.07

Topic: E.04. Voluntary Movements

Support: NIH Grant K01 NS119372

Title: Local and long-range action selection mechanisms across a cortico-collicular circuit

Authors: *A. THOMAS[1], S. LAASYA TIPPARAJU[1], G. CHEN[1], W. YANG[1], K. SWIEKATOWSKI[1], M. TATAM[1], C. GERFEN[2], N. LI[1];
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Abstract: Prior to movement, competing motor representations are intermingled and distributed across a multi-regional circuit, including frontal cortex and superior colliculus (SC). Preparatory activity in these regions is considered a correlate of action selection, yet the circuit mechanisms mediating this process remain poorly understood. Traditional models posit that cortex selects upcoming behaviors, then sends a motor command to SC. However, recent data implicate SC feedback in action selection. We employed anatomical tracing, optogenetic manipulation and electrophysiological recordings to investigate action selection mechanisms in mice. Our anatomical tracing experiments reveal that the lateral SC receives direct input from ipsilateral frontal cortex (anterior lateral motor cortex, ALM) and projects back to ALM through medio-dorsal and ventral-medial thalamus, forming a loop. To define SC contributions to behavior, we performed optogenetic manipulation during a delayed response task. Animals used their whiskers to discriminate the position of an object during a sample period. After a delay period (1.3 s), the animals reported their choice via directional licking (left or right). Unilateral activation of SC evoked contralateral licking, and bilateral inactivation blocked licking altogether. Unilateral inactivation of the SC during the delay and response periods (but not the sample period) produced ipsilateral choice biases. Even transient inactivation of SC during the early delay was sufficient to bias upcoming licking, suggesting that the SC participates in the selection of future movements. Electrophysiological recordings revealed topographically organized action representations where SC preparatory activity is localized to the ALM projection zone. Intermingled neurons in both SC and ALM encoded competing representations for ipsilateral and contralateral licking prior to movement. Combined SC inactivation and ALM recording reveal that a local SC mechanism was sufficient to drive action selection dynamics across the ALM-SC loop prior to movement. Optrode tagging experiments within SC show that SC GABA neurons preferentially encoded ipsilateral licking and locally inhibited contralateral licking representations. Stimulating SC GABA neurons was sufficient to mediate competition between motor representations and bias upcoming behavior. Our findings argue against action selection models that relegate SC function to a motor relay. Instead, the SC is implicated as a locus for behavioral competition and a driver of cortical action selection dynamics.

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Poster

P537. Cortical Planning and Execution: Premotor and Motor Cortex: Neurophysiology: Animal II

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Program #/Poster #: P537.08

Topic: E.04. Voluntary Movements

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Title: Speed Prediction and Temporal Relationship Across Multiple Motor Association Areas During a Precision Reaching Task

Authors: *K. S. CHATLEY[1], A. G. ROUSE[2];
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Abstract: While many studies have examined how the cortex encodes movement by recording directly from neurons, most have recorded from only one or two cortical areas simultaneously and during similar magnitude movements. Less is known about how neural activity is coordinated across multiple motor-related areas during a wide range of movement speeds including low speed, precise movements. We recorded from four different cortical areas and used a linear regression model to predict speed from neural activity to assess their time preceding movement as well as in their information about the specific timing and magnitude of speed peaks.

Two *Rhesus Macaques* were trained on a precision center out reaching task that often required corrective movements. The animals were implanted with electrode arrays in primary motor (M1), dorsal premotor (PMd), ventral premotor (PMv), and Parietal Reach Region (PRR) to simultaneously record the neural activity as they performed the task. The timing and magnitude for neurally predicted speed was compared to the actual movement. A linear regression model was used to separately analyze the initial reach period on each trial and any subsequent corrective submovements.

First, the time lag with the best R-squared between movement speed and speed predicted from neural activity was identified. For all regions analyzed, corrective movements had a shorter time lag than initial movements. For corrective movements, the longest time lag was in PMd at 94ms-121ms, then M1 at 80ms-87ms, then PRR at 63ms-42ms while initial movements tended to have a lag of 10-30ms longer in each area. The timing of speed peaks was generally well predicted with a standard deviation of 50-75ms for all individual areas. The peak magnitude, however, was considerably smaller with predicted amplitude being only 40-85% of the true magnitude.

Linear regression models for speed prediction provide valuable information about neural activity across cortical areas. In this analysis, an important temporal relationship between multiple motor-associated areas is demonstrated with a progression from premotor to M1 then, surprisingly, to PRR. When comparing initial to corrective movements, a shorter time lag is observed with corrective movements. Additionally, this analysis demonstrates that the timing of peaks can be well predicted via a simple linear regression model, especially in initial peaks. Linear models fall short, however, in accurately predicting the magnitude of these peaks. Further refinement in these models and potentially machine learning algorithms may better delineate these relationships, especially with low speed corrective movements.

Disclosures: K.S. Chatley: None. **A.G. Rouse:** None.

Poster

P537. Cortical Planning and Execution: Premotor and Motor Cortex: Neurophysiology: Animal II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P537.09

Topic: E.04. Voluntary Movements

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Title: Complexity and high-dimensionality of motor cortex activity during a simple task

Authors: *E. AMEMATSRO, N. MARSHALL, E. TRAUTMANN, L. ABBOTT, M. M. CHURCHLAND;
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Abstract: Single-neuron responses in motor cortex (M1) are complex and heterogeneous, yet the nature and implications of that complexity have yet to be fully characterized. For a limited set of behaviors, a handful of dimensions is typically sufficient to capture most response variance. But are the smaller signals in higher dimensions effectively noise, or are they computationally relevant? Are the dimensions that contain large signals consistent across behaviors? How does cortical heterogeneity compare with that of the motoneurons?
We trained rhesus macaques to generate one-dimensional isometric forces with diverse temporal profiles. We recorded from 1257 M1 neurons using Plexon S-probes and primate-optimized Neuropixels. We recorded the activity of 134 motor units, allowing direct comparisons between two populations of spiking neurons (M1 and motoneuron). Consistent with prior findings, M1 responses were highly heterogeneous. Although motor output was simple (a 1D force), almost no neurons had activity resembling force or its derivative. While force could still be reliably decoded, the decoding dimension captured <5% of the response variance. Those dimensions that did capture considerable variance were behavior specific - e.g., they differed for slowly versus rapidly changing forces. Consequently, it took ~80 dimensions to capture 90% of the variance across all force profiles. Even dimensions past those 80 contained structure that was reliable across trials. Using a novel method, we estimated that the M1 population contained >300 independent signals. Decoding of simultaneously recorded motor units confirmed that low-

variance dimensions contained useful signals. Despite this complexity, M1 trajectory tangling remained low (far lower than for the motor units) due to structure in the higher-variance dimensions.
Thus, even when controlling a one-dimensional force, the motor cortex population response was remarkably complex and high-dimensional. This finding does not invalidate the use of low-dimensional projections, which often discriminate amongst model predictions. Yet our results argue that it is equally important for candidate models to account for the behavioral specificity of the high-variance features, and for the large number of reliable but low-variance features.

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Poster

P537. Cortical Planning and Execution: Premotor and Motor Cortex: Neurophysiology: Animal II

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Title: Network information flow in macaque motor cortex during manual interception

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Abstract: Interaction with the evolving world largely relies on dynamic interplay between sensory inflow and motor outflow in central nervous system, but network mechanism underlying dynamic sensorimotor control is still unclear. In the present study, we aim to reveal information flow between premotor cortex (PMd) and primary motor cortex (M1) from a network perspective. We calculated functional connectivity of neuronal networks in monkeys performing a manual interception task. The Utah microelectrode arrays were implanted in PMd and M1 of two macaques. Cross-correlation histograms (CCHs) between pairwise spike trains in task epochs were computed. Then, a cluster-based surrogate test was used to find significant peaks or troughs. Based on this analysis, functional connectivity matrices were calculated. Consistent with

previous studies, we found the small-world and scale-free (power-law distribution) characteristics of functional networks under our task. Since the complexity of computing the CCHs between the pairwise neurons is $O(n^2)$ and surrogate CCHs are 1000 times, a point process and GPU-based algorithm was developed and tremendously reduced calculation cost, allowing us to calculate dynamic functional networks under different task epochs. By comparing the directed functional network in the pre- and peri-movement periods, we found a significant increase in connectivity from PMd to M1, indicating a rise in information flow from PMd to M1 and a temporal hierarchy between the two areas. Furthermore, the connectivity from PMd to M1 during intercepting moving target is stronger than the reaching to the static target. Finally, calculations of the network entropy show a significant drop in PMd from pre- to peri-movement period for the static target condition, while M1 does not. In contrast, the network entropy of both brain regions was consistently maintained at a high level for moving targets. This indicates that the local functional network in PMd demands an enriched intrinsic information processing in dynamic environment.

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Poster

P537. Cortical Planning and Execution: Premotor and Motor Cortex: Neurophysiology: Animal II

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Wu Tsai Neurosciences Institute at Stanford

Title: Feedback control dynamics explain motor cortical activity

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Electrical Engin., [7]Bio-X Inst., [8]Departments of Bioengineering and Neurobio., Stanford Univ., Stanford, CA; [9]Bio-X Program, Stanford, CA

Abstract: Even simple reaching movements require sophisticated cortical computations that give rise to complex, spatiotemporally patterned muscle activity. Current theories of motor cortex suggest that these computations are carried out by a neural dynamical system that, once initialized during motor preparation, uses self-propagating dynamics to generate patterns of motor output. While this theory is attractive, it cannot yet explain a fundamental property of motor control: how sensory feedback is seamlessly integrated throughout a movement to correct unexpected errors (i.e., "feedback control"). Here, we combine the computation-through-dynamics perspective with a feedback control approach to propose a new model of how motor cortex controls movement. Critically, this model was enabled by new methods for training and dissecting high-fidelity neural network models that control simulated musculoskeletal arms. We identify three dynamical motifs that consistently emerged in neural networks trained to control arms: (1) integration dynamics that are used to counteract persistent visual and haptic perturbations, (2) oscillating dynamics that compensate for long feedback delays, and (3) gating dynamics that prevent goal information from reaching the feedback control subsystem when movement must be inhibited. In our model, preparatory neural dimensions serve as dynamically gated 'input' dimensions through which goal information flows into a low-level feedback control subsystem. This model reproduces many neural phenomena of recent interest, such as the existence of preparatory activity, the appearance of rotatory neural dynamics after the go cue, and small reaction time penalties when preparatory activity is disrupted - but it does so through neural mechanisms that are shaped by the need to perform robust feedback control in the presence of unpredictable perturbations and long feedback delays. Moving forward, we envision that high-fidelity neural network models of feedback control will play a key role in generating new hypotheses about neural mechanisms, understanding whether there are alternate explanations for observed neural phenomena, and designing experiments that are capable of distinguishing certain models from others based on neural recordings.

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Poster

P562. Posture and Gait: Higher Order Control, Multi-Task Integration, and Theory

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P562.01

Topic: E.06. Posture and Gait

Title: Continuous and difficult cognitive tasks do not improve gait control compared to a discrete and easier task in healthy young adults

Authors: *L. MICHAUD, J. DENNIS, C. LEBEAU, M. PELLETIER, A. ZEVNIK, Y. LAJOIE;
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Abstract: Background**:** It is believed that gait is not purely automatic since multiple studies found a decrease in gait performance (slower, more variable, etc.) under dual task conditions which might indicate a competition for attentional resources (Yogev et al., 2005). However, based on the constrained action theory (Wulf, 2013), dual task conditions could in fact enhance motor performance due to a shift of attention away from the primary task (e.g. walking) that promotes a more automatic and efficient control. In a recent study on quiet standing, it was observed that continuous and more difficult cognitive tasks as secondary tasks were better at improving postural control compared to discrete and easier cognitive tasks since more engaging secondary tasks might favor a shift of attention and, thus, automaticity (Lajoie et al., 2016). The objective of this study was to assess if automaticity could be promoted for walking using continuous and more difficult cognitive tasks as secondary tasks.
Method**:** Twenty healthy young adults (25.3 ± 2.8 years) were recruited. The experiment involved walking in a straight line for eight meters while performing different cognitive tasks or no secondary task (Baseline). The tasks were: a serial reaction time (SRT), counting backward by seven (7down) and identifying the total number of times that two predetermined digits occurred during a sequence of three-digit numbers (Recall). Each conditions was performed five times. Gait speed and stride variability and length were recorded using 3-D kinematic data from markers on the right and left foot with the Vicon512^{TM} System at 200 Hz. Two-way repeated ANOVAs were computed to assess changes between leg side (left and right) and the four conditions (Baseline, SRT, 7down and Recall).
Results**:** Statistical analysis revealed a main effect of condition for gait speed and stride length. Gait speed was reduced during 7down and Recall compared to SRT and Baseline. Stride length was also reduced during 7down compared to all other conditions and during Recall compared to Baseline and SRT.
Conclusion**:** The absence of changes for stride variability and a decrease in gait speed and stride length for 7down and Recall suggest that more difficult and continuous secondary tasks interfered with the primary task of walking. Thus, our results do not support that more engaging tasks promote a more automatic gait control.

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Poster

P562. Posture and Gait: Higher Order Control, Multi-Task Integration, and Theory

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Program #/Poster #: P562.02

Topic: E.06. Posture and Gait

Title: Prefrontal cortex activity during gait in the recently concussed adults

Authors: *D. MARTINI[1], M. MANCINI[2], R. VITORIO[3], L. A. KING[2];
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Abstract: A concussion results in transient symptoms (e.g. cognitive, somatic, effective) that are tied to the metabolic energy crisis stemming from the need to reestablish neurophysiological homeostasis. This post-concussion symptomatic period is associated with gait dysfunction, the cortical underpinnings of which are poorly understood. Assessing prefrontal cortex activity during gait may provide insight into the altered neurophysiology during gait in the concussion recovery phase. The purpose of this preliminary study is to quantify gait and prefrontal cortical activity in symptomatic individuals with a recent concussion. We hypothesize that worse symptoms will relate to increased cortical activity, and that increased cortical activity will correlate with worse gait performance. We present data from 7 symptomatic, recently concussed (32 years old; 5 female; 36 days from concussion (min = 12; max = 66)). The Neurobehavior Symptom Inventory (NSI) was used to characterize the self-reported number and severity of concussion symptoms. The NSI provides a somatic, affective, and cognitive symptom sub-score, with a total possible score of 88; higher scores are worse. Prefrontal cortex activity was quantified using a portable functional near-infrared spectroscopy system, which uses near-infrared light to determine changes in concentration of oxygenated (HbO2) and deoxygenated hemoglobin. Gait was quantified using 6 inertial sensors (2 feet, 1 lumbar, 1 sternum, 2 wrists), which provide spatiotemporal and turning variables of gait. Participants walked for 80 seconds, back and forth over a 9-m straight path with a 180° turn at each end. The average NSI total score was 19.1 (s.d. = 11.1). The HbO2 during gait significantly correlated with the NSI total symptom score ($r = 0.93$; $p = 0.002$), NSI somatic score ($r = 0.80$; $p = 0.03$), NSI affective score ($r = 0.86$; $p = 0.01$), and NSI cognitive score ($r = 0.94$; $p = 0.002$). Further, the HbO2 during gait significantly related to the medial-lateral lumbar range of motion ($r = 0.83$; $p = 0.02$), anterior-posterior lumbar range of motion ($r = 0.79$; $p = 0.03$), and turn velocity ($r = 0.89$; $p = 0.007$). These preliminary data provide the first evidence of prefrontal cortex activity being related to symptom severity and gait performance in the recently concussed. Quantifying cortical activity during gait in this population may provide greater insight into identifying individuals more likely to have a prolonged recovery timeline. Future work needs to replicate these preliminary results in a larger group of recently concussed and compare gait related changes in HbO2 to a control group to verify the changes in HbO2 observed herein are concussion-specific.

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Poster

P562. Posture and Gait: Higher Order Control, Multi-Task Integration, and Theory

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Program #/Poster #: P562.03

Topic: E.06. Posture and Gait

Title: Synergic relations between eye and postural movements in searching to locate objects in room inside houses

Authors: *C. T. BONNET[1], J. A. BARELA[2], T. SINGH[3];
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Abstract: Although people continuously sway in the standing position, they still succeed in visually exploring their environment and performing precise visual tasks efficiently. Based on this knowledge, our team published a synergic model of postural control in 2016 suggesting that the brain should continuously adapt postural control to the precision and difficulty of performed gaze shifts. In the present study, our objective was to test the existence of behavioural synergic relations between eye and postural movements in a goal-directed, precise, visual search task (locate target objects in large images). Twenty young participants performed a search task and a control free-viewing task. The method was rigorous with calculation of the sample size and the use of a validated, already published, methodology to analyse data; this was a confirmatory study. Negative correlations between the recorded eye and postural movements were assumed to show synergic, stabilizing, relations. In fact, they showed that postural sway was better reduced to perform and succeed in longer, more difficult, gaze shifts. In other words, they showed that the further the eyes moved, the less people swayed. In contrast, positive correlations were assumed to show destabilizing relations for the opposite reason. Our results showed that young adults exhibited significant negative correlations between eye and postural (head and upper back) movements in the precise visual task. They also did not exhibit any significant correlation in the free-viewing task. In conclusion, these results confirmed that the brain adjusts postural control online to succeed in subtle and continuous precise gaze shifts.

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Poster

P562. Posture and Gait: Higher Order Control, Multi-Task Integration, and Theory

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Topic: E.06. Posture and Gait

Support: Temple University CPH Visionary Research Fund

Title: The impact of attention on sensory integration for postural control as characterized by behavioral and cortical changes.

Authors: *L. MA[1], P. J. MARSHALL[2], W. G. WRIGHT[1];
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Abstract: Postural control requires accurate integration of multiple senses to maintain body orientation and equilibrium. However, it is unclear how changes in focus of attention and instruction may influence the underlying sensorimotor mechanisms for postural control. By studying how directing attention can influence sensory processes and postural control, we can better understand how to improve balance and prevent falls. The present study investigates how different types of attentional focus specifically affect visual integration in maintaining posture. Fifteen healthy adults (>20 years old) underwent an experimental paradigm where they balanced on a rocker board while wearing a virtual reality head-mounted display (HMD) and a 32-channel electroencephalography (EEG) cap. The HMD displayed a virtual world of standing on a boat that tilted from side to side in the mediolateral direction at a frequency of 0.2Hz and amplitudes of 2 ± 0.2 degrees. All participants were first given the instruction of looking straight ahead and keeping their body as still as possible (control condition). Using random assignment, participants were then either instructed to focus on keeping the rocker board flat to help them keep still (external focus condition; n=8) or instructed to focus on keeping both feet leveled to each other to help them keep still (internal focus condition; n=7). The dependent variables were the EEG alpha power response (9-13Hz) of independent components clustered in the occipital cortex and the gain response of the participant's postural sway relative to the 0.2Hz visual stimulus. Results revealed a significant ($p<0.05$) increase in alpha power over the occipital cortex during both external and internal focus conditions when compared to control condition, with external focus showing a trend towards higher alpha power than internal focus. This suggests that both types of focus may influence brain responses to visual input during postural control. Interestingly, participants instructed to focus externally showed a significant decrease in visual influence on postural sway relative to the control condition, while participants instructed to focus internally showed no change. This suggests that external focus may be better than internal focus at resolving sensory mismatches for postural control. Findings from this study may help guide clinicians and researchers on how best to provide instructions to direct attention during balance control and explain why external focus has been shown to be more effective for balance performance in prior studies.

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Poster

P562. Posture and Gait: Higher Order Control, Multi-Task Integration, and Theory

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Topic: E.06. Posture and Gait

Support: American Heart Association Fellowship 13PRE14780094

Title: Visual shift in normal standing affects the direction of loss of balance during perturbation.

Authors: *B. TAHAYORI[1], D. M. KOCEJA[2], B. TAHAYORI[3];
[1]Univ. of St. Augustine, Coral Gables, FL; [2]Indiana Univ., Bloomington, IN; [3]Florey Dept. of Neurosci. and Mental Hlth., The Univ. of Melbourne, Melbourne, Australia

Abstract: Controlling upright posture entails acute adjustments by the neuromuscular system to keep the center of mass (COM) within the limits of a relatively small base of support. Several sensory systems interact with each other to provide accurate information for maintaining upright posture. In this experiment we shifted the visual field of 15 healthy, young individuals by using a 25 diopter prism goggle, and recorded their Centre of Pressure (COP) movement. While subjects were standing, we stimulated the posterior tibial nerve by a constant current stimulator and thereby, simulated a perturbation or loss of balance. We therefore, tested the balance reaction of our subjects in 4 different conditions of normal vison (NMV), no vision (NOV), visual shift to the right (VSR) and visual shift to the left (VSL). With each nerve stimulation, the COP would follow a relatively consistent pattern of immediate frontward displacement followed by backward movement and several oscillations to bring the body back to stability. When stimulating the right leg, COP would deviate to the right and vice versa, when stimulating the left leg, COP would move toward the left. This was manifested by calculating a vector for each COP displacement (Vector origin as the location of COP just before stimulation and endpoint as the deflection in COP trajectory).COP deviation during normal vision, caused on average 16.7±2.9 degrees relative to midline. In the VSL condition, left side stimulation caused 20.12±1.5 degrees of deviation. In VSR condition, stimulating the right leg caused 15.8±2.1 shift to the right. Visual information is a major sensory source for maintaining balance during quiet standing. However, these findings suggest that during loss of balance, visual sensory input plays an important role in the recovery mechanism of regaining balance. It was shown that a change in the directional change of visual information proportionally affects the direction of COP trajectory displacement. Understanding the neural and mechanical events occurring during loss of balance (LOB) are helpful to understand the underlying mechanisms of balance control, loss of balance and regaining balance after a perturbation.

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Poster

P562. Posture and Gait: Higher Order Control, Multi-Task Integration, and Theory

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Program #/Poster #: P562.06

Topic: E.06. Posture and Gait

Support: PDE_1_2019
20020170100729BA

Title: Uncovering hidden organization of apparent random fluctuations in human gait

Authors: J. MAZZEO[1], F. WAGNER[1], G. CAMPIGLIO[1], *V. DELLA-MAGGIORE[2]; [1]Inst. de Ingeniería Biomédica, Univ. de Buenos Aires, Buenos Aires, Argentina; [2]IFIBIO Houssay, Sch. of Medicine, UBA, Buenos Aires, Argentina

Abstract: The classical model of gait describes it as a quasi periodic process based on “cycles”. In this framework, differences between cycles or "fluctuations" are considered to lack information and, thus, are often disregarded. The present study focused on the analysis of these apparently random fluctuations under the hypothesis that they can characterize operating modes of the motor control system that remain hidden under the classical view. From the theoretical framework of stochastic processes, here we analyzed the variability of step times (ST) obtained experimentally under four different conditions: free gait, treadmill, metronome, and obstacles. Based on ST, we estimated the Hurst index (H) or "scaling index", a metric of long-term memory in time series, which reflects the statistical dependence of steps as the lag between them increases from short (3-10 steps) to long (300-400 steps) time scales. An H index of 0.5 would be consistent with the occurrence of random fluctuations in ST, whereas H> 0.5 would reflect the presence of positive autocorrelation between ST, and H<0.5 would reflect the presence of negative autocorrelation. We obtained continuous 40 min recordings of foot pressure under the heel from normal subjects, while they walked in a gym under the four conditions described above. Preliminary data yielded differences in the H index depending on the experimental condition ($p = 0.01$). Free gait, obstacle gait and treadmill gait were characterized by positive autocorrelation (mean = 0.79, SD = 0.049, $p < 0.01$) both at short and long time scales. This finding is consistent with a long-term memory process. In contrast, metronome gait was also characterized by positive autocorrelation at short-time scales (mean = 0.68, SD = 0.01), but by negative autocorrelation at long time scales (mean = 0.29, SD = 0.02). The latter may reflect the fact that as subjects adjust their gait to the pace of the metronome they produce ST that fall either above or below its period. Interestingly, however, the operation at the lowest temporal scales remained similar to that of the free gait. Our preliminary findings may reflect the existence of two different endogenous mechanisms controlling gait at the short-and long time scales.

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Poster

P562. Posture and Gait: Higher Order Control, Multi-Task Integration, and Theory

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Program #/Poster #: P562.07

Topic: E.06. Posture and Gait

Title: Step length synergy is reduced with increasing demands on foot placement during adaptive gait

Authors: *A. KULKARNI, C. CUI, S. RIETDYK, S. AMBIKE;
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Abstract: During community ambulation, forward motion and stability are maintained while gait is adapted to navigate hazards. In order to maintain stability, foot placement is modified to control the center of mass (COM) motion. However, foot placement must also be modified to avoid tripping on a hazard. Previously, we have shown that the step length around an obstacle is stabilized (i.e., *step length synergy* exists), indicating that foot placement is controlled not only with reference to the obstacle but also to stabilize the step length, which in turn controls the COM motion. In the current work, we increased the foot placement demands by adding step targets while approaching the obstacle. These targets allow us to determine how foot placement demands affect the step length synergy, indicating priorities of the nervous system in challenging environments. Fifteen healthy participants (24$\pm$3 yrs) walked on a 6 m walkway and stepped over an obstacle (25% of leg length, about 24 cm high). Conditions included baseline (no target) and five target locations projected onto the walkway (1, 2, 3, 4, or 5 steps before obstacle). When a target was present, participants stepped on the target and continued walking. The across-trial variance in the horizontal distances between trail and lead heel from the obstacle for the crossing step was analyzed using the uncontrolled manifold analysis to determine step length synergy. We hypothesized that (1) foot targets will reduce the step length synergy, and (2) targets closer to the obstacle will lead to greater reduction in synergy. A weaker synergy indicates greater priority for foot placement relative to external cues, compared to step length stability. We observed that the step length was stabilized for all target locations ($p < .01$). That is, the front and back heel distances relative to the obstacle's edge co-varied synergistically for all target locations. However, the strength of the step length synergy was affected by target location ($p = .01$). Synergies were up to 66% lower when the step target was closer to the obstacle, as hypothesized with an exception: when the target was two steps before the obstacle, the step length synergy was not different from baseline. This exception may be related to the observation that people visually fixate on the obstacle to gather obstacle position information when they are two steps away from the obstacle; this will be examined further in the future. Overall, young adults responded to increased challenges by prioritizing foot placement relative to external cues over stabilizing step length. Yet, they maintained step length synergies, demonstrating the relevance of this strategy during adaptive gait.

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Poster

P562. Posture and Gait: Higher Order Control, Multi-Task Integration, and Theory

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Topic: E.06. Posture and Gait

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Title: The rat hindlimb sensorimotor cortex encodes for postural perturbations after complete spinal cord injury

Authors: *G. DISSE[1], J. B. DOUGHERTY[2], N. R. BRIDGES[3], K. A. MOXON[4];
[1]Univ. of California Davis, Sacramento, CA; [2]Biomed. Engin., [3]Sch. of Biomed. Engin. Sci. and Hlth. Systems, Drexel Univ., Philadelphia, PA; [4]UC Davis, Davis, CA

Abstract: While descending cortical signals are not critical for maintaining postural control in the normal animal, the role of the cortex after spinal cord injury is unknown. To assess how the cortex responds to unexpected postural perturbations, adult rats were placed on a tilting platform, and the firing patterns of single neurons in the hindlimb sensorimotor cortex were recorded with microelectrode arrays as the platform rotated with different tilt profiles (e.g. varying velocities, maximum angles, directions, and durations). For a subset of animals, limb ground reaction forces were recorded as well throughout the tilts. This procedure was repeated 1-3 weeks after animals underwent a complete mid-thoracic spinal cord transection. Confirming previous studies, neurons in the hindlimb sensorimotor cortex of pre-injury animals fired in response to tilts and the magnitude and timing of the response was modulated with different tilt profiles. Additionally, significant mutual information existed between cortical neuron firing patterns and tilt types, suggesting that the cortex conveys significant information about postural perturbations. After an injury severing all ascending and descending tracts at the mid-thoracic spinal cord, clear behavioral deficits were observed in the limb ground reaction forces. However, the cortex continued to display a similar pattern of response modulation to different tilts -albeit with a slight increase in latency. Additionally, there was a decrease in mutual information after injury. Despite this, it remained significantly above random information as estimated through bootstrapping. These results suggest that the cortex not only continues to respond to and encode for postural perturbations after SCI, but that it conveys enough information to discriminate tilt types from both stance (tilt sensitivity) and other tilts (tilt specificity). This is likely due to remaining forelimb and trunk afferents from above the lesion, and suggests that the cortex is an viable target for restoring independent, functional movements after spinal cord injury.

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Poster

P562. Posture and Gait: Higher Order Control, Multi-Task Integration, and Theory

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Title: Stride-by-stride analyses of anterior cingulate cortex activity during perturbed recumbent stepping

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Abstract: Theta band (3-8 Hz) synchronization in the anterior cingulate cortex is commonly associated with error monitoring during motor learning. During seated recumbent stepping, applying brief increases in stepping resistance during each stride perturbs the stepping motion, generating an error. In healthy young adults, these perturbations elicited increased anterior cingulate theta spectral power around the onset of the perturbation. In this study, we performed stride-by-stride analyses on the motor error and anterior cingulate cortex responses from our previous perturbed recumbent stepping study with young adults (n=17). In our experimental protocol, there were 4 tasks based on the perturbations being applied, which was either at right/left either leg extension onset or mid-extension (4 tasks = 2 legs x 2 timings). There were also “catch” trials where no perturbation was applied. These catch trials occurred randomly once within every 5 strides. The stride-by-stride approach allowed us to explore whether increasing predictability influenced anterior cingulate responses. We hypothesized that the maximum anterior cingulate theta power and motor errors (pacing errors and stepping errors) would decrease as perturbations became more predictable. We computed the theta-band event related spectral power (ERSP) for each stride and binned each stride relative to a preceding catch stride as catch+1, catch+2, catch+3, etc. Perturbations more distant from the catch are more predictable as they are preceded by more perturbations. For the group averaged theta-band ERSPs for left and right mid-extension and left extension onset perturbations, the perturbed stride immediately after the catch, catch+1, the unexpected and less predictable stride had the largest peaks. Individual subject linear fits of the average maximum theta-band ERSP on bin positions, however, showed that only 6 out of 14 subjects had a negative slope in each perturbation, suggesting that more predictable perturbations did not correlate with less anterior cingulate theta power in most subjects. Individual subject linear fits of the average stepping error on bin positions showed that most of the subjects had a negative slope, suggesting that stepping errors decreased as perturbations became more predictable. These preliminary results suggest that anterior cingulate activity does not reliably correlate with perturbation predictability, even though stepping errors decreased with increasing perturbation predictability.

Disclosures: L. Richardson: None. **S. Shirazi:** None. **H.J. Huang:** None.

Poster

P562. Posture and Gait: Higher Order Control, Multi-Task Integration, and Theory

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P562.10

Topic: E.06. Posture and Gait

Title: Vision and walking: how reliance on feedback and feedforward visual information changes during locomotor learning

Authors: *A. CATES[1], J. DEMBSKY[1], K. E. GORDON[1,2];
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Abstract: Vision plays a vital role in executing controlled walking. Feedforward fixations (i.e. fixations greater than 1.5 steps ahead) are used to make feedforward motor plans while feedback fixations (i.e. fixations less than 1.5 steps ahead) are used to make corrective actions to the current step. The present study examined how healthy young adults (n=9) change their reliance on feedback and feedforward visual information during a locomotor learning task. Participants completed a treadmill-based target stepping task in which they were asked to accurately step on small moving targets projected onto the treadmill surface. To determine an individual's reliance on feedforward and feedback visual information, we compared stepping performance across 3 different visual conditions: full vision, no feedback vision where the targets were not visible within 1.5 steps of the individual, and no feedforward vision where the targets were not visible greater than 1.5 steps away from the individual. Visual reliance was defined as the decrease in step accuracy when the targets related to feedforward or feedback visual information were removed. We hypothesized that participants would rely more on feedback information during their initial exposure to the novel walking task and shift to a greater reliance on feedforward information as they learned the task. Our results do not support our hypothesis. Instead, we found that participants increased reliance on feedback information as they improved at the task while their reliance on feedforward visual information remained unchanged. One possible explanation may be an increased emphasis on the temporal coordination between vision and locomotor execution. Specifically, previous research has shown better target stepping performance by fixating on the target until heel strike. Participants may have adopted a similar strategy as their performance improved in the present study. Future research should focus on the interplay of spatial and temporal relationships between gaze and walking.

Disclosures: A. Cates: None. **J. Dembsky:** None. **K.E. Gordon:** None.

Poster

P562. Posture and Gait: Higher Order Control, Multi-Task Integration, and Theory

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P562.11

Topic: E.06. Posture and Gait

Support: NRF-2020R1A2C3010304
IITP-2021-2020-0-01821

Title: Changes in Cortical Activity during Preferred and Fast Speed Walking under Single and Dual Task in the Young-old and Old-old

Authors: *J. KIM[1,3], J. KIM[3], E. KIM[3,2], S.-H. LEE[3], G. LEE[3], J. LEE[3], S. SHIN[3], W. CHANG[3], Y.-H. KIM[1,3,2];
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Abstract: In the elderly, walking performance starts to decline with aging. Furthermore, walking while simultaneously engaging in other activities become more and more difficult with severe aging. This study aimed to examine the changes in cortical activity during walking with aging. We especially try to reveal differences of cortical activities during the dual task walking between the young-old and old-old persons. Fifty elderly participants without history of neurological or psychiatric symptoms participated in this cross-sectional study. Prior to the experiment, evaluations such as the short physical performance battery (SPPB), the four-square step test (FSST), the timed up and go test (TUG), the 10-meter walking test (10MWT) were performed to assess function. Each subject completed four walking tasks on the treadmill, 2x2 design; two normal walking conditions with self-selected walking speed (NW-SSWS) and fast walking speed (NW-FSW), and two dual task walking conditions with self-selected walking speed (DTW-SSWS) and fast walking speed (DTW-FWS). The fast speed was set to 130% of the self-selected speed and for the dual task, the phonemic verbal fluency task was given. Functional near-infrared spectroscopy (fNIRS) was applied for the measurement of regional cerebral oxyhemoglobin (oxyHb) concentration during each walking condition. The fNIRS data were analyzed using the statistical parametric mapping (SPM) software and time-series analysis. Region of interests were the primary leg motor cortex (M1-leg), the pre-motor area (PM), the supplementary motor area (SMA), the posterior parietal cortex (PPC), and the dorsolateral prefrontal cortex (dlPFC). Twenty-seven participants were divided into the young-old (under 75-yrs old; 70.2 ± 3.0 years) and 23 into the old-old (over 75 yrs old; 78.0 ± 2.3 years) groups. There were significant functional differences between the two groups in the SPPB, the FSST, the 10MWT, and the TUG. In the SPM analysis, cortical activities were increased during the two DTW conditions than the NW conditions, but decreased during the two FWS conditions than the SSWS conditions on the bilateral M1-leg, the SMA, and the dlPFC in both old groups. These oxyHb

concentration changes were significantly less prominent in the old-old group than in the young-old group, which reflected diminished adaptive plasticity of cortical activity with severe aging. This study demonstrated that degree of cortical activity changes during the dual task walking was decreased in the old-old than the young-old group. These different profiles between the two groups of elderly people can explain impaired compensation strategies and increased fall risk with aging.

Disclosures: J. Kim: None. **J. Kim:** None. **E. Kim:** None. **S. Lee:** None. **G. Lee:** None. **J. Lee:** None. **S. Shin:** None. **W. Chang:** None. **Y. Kim:** None.

Poster

P562. Posture and Gait: Higher Order Control, Multi-Task Integration, and Theory

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P562.12

Topic: E.06. Posture and Gait

Support: NSF CRCNS 1822568

Title: Flexible Recruitment of Balance Mechanisms to Environmental Constraints During Walking

Authors: T. D. FETTROW[1], **S. DIBIANCA**[2], F. VANDERLINE DOS SANTOS[3], J. REIMANN[2], J. JEKA[2];
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Abstract: Flexible Recruitment of Balance Mechanisms to Environmental Constraints During Walking
Authors: T. Fettrow, S. DiBianca, F. Vanderlinde dos Santos, H. Reimann, and J. Jeka
Disclosures:Tyler Fettrow, None**. Stephen DiBianca,** None**. F. Vanderlinde dos Santos,** None**. Hendrik Reimann,** None**. John Jeka,** None**.**
Abstract: Walking is a complex task, especially in an environment that constrains a person to where they can step. Constant active control must be modulated to maintain an upright and balanced position. We do not have a comprehensive theory for how humans regulate balance during walking especially when there is awareness of a known navigational restraint. We have previously identified three balance mechanisms utilized by healthy young adults to maintain bipedal locomotion: 1) The ankle roll mechanism, a modulation of ankle inversion/eversion; (2) The foot placement mechanism, a shift of the swing foot placement; and (3) The push-off mechanism, a modulation of the ankle plantarflexion angle during double stance. This study seeks to observe changes in this neural control paradigm when presented with an environmental constraint. Twenty healthy young subjects walked on a self-paced treadmill while immersed in a virtual environment displayed via an Oculus Rift. The virtual scene provided three different

colored pathways. Subjects were instructed not to step in the No-Step Zone, which appeared either on the right or left side of the subject. While walking, subjects received balance perturbations in the form of galvanic vestibular stimulation, providing the sensation of falling sideways, either toward the No-Step zone or toward the Neutral zone on the other side. The results indicate that the use of the balance mechanisms are altered depending on whether the perceived fall is toward the No-Step or the Neutral zone. Participants increased the use of the lateral ankle mechanism and foot placement mechanisms for No-Step stimuli resulting in an overall larger shift of the center of mass (CoM) compared to the Neutral stimuli. This difference proposes a supraspinal tuning bias of the balance response imposed by the external constraint. Although a stronger CoM shift is expected for a response away from a perceived constraint, we expected this shift to be generated by the ankle and push-off mechanisms. We hypothesized that perceived falls in the direction of the No-Step zone would constrain the foot placement mechanism, resulting in a decoupling of the balance mechanisms. Instead, this non-intuitive increase in foot placement suggests that an overall balance response is driven via supraspinal mechanisms, which is coordinated throughout the gait cycle.
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Disclosures: T.D. Fettrow: None. **S. DiBianca:** None. **F. Vanderline Dos Santos:** None. **J. Reimann:** None. **J. Jeka:** None.

Poster

P562. Posture and Gait: Higher Order Control, Multi-Task Integration, and Theory

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P562.13

Topic: E.06. Posture and Gait

Title: Cortical brain dynamics associated with gait adaptation to an active ankle exoskeleton

Authors: *J. BRADFORD[1], S. SONG[1,2], C. HAYNES[1];
[1]US DEVCOM Army Res. Lab., Aberdeen Proving Ground, MD; [2]Texas A&M Univ., College Station, TX

Abstract: Adaptive exoskeletons have been shown to reduce the metabolic cost of walking in some individuals. While this is an important metric for exoskeleton performance, little research has been performed to understand how the human brain and body respond and adapt to these devices. The goal of this study was to uncover cortical brain signals related to the user's adaptation to an autonomous, bilateral ankle exoskeleton. 22 subjects walked on a treadmill while wearing a powered bilateral ankle exoskeleton that assisted with plantarflexion at toe off. We measured EEG, EMG, kinematics, and ground reaction forces during early and late adaptation periods. We found reduced alpha and beta band power in sensorimotor brain areas

during late adaptation. We found that these changes in brain dynamics corresponded with reduced EMG activity and reduced step width during late adaptation.

Disclosures: J. Bradford: None. **S. Song:** None. **C. Haynes:** None.

Poster

P562. Posture and Gait: Higher Order Control, Multi-Task Integration, and Theory

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P562.14

Topic: E.06. Posture and Gait

Support: NSF CRCNS 1822568

Title: A neuromuscular model of human locomotion that combines flexibility and stability

Authors: *R. RAMADAN[1], G. SCHONER[2], J. J. JEKA[3], H. REIMANN[4];
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Abstract: Human locomotion is amazingly flexible. We walk effortlessly in varied environments, choose gaits and avoid obstacles of different sizes and shapes. Achieving that flexibility requires sophisticated mechanisms of stabilization as the human body is mechanically unstable during locomotion and the body's configuration changes throughout thegait-cycle. Combining flexibility and stability, therefore, is a challenge. Avoiding an obstacle, for instance, involves not only steering the foot over the obstacle, but also stabilizing the gait pattern against the perturbation caused by the avoidance maneuver. This requires estimation of the state of the body in space from sensory information and updating the movement plan by adjusting motor commands that integrate constraints from biomechanics, muscle dynamics and reflex pathways of the spinal cord. Here, we present a neural process model of human locomotion that is able to generate updatable movement plans for steady state locomotion. The model organizes movement plans at the task level. During swing, vestibular feedback is used to determine a target joint angle configuration for foot placement on each step that generates stable walking. The movement plan to reach that target configuration is generated as a minimal jerk trajectory and updated during movement based on sensory information. The supraspinal controller is based on an inverse internal model of the biomechanics, muscles, and spinal reflexes and generates descending commands that implements the task-level movement plan. At the spinal level, the descending commands are integrated with the stretch reflex during the swing phase. Stance leg control relies on dedicated spinal reflex pathways that are adopted from [1].The biomechanical model comprises 8 DoFs, realistic moment arms, and 22 Hill-type muscles. Muscles are innervated by α-motorneurons and are controlled by descending activations and reflex circuits. Our model is

able to combine stable locomotion at different speeds with varied movements of the swing leg, that enable stepping over obstacles of different heights and to resist external pushes. The swing leg by itself can execute arbitrary movements within the reachable workspace.
[1] Song, Seungmoon, and Hartmut Geyer. "A neural circuitry that emphasizes spinal feedback generates diverse behaviours of human locomotion."

Disclosures: R. Ramadan: None. **G. Schoner:** None. **J.J. Jeka:** None. **H. Reimann:** None.

Poster

P587. Brain Circuits Underlying Social Behavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P587.01

Topic: F.02. Behavioral Neuroendocrinology

Support: NIMH (K99/R00)

Title: Neural circuits for social behavior adaptations to environmental temperature.

Authors: *Z. ADAHMAN[1], R. OYAMA[1], T. ABE[2], J. S. RICEBERG[3], I. CARCEA[1]; [1]Pharmacology, Physiol. and Neurosci., Rutgers, The State Univ. of New Jersey, Newark, NJ; [2]Neurosci. and the Mortimer B. Zuckerman Mind Brain Behavior Inst., Columbia Univ., New York, NY; [3]Neurosci., Mount Sinai Sch. of Med., New York, NY

Abstract: Thermoregulation is essential for survival. In mammals, changes in environmental temperature induce both autonomic and behavioral thermoregulatory responses. Particularly in the case of maternal behavior, environmental temperature influences the dams' behavior response in providing maternal care to poikilothermic neonates. We characterized these changes in dams at several levels: nest building, pup retrieval, and co-housing with another adult female and pups. Compared to room temperature, dams build larger nests in the cold and smaller nests in warm temperature (nest area at room temperature is 69.53±6.48 cm^2, cold temperature is 85.77±7.51 cm^2, and warm temperature is 38.38±2.75 cm^2, p=0.0004, N=16). Although dams retrieve acutely isolated pups similarly at all temperatures, during prolonged episodes (4 hours) at warm temperature they let pups disperse (average area covered by pups dispersed at room temperature is 84.78±2.95 cm^2, cold temperature is 35.51±1.70 cm^2, and warm temperature is 138.10±8.31 cm^2, p for time=0.14, p for temperature = 0.0003, p-value of time and temperature interaction=0.024, N=3). In the co-housing assay, dams and non-maternal females cooperate to spend more time together in nest at cold temperature (CT), compared to room (RT) and warm temperatures (WT) (p-value is 0.0001, 0.0001 and 0.02 for CT versus RT, CT versus WT, and RT versus WT respectively).
To understand the neural circuits controlling temperature-modulated social behavior, we investigated the activity of hypothalamic neurons that secrete the vasopressin neuropeptide.

Vasopressin plays important roles in maternal care, thermoregulatory processes, and other social behaviors. Using an activity reporter mouse line, we find elevations in ambient temperature activate vasopressin neurons in the paraventricular nucleus of the hypothalamus. Synaptic anterograde tracing from vasopressin neurons shows connections with structures implicated in thermoregulatory responses (lateral parabrachial nuclei), maternal behavior (periductal gray area), and other social behaviors (cingulate cortex, ventral tegmental area, dorsal raphe nuclei). Taken together, we hypothesize that warmer environmental temperatures control several components of social aggregation and nest-building by modulating the firing of vasopressin hypothalamic cells. Using a combination of activity-dependent gene expression and retrograde tracing, we will investigate the neural circuit mechanisms by which temperature information might reach vasopressin cells, and the extent to which these circuits control changes in social behaviors response to environmental temperature.

Disclosures: Z. Adahman: None. **R. Oyama:** None. **T. Abe:** None. **J.S. Riceberg:** None. **I. Carcea:** None.

Poster

P587. Brain Circuits Underlying Social Behavior

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Program #/Poster #: P587.02

Topic: F.02. Behavioral Neuroendocrinology

Support: NIH award DP2OD026143
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Dana Foundation

Title: Interbrain correlation in prairie voles scales with degree of social interaction and pair bond status

Authors: *A. D. MCTIGUE[1], K. MURPHY[1], *L. E. BRUSMAN[1], Z. R. DONALDSON[2]; [1]Psychology and Neurosci., Univ. of Colorado Boulder, Boulder, CO; [2]MCBD/Psychology & Neurosci., Univ. of Colorado, Boulder, Boulder, CO

Abstract: Simultaneously recording neural activity from multiple individuals provides an opportunity to ask how the neural basis of complex behavior is shaped in real time during evolving social interactions. Previous work in humans has shown that, during interaction, oscillatory neural activity becomes synchronized. The degree of synchrony between two individual scales with relational variables - with romantic partners exhibiting stronger synchrony than individuals who do not know each other. This phenomenon, known as interbrain correlation, is not limited to humans, and has been documented in mice and bats. However, these species do not form the exclusive, monogamous pair bonds that are central to human experience. Here, we

asked whether interbrain correlation is observed in monogamous prairie voles and whether the correlation scales as a function of pair bonding. Using fiber photometry and GCaMP, a molecular sensor for calcium and proxy for neural activity, we found consistently increased interbrain correlations when two animals are interacting compared to when they are separated by a divider. The degree of correlation further scales with social engagement with the strongest correlations occurring during active dyadic interactions. Finally, we found that pair bonded animals exhibited stronger interbrain correlation with their partner than a novel animal. These data indicate that prairie voles provide an ideal model for examining the role of interbrain correlation in mediating attachment.

Disclosures: A.D. McTigue: None. **K. Murphy:** None. **L.E. Brusman:** None. **Z.R. Donaldson:** None.

Poster

P587. Brain Circuits Underlying Social Behavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P587.03

Topic: F.02. Behavioral Neuroendocrinology

Support: Biobehavioral Health Department

Title: Posterior hippocampus stress-regulating gene expression is associated with rodent behavioral inhibition and social boldness

Authors: E. K. MCMAHON[1], ***S. FARHAN**[3], *S. A. CAVIGELLI[2];
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Abstract: Temperament suggests consistent individual responses to environmental conditions. For example, social boldness (SB) is defined as an individual's willingness to take risks and interact with social novelty. Another behavioral trait commonly studied is behavioral inhibition (BI), defined as avoidance and fear of the unfamiliar. The effects of BI in children include increased vulnerability for later-onset anxiety, cognitive disruption, and withdrawal (Biederman et al, 1993; Polivy, 2009), while long term effects of SB are not well understood. Characterizing the underlying mechanisms associated with these complex temperaments can improve our understanding of individual variation in social behavior and health trajectories. Behaviors related to SB and BI are associated with glucocorticoid (GC) production, however, changes in central nervous system function associated with these temperaments are less understood (Kabelik et al. 2021). We analyzed constitutive gene expression in the posterior hippocampus and stress-induced GC levels from blood collected immediately after acute restraint stress in 20 male Sprague-Dawley rats. Rat SB & BI temperament were assessed through repeated behavior

testing in the Novel Social, Novel Physical, and Partner Preference tests. Rats with regularly higher than median latency to approach social or physical novelty were categorized as high BI, and those with higher than median scores on a social boldness factor calculated from behavior in the novel social and partner preference test were categorized as high SB. We profiled genes involved in stress regulation (glucocorticoid receptor (*GR*), *FKBP5*, and *ADCYAP1*) and novelty reactivity (*neuroligin-2*) using qPCR, and compared gene expression to temperament (SB, BI) using regression analyses. We found that *GR* expression was a weak predictor of SB and BI. However, low *ADCYAP1* expression predicted greater SB, and a regression model with both *ADCYAP1* expression and stress-induced GC levels predicted BI better than each of these two variables independently. This combined model was not a better predictor of SB. GC production has been associated with both SB and BI in a variety of animal models and humans, but mechanisms driving this relationship are not clear. However, *ADCYAP1* in the hippocampus encodes the PACAP protein, which mediates the stress response and has been shown to regulate social behaviors. These results demonstrate a possible pathway by which glucocorticoid stress reactivity may affect temperaments that are associated with the tendency to approach and avoid novelty.

Disclosures: E.K. McMahon: None. **S. Farhan:** None. **S.A. Cavigelli:** None.

Poster

P587. Brain Circuits Underlying Social Behavior

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Program #/Poster #: P587.04

Topic: F.02. Behavioral Neuroendocrinology

Support: DP2MH126375
R01MH126035
Alfred P. Sloan Fellowship

Title: Multisite recordings of a highly recurrent hormone-sensitive macrocircuit across diverse social behaviors

Authors: *E. M. GUTHMAN, J. M. IRAVEDRA-GARCÍA, T. D. PEREIRA, A. L. FALKNER;
Princeton Neurosci. Inst., Princeton Univ., Princeton, NJ

Abstract: Social behavior is a fundamental aspect of animal life across species. Animals flexibly adjust their social behavior across a variety of external contexts and internal states to achieve access to resources, protect their territory, find mates, and raise offspring. These behaviors are controlled by the social behavior network (SBN), a highly-conserved and recurrent multiregional brain macrocircuit. SBN neural populations are united by their common expression of estrogen

receptor alpha (ERα), rendering them sensitive to the gonadal hormones estradiol and testosterone. Although dysregulated social behavior is a common symptom of many mental illnesses, the ways in which social context and internal state together influence SBN activity to change social behavior remain unknown. Here we discuss our development of novel tools and methods that we use to investigate how social context and gonadal hormone signaling influence SBN control of social behavior across sexes. Ongoing work combines novel method development for two-color, customizable full-brain fiber photometry recordings with automated tools for behavior discovery, classification, and quantification across context and hormone state. Specifically, using a combinatorial Cre-dependent and Cre-excluding viral strategy in the Esr1-Cre mouse line, we target ERα+ and ERα- SBN populations with different Ca2+ indicators. With these viral tools and our novel recording strategy, we are able to simultaneously track the activity of hormone-sensitive and insensitive populations during social behavior. We quantify behavior with an unsupervised approach, using the deep neural network, SLEAP, to track mouse posture dynamics. From these posture dynamics, we generate a set of behavior feature variable time-series representing social interaction between mice and cluster these feature variables for behavior classification. After two weeks of behavioral and neural recordings across 15 unique social contexts, mice are gonadectomized and experiments are repeated. Behavioral expression and SBN activity dynamics during behavior will be compared across sex, social context, and hormone state. Here, we present proof-of-principle data demonstrating the effectiveness of our methods, as well as findings demonstrating how mouse social behavior changes across social context and hormone state.

Disclosures: E.M. Guthman: None. **J.M. Iravedra-García:** None. **T.D. Pereira:** None. **A.L. Falkner:** None.

Poster

P587. Brain Circuits Underlying Social Behavior

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Topic: F.02. Behavioral Neuroendocrinology

Support: NIH Grant MH110212
NIH Grant MH122622

Title: Social defeat, but not subordinate status, reduces social reward and mesolimbic activation in Syrian hamsters (Mesocricetus auratus)

Authors: *E. WILLIAMS, K. L. HUHMAN, H. E. ALBERS;
Georgia State Univ., Atlanta, GA

Abstract: Social stress plays an important role in the etiology of many neuropsychiatric disorders that are characterized in part by behavioral deficits such as social withdrawal. Despite the potent effects of social stress in the development of neuropsychiatric disorders, little is known about the underlying mechanisms responsible for these effects. One way that social stress may contribute to psychiatric disorders is by reducing the rewarding properties of social interactions, thereby reducing social motivation. We investigated the impact of varying types of social encounters on social reward by comparing the reinforcing properties of social interactions in dominant, subordinate, and socially defeated Syrian hamsters. Social status was established during training over five consecutive days of five min parings with a same-sex conspecific. Dominant and subordinate hamsters were stable, age- and weight-matched dyads, while defeated hamsters were paired daily with a novel, larger resident aggressor (RA). All behavior, including duration of aggression, during training was quantified to determine if the "dose" of aggression received impacted social reward. Using an Operant Social Preference (OSP) task, we found that socially defeated hamsters made significantly fewer entries and had longer latencies to enter chambers containing novel, same-sex conspecifics compared to dominant and subordinate hamsters. Surprisingly, there was no difference in entries into the social chamber by hamsters with dominant versus subordinate status. Furthermore, subordinate hamsters experienced more aggression during training than did defeated hamsters. Therefore, the "dose" of prior aggression was not directly related to rewarding properties of subsequent social interactions. These data suggest that social instability may be more stressful than being submissive in a stable, albeit aggressive dyad. We also examined the resulting neural activation using cFos immunohistochemistry in brain regions associated with motivated behavior and reward, including medial prefrontal cortex (mPFC), nucleus accumbens (NAc), and ventral tegmental area (VTA). While mPFC activation was similar across all three groups, defeated hamsters had less activation in the VTA and NAc compared to dominant and subordinate hamsters. This difference in neural activation following social conditioning mirrors the behavioral differences seen in the OSP task. Thus, social defeat stress, but not stress associated with a stable subordinate status, reduces social reward and mesolimbic activation.

Disclosures: E. Williams: None. **K.L. Huhman:** None. **H.E. Albers:** None.

Poster

P587. Brain Circuits Underlying Social Behavior

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UofT/Medicine by Design

Title: Release from social suppression initiates transcriptomic differentiation of the hypothalamic-pituitary-gonadal axis of the naked mole-rat

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Abstract: Pubertal timing is highly heritable yet influenced by environmental conditions including psychosocial stressors. Pubertal timing, its susceptibility to social modulation, and associated health outcomes, all differ between males and females. Thus, understanding sex-specific mechanisms controlling pubertal timing has tremendous importance for human health. The naked mole-rat (NMR) is a unique rodent residing in large colonies of adults who remain in a pre-pubertal state due to the presence of a single, dominant breeding pair. How puberty is suppressed in adult NMRs is unknown, though sex differences in mechanism likely exist (e.g., males show reduced suppression). NMRs removed from the colony will enter puberty within a week. Here, we perform transcriptomic profiling of the hypothalamic-pituitary-gonadal axis in NMRs within the colony (breeders and subordinates) to animals that have been removed from their colony for 1 or 4 weeks (ex-subordinates), which triggers pubertal onset. The hypothalamus was separated into dorsal and ventral portions to separate populations relevant to stress input and reproductive output, respectively. By 1 week post-removal, we observe changes in the ventral hypothalamus of females, but not males. This includes the upregulation of *Trh*, the regulator of thyroid hormone activity and a key hormone related to the metabolic control of puberty. We found changes within the ovaries within a week which led us to focus on how the transcriptome changes and in which cell populations these changes occur in the hypothalamic-pituitary-gonadal axis of females during their first week of transition using single-cell RNA sequencing. Animals were removed from colony for 1 day, 4 days or 1 week (ex-subordinates) and compared to in-colony subordinates and breeders. To put these single cell gene expression changes in context we compared our data to single cell RNA-seq experiments performed in the mouse hypothalamus before and after puberty. Using canonical marker genes we are able to find broad cell populations shared between the NMR and mouse. Notably, within common cell populations of KNDy, AGRP/NPY+ and POMC+ neurons, we identified significantly higher levels of *Crhbp* relative to mice. CRHBP is a negative regulator of CRH expression and is an interesting candidate gene could related to psychosocial stress mediated pubertal suppression in NMRs. Here we will describe these results and ongoing work to elucidate the molecular mechanisms underpinning the striking pubertal suppression phenotype in NMR.

Disclosures: M. Faykoo-Martinez: None. **D.J. Sokolowski:** None. **K. Yuki:** None. **C. Chan:** None. **H. Hou:** None. **M. Palmert:** None. **T. Pugh:** None. **M.D. Wilson:** None. **M.M. Holmes:** None.

Poster

P587. Brain Circuits Underlying Social Behavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P587.07

Topic: F.02. Behavioral Neuroendocrinology

Support: NIH Grant R01MH115094-01A1

Title: Brain wide synaptic inputs to aromatase expressing neurons in the medial amygdala suggest complex circuitry for modulating social behavior

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Abstract: Social behaviors are critical for survival and depend on the integration of external and internal sensory cues. In mice, chemosensory information provides the primary sensory input to a conserved network of brain regions dedicated to executing social behaviors. The core brain regions that make up the social behavior network are established and conserved for all vertebrate species examined thus far. However, the cellular heterogeneity and overlap of functional circuits within many social brain regions has presented challenges for identifying the specific neural circuits require to execute social behaviors.
The medial amygdala (MeA) is a central node in the social behavior network and receives sensory input directly from the accessory olfactory bulb. The MeA integrates chemosensory signals with modulatory input from other brain regions and circulating hormones to guide behavioral responses through its projections to efferent behavioral centers. Aromatase, an enzyme that converts testosterone to estradiol, is expressed by a robust subset of neurons in nearly all brain regions of the SBN. Aromatase expressing neurons in the MeA are critical for aggression in both sexes and play an essential role in shaping the representation of social stimuli in the MeA.
Here, we used rabies-based circuit mapping from the specific aromatase expressing MeA cell population and CLARITY-based light-sheet microscopy to reveal an unbiased view of the brain regions that provide synaptic input to MeA aromatase expressing neurons. We observed both well-established connections (e.g., bed nucleus of the stria terminalis and accessory olfactory bulb), as well as novel and surprising inputs from areas involved in parenting and aggression, metabolism, fear and anxiety, and memory and cognition. These results confirm the central role

of the medial amygdala in social recognition behavior and point to an expanded role for its aromatase expressing neurons in the integration of multiple sensory and homeostatic factors, which can be used to modulate many other social behaviors.

Disclosures: J. Dwyer: None. **D.A. Kelly:** None. **J.F. Bergan:** None.

Poster

P587. Brain Circuits Underlying Social Behavior

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Program #/Poster #: P587.08

Topic: F.02. Behavioral Neuroendocrinology

Support: NIH RO1MH110212

Title: Effects of chemogenetic activation and inhibition of the anterior hypothalamus on social communication

Authors: *Z. GRIEB[1], J. VANDER VELDEN[1], K. L. HUHMAN[1], V. MICHOPOULOS[2], H. ALBERS[1];
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Abstract: Social communication deficits are a hallmark of many neuropsychiatric disorders, such as autism spectrum disorders. Social communication is controlled, in part, by the anterior hypothalamus (AH), making the AH a potential pharmatherapeutic target for alleviating social deficits in neuropsychiatric disorders. In Syrian hamsters, social communication includes a sterotyped flank marking behavior that is controlled by vasopressin V1a receptor activation in the AH. Injection of vasopressin into the AH induces flank marking and lesions of the AH or injections of V1a receptor antagonists decrease the expression of flank marking. Given these results, we hypothesized that activating or inhibiting the AH with chemogenetics would regulate flank marking in an opposing manner. Specifically, we predicted that chemogenetic activation of the AH would increase flank marking, while chemogenetic inhibition of the AH would decrease flank marking. To examine our hypothesis, we transduced adeno-associated viruses expressing either hM3Dq (i.e., Gq-linked DREADD; excitatory; $n = 9$), hM4Di (i.e., Gi-linked DREADD; inhibitory; $n = 7$), or mCherry (i.e., control virus; $n = 7$) into the AH of male hamsters. Three weeks after transduction, subjects were injected I.P. with either clozapine (0.1 mg/kg diluted in saline; note: dose of clozapine did not affect flank marking in controls) or vehicle in a counterbalanced design and then placed into either a clean cage or a cage recently marked by a same sex conspecific. Flank marking was then scored for 30 mins. We found, contrary to our predictions, that excitation of the AH led to decreased flank marking compared to controls and that inhibition of the AH led to increased flank marking compared to controls, independent of whether testing occurred in a clean cage or a cage containing conspecific odor. These data

suggest that the ultimate output of the AH has inhibitory action on flank marking and that V1a activation in the AH might act to inhibit that output. These results extend our previous data supporting a critical role of the AH in regulating social communication and further our understanding of the AH circuitry involved.

Disclosures: Z. Grieb: None. **J. Vander Velden:** None. **K.L. Huhman:** None. **V. Michopoulos:** None. **H. Albers:** None.

Poster

P587. Brain Circuits Underlying Social Behavior

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Program #/Poster #: P587.09

Topic: F.02. Behavioral Neuroendocrinology

Support: HHMI
Simons Collaboration on the Global Brain

Title: Mesoscale hypothalamic network dynamics in social behavior

Authors: S. STAGKOURAKIS, G. SPIGOLON, J. KIM, M. FEYDER, *D. J. ANDERSON; Caltech, Pasadena, CA

Abstract: In the animal kingdom, aggression holds a unique position on the spectrum of social behaviors, as its expression has high metabolic costs and potentially life-threatening consequences. Whether the brain utilizes an exclusive strategy for eliciting this risky behavior remains unknown. Previous work has identified features in the control of this behavior, such as the finding that silencing of different hypothalamic nuclei halts the expression of aggression, raising questions over the operating principles of the functional network underlying its expression. Here, we performed simultaneous neural activity measurements throughout the hypothalamus and identified that aggression correlates with high levels of inter-region synchrony, not observed in other social behaviors. Following the expression of attack, recurrent aggressive episodes correlate with lower activity levels, albeit highly synchronous. Surprisingly, pan-hypothalamic synchrony persists even after the end of an aggressive encounter suggesting this feature as a marker of a generalized aggressive state. These results reveal the unique neural encoding properties of social behaviors at the level of the hypothalamus, and identify specialized features in control of an animal's most risky social behavior expression.

Disclosures: S. Stagkourakis: None. **G. Spigolon:** None. **J. Kim:** None. **M. Feyder:** None. **D.J. Anderson:** None.

Poster

P587. Brain Circuits Underlying Social Behavior

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Program #/Poster #: P587.10

Topic: F.04. Stress and the Brain

Support: NIDA Grant R00DA045795
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Title: Zfp189-mediated transcription in prefrontal cortex controls social behavior and promotes mature pyramidal neuron morphology

Authors: N. L. TRUBY[1], ***G. M. SILVA**[1], J. A. PICONE[1], H. E. WOOLARD[1], R. L. NEVE[2], X. CUI[1], P. J. HAMILTON[1];
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Abstract: Earlier work has correlated the expression of *Zfp189* within mouse prefrontal cortical (PFC) neurons with enhanced behavioral resilience to chronic social stress. However, the exact mechanisms through which *Zfp189* is able to manifest stress resilience remains unclear. The *Zfp189* gene product is a Krüppel associated box (KRAB) zinc finger transcription factor of unknown function. To directly interrogate the transcriptional function and gene targets of ZFP189, we reprogrammed the endogenous ZFP189^{WT} by replacing the repressive KRAB domain with an enhanced transcriptional activation domain (VP64-p65-Rta (ZFP189^{VPR}) or by removing the functional moiety entirely (ZFP189^{DN}). We demonstrate divergent transcriptional regulation at a *luciferase* target gene, *in vitro*. Upon packaging these ZFP189 variant constructs in herpes viral vectors (HSVs) and delivering to mouse PFC, we observe that mice infected with HSV-ZFP189^{VPR}, the variant with inverted transcriptional function relative to the pro-resilient ZFP189^{WT}, demonstrate dramatically reduced social behaviors when exposed to a subthreshold defeat stress. Other stress-related behavioral metrics, like elevated plus maze, remain unaffected. The infected PFC tissues were then microdissected and subjected to RNA-sequencing, providing insight into which ZFP189-gene targets might be mediating these effects. Lastly, we investigated the consequences of altered ZFP189-mediated transcriptional function on spine density and morphology in cortical pyramidal neurons. Mice were transcardially perfused with 4% paraformaldehyde, brain tissue was extracted and sectioned, and was then imaged on a Zeiss 880 Airyscan confocal microscope. We observed pyramidal neurons infected with either ZFP189^{WT} or ZFP189^{VPR} possessed greater spine density and mature spine morphology relative to the transcriptionally inert ZFP189^{DN} and GFP controls. Collectively this body of work indicates that *Zfp189* mediates stress resilience by acting as a transcriptional repressor at key target genes in PFC cortical neurons, which drive dendritic spine maturity and pro-social behaviors.

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Poster

P587. Brain Circuits Underlying Social Behavior

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Topic: F.02. Behavioral Neuroendocrinology

Support: JP21dm0207065

Title: Development of a novel automated behavior analysis system for multiple freely-moving marmosets

Authors: *T. YURIMOTO, T. INOUE, W. KUMITA, K. SATO, Y. KUROTAKI, E. SASAKI; Central Inst. For Exptl. Animals, Kawasaki-Shi, Japan

Abstract: Many behavioral abnormalities are caused by symptoms of many neurological disorders. The common marmosets (*Callithrix jacchus*) social behaviors are similar to those of humans than rodents, such as family-based herding and food-sharing behaviors. Recently, various genetically manipulated marmoset models of neurodegenerative diseases have been created. However, it is unclear when and how the new genetically modified marmoset models will develop symptoms. Therefore, an automated behavioral analysis system that accurately detects and quantifies the lifelong behaviors of the model animals is necessary. Using this system to detect behavioral changes before and after the onset of disease and abnormal signs in the very early stages. However, there are no reports of behavior analysis systems for multiple marmosets or large cages that can house marmoset families. This study developed a behavior analysis system that combines light detection and ranging (LiDAR), video tracking, and deep learning for automatically tracking and analyzing individual behaviors of multiple marmosets in a family cage were tracked and their behaviors detected. Three wild-type family marmosets were kept in a family cage (W820 × D610 × H1578 mm) in accordance with EU guidelines. At the front and back of the cage were installed acrylic panels for acquisition of images from cameras and LiDAR. Four LiDAR and four cameras were set up in front of the cage. LiDAR and video tracking by deep learning were used to detect the positions of individual marmosets and create trajectories for each scene. Each trajectory was identified by face recognition using deep learning, and over one month simultaneous tracking of multiple animals. In addition, the animal behaviors were detected by deep learning of video frames to determine their content and location. The behavior information was matched with the tracking information to identify which animal performed the behaviors. So far, the face recognition accuracy is more than 85%. In addition, we have confirmed that the creation of trajectories and the detection of grooming behaviors of three animals can be accurately done by comparing with visual inspection.

Currently, video frames of typical behaviors such as drinking, feeding, mating, lying down, stretching, purring, jumping, tongue flicking, and marking are collected as training data for deep learning. Using these training data, not only the individual trajectories but also the animals behaviors will be automatically classified.

Disclosures: T. Yurimoto: None. **T. Inoue:** None. **W. Kumita:** None. **K. Sato:** None. **Y. Kurotaki:** None. **E. Sasaki:** None.

Poster

P587. Brain Circuits Underlying Social Behavior

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P587.12

Topic: F.01. Neuroethology

Support: NINDS F32NS098809
HHMI Investigator Award

Title: An interneuronal neuropeptidergic circuit modulates birdsong stability

Authors: *B. M. COLQUITT, M. S. BRAINARD;
Dept. of Physiol., UCSF Ctr. For Integrative Neurosci. and Howard Hughes Med. Inst., San Francisco, CA

Abstract: A hallmark of skilled behavior is consistent performance across renditions. Although the neural mechanisms that maintain stable performance have been extensively studied at systems and physiological levels, little is known about how the molecular properties of motor circuits influence skill stability. Songbird courtship song, a model for skilled behavior, is a highly structured vocalization that is learned and adaptively maintained throughout adulthood. To identify molecular pathways that influence song stability, we developed a dense gene expression profiling approach to characterize transcriptional responses across song neural circuitry to song destabilization following deafening. Song destabilization induced the largest expression changes in the motor output nucleus RA, including a strongly downregulated module of genes enriched in Pvalb and Sst-class GABAergic interneurons. Furthermore, this module showed an interaction in RA between song destabilization and singing rate, suggesting that the expression of these genes is coupled to both song stability and the amount of singing. The most highly modulated gene among these was corticotropin releasing hormone binding protein (CRHBP), whose product binds the neuropeptide corticotropin releasing hormone (CRH), a pathway previously implicated in neuronal excitability and plasticity. The CRH signaling pathway is expressed in non-overlapping neuronal populations in each song motor region and therefore could act as a local neuromodulatory pathway that influences song stability. Consistent with this role, genetic and pharmacological manipulation of the CRH pathway in the song motor

pathway resulted in bidirectional modification of song variability with CRHBP acting to maintain low variability and CRH acting to increase variability. These data indicate that singing engages an interneuronal neuropeptidergic pathway in motor circuits that maintains the consistency of song, thus providing a molecular mechanism to couple motor performance and stability.

Disclosures: B.M. Colquitt: None. **M.S. Brainard:** None.

Poster

P590. Vocal/Social Communication: Non-Avian I

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Program #/Poster #: P590.01

Topic: F.01. Neuroethology

Support: Genentech Fellowship
George A. and Marjorie H. Anderson Fellowship

Title: An amygdalocortical circuit for the integration of multisensory social cues

Authors: *A. NOWLAN, A. CORONA, C. KELAHAN, S. D. SHEA;
Cold Spring Harbor Lab., Cold Spring Harbor, NY

Abstract: Many species, including mice, communicate using multisensory signals to coordinate social behaviors essential for survival. How social experience influences multisensory integration and supports dynamic behavioral goals is not well understood at the circuit level. For example, when newborn pups become separated from the nest, a mother relies on olfactory and auditory cues to motivate her retrieval of pups back to the nest. Co-housing a virgin female with a dam and her pups is sufficient to provoke maternal retrieval behavior and olfactory-auditory integration without pregnancy. In mothers and experienced virgins ('surrogates'), pup odor modifies activity within the auditory cortex (AC) to enhance the detection of pup distress calls and potentially improves retrieval performance. However, the underlying circuitry involved in olfactory-auditory integration and maternal behavior remains incomplete.Here we reveal that projection neurons from the basal amygdala (BA) target the AC, detect olfactory cues, and evoke experience-dependent changes to the neural activity within the AC. Using fiber photometry, we show that BA-AC projection neurons are sensitive to pup odor and are primarily active while the surrogate mother searches for an isolated pup. We find that optogenetic activation of the BA-AC terminals during the presentation of auditory stimuli elicits complex changes in the activity of auditory neurons. Both ongoing and auditory evoked activity was preferentially suppressed in pup-naïve females but enhanced in surrogate mothers. Our results suggest that relaying olfactory cues through the amygdala may enhance the perception of pup distress calls by conveying contextual features about the pup stimuli to the AC. These features might comprise the perceived

salience or valence of individual social cues or the animal's overall affective or behavioral state, however, these details remain undetermined. Nevertheless, the BA-AC circuit represents a novel avenue by which the amygdala modulates primary sensory cortex that warrants further exploration and may be particularly relevant to anxiety and autism spectrum disorders.

Disclosures: A. Nowlan: None. **A. Corona:** None. **C. Kelahan:** None. **S.D. Shea:** None.

Poster

P590. Vocal/Social Communication: Non-Avian I

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Program #/Poster #: P590.02

Topic: F.01. Neuroethology

Support: NIMH R01MH122752
NIH P20GM1036653

Title: Knockout of the autism-associated gene Shank3B leads to alterations in social and vocal behavior in female mice

Authors: *R. S. CLEIN[1], M. R. WARREN[2,1], J. P. NEUNUEBEL[1];
[1]Univ. of Delaware, Newark, DE; [2]Emory Univ., Atlanta, GA

Abstract: Autism spectrum disorder (ASD), a highly heritable neurodevelopmental disorder, is characterized by challenges in social interaction, verbal and non-verbal communication, and enhanced repetitive behaviors (Faras et al., 2010). While the exact etiology of ASD is unknown, multiple genetic alterations have been associated with autism-like behavior. For example, more than 50% of patients with Phelan-McDermid syndrome (22q13.3 deletion syndrome) have a syndromic form of ASD, and most have a chromosomal deletion on the *SHANK3* gene (Phelan & McDermid, 2012). Mice with a *Shank3B* deletion (Peca et al., 2011) show physiological and behavioral abnormalities, providing a promising model for the study of ASD. However, although equal proportions of males and females carry a 22q13 deletion (Tabet et al., 2017), most behavioral studies using mouse models of *Shank3B* deletion focus solely on males. Therefore, direct quantification of the innate social and vocal behavior of female mice with genetic modifications to the *Shank3b* gene is lacking. To that end, we used a sound source localization system to simultaneously record the social and vocal behavior of females and males in mixed-sex groups (2 males and 2 females per group) of either *Shank3b* knockout mice (KO; n = 6 groups, 24 mice) or wild-type controls (WT; n = 7 groups, 28 mice). Surprisingly, pairs of KO females interacted with each other more often than did pairs of KO males (Mann-Whitney, $U = 21$ $p < 0.01$). This pattern was not observed in WT animals (Mann-Whitney, $U = 66$, $p = 0.09$). When quantifying the time females spent interacting with each other, KO females interacted for longer than WT females (Mann-Whitney, $U = 28$, $p < 0.05$). Even during interactions between females

and males, the duration was longer in KO than WT mice (Mann-Whitney, $U = 22$, $p < 0.05$). Moreover, the inter-individual distances between pairs of KO females were significantly smaller than WT females, indicating that KO females were physically closer together during social interactions than their WT counterparts (Mann-Whitney, $U = 68$, $p < 0.05$). However, the inter-individual distances between females and males were similar in KO and WT mice (Mann-Whitney, $U = 49$, $p > 0.05$). Additionally, during social interactions, the vocal rate of WT females was higher than KO females (Kruskal-Wallis, $H = 20.67$, $p < 0.01$, Dunn-Sidak post hoc correction). Taken together, these results demonstrate altered vocal and social behavior in female mice with known modifications in the *Shank3b* gene and suggests that genetic alterations to the *Shank3B* gene differentially affect behavior in males and females.

Disclosures: R.S. Clein: None. **M.R. Warren:** None. **J.P. Neunuebel:** None.

Poster

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Program #/Poster #: P590.03

Topic: F.01. Neuroethology

Support: NIMH R01MH122752
NIH P20GM1036653

Title: Investigating the acoustic directionality of mouse ultrasonic vocalization

Authors: *Y. MA[1], M. R. WARREN[1,2], J. P. NEUNUEBEL[1];
[1]Univ. of Delaware, Newark, DE; [2]Emory Univ., Atlanta, GA

Abstract: Across the animal kingdom, ultrasonic vocalizations (USVs) are used as a means of communication, navigation, and predation, and in some species, the directionality of the signal facilitates these processes (Jakobsen et al., 2015; Portfors et al., 2014). Previous research has shown that some species of bats and whales produce highly directional ultrasound beams to echolocate (Au, 1993; Surlykke et al., 2009). While numerous studies have demonstrated that mice produce USVs in a variety of social contexts (e.g., Warren et al., 2021), the directionality of these signals is less clear, as identifying the vocalizer in group settings is challenging. To that end, we used an 8-channel microphone array to localize and assign USVs to individual mice that were either in isolation (n=28) or socially interacting (male-female dyads, n = 13; 4-mouse, mixed-sex groups, n = 11). Across recordings, males emitted 82 – 100% of assigned vocalizations. To assess directionality, we used the position of the mice, position of the microphones, and the sound intensity of signals at each microphone. For each vocalization, we mapped the microphone positions on a Cartesian plane with the position of the vocalizer at the origin. We then computationally rotated the map such that the head of the mouse always faced 0

degrees, thus aligning the positions of the microphones relative to the source of the vocalization. The resulting maps were subsequently sectioned into 72 non-overlapping, equally-sized wedges and the average sound intensity was calculated for each wedge. USV intensity was strongest in the direction that the mouse was facing, and weakest in the opposite direction, suggesting a directional pattern. We next calculated a directional index (DI) for the male vocalizers in each recording, where a DI of 0 indicated an omnidirectional vocalization. On average, male mice produced USVs with DIs of 48.9, 23.7, and 24.3 when in isolation, dyads, and mixed-sex groups, respectively. To determine the DI values expected by chance, we conducted a permutation test. We demonstrated that across all behavioral contexts, the directionality of male-emitted USVs was significantly greater than chance levels (Z-test, all p's < 0.001). Overall, these findings directly demonstrate that male mouse USVs are directional and may provide additional insight on the communicative function of mouse vocalizations.

Disclosures: Y. Ma: None. **M.R. Warren:** None. **J.P. Neunuebel:** None.

Poster

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Program #/Poster #: P590.04

Topic: F.01. Neuroethology

Title: Neural circuits underlying the effects of acute isolation on vocal and non-vocal social behaviors in female mice

Authors: *X. ZHAO, Y. CHAE, S. RABINOVICH, T. GRINBERG, S. CHU, Z. HE, A. SADANGI, K. A. TSCHIDA;
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Abstract: Social isolation is a potent stressor that adversely impacts both mental and physical well-being. Despite a wealth of studies demonstrating the impacts of chronic isolation on social behavior, it remains less clear how acute social isolation impacts the brain and behavior and whether these effects vary according to sex and social context. To investigate this question, we established a behavioral paradigm in which mice that were either group-housed or single-housed for three days interacted with a novel group-housed visitor, and their ultrasonic vocalizations (USVs) and non-vocal social behaviors were examined in three different contexts (female-female, male-female, and male-male interactions). In female-female trials with a single-housed resident, we observed a dramatic increase in the production of USVs, increased social interaction time, and the emergence of mounting accompanied by USV production as compared to trials with group-housed residents. In contrast, acute isolation drove only subtle effects on social behavior during male-female and male-male encounters. To explore the neural circuit basis for the effects of acute isolation on female social behavior, we are currently comparing c-Fos

activation in group-housed and single-housed females to test whether acute isolation enhances the activity of different populations of forebrain and midbrain neurons, including the preoptic area of the hypothalamus, the bed nucleus of the stria terminalis, the medial amygdala, and the periaqueductal grey (PAG). We are also combining these Fos immunostaining experiments with retrograde labeling from the PAG to identify forebrain neurons whose activity is potentiated by acute isolation and that project to the region of the PAG that contains neurons necessary to USV production. We anticipate that our findings will shed light on the forebrain-to-midbrain circuits that regulate the production of social vocalizations in female mice, as well as how acute isolation acts on these circuits to affect social behavior.

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Poster

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R21 DC016135 (PI: Kelm-Nelson)
T32 DC009401 (PI: Thibeault)

Title: Social isolation changes ultrasonic vocalizations and reduces dopamine and serotonin ventral tegmental area of healthy rats

Authors: *C. BROADFOOT[1], C. LENELL[2], C. A. KELM-NELSON[3], M. R. CIUCCI[2]; [2]Surgery-Otolaryngolgy, [1]Univ. of Wisconsin Madison, Madison, WI; [3]Dept. of Surgery, Div. of Otolaryngology, Univ. of Wisconsin Madison Dept. of Surgery, Madison, WI

Abstract: Vocal communication plays an important role in health and wellness. Of interest to our laboratory, are the effects of social isolation on vocal communication in disease states, such as Parkinson disease, and the ability of social enrichment to treat these deficits. However, the effects of social context such as isolation and enrichment, and the neural mechanisms governing these complex behaviors, is relatively unknown. As a crucial first step, we studied socially-motivated ultrasonic vocalizations (USV) in wild-type (*i.e.* healthy) rats. The catecholaminergic systems of the ventral tegmental area (VTA) are associated with social and reward processing and are suseptible to changes in social stress (isolation). The central hypotheses for this study

were that social condition would alter vocal communication by changing USV acoustics, non-motor behaviors (anxiety, anhedonia, and cognitive function) and affect catecholamine concentrations in the VTA. Specifically, we hypothesized that social isolation would decrease vocal function (loudness), increase the presence of anxiety and anhedonia, and decrease cognitive function. To test these hypotheses, twenty-four male wild type (WT) rats were randomized into 3 groups: 1) control, housed with a cage-mate, 2) socially-isolated, housed individually, and 3) socially-enriched, housed with a cage-mate and underwent social enrichment. Social enrichment involved grouping rats in a larger 'socialization' cage with another pair of cage mates 5x per week. USVs were recorded at 2 (baseline-prior to randomization), 4, 6, 8 and 10 month time points using an established elicitation mating paradigm. At each timepoint, all animals were also assessed for cognitive skills (novel object recognition test), anxiety levels (elevated-plus maze), and presence of anhedonia (sucrose preference test). Micro punches of the VTA were assayedfor levels of dopamine, norepinephrine, and serotonin and associated metabolites using high-performance liquid chromatography. A repeated measures ANOVA with factors of social condition and timepoint revealed that vocalizations of rats that underwent social enrichment produced calls that were louder ($p<0.01$) with greater duration ($p<0.001$) compared to the control and isolation rats. The results of the non-motor behavior analyses will be presented. Additionally, rats in the social isolation group had significant increases in levels of dopamine ($p<0.001$), HVA ($p<0.05$), and 3MT ($p<0.001$) as well as 5-Ht ($p<0.001$) in the VTA. This study suggests that chronic changes to social condition in adulthood affects both behavioral outcomes and neural mechanisms in healthy rats.

Disclosures: C. Broadfoot: None. **C. Lenell:** None. **C.A. Kelm-Nelson:** None. **M.R. Ciucci:** None.

Poster

P590. Vocal/Social Communication: Non-Avian I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P590.06

Topic: F.01. Neuroethology

Support: NIMH R01MH122752
NIH P20GM103653

Title: Evaluating ultrasonic vocalizations as predictors of social behavior during group interaction

Authors: *C. M. KAWAR[1], R. S. CLEIN[1], M. R. WARREN[1,2], J. P. NEUNUEBEL[1]; [1]Univ. of Delaware Grad. Program In Behavioral Neurosci., Newark, DE; [2]Emory Univ., Atlanta, GA

Abstract: During social interaction, animals integrate a variety of sensory information, which influences future behavioral decisions. For example, vocalizations can alert conspecifics to predators or reproductive opportunities and then elicit distinct responses (Seyfarth et al., 1980; Macedonia, 1990; Evans, 1993). Mice produce ultrasonic vocalizations (USVs) associated with discrete behaviors (Sangiamo et al., 2020), however, it is unclear if USVs foreshadow specific actions. To investigate the predictive power of mouse USVs on social behavior, we recorded mixed-sex groups of freely interacting adult mice (n = 11, 2 males and 2 females per group; 12-21 weeks old, B6.CAST-Cdh23Ahl+/Kjn) for 5 hours using a sound source localization system. Though it has traditionally been challenging to identify individual vocalizers while mice are socially interacting, this 8-channel microphone array system allows us to assign vocalizations to individual mice. Using machine learning-based approaches (Kabra et al., 2013) to automatically categorize vocalizations and extract distinct behaviors in which two mice have unique roles as actor or recipient (i.e., approach, flee, investigate, chase, and mount), we found a significant interaction between particular social behaviors and the vocalization types that preceded (2-way ANOVA, $F = 4.78$, $p < 0.001$). A permutation test showed that specific vocalization types were produced at above-chance proportions when they preceded dominant behaviors (involving an actor courting or behaving aggressively toward a recipient), while other types were emitted at above-chance proportions when preceding submissive behaviors (involving an actor avoiding a recipient) (Z-tests, p-values < 0.001). We then trained multi-class support vector machines to distinguish behavioral role (actor versus recipient) based on the proportion of vocalization types the participants produced preceding the behaviors. While classifiers were unable to accurately distinguish the behavioral role of the mouse at above-chance levels for some behaviors (flee, approach, male-investigate-male, mount, female-investigate, and fight) we found accuracies significantly above chance for dominant behaviors (male-chase-male, male-chase-female, male-investigate-female; Z-tests, p-values < .0005). In sum, our results demonstrate that mouse vocalizations may predict specific subsequent behaviors.

Disclosures: C.M. Kawar: None. **R.S. Clein:** None. **M.R. Warren:** None. **J.P. Neunuebel:** None.

Poster

P590. Vocal/Social Communication: Non-Avian I

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P590.07

Topic: F.01. Neuroethology

Support: NIH Grant 1U19NS107616-01
NSF GRFP Grant 1650114

Title: A novel, open-source, high-throughput monitoring system for maternal behavior in mice

Authors: *L. F. SCHUSTER[1], D. C. ANANTH[3], A. C. MAR[2], R. C. FROEMKE[1];
[1]Grossman Sch. of Medicine, Skirball and Neurosci. Institutes, Departments of Otolaryngology, [2]Neurosci. and Physiol., New York Univ., New York, NY; [3]Northeast Ohio Med. Univ., Rootstown, OH

Abstract: Understanding the neural basis of maternal behavior requires a comprehensive analysis of the repertoire that parents express, under conditions that enable high-resolution monitoring of most if not all behaviors 24 hours/day for weeks to months. To achieve this, we designed a semi-automated habitat for mice using open-source tools, where behavioral data can be collected and saved near-continuously over 4+ months. Our system is soundproofed, temperature- and humidity-controlled, and supports neural recordings and gain-of-function and loss-of-function interventions. Our overall goal is comprehensive behavioral phenotyping throughout life and over generations.
To date we have monitored 10 wild type (WT) dams across 2-4 litters apiece, examining changes across different aspects of maternal care from litter to litter. While infanticide is quite common in first-time mothers, the incidence decreases in most dams with the second and subsequent litters. We found that even before birth of the first litter, quality of nest-making predicted degree and perseverance of infanticide. Those dams that continued to be infanticidal stopped this behavior when co-housed with experienced dams that were not infanticidal.
Previously we found that the neuropeptide oxytocin promotes long-term plasticity and mediates experience-dependent changes in neural circuits that increase the sensitivity to pup cues, thereby facilitating behavioral responses to pup distress. To understand the effects of genetic disruption of oxytocin signaling in maternal care, we monitored the first pregnancies of 2 oxytocin receptor knockout (OXTR KO) dams. These OXTR KO animals built subpar nests, spent less time interacting with their pups, and had high pup mortality rates compared to WT mice who raised successful first litters. Interestingly, WT dams who neglected or cannibalized their pups showed similar nest-building patterns and time spent in the nest as the OXTR KO mice.

Disclosures: L.F. Schuster: None. **D.C. Ananth:** None. **A.C. Mar:** None. **R.C. Froemke:** None.

Poster

P590. Vocal/Social Communication: Non-Avian I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P590.08

Topic: F.01. Neuroethology

Support: NIH 2R01 DC012087

Title: Within-neuron comparison demonstrates context-dependence of natural social signal processing in primate prefrontal cortex

Authors: *V. JOVANOVIC[1], L. A. DE LA MOTHE[2], K.-F. LEE[3], C. T. MILLER[1];
[1]Cortical Systems and Behavior Lab., Univ. of California San Diego, La Jolla, CA; [2]Psychology, Tennessee State Univ., Nashville, TN; [3]PBL-L, Salk Inst., La Jolla, CA

Abstract: Communication is an active exchange of social signals between individuals and yet the neural basis of primate communication has primarily been studied in head-restrained paradigms in which social signals are entirely divorced from the contexts in which they naturally occur. The few studies to examine the neural basis of vocal communication in freely-moving monkeys have yielded patterns of results that diverge notably from parallel experiments using more traditional, head-restrained approaches. Here we sought to directly test - within neuron - whether these observations reflect differences in experimental design or more fundamental insight into how the primate brain functions under natural conditions. We recorded the responses of the same individual neurons in marmoset prefrontal cortex (PFC) to acoustic stimuli, including vocalizations, across a series of contexts ranging from head-restrained passively-listening animals to freely-moving monkeys engaged in interactive conversational exchanges (i.e. active, natural communication). Consistent with previous reports in rhesus macaques, we observed robust responses to acoustic stimuli in PFC neurons when marmosets were in a traditional head-restrained context. When presenting the identical set of acoustic stimuli to freely-moving marmosets, PFC neurons likewise exhibited robust responses. However, the same individual neurons exhibited little to no response to the vocalization stimuli when subjects were engaged in their natural communicative exchanges. These results suggest that while mobility had little impact on the pattern of neural responses in primate PFC, actively engaging in natural communication profoundly affected the responses of these neurons. Further analyses indicated that contextual differences were not limited to stimulus firing rate, but rather several characteristics of neural activity distinguished PFC activity during natural communication from the other behavioral contexts. This suggests that key facets of active communication may not be replicable using the more reductionistic, head-restrained paradigms that typify previous experiments investigating neural basis of primate communication. Thus, we show the assumption that data in traditional, restrained paradigms of social signal processing reflect the natural analog is not strictly true. Explicating the neurobiology of communication in the primate brain likely necessitates that we examine neural activity as individuals actively engage in natural communication exchanges.

Disclosures: V. Jovanovic: None. **L.A. de la Mothe:** None. **K. Lee:** None. **C.T. Miller:** None.

Poster

P590. Vocal/Social Communication: Non-Avian I

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P590.09

Topic: F.01. Neuroethology

Support: NIH Grant R25 NS 80686

Title: Meta-analysis of how sex and dominance interact with social position to affect prosocial behaviors in marmosets (Callithrix jacchus)

Authors: *S. RIVERA[1], O. MEISNER[2], S. PARK[2], M. P. JADI[3], A. S. NANDY[4], S. CHANG[5]; [1]Hunter Col., New York City, NY; [3]Dept. of Psychiatry, [4]Dept. of Neurosci., [5]Dept. of Psychology, [2]Yale Univ., New Haven, CT

Abstract: Research in prosocial behaviors garnered much interest in the field of social neuroscience over recent years, but human research is limited in its ability to address the underlying neural mechanisms of these behaviors. Research in this domain using rodent models has furthered our understanding of the neural circuit mechanisms of social behaviors. However, the rodent model also faces limitations when it comes to modeling human social behaviors. For example, the fundamental mode of social communication between rodents (olfactory) is much different than the largely visual communication used by primates. Primate models have been beneficial to social neuroscience due to their complex social communication and environments that more closely resemble that of humans. More recently, the common marmoset (*Callithrix jacchus*) has emerged as a model system for investigating the neural bases of primate social behaviors. Despite evolutionarily diverging much earlier from humans compared to macaques or capuchins, they are cooperative breeding and a highly prosocial species that is small in size and easy to maintain in captivity. Though marmosets are potentially an excellent comparative model for human cooperative prosociality, their complex group dynamics also can stand as an obstacle. In this meta-analysis, we will investigate how sex and dominance interact with social role to affect prosocial behaviors in the common marmoset. As the identity of individual monkeys dictates much of marmoset social life, we will investigate how the sex, position within dominance hierarchy, and social role dictate which members of a colony are more likely to perform prosocial behaviors and if a correlation exists across this species. We are exploring previous research where prosocial and cooperative behaviors demonstrated and identify which individuals successfully display different types of prosocial behaviors in these experiments. As social experiments in marmosets tend to successfully elicit cooperative social behaviors in a subset of individuals, we aim to identify which individuals are most likely to engage in these behaviors. As the allocation of prosocial behaviors among members is variable, this investigation hopes to shed light on the complex social structures of marmosets to help inform future social neuroscience research in this species.

Disclosures: S. Rivera: None. **O. Meisner:** None. **S. Park:** None. **M.P. Jadi:** None. **A.S. Nandy:** None. **S. Chang:** None.

Poster

P590. Vocal/Social Communication: Non-Avian I

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NIH grant R01AG060931
NIH grant R00AG051764

Title: Social connections predict brain structure in a multidimensional free-ranging primate society

Authors: *C. TESTARD[1], L. J. N. BRENT[2], J. ANDERSSON[3], K. L. CHIOU[4], J. E. NEGRON DEL VALLE[4], A. R. DECASIEN[5], A. ACEVEDO-ITHIER[1], M. K. STOCK[6], S. C. ANTON[5], J. E. HORVATH[7], N. R. CAMPO[8], K. MILLER[3], O. GONZALEZ[9], C. S. WALKER[10], S. BAUMAN[8], A. V. RUIZ-LAMBIDES[8], M. I. MARTINEZ[8], P. J. H. SKENE[11], J. P. HIGHAM[5], N. SNYDER-MACKLER[4], M. J. MONTAGUE[1], J. SALLET[3], M. L. PLATT[1];
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Abstract: Reproduction and survival in most primate species reflects management of both competitive and cooperative relationships. Here we investigated the links between neuroanatomy and sociality in free-ranging rhesus macaques. In adults (n=68), the number of social partners predicted morphology of mid-superior temporal sulcus and anterior insula, implicated in social decision-making and empathy, respectively. We found no link between brain structure and other key social variables such as status or indirect connectedness in adults, nor between maternal social networks or status and dependent infant (n=20) brain structure. Our findings demonstrate the size of specific brain structures varies with the number of direct social connections, and suggest this relationship arises during development. These results reinforce proposed links between social network size, biological success, and the expansion of specific brain circuits.

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Poster

P595. HPG Axis

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P595.01

Topic: F.03. Neuroendocrine Processes

Support: NIEHS 1R01ES025547

Title: Impact of bisphenol-A and synthetic estradiol on brain, behavior, gonads, and sex hormones in a sexually labile coral reef fish

Authors: *A. R. HISTED[1], J. A. GONZALEZ[1], S. E. CRAIG[1], E. NOWAK[1], D. LANGE[1], C. G. PARKER[1,2], A. KAUR[1], S. BHUVANAGIRI[1], K. J. KROLL[4], C. J. MARTYNIUK[4], N. D. DENSLOW[4], C. S. ROSENFIELD[5], J. S. RHODES[1,2,3];
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Abstract: Endocrine disrupting chemicals, such as bisphenol A (BPA) and ethinylestradiol (EE2), are detected in the marine environment from plastic waste and wastewater effluent. However, their impact on reproduction in sexually labile coral reef fish is unknown. The objective of this study was to determine impacts of environmentally relevant concentrations of BPA and EE2 on behavior, brain gene expression, gonadal histology, sex hormone profile, and plasma vitellogenin (Vtg) levels in the anemonefish, *Amphiprion ocellaris*. *A. ocellaris* display post-maturational sex change from male to female in nature. Sexually immature, male fish were paired together and fed twice daily with normal food (control), food containing BPA (100 µg/kg), or EE2 (0.02 µg/kg) (n=9 pairs/group). Aggression towards an intruder male was measured at 1, 3, and 6 months. Blood was collected at 3 and 6 months to measure estradiol (E2), 11-ketotestosterone (11-KT), and Vtg. At the end of the study, fish were euthanized to assess gonad morphology and to measure expression of known sexually dimorphic genes in the brain. Relative to control, BPA decreased aggression, altered brain transcript levels, increased non-vitellogenic and vitellogenic eggs in the gonad, reduced 11-KT, and increased plasma Vtg. In two BPA-treated pairs, both individuals had vitellogenic eggs, which does not naturally occur. EE2 reduced 11-KT in subordinate individuals and altered expression of one transcript in the brain toward the female profile. Results suggest BPA, and to a lesser extent EE2, pollution in coral reef ecosystems could interfere with normal reproductive physiology and behavior of the iconic sexually labile anemonefish.

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Poster

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Program #/Poster #: P595.02

Topic: F.03. Neuroendocrine Processes

Support: NSERC Grant
Ontario Graduate Scholarship
Ontario Veterinary College Doctoral Scholarship

Title: The effects of Bisphenol A and Bisphenol S exposure on stress and sex steroid-sensitive protein expression in the murine hippocampus.

Authors: *K. NICHOLSON[1], J. WOODMAN[1], E. CHOLERIS[2], N. J. MACLUSKY[1];
[1]Biomed. Sci., [2]Psychology, Univ. of Guelph, Guelph, ON, Canada

Abstract: Bisphenol A (BPA) is a potent endocrine disrupting chemical that interferes with the mechanisms of gonadal hormones such as estradiol and adrenal hormones such as glucocorticoids. Specifically, BPA is known to disrupt estradiol-mediated activation of intracellular cascades including the extracellular signal-regulated (ERK) and c-Jun N-terminal (JNK) kinases. These kinase cascades are also known to be glucocorticoid-sensitive while playing important roles in hippocampal protein synthesis, synaptogenesis, learning, and memory. Therefore, many industrial processes have since replaced BPA with structural analogues such as Bisphenol S (BPS). However, the effects of BPS exposure on these glucocorticoid and estradiol-sensitive mechanisms within the mammalian hippocampus remain unknown. This work aimed to compare the effects of BPA and BPS exposure on hippocampal expression of ERK, JNK, mitogen-activated protein kinase phosphatase 3 (MKP3-a phosphatase implicated in stress and depression), glial fibrillary acidic protein (GFAP), and postsynaptic density-95 (PSD-95). For 10 consecutive days, gonadally intact male and female CD-1 mice (2-3 months of age) were administered vehicle, BPA, or BPS-containing peanut butter at the tolerable daily intake (TDI) dose of 50 μg/kg/day as defined by the American Environmental Protection Agency. Female estrous cycles were tracked by daily vaginal lavages. On day 10, mice were sacrificed 4 hours after their final treatment for hippocampal extraction (n=9/group). Hippocampi were homogenized and analyzed through western blotting for changes in ERK, JNK, MKP3, GFAP, and PSD-95 protein expression. Significant effects of treatment and sex were observed for expression of 54 and 46 kDa isoforms of JNK, respectively. BPA-treated females exhibited a stress-like reduction in MKP3 expression in comparison to BPA-treated males. Females treated with BPA and BPS exhibit reduced total MKP3 expression in comparison to vehicle-treated female controls. ERK, GFAP and PSD-95 levels were unaffected by BPA or BPS treatment. Plasma corticosterone levels were measured using ELISAs (n=7-9/group). Females exhibited

higher plasma corticosterone concentrations than males, although no effects of treatment were detected. These findings may have important implications for sexually differentiated cellular and stress response mechanisms of bisphenols within the mammalian hippocampus. Our ongoing studies will further investigate the cellular interactions between glucocorticoids, estradiol, and bisphenols using in vitro and in vivo models.

Disclosures: K. Nicholson: None. **J. Woodman:** None. **E. Choleris:** None. **N.J. MacLusky:** None.

Poster

P595. HPG Axis

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Program #/Poster #: P595.03

Topic: F.03. Neuroendocrine Processes

Support: T32 MH019524

Title: Oxytocin and lactationally-triggered embryonic diapause in mice

Authors: *J. L. MINDER[1,3,4], M. V. CHAO[1,3,4], R. C. FROEMKE[2,3,4];
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Abstract: Embryonic diapause is a state in which the development of the pre-implantation embryo is halted until conditions become favorable for pregnancy progression (Fenelon et al., 2014). It is mediated by the hypothalamic-pituitary axis and results in the pausing of embryonic development at the blastocyst stage, with implantation into the uterine endometrium being temporarily blocked (Stouffer and Hennebold, 2014). Development resumes as normal upon a return to homeostasis. Diapause has been reported in many mammalian species, including suggestive reports in humans that underwent in vitro fertilization (Grinsted and Avery, 1996). Despite the long history of study and potential importance for fetal development, little is known about the initiating factors for diapause in rodents. We have been examining whether oxytocin (OT) might be involved in lactationally-triggered facultative diapause in C57 mice. OT is released from the posterior pituitary during the sensory stimulus of suckling, resulting in peripheral OT surges reaching the womb. First, we identified oxytocin receptor (OXTR) expression on mouse blastocysts. We also found that embryo implantation in the presence of OT is delayed in comparison to saline (n=79 embryos, p=0.009) using an in vitro embryo culture model of implantation (Bedzhov et al, 2014). When pregnancy ensues coincident with nursing a previous litter, we found that gestational times were extended by one week (n=3), as compared to gestation in the absence of nursing (n=12; p=0.004). A drop in estrogen levels in the mother is thought to contribute to halting embryo implantation (Stouffer and Hennebold, 2014). After

treating the hypothalamic cell line GT1-7 with the estrogen receptor antagonist 4-hydroxytamoxifen, OT mRNA levels were increased relative to saline-treated controls (n=11, p=0.005). This suggests that OT levels are increased in the mother during nursing. Finally, we found that without the OXTR, embryos may fail to implant at the right time and become reabsorbed. Only 5% of OXTR knock-out pups were born in litters that experienced diapause (n=3), as opposed to 33% of OXTR knock-out pups being born from normal pregnancies (n=9). We conclude that nursing induces diapause in mice, and that maternal OT levels increase during this state. Given that fewer OXTR KO pups were born in litters that experienced diapause, we hypothesize that OT levels serve as a timing cue for the embryo to enter a paused state in order to properly coordinate with the maternal uterine environment and to survive diapause. Understanding how OT levels might regulate reproductive state and embryonic development has major implications for reproductive medicine.

Disclosures: J.L. Minder: None. **M.V. Chao:** None. **R.C. Froemke:** None.

Poster

P595. HPG Axis

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Program #/Poster #: P595.04

Topic: F.03. Neuroendocrine Processes

Support: NSERC Discovery 2020-04302

Title: Lps treatment alters gad67 mrna expression and gonadotropins concentrations in a sex- and age-dependant manner

Authors: *K. B. SMITH[1], J. ZHONG[1], N. ISMAIL[2];
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Abstract: Puberty is a period of sexual and reproductive development, and neuronal reorganization and remodeling. Exposure to stress during this period can result in enduring effects on reproductive behaviour. Stress-induced HPA axis activation causes inflammatory cytokine release which contributes to downregulating the hypothalamic pituitary gonadal (HPG) axis. This downregulation may be caused by GABAergic inhibition to the pituitary gland, preventing the secretion of gonadotropins. The current study examined the acute and enduring effect of LPS treatment on GAD67 mRNA expression in the pituitary gland, and LH, FSH, interleukin-6 (IL6), and tumor necrosis factor alpha (TNFα) concentration in the blood of male and female mice. 160 male and female mice were treated with either saline or LPS at 6 (puberty) or 10 (adulthood) weeks of age and euthanized either 8 hours (acute effects) or 4 weeks (enduring effects) post-treatment. LPS treatment in adult males and females increased GAD67 mRNA expression and TNFα concentration, and decreased LH concentration at 8 hours

following treatment. LPS-induced upregulation of GAD67 mRNA expression was long-lasting at it was still present 4 weeks following treatment. LPS treatment in pubertal males and females increased IL6 concentration and decreased LH concentration only at 8 hours following treatment. We also identified sex differences in gonadotropin expression, where LPS-treated pubertal and adult females displayed less FSH concentration compared to males at 8 hours and 4 weeks following treatment. LPS-treated adult females also expressed lower LH concentration compared to male counterparts. Lastly, LPS-treated pubertal and adult males displayed greater IL6 concentration compared to females 8 hours post-treatment. Our data show important age and sex differences in stress-induced downregulation of the HPG axis. Moreover, although the upregulation of GAD67 mRNA persisted 4-weeks post LPS treatment in adult mice, it does not impact LH concentrations. Taken together, our findings show that the GABAergic pituitary inhibition is an important mechanism in stress-induced downregulation of the HPG axis, but in an age- and sex-specific manner. The presence of these sex and age differences implies that sex hormones may play an influential role upstream to the pituitary control of gonadotropin secretion.

Disclosures: K.B. Smith: None. **J. Zhong:** None. **N. Ismail:** None.

Poster

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Topic: F.03. Neuroendocrine Processes

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PECASE
NIH BRAIN RF1MH117069
Heritage Medical Research Institute
Chen Institute at Caltech
Caltech BBE Postdoctoral department fellowship
The Hebrew University postdoctoral fellowship for women, Israel

Title: The role of suprachiasmatic VIP neurons in circadian rhythm regulation of the estrous cycle of female mice

Authors: *A. KAHAN, G. M. COUGHLIN, M. BORSOS, J. E. ROBINSON, V. GRADINARU;
Caltech, Pasadena, CA

Abstract: Female mice ovulate every 4-5 days at the beginning of the dark phase. The temporal specificity of this process points to a role for the circadian clock – and thereby the

suprachiasmatic nucleus (SCN), the circadian rhythm pacemaker – in ovulation. One key neuronal population in the SCN expresses vasoactive intestinal peptide (VIP); these neurons have both direct and indirect effects on gonadotropin-releasing hormone (GnRH) neurons, which control the release of reproductive hormones such as luteinizing hormone (LH). Although signaling from SCN^{VIP} to GnRH neurons has been hypothesized to control estrous cycle timing, it is unknown whether the activity of SCN^{VIP} neurons depends on the estrous state and how SCN^{VIP} neuronal signaling is relevant for ovulation. To address the first question, we recorded calcium-dependent fluorescence signals via fiber photometry (FP) from SCN^{VIP} neurons *in vivo* in female mice. In one data set, we recorded SCN^{VIP} FP activity over 24h (10 min/hr) for multiple days. In a separate data set we recorded SCN^{VIP} FP activity for 30 minutes at ZT10 (zeitgeber time), which corresponds to the LH surge onset, and for two hours during the transition from light to dark, ZT11-13. FP data showed that over the course of 24 hours, SCN^{VIP} activity is not dependent on the estrous cycle. However, longer recordings around ZT10-13 show estrous cycle dependency; at ZT11 these neurons are active at the rate of 0.7 ± 0.1 events/min while females are in estrus, higher by a factor of 1.4 compared to recordings from females in proestrus or diestrus ($P<0.05$), as confirmed by slice electrophysiology. These results suggest that SCN^{VIP} neurons are under hormonal control. To investigate the importance of SCN^{VIP} neurons for the regulation of estrous cycles, we experimentally manipulated the circadian rhythm by ablating SCN^{VIP} neurons through viral-mediated expression of Caspase-3, which triggers cell-autonomous apoptosis. We observed a significant decrease in estrous cycle regularity ($P<0.001$). In addition, dark-dark conditions with strict light exposures reduced fertility, as shown in reduced regularity of the estrous cycle ($P<0.005$) and oocyte release ($P<0.05$). When light was applied specifically at the LH onset, the fertility profile was rescued. Likewise, optogenetic and chemogenetic activation of SCN^{VIP} neurons during the same time window were able to partially rescue the estrous cycle irregularity arising from dark-dark conditions. These experiments provide new insights into the role of SCN^{VIP} neurons as regulators of ovulation occurrence, and suggest that SCN^{VIP} neuronal activity is also affected by the estrous cycle.

Disclosures: A. Kahan: None. **G.M. Coughlin:** None. **M. Borsos:** None. **J.E. Robinson:** None. **V. Gradinaru:** None.

Poster

P595. HPG Axis

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Program #/Poster #: P595.06

Topic: F.03. Neuroendocrine Processes

Title: The role of sex and ovarian cycles in trait variability across timescales and subjects

Authors: B. N. MARTIN[1], K. J. MOSHOFSKY[1], *A. BEERY[2,1];
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Abstract: Exclusive use of male subjects is common in animal research, based in part on the long-held assumption that females are more variable than males because of estrous cyclicity. Recent surveys of rat and mouse studies have found no evidence of different variability between the sexes for most outcomes, with males more variable slightly more often than females. These studies leave unresolved whether the sources of variability in males and females are similar. Similar variability could arise from high variability across days in females and high variability across individuals in males, or, males and females could have similar variability across timescales. There are indications that trait variability differs in male and female mice for body temperature but not food intake (Smarr et al. 2017, 2019). In the present study, we investigated the time-course of variability across several behavioral and physiological measures over 8 consecutive days of testing.
Female Syrian hamsters (*Mesocricetus auratus*) exhibit robust, precisely 4-day estrous cycles. Ovariectomized female hamsters were compared to intact (or sham operated) females and males. Measures included exploratory behavior and locomotion in an open field, novel object preference, sucrose preference and fluid intake, body mass, and internal body temperature. Sex differences were present in multiple traits, but as in other rodent species tested, there was no sex difference in the extent of inter-individual variability (assessed as coefficient of variation) between individuals in males and females. We also assessed the mean CV of each individual over time (intra-individual CV). Patterns of variation were trait specific rather than sex-specific; for example, males exhibited higher inter-individual variability in body temperature than females, but females had higher inter-individual variability in sucrose preference. Staging by estrous cycle phase did not reduce variability except for the most cycle-dependent trait (reproductive behavior), which was used as a positive control. This study extends the finding that females are not more variable than males to hamsters, and supports no consistent sex-specific patterns of variability within or across individuals in a variety of traits.

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Poster

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Title: Influence of gonadal steroids on cortical surface area in infancy

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Abstract: Sex differences in the human brain emerge as early as mid-gestation and have been linked to sex hormones, particularly testosterone. We analyzed markers of early sex hormone exposure (polygenic risk score (PRS) for testosterone, salivary testosterone level around 3 months of age, number of CAG repeats in the androgen receptor (AR) gene, digit ratios and PRS for estradiol) in relation to cortical surface area expansion in a longitudinal cohort of 722 infants. We focused on surface area because it shows strong sex differences in infancy with males exhibiting larger surface area, on average, at birth and growing faster than females. We used an orthogonal polynomial with order 2 for mixed model regression analysis between our sex hormone markers and surface area. Analyses were performed separately in males and females. We found PRS for serum testosterone and right-hand digit ratio to be significantly associated with surface area, but only in females. PRS for testosterone in the most stringent p value threshold was associated with increased surface area expansion over time. Higher right hand digit ratio, which is indicative of low prenatal testosterone levels, was associated with reduced surface area throughout infancy. This is the first study to explore potential associations between gonadal steroid exposure during early life and infant brain surface area. The current work suggests that variation in testosterone levels during both the prenatal and postnatal period may contribute to cortical surface area development and predicts inter-individual variation in females, but not in males.

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Poster

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Title: Reproductomics: regulation by neuropeptide processing

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Abstract: The reproductive axis has established layers of regulation: the gonadotropin releasing hormone (GnRH) neuron is controlled by Kisspeptin (Kiss), neurokinin B (NKB) and dynorphin A (DynA) which together comprise KNDy neurons in the hypothalamus. As neuropeptides, Kiss and DynA must be modified enzymatically in the extracellular space to modulate downstream signaling, though the enzyme(s) underlying this process remain elusive. Furthermore, Phoenixin (Phe), a hypothalamic neuropeptide plays a novel role in the regulation of the reproductive axis. Phe increases GnRH expression, GnRH receptor and Kisspeptin mRNAs, which ultimately supports GnRH secretion and ovarian cyclicity. We posit that EP24.15 (EC 3.4.24.15, thimet oligopeptidase, EP24.15) can serve the role as the metallo-endopeptidase for these neuropeptide regulators. EP24.15, expressed throughout the hypothalamus and other brain regions is best known for regulating GnRH by inactivation of binding to its cognate receptor. To validate the biochemical and physiological regulation of Kiss and DynA, we utilized a systems biology approach to hormone regulation by a high throughput pipeline including substrate scanning analyses to identify peptides, potential active site binding and cleavage sites by mass spectrometry, enzymatic kinetic parameters and *in silico* molecular modeling. Finally, we determined that EP24.15 and both Kiss and DynA immunoreactivities are co-expressed in the brain, identifying a functional neuroendocrine circuit whereby EP24.15 could regulate Kiss- or DynA -induced reproductive hormone secretion. Kiss and DynA immunoreactivities are co-expressed and localized with EP24.15 in the hypothalamus. To build upon mass spectrometry and kinetic analyses of EP24.15 cleavage of Kiss, DynA and Phoenixin we established high resolution 3D structural models of enzyme and substrate. Together, these data demonstrate that EP24.15 cleaves Kiss, DynA, and Phoenixin *in vivo* as an additional layer on the exquisite control mechanism of the mammalian reproductome. Similar analyses of Neurokinin B (NKB) demonstrated that while they are colocalized, NKB was not a substrate reinforcing the regulatory specificity of the enzyme in KNDy neurons and allowing for finer tuning of reproduction by EP24.15. Further investigating EP24.15 in neuropeptide metabolism and reproductive physiology will provide targets for modulating normal and aberrant reproductive function validating the high throughput reproductomics approach.

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Poster

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Topic: F.03. Neuroendocrine Processes

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Title: Reciprocal changes in voltage-gated potassium and subthreshold inward currents help maintain firing dynamics of anteroventral-periventricular (AVPV) kisspeptin neurons during the estrous cycle.

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Abstract: The brain initiates ovulation via a gonadotropin-releasing hormone (GnRH) surge, a process that requires estradiol action on central circuits. This surge is induced by estradiol positive feedback, induced when high estradiol levels cause a switch from a pulsatile pattern, regulated by estradiol negative feedback, to a surge of GnRH secretion. This occurs on the afternoon of proestrus in rodents. Positive feedback is thought to be conveyed to GnRH neurons by kisspeptin-expressing neurons in the AVPV. Kisspeptin is a potent GnRH secretagogue, and AVPV neurons express higher amounts of kisspeptin mRNA on proestrus compared to cycle phases, such as diestrus, when estradiol negative feedback prevails. AVPV kisspeptin neurons also fire more action potentials and more action potential bursts (associated with peptide release) on the afternoon of proestrus. Combined, these factors are thought to increase excitatory signaling to GnRH neurons and thus stimulate a GnRH surge. The mechanisms facilitating the increase in AVPV kisspeptin neuron electrical activity on proestrus are not fully understood. We hypothesized changes in voltage-gated potassium (K^+) currents help regulate the excitability of these cells in a cycle-dependent manner. Whole-cell voltage-clamp recordings of GFP-identified AVPV kisspeptin neurons in brain slices from diestrous and proestrous mice revealed three sub-components of the voltage-sensitive K^+ current: fast-inactivating (A-type), slow-inactivating, and residual. During proestrus, the V1/2 of inactivation of the fast current was depolarized and the amplitude of the slow-inactivating component was reduced compared to diestrus; the residual component was consistent across both stages. Computational models were fit to experimental data, including published estrous-cycle effects on other voltage-gated currents. Surprisingly, computer simulations suggest proestrus-typical K^+ currents are suppressive compared to diestrus.

Interestingly, larger T-type, persistent-sodium, and hyperpolarization-activated currents during proestrus compensate for this suppressive effect by increasing excitability and enabling post-inhibitory rebound bursting. These findings suggest modulation of both voltage-gated K^+ and multiple subthreshold depolarizing currents across the negative to positive feedback transition maintain AVPV kisspeptin neuron excitability in response to depolarizing stimuli. These changes also enable firing following hyperpolarization, which may attenuate or reverse effects of inhibitory input, thus contributing to activation of this population leading up to the preovulatory GnRH surge.

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Poster

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Title: Relationship between action potentials and hormone release in gonadotropin-releasing hormone (GnRH) neurons is modified by gonadal feedback

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Abstract: In vertebrates, the pulsatile release of GnRH from neurons in the preoptic area (POA) and hypothalamus plays a critical role in triggering secretion of the gonadotropins, luteinizing hormone (LH) and follicle-stimulating hormone (FSH) from the anterior pituitary. LH and FSH activate steroidogenesis, and steroids in turn exert feedback on GnRH release. Although long-term changes in episodic firing pattern of GnRH neurons resemble the pulsatile release of GnRH and LH, the relationship between short-term GnRH neuron firing and release, and how the relationship is influenced by gonadal feedback are unknown. We used voltage-clamp to control GnRH neuron membrane potential while monitoring GnRH release by fast-scan cyclic voltammetry in sagittal brain slices from gonad-intact (sham) and orchiectomized (ORX) male GnRH-GFP mice. GnRH release was detected in the POA at appositions of pairs of GFP-identified cells/processes. A train of 2, 5 or 15 action potentials (APs) with a constant interval of 213.9ms (median intraspike interval of spontaneous bursts) was used as command voltage to manipulate cell firing. To test if there was a refractory period for induced release, a second identical train was given to the cell four minutes after the first one. AP-triggered GnRH release began 0.6 to 79.4s after stimulation (mean 30 ± 3s, n=39 releases). GnRH release was trigged by

2APs in 50% of sham (4/8) and 70% of ORX (7/10) trials; the peak of the first triggered release was similar between sham (6.6±1.4 mM n=4) and ORX (7.2±0.6 mM n=5) groups. 5APs triggered release in 90% (9/10) of sham and 100% (10/10) of ORX trials. Preliminary data suggest amplitude of the first 5AP-triggered GnRH release may be increased by ORX (sham 10.7±2.6mM, n=5, ORX 20.6±2.9mM, n=6, p=0.09). In ORX mice, more GnRH was released after 5 than after 2 APs ($p<0.05$). All 15AP stimulations triggered GnRH release in both groups. The first 15AP-triggered release was greater than that following 5APs in sham (19.1±6.2 mM, n=4) but not ORX (19.8±3.2 mM, n=3) groups. The 2nd stimulation-triggered GnRH release was lower than the 1st in 77.7% (7/9) of cells given 5AP stimulation and 85.7% (6/7) of cells given 15AP stimulation, suggesting some depletion of releasable pool. In addition to triggered release, spontaneous releases were observed before (8-135s) and well after (105-445s) stimulations; these releases may be AP-independent (Endocrinology 156: 231) or arise from the un-patched cell of the pair. These results suggest coupling between GnRH neuron firing and release is stronger with increased stimulation, that a relative release refractory period may exist, and that ORX increases sensitivity of secretory mechanisms to APs.

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Poster

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Topic: F.03. Neuroendocrine Processes

Title: Serotonin stimulates preoptic area kisspeptin neuron activity in female mice

Authors: *R. PIET, A. ANELLO, C. BUO, J. DAKIN, R. BEARSS;
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Abstract: Ovulation is controlled by brain circuits that regulate the secretion of gonadotropin-releasing hormone (GnRH) from the hypothalamus and luteinizing hormone (LH) from the anterior pituitary. Kisspeptin (Kiss1) neurons, which project to GnRH neurons, have emerged as key regulators of GnRH neuron activity and LH secretion. Kiss1 neurons located in the preoptic area (POA^{Kiss1}) of the hypothalamus are involved in generating the preovulatory surge in LH secretion that triggers ovulation. In addition to Kiss1, other neurotransmitters and neuropeptides likely regulate the preovulatory LH surge. Previous studies have indicated that serotonin (5HT) substantially contributes to generating the estrogen-induced LH surge in ovariectomized female rodents (Dow et al 1995, Psychoneuroendocrinology). Here, we hypothesized that the impact of 5HT on the LH surge results from excitatory effects on POA^{Kiss1} neuron activity. To address this question, we used mice that express Cre recombinase in Kiss1 cells and a Cre-dependent, genetically-encoded fluorescent calcium indicator GCaMP6f. In brain slices obtained from these

mice, we used live-cell epifluorescence imaging to determine changes in intracellular calcium concentrations $[Ca^{2+}]_i$ in multiple POA^{Kiss1} neurons simultaneously, as an index of their activity. In brain slices from female mice, 10 μM 5HT increased $[Ca^{2+}]_i$ (3.1±0.4% change in fluorescence) in 89% of POA^{Kiss1} neurons (n=74 cells in 7 slices from 4 mice). This effect was smaller in male mice where 48% of POA^{Kiss1} neurons showed $[Ca^{2+}]_i$ rises (0.5±0.3% change in fluorescence; n=54 cells in 5 slices from 3 mice). We next investigated if the effect of 5HT was mediated directly at POA^{Kiss1} neurons or via an indirect mechanism. Two successive 5HT applications evoked similar responses in POA^{Kiss1} neurons and the amplitude of the response to the second application was on average 83.1±12.4% of that of the first one, indicating that the effect of 5HT is repeatable. We then tested the effect of blocking action potentials in the slice using tetrodotoxin (TTX, 0.5 μM). 5HT-induced rises in POA^{Kiss1} neuron $[Ca^{2+}]_i$ in the presence of TTX were comparable to those evoked without TTX (n=68 cells in 7 slices from 4 mice; $p>0.37$, Wilcoxon test) and the response ratio was similar to that measured in control experiments (83.5±15.5%, $p>0.99$ Mann-Whitney test). Together these data indicate that 5HT stimulates the activity of POA^{Kiss1} neurons, likely through a direct mechanism. This effect is more prominent in females than in males. Our data suggest a potential mechanism through which 5HT might regulate the preovulatory surge in female rodents.

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Poster

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Title: Evidence that elevated agouti-related peptide signaling during undernutrition inhibits arcuate kisspeptin neurons in male sheep

Authors: S. L. SHUPING[1], A. N. RENWICK[2], K. HARLOW[1], J. R. SOMMER[1], *C. C. NESTOR[1];
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Abstract: Proper energy balance is important to ensure reproductive success. While it is known that nutrient restriction impairs hypothalamic-pituitary function, the central mechanism whereby undernutrition inhibits reproduction remains to be fully elucidated. Given recent evidence in

sheep and mice that the majority of arcuate kisspeptin neurons express melanocortin 3 receptors (MC3R), anorexigenic neurons which produce agouti-related peptide (AgRP), an endogenous MC3R antagonist, may play an important role in relaying energy status to the reproductive axis. Since kisspeptin neurons are essential for reproduction and undernutrition increases AgRP expression, we hypothesized feed restriction would increase AgRP input to kisspeptin neurons and inhibit kisspeptin expression in MC3R-expressing neurons. We selected tissue containing the arcuate nucleus from wethers that were fed to maintain pre-study body weight (FM; n=4) or feed-restricted to lose 20% of pre-study body weight (FR; n=4). Using left brain hemisections (n=2/animal), we conducted dual-label immunofluorescence with primary antisera for kisspeptin and AgRP. Using right brain hemisections (n=2/animal), we conducted RNAscope for mRNA expression of kisspeptin and MC3R. Using immunohistochemistry, we observed significantly more AgRP close-contacts onto ARC kisspeptin cells in FR wethers (5.96 $\pm$ 0.9 contacts/cell) than in FM wethers (3.33 $\pm$ 0.2 contacts/cell). RNAscope analysis revealed significantly fewer total kisspeptin cells in FR animals (91 $\pm$ 15 cells) compared to FM animals (164 $\pm$ 17 cells), while there was no significant difference for total MC3R cells between groups (FM, 193 $\pm$ 21 cells; FR, 180 $\pm$ 30 cells). In addition, we observed significantly fewer cells co-expressing mRNA for kisspeptin and MC3R in FR animals (37.1 $\pm$ 6 cells) compared to FM animals (104.8 $\pm$ 9 cells). Furthermore, we observed that the integrated density (mRNA per cell) for kisspeptin in kisspeptin/MC3R cells tended to be lower ($p = 0.052$) in FR wethers (920737 $\pm$ 431678 AU) compared to FM wethers (2128366 $\pm$ 380764 AU). Together, we suggest that AgRP acts to inhibit kisspeptin neurons and plays a key role in relaying low energy status to reduce GnRH/LH secretion and ultimately impair reproduction.

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Poster

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Title: Study on the method of gene delivery to the hypothalamic arcuate nucleus using goats and newly established goat kisspeptin neuron cell lines

Authors: *F. MATSUDA[1], K. OKUDA[2], Y. SUETOMI[2], K. KOBAYASHI[3], S. OHKURA[2]; [1]Dept. of Vet. Med. Sci., The Univ. of Tokyo, Tokyo, Japan; [2]Grad. Sch. of Bioagricultural Sci., Nagoya Univ., Nagoya, Japan; [3]Section of Viral Vector Develop., Natl. Inst. For Physiological Sci., Okazaki, Japan

Abstract: Kisspeptin neurons located in the hypothalamic arcuate nucleus (ARC) is known to be the key regulator of pulsatile gonadotropin-releasing hormone (GnRH) secretion and the following gonadotropin pulses that stimulate gonadal activity in mammals. The goat is a good experimental animal not only for having similar characteristics with cattle but for the usefulness in physiological analyses. Important studies related to the regulation mechanism of hypothalamus-pituitary-gonadal (HPG) axis including the function of ARC kisspeptin neurons have been reported using goats. Optogenetic regulation of specific neurons have recently been developed, which is expected to be also effective to artificially regulate kisspeptin neurons in goats to further clarify their functions. In this study, we aimed to establish the method to introduce a light-gated ion channel gene into the hypothalamus using goats and newly established goat kisspeptin neuron cell lines. Experiment 1: In vivo analyses to establish the method to deliver genes into the goat ARC by adeno-associated virus (AAV) vector. AAV of four serotypes (AAV-1, 2, 5, and DJ), loading a light-gated ion channel (channelrhodopsin-2, ChR2) gene and a fluorescent protein gene (tdTomato), was generated. The AAV of each serotype was injected into four different parts of the ARC of castrated male Shiba goats (n = 3) with volume of 0.3 μl and flow rate of 0.03 μl/min. Three to four weeks later, brains were obtained and tdTomato fluorescence and immunoreactivity of kisspeptin in the ARC were examined. tdTomato fluorescence and kisspeptin-immunoreactivity were detected in the equivalent area of the ARC in adjacent sections, suggesting that ChR2 gene might be introduced into ARC kisspeptin neurons. tdTomato fluorescence was relatively stable and strong in AAV-1 and DJ serotypes, suggesting the advantage of these serotypes in introducing genes into goat brains. Experiment 2: In vitro analyses to evaluate the efficiency of gene delivery by AAV using immortalized kisspeptin neuron cell lines derived from goat ARC, GA28 and GA64. The AAV of four serotypes used in the Experiment 1 was added to GA28 and GA64 and after the 14 days of culture, cells with tdTomato fluorescence were counted by fluorescent activated cell sorting (FACS). As a result, the numbers of fluorescent cells were higher in AAV-2, 5, and DJ serotypes compared with control and AAV-1. Taken together, AAV-DJ was likely to be the most suitable serotype to deliver genes to ARC kisspeptin neurons in goats. The method introducing ChR2 into the goat hypothalamus by AAV we established in this study will contribute to further elucidate the function of ARC kisspeptin neurons.

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Poster

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Title: In vivo calcium imaging of arcuate nucleus KNDy neurons reveal the GnRH pulse generator in awake, mobile female mice

Authors: *A. M. MOORE, L. M. COOLEN, M. N. LEHMAN;
Biol. Sciences, Brain Hlth. Res. Inst., Kent State Univ., Kent, OH

Abstract: Reproduction is critically dependent on luteinizing hormone (LH) pulses driven by episodic release of gonadotropin-releasing hormone (GnRH). It is hypothesized that neurons in the arcuate nucleus (ARC) expressing kisspeptin, neurokinin B and dynorphin (KNDy cells) are capable of synchronized, episodic activity driving GnRH pulses, and thus constitute the so-called "GnRH pulse generator". However, without *in vivo* visualization of KNDy cell activity at a single-cell level this hypothesis remains unproven. Using *in vivo* calcium imaging we examined the activity of KNDy neurons at single-cell resolution in awake, freely moving ovariectomized female mice. Kiss1-Cre mice were stereotaxically injected with a Cre-dependent AAV containing the calcium indicator GCaMP6s into the unilateral ARC. A 500µm diameter Gradient-Index (GRIN) lens (Inscopix Inc) was inserted above the ARC and secured in place by attachment of an integrated baseplate to the skull. After 5 weeks of habituation, an Inscopix Miniscope was mounted to the baseplate and recordings were performed at 10Hz for 1 hour (n=5 mice). Serial blood samples were collected from the tail-tip every 3-4 minutes for analysis of LH release. Fluorescent traces were extracted from 12.2 ± 1.2 cells per animal using the PCA-ICA cell detection algorithm. The KNDy population exhibited episodic periods of elevated fluorescence across the recording period, with a duration of 77.0 ± 6.8 seconds and an interval of 4.1 ± 0.7 min. Our analysis showed that 75.2 ± 5.7% of these episodic events involved activation of all recorded cells, depicting complete synchronization of the population. All other events, in which only 7.1-71.4% of the recorded cells were activated, exhibited significantly lower amplitude and duration compared to events where 100% of cells were recruited ($p<0.05$). Importantly, LH pulses were always preceded by an event (within 3.1 ± 0.3 min) in which all cells were synchronously activated. Analysis of the order of activity onset and peak amplitude during synchronized events (from an average of 11.2 ± 0.9 synchronized events per animal) revealed a small population of cells that slowly activate to begin an event, followed by rapid activation of a separate population of cells that reach peak amplitude first. Cells first to activate

or first to reach peak amplitude comprised a separate population from those that did so later. Together, these data support that synchronized activation of KNDy cells drives pulsatile LH release and suggest the existence of discrete subsets of leader and follower cells that are activated during individual pulses.

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Poster

P638. Sleep: Systems

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Title: Locus coeruleus noradrenergic control of non-REM sleep substates

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Abstract: Noradrenergic neuromodulation of the mammalian brain is wake- and attention-promoting. Noradrenaline (NA) is released into the forebrain by the pontine locus coeruleus (LC). However, the LC is also active during non-REM sleep, playing roles in sleep spindle activity (Aston-Jones et al., 1981; Swift et al., 2018), sensory arousability (Hayat et al., 2019) and sleep-dependent memory consolidation (Gais et al., 2011). Still, how much NA is released during non-REM sleep, where it acts and what its roles are for non-REM sleep has remained under-investigated. We used the newly developed NA biosensors $GRAB_{NE1h}$ or $GRAB_{NE1m}$ (Feng et al., 2019) in combination with polysomnographic monitoring of natural sleep in C57BL/6J mice (either sex, 5-9 weeks-old). We observed recurrent fluctuations of the wake-promoting neurotransmitter NA in the primary thalamic somatosensory nucleus on the infraslow, ~50-seconds time-scale. These fluctuations occurred around mean NA levels greater than the ones of quiet wakefulness, while they declined steeply in REM sleep. Low NA levels coincided with a clustering of sleep spindle rhythms in the forebrain and with low heart rate, whereas high NA levels occurred when sleep spindles were rare and when heart rate increased. We addressed the origins of these fluctuations by using a) local thalamic pharmacology to antagonize noradrenergic signaling and b) closed-loop optogenetic LC activation or inhibition timed to NREMS or to moments of low and high spindle activity. Pharmacological blockade of noradrenergic signaling in thalamus suppressed infraslow variations in sleep spindle clustering

while leaving spindle properties intact. Using optogenetic LC activation or inhibition in dopamine-β-hydroxylase-Cre mice expressing hChR2(H134R) or Jaws in a Cre-dependent manner, we could suppress, lock or entrain sleep spindle clustering or heart rate variations. Noradrenergic modulation of thalamic but not cortical circuits seemed primarily required for sleep spindle clustering. Whole-cell patch-clamp recordings showed that local LC fiber-mediated NA release onto reticular thalamic nuclei or thalamic sensory neurons activated α1- or β-adrenergic receptors, respectively, to depolarize membrane potentials over the tens-of-seconds time scale. Noradrenergic signaling by LC thus divides non-REM sleep into substates that differ in terms of central (spindles) and autonomic (heart rate) physiological correlates. As these correlates parallel varying sensory arousability from non-REM sleep (Lecci et al., 2017; Cardis et al., 2021), we propose that LC signaling renders non-REM sleep more 'wake-like' on the close-to-minute-time scale

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Poster

P638. Sleep: Systems

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P638.02

Topic: F.08. Biological Rhythms and Sleep

Support: NIH Grant 1R01GM116916
Tiny Blue Dot Foundation

Title: Bilateral optogenetic stimulation of posterior parietal cortex leads to EEG activation and recovery of the righting reflex in a mouse model of coma

Authors: *R. MAO, M. CAVELLI, K. A. DRIESSEN, G. FINDLAY, G. TONONI, C. CIRELLI;
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Abstract: Years of experimental evidence points to the cerebral cortex as an essential brain structure in supporting conscious experience. Despite this, we still do not know whether posterior or frontal cortical areas alone are sufficient for consciousness, nor do we know the precise cortical mechanisms at work. To address these questions we first optimized in mice a protocol previously validated in rats, to induce a reversible state of profound, coma-like unresponsiveness via muscimol injections in the midbrain (Lanir-Azaria and Meiri et al., 2018). We then tested whether direct optogenetic cortical stimulation can reverse this coma-like state. CaMKIIα-Cre mice (3 males) were injected with rAAV5-Ef1a-DIO-hChR2(C128S/D156A-

EYFP) in the posterior parietal association area (PPtA, left or bilateral; from bregma, A/P -2.10, M/L ±1.50, D/V -0.60). Other CaMKIIα-Cre mice were crossed with Cre-dependent ChR2(H134R)/EYFP mice and adult offspring (CaMKIIα::ChR2) of both sexes were used (8 mice, 3 females). All mice were implanted with bilateral guide cannulas for muscimol injection in the midbrain, left or bilateral optic fibers for optogenetic stimulation of PPtA, EEGs and muscle electrodes, and cortical laminar silicon probes. In all mice (n = 11), muscimol injections in the midbrain induced a coma-like state characterized by loss of righting reflex, significantly reduced responsiveness to a battery of stimuli, and an EEG pattern dominated by slow waves and a high ratio of delta (0.5-4.5 Hz) to gamma (32-100 Hz) band power. All mice spontaneously exhibited recovery of righting reflex (RORR) after 1.5-4.5 hours and behaved normally the next day. In CaMKIIα::ChR2 mice the bilateral optogenetic stimulation of PPtA (0.3-4.7mW at cortex) during the coma-like state consistently led to EEG activation (decreased low frequencies, increased high frequencies) and full RORR. In the same mice, the same or stronger stimulation never resulted in EEG and behavioral activation under deep sevoflurane- dexmedetomidine anesthesia (1.0-1.5% sevoflurane, 70-100ug/kg dexmedetomidine), while brief stimulations were sufficient to wake up the animals from NREM sleep or REM sleep. In the mice that received unilateral stimulation during the coma-like state, full RORR or RORR attempts with partial EEG activation were observed. Unilateral stimulation was always able to wake up the mice from sleep. These results suggest that the direct activation of cortical pyramidal neurons in mouse posterior parietal area alone is sufficient for the recovery of consciousness.

Disclosures: R. Mao: None. **M. Cavelli:** None. **K.A. Driessen:** None. **G. Findlay:** None. **G. Tononi:** None. **C. Cirelli:** None.

Poster

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Topic: F.08. Biological Rhythms and Sleep

Support: NIH Grant 1R01GM116916
Tiny Blue Dot Foundation

Title: Sleep/wake changes in perturbational complexity in mice and rats

Authors: *M. CAVELLI, R. MAO, G. FINDLAY, K. DRIESSEN, T. BUGNON, G. TONONI, C. CIRELLI;
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Abstract: The Perturbational Complexity Index (PCI) and the related PCI^{ST} (ST = “state transitions”) are causal measures of spatiotemporal complexity in cortical event related potentials

(ERPs). In human subjects receiving transcranial magnetic or deep intracranial electrical stimulation, PCI and PCI^{ST} show reliable changes depending on the presence or absence of consciousness: they are high during wake and REM sleep, and low during dreamless NREM sleep and slow wave anesthesia (Casarotto et al., 2016; Comolatti et al., 2019). Here, we tested whether these measures also change across the sleep/wake cycle in mice and rats. Moreover, we tested the assumption that low PCI^{ST} values are linked to cortical bistability, which could disrupt causal interactions and lead to stereotypical evoked responses by promoting the occurrence of a large OFF period in response to stimulation. All animals were chronically implanted with cortical laminar probes (NeuroNexus or Neuropixels). The offspring of CaMKIIα-Cre mice crossed with Cre-dependent ChR2 (H134R)/EYFP mice (6 mice, both sexes) received optogenetic stimulation in the posterior parietal association area, and ERPs were recorded in posterior cortex. Rats (3 males) received electrical stimulation in primary sensory, motor and posterior associative cortex, and ERPs were recorded in ipsilateral or contralateral cortex. In each animal PCI^{ST} values were lower in NREM sleep than in wake or REM sleep (% relative to wake = 100: NREM sleep = 53.0 ± 22.2%; REM sleep = 87.7 ± 28.3%; ANOVA $F_{(2.4\ 17.4)} = 21.2$, $p < 0.0001$; NREM vs wake and NREM vs REM, both $p < 0.05$ after correction for multiple comparisons). In the same animals PCI^{ST} values were also low during deep anesthesia induced by sevoflurane and dexmedetomidine (29.5 ± 23.6% of wake). Consistent with results in humans, PCI^{ST} values and the Phase Locking Factor (PLF), which measures how long the evoked response remains phased locked to the original stimulus, were positively correlated ($r = 0.5$; $p = 5.9e^{-4}$). In both rats and mice, unit analysis showed that the early cortical response to stimulation is an OFF period that is most prominent during slow waves states. Together, these results show that PCI^{ST} values in rodents change across the sleep/wake cycle in a manner consistent with results obtained in humans. Low PCI^{ST} values are linked to the occurrence of OFF periods and short PLFs, which are believed to reflect the breakdown of causal interactions necessary for consciousness (Pigorini et al., 2015; Tononi et al., 2016).

Disclosures: M. Cavelli: None. **R. Mao:** None. **G. Findlay:** None. **K. Driessen:** None. **T. Bugnon:** None. **G. Tononi:** None. **C. Cirelli:** None.

Poster

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Program #/Poster #: P638.04

Topic: F.08. Biological Rhythms and Sleep

Support: U.S. Department of Defense MURI grant W911NF1910280
Tiny Blue Dot Foundation

Title: Sleep homeostasis in rat cortex and hippocampus

Authors: *G. FINDLAY[1], M. CAVELLI[1], W. MARSHALL[2], G. TONONI[1], C. CIRELLI[1]; [1]Psychiatry, Univ. of Wisconsin-Madison, Madison, WI; [2]Mathematics and Statistics, Brock Univ., St. Catharines, ON, Canada

Abstract: Slow waves are a defining feature of NREM sleep and reflect sleep need: cortical slow wave activity (CX-SWA) increases with time spent awake and decreases in the course of NREM sleep. Hippocampal slow wave activity (HC-SWA) and sharp-wave ripples, on the other hand, are hallmarks of NREM sleep but also occur in wake, and whether they reflect sleep need remains unclear. HC-SWA in cats (Lancel et al., 1992) and ripple occurrence in rats (Ponomarenko et al., 2003) increases after sleep deprivation (SD), but sharp waves (SPWs) were not analyzed. Although they often co-occur, SPWs and ripples are independent events produced by different synaptic networks (Buzsaki, 2015). Here we studied in rats how CX-SWA, HC-SWA, and SPWs change after SD.

Rats (6 males) were implanted with cerebellar-referenced Neuropixel probes in medial parietal association cortex (MPtA) and dorsal hippocampus. After 24h of baseline recording, rats underwent 6h SD by exposure to novel objects, followed by recovery sleep. SPWs were detected as large current sinks (>2.5 std. dev. above the mean current source density) in the stratum radiatum. Cortical and hippocampal signals were re-referenced to the white matter between MPtA and CA1 before computing CX-SWA and HC-SWA (0.5-4 Hz power spectral density). CX-SWA and HC-SWA in each rat were expressed as percentages of their respective baseline NREM medians.

CX-SWA increased in the first 2h after SD compared to circadian-time-matched baseline sleep ($n = 5$; $p = 0.001$, paired sample t-tests throughout) and declined from early to late recovery sleep ($n = 6$, $p = 0.002$). HC-SWA showed similar changes ($n = 5$, rebound, $p = 0.0002$; $n = 6$, decline, $p = 0.0002$), and increased from early to late SD ($n = 6$, $p = 0.002$ c.f. $p=0.102$ in cortex). HC-SWA rebound and decline were smaller than for CX-SWA ($n = 5$, relative rebound, $p = 0.006$; $n = 6$, relative decline, $p = 0.016$). Rate and amplitude of SPWs were elevated in the first 2h after SD compared to circadian-time-matched baseline sleep ($n = 5$, rate, $p = 0.003$; amplitude, $p = 0.013$), declined during recovery sleep ($n = 6$, rate, $p = 0.001$; amplitude, $p = 0.001$), with rate also increasing from early to late SD ($n = 6$, $p = 0.016$ c.f. $p = 0.074$ for amplitude). Although CX-SWA rebound and decline were strongly correlated ($n = 5$, Pearson's $r = 0.95$, $p = 0.012$), no similar correlation was present in the hippocampus, and hippocampal changes showed no consistent correlation with cortical changes.

Together, these results show that both HC-SWA and SPWs increase with time spent awake and decline during sleep. Their homeostatic behavior, however, seems weakly correlated with- and blunted relative to homeostasis of CX-SWA.

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Poster

P638. Sleep: Systems

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P638.05

Topic: F.08. Biological Rhythms and Sleep

Support: University of Michigan Medical School: Startup Funding

Title: A novel rhythm within REM sleep: hippocampal theta frequency, rapid movements and neocortical laminar LFP co-fluctuate in infraslow cycles

Authors: L. S. BUENO-JUNIOR, M. RUCKSTUHL, B. O. WATSON;
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Abstract: Rapid eye movement sleep (REM) has classically been divided into tonic and phasic microstates. To more finely determine this intra-REM structure, we recorded electrophysiologically from across the mouse S1 cortex and hippocampus at once using a linear silicon probe, while filming whisker and eye movements during sleep. We observed that rapid eye and whisker movements indeed burst in delimited periods, similar to the concept of phasic REM. However, hippocampal theta frequency (5-10 Hz, commonly used to delineate REM in rodents) was found to fluctuate in a non-binary fashion, contrary to what we would expect from the dichotomous concept of tonic REM vs. phasic REM. Interestingly, those fluctuations were at 0.02-0.06 Hz, an increasingly recognized brain rhythm known as infraslow. Rapid movements occurred preferentially at the crest of infraslow cycles, supporting this underlying periodicity. We additionally found that cross-frequency coupling between theta and high-frequency oscillations (HFO, 100-160 Hz) also fluctuates continuously in the infraslow range. This phenomenon varied in strength across our recording probe profile, being more evident in the hippocampus and either superficial or deep, but not intermediate cortical channels. Therefore, we expand the characterization of rodent REM to include co-fluctuations that span multiple frequency bands, and are anatomically uneven along cortical-hippocampal depths. We also demonstrate that the temporal structure of rodent REM is organized in continuous cycles of dozens of seconds, which could be speculated to govern mnemonic and homeostatic processes. Investigating these rhythms in other species may enable better dissection of REM and related disorders.

Disclosures: L.S. Bueno-Junior: None. **M. Ruckstuhl:** None. **B.O. Watson:** None.

Poster

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Program #/Poster #: P638.06

Topic: F.08. Biological Rhythms and Sleep

Support: University of Michigan Medicine: Startup Funding

Title: Rem sleep microstates vary in neuronal activity across neocortex and hippocampus

Authors: *M. S. RUCKSTUHL, L. S. BUENO-JUNIOR, B. O. WATSON;
Dept. of Psychiatry, Univ. of Michigan Med. Sch., Ann Arbor, MI

Abstract: Traditionally, REM sleep is divided in human studies into phasic and tonic REM based on eye movement, and these microstates are thought to differ with regard to sensitivity to sensory stimuli. These microstates are believed to exist in rodents as well, as reflected in the heterogeneity of peak theta frequency during REM. However, we have found that peak theta frequency actually fluctuates continuously at an infraslow rhythm, suggesting that phasic and tonic REM exist on a continuum. Here we examine the impact of that continuum on neuronal activity across somatosensory cortex and hippocampus. We found that most neurons exhibit a phase preference for the peak of the infraslow rhythm, when theta frequency is highest. This concords with prior findings that firing rate tracks with theta frequency in awake animals and explains greater population activity during what is conventionally defined as phasic REM. We find that this is true across cell types and layers of neocortex and is not predicted by the baseline firing rate of a neuron. To better understand how cortical neurons work in concert, we performed a complementary analysis of assembly activity during REM sleep. We found that groups of consistently co-firing neurons tend to peak in activity during periods of high-frequency theta, and that assemblies detected during these periods are maximally dissimilar to assemblies detected during low-frequency theta. This exploration of neuronal activity during REM microstates informs our ongoing investigation of the structure of REM and sleep in general. This may lead to later insight about the impact of REM sleep on homeostasis and memory.

Disclosures: M.S. Ruckstuhl: None. **L.S. Bueno-Junior:** None. **B.O. Watson:** None.

Poster

P638. Sleep: Systems

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Program #/Poster #: P638.07

Topic: F.08. Biological Rhythms and Sleep

Title: The role of OLM interneurons in REM sleep dependent memory consolidation

Authors: *M. FRAZER, G. POE;
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Abstract: Forming and maintaining accurate spatial and contextual memories is important to an animal's ability to adapt to its environment. A growing body of evidence suggests that sleep is necessary for the proper consolidation of memory. Disordered sleep has numerous negative

effects on cognitive function, and is comorbid with nearly every common psychiatric illness. Thus, understanding why sleep is important to memory function is necessary for a full understanding of disorders in which sleep is affected. The role of rapid eye movement (REM) sleep, in particular, is contested and largely undetermined. Correlative studies suggest that depriving animals of REM results in deficits in spatial memory, and point to the necessity of the theta rhythm of REM. In waking, the theta rhythm is thought to temporally segregate inputs into the CA1, allowing for both encoding and retrieval; the role of REM theta, however, remains unknown. Hippocampal theta is driven by interactions between medial septal projections, pyramidal cells, and a diverse group of inhibitory cells. One such interneuron is the somatostatin-expressing oriens-lacunosum moleculare (OLM) cell, which fires spontaneously at the theta rhythm, and is phase-locked to theta peaks. Recent studies find that normal OLM cell activity is necessary for encoding spatial and contextual information during waking; the role they play in sleep-dependent consolidation, however, is unknown. To investigate the role of local inhibition in sleep-dependent consolidation, we have optogenetically silenced OLM interneurons selectively during REM sleep episodes, using a recently-developed mouse line specifically targeting OLM cells. We find that modulating OLM activity during REM sleep following acquisition of novel object place recognition (NOPR) tasks and contextual fear conditioning (CFC) disrupts typical consolidation. This suggests a role for OLM cells in consolidating hippocampal-dependent contextual tasks during REM sleep.

Disclosures: M. Frazer: None. **G. Poe:** None.

Poster

P638. Sleep: Systems

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Title: A role for the cortex in sleep-wake regulation

Authors: *L. B. KRONE[1,2], T. YAMAGATA[3,2], C. BLANCO-DUQUE[5,2,1], M. C. C. GUILLAUMIN[6,3], M. C. KAHN[5,1,2], V. VAN DER VINNE[7,1,2], L. E. MCKILLOP[2,1], S. K. E. TAM[2,3], S. N. PEIRSON[2,3], C. J. AKERMAN[4], A. HOERDER-SUABEDISSEN[1], Z.

MOLNAR[1], V. V. VYAZOVSKIY[1,2];
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Abstract: Sleep is vital yet its regulation in the mammalian brain is poorly understood. The conventional view is that the cortex generates state-dependent brain oscillations, while subcortical brain structures control global sleep-wake switching. However, neocortex can locally generate sleep-like activity when homeostatic sleep pressure is high and bistable firing patterns in neocortex foreshadow sleep onset. In this study we tested if cortex contributes to vigilance state control and global sleep homeostasis. We performed chronic electrophysiological and passive infrared recordings to assess sleep and circadian rhythms in a transgenic mouse line (Rbp4-Cre;Ai14;Snap25$^{fl/fl}$), in which a subset of neocortical layer 5 pyramidal and archicortical dentate gyrus granule cells are functionally silenced by ablation of the synaptosome associated protein 25 (SNAP25). Laminar probe recordings from primary motor cortex showed layer-specific changes in neocortical dynamics during slow wave sleep in male cortical SNAP25-ablated mice. In addition, these conditional knockout mice (cKO, n=9) spent more time awake (Mean 13.8h ± SEM 0.4h) compared to Cre-negative controls (10.4 ± 0.4h, n=7, p<0.001, unpaired t-test, Cohen's d=2.96) and presented longer individual wake episodes. Following sleep deprivation, the level of frontal electroencephalographic (EEG) slow wave activity (SWA) during non-rapid eye movement (NREM) sleep, a measure of sleep intensity, was significantly lower in cortical SNAP25-ablated mice compared to controls (8 cKO mice: 136.77% ± 3.98%; 5 controls: 180.57% ± 5.13%; p<0.001, unpaired t-test, Cohen's d=-3.87). Circadian measures including period length, periodogram power, and the phase shift after a phase-delaying light pulse showed no significant differences between genotypes. Taken together, our study reveals an essential role for the cortex in sleep-wake regulation. Our data demonstrate that cortical SNAP25 ablation leads to a diminished homeostatic sleep drive without affecting core circadian parameters. These results support the possibility that cortical structures generate sleep drive locally, in an activity-dependent fashion, and raise the question of which mechanisms the cortex could use to generate, sense and/or integrate signals of sleep need. The next step will be to delineate these molecular mechanisms and to understand how the cortex interacts with established circuits of sleep–wake control. Further research on the cortical control of sleep will pave the way for a more comprehensive understanding of sleep regulation in the mammalian brain and help identify novel targets for the treatment of sleep disorders.

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Poster

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Topic: F.08. Biological Rhythms and Sleep

Support: VA BX000798
VA 1K6BX004216

Title: Zona incerta Lhx6 neurons are most active during NREM and REM sleep and after prolonged waking

Authors: C. A. BLANCO-CENTURION[1], A. VIDAL-ORTIZ[2], ***P. SHIROMANI**[2];
[1]Psychiatry, Med. Univ. of South Carolina, Charleston, SC; [2]Res., Ralph H Johnson VA Med. Ctr., Charleston, SC

Abstract: Emerging data shows that GABA neurons in the zona incerta (ZI) play a prominent role in regulating sleep. Transfer of the orexin gene into ZI neurons blocks cataplexy in narcoleptic orexin-knockout mice (Liu et al., JNeurosci, 2011) and vGAT GABA neurons in the ZI anticipate onset of NREM (Blanco-Centurion et al., SLEEP, 2021). To identify the subtype of GABA neurons regulating sleep, we examine Lhx6 neurons. This study tests the hypothesis that Lhx6 neurons show peak calcium fluorescence during sleep, and that the fluorescence is further increased after prolonged waking. In Lhx6-cre mice (mice=4; all females; 4-5 mo), rAAV-DIO-GCaMP6s was delivered stereotaxically to the ZI (isofluorane anesthesia) and a GRIN lens, along with EEG and EMG electrodes were implanted. 21d later a miniscope (INSCOPIX) was attached, and after 3d of adaptation, sleep and fluorescence in individual Lhx6 neurons were recorded for 4h (baseline). On another day, the mice were kept awake for 6h (gentle handling; 9a-3p) and fluorescence in Lhx6 neurons was recorded for 2h during recovery sleep. The imaged data from the two recording periods (baseline and recovery sleep) was combined into a single data file and the change in fluorescence was determined against the mean image frame (F0). Previously, we and others found that the GCamP calcium fluorescence is a direct measure of action potentials and serves as a marker of activity. 97 neurons were automatically extracted (PCA-ICA analysis; blinded without knowledge of sleep state). In 66 neurons (68%) the average fluorescence was significantly higher during REM, NREM or both, compared to waking (Mixed Model ANOVA; SPSS25; $P<0.01$). In this population the fluorescence was significantly higher during recovery sleep compared to baseline ($P<0.001$) indicating increased activity of the sleep-active neurons during recovery sleep. With ensuing sleep, the increase in fluorescence gradually returned to baseline levels, attesting to the fluorescence as a marker of homeostatic sleep pressure. In 14 neurons (14%), fluorescence was highest in waking as compared to the other states during baseline, and in these neurons, fluorescence did not increase after sleep loss. Interestingly, six neurons (6%) were most active in waking and NREM but silent in REM (REM-off). This is the first study to measure fluorescence in individual neurons after sleep loss. We find that the fluorescence in two-thirds of ZI Lhx6 neurons is tightly linked to sleep, and that the

average fluorescence is further increased after prolonged waking. Microendoscopy is superior to indirect measures such as c-FOS or photometry in gauging sleep pressure in individual neurons.

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Poster

P638. Sleep: Systems

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Program #/Poster #: P638.10

Topic: F.08. Biological Rhythms and Sleep

Title: Medulla glutamatergic neurons control weak-sleep transition

Authors: *S. TENG[1], F. ZHEN[1], J. C. SCHALCHLI[2], X. CHEN[1], H. JIN[2], L. WANG[2], Y. PENG[1];
[1]IGM, [2]Zuckerman Inst., Columbia Univ., New York, NY

Abstract: The transition from wakefulness to sleep is an initiation process of sleep behavior. Understanding the underlying neural mechanisms is particularly relevant for the development of new treatments for insomnia and other sleep disorders. Yet, brain circuits controlling this process remain poorly understood. Here we identified a novel population of glutamatergic neurons in the medulla that project to the preoptic area (POA), a prominent sleep-promoting region. Chemogenetic silencing of POA-projecting medulla neurons disrupts the transitions from wakefulness to Non-Rapid Eye Movement (NREM) sleep, whereas chemogenetic activation of these neurons promotes NREM sleep. Moreover, we show that optogenetic activation of medulla glutamatergic neurons or their projections in the POA reliably initiates NREM sleep in awake mice. Together, our findings uncover an excitatory brainstem-hypothalamic circuit that controls the wake-sleep transitions.

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Poster

P652. Threat/Fear: Acquisition

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Program #/Poster #: P652.01

Topic: G.01. Fear and Aversive Learning and Memory

Support: R01-MH117791

Title: Diversity and temporal structure of rat behavior across Pavlovian fear discrimination

Authors: *A. CHU[1], C. B. MICHEL[2], N. T. GORDON[2], A. M. DUBOIS[2], D. C. WILLIAMS[2], M. A. MCDANNALD[2];
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Abstract: In Pavlovian fear conditioning, a neutral cue is paired with an aversive stimulus, such as foot shock delivery. One result of this predictive relationship is that cue presentation will elicit a suite of defensive behaviors. The most commonly studied defensive behavior is freezing, a 'passive' defensive behavior defined by the absence of movement. Recent studies have revealed that shock-associated cues can also elicit 'active' defensive behaviors, such as darting, characterized by increased movement. Thorough ethograms of rodent defensive behavior have been confined to contextual fear conditioning procedures and lack temporal specificity. The goal of the current study was to construct a complete ethogram of rat behavior across Pavlovian fear discrimination. To do this, we tested 24 rats (12 females and 12 males) in a conditioned suppression procedure. Briefly, rats were trained to nose poke for food rewards. Rats then received 16 fear discrimination sessions in which three auditory cues predicted unique foot shock probabilities: danger (p = 1.00), uncertainty (p = 0.25), and safety (p = 0.00). Poke-reward and cue-shock contingencies were independent. During each trial, we captured individual images at a rate of 5 frames/s before and during cue presentation (2 s pre-cue, 10 s during cue, 12 s total). Initial observations show that rats exhibit a mixture of freezing and active behaviors such as locomotion during danger and uncertainty cue presentation. We will present a full analysis of behaviors such as freezing, locomotion, grooming, nose poking, and eating across cue presentation and across the 16 fear discrimination sessions. Additionally, we will scrutinize our findings for sex differences and for quantitative and qualitative differences in behavior elicited by danger and uncertainty cues. Our hand-scoring efforts will not only reveal fine organization of behavioral responding, but form the basis of a data set to train neural networks for behavior classification. Successful application of neural networks to behavior classification will permit highly-detailed, large-scale analysis of behavior to more completely reveal neural circuits supporting diverse threat behavior.

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Poster

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Support: P50MH115874
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Title: The effect of sleep deprivation on fear extinction learning and memory

Authors: *A. R. FOILB[1,2], E. M. TAYLOR-YEREMEEVA[1], G. MISSIG[1,2], S. A. MADDOX[1,2], Y. DAI[3], K. J. RESSLER[1,2], W. A. CARLEZON, Jr.[1,2];
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Abstract: Sleep has a reciprocal relationship with fear- and trauma-related disorders. Sleep problems, including nightmares, difficulty falling or staying asleep, and restless unsatisfying sleep, are part of the diagnostic criteria for conditions such as Generalized Anxiety Disorder (GAD) and Post-Traumatic Stress Disorder (PTSD). Further, abnormalities in sleep patterns may contribute to, or exacerbate, the symptoms of these conditions and other co-morbid disorders. Existing research in rodent models demonstrate significant deficits in sleep as a result of fear learning, as well as improvements in sleep after extinction training. Prior studies on the inverse relationship, the impact of sleep on fear learning and extinction, have largely focused on disrupting the consolidation of memories immediately following learning. Here, we implanted male and female C57BL/6J mice with wireless telemetry transmitters (DSI) that allow for continuous collection of EEG, EMG, body temperature and locomotor activity. Vigilance states were tracked throughout fear conditioning, extinction training and recall testing. We used gentle handling to produce sleep deprivation, keeping mice awake for six hours prior to extinction training. Surprisingly, sleep deprived mice displayed enhanced extinction learning with reduced freezing to cue presentations during extinction and recall testing. Analysis of sleep data showed that sleep deprivation led to rebound slow wave sleep (SWS) and rapid eye movement (REM) sleep after extinction training, and that the retention of extinction memory correlated with the duration of rebound SWS in sleep deprived animals. In all animals, fear conditioning resulted in increased SWS and REM sleep during the dark phase, when mice are typically awake and active. Ongoing analyses will further investigate hourly changes in activity and sleep, as well as explore sex differences and a role of estrous cycle. Sleep is an ideal endpoint for preclinical neuroscience work, as it can be defined and assessed the same in the same ways in humans and animal models. Understanding the impact of stress and trauma on sleep will enable better alignment of neuroscience research and clinical studies and better treatments for a wide variety of psychiatric conditions.

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Poster

P652. Threat/Fear: Acquisition

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P652.03

Topic: G.01. Fear and Aversive Learning and Memory

Support: DANA Foundation
R37 MH065635
T32-MH019524 27
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T32AG052909

Title: Parvalbumin cells in the hippocampus are required for infantile memory formation in rats

Authors: *J. MIRANDA[1], E. CRUZ[1], B. BESSIÈRES[3], C. M. ALBERINI[2];
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Abstract: Episodic memories formed during infancy are rapidly forgotten, a phenomenon associated to infantile amnesia, the inability of adults to recall infantile memories. We previously showed that contextual experiences in infant rats are stored as latent memories and can be reinstated by reminders given later in life. We also found that infantile memory formation requires the dorsal hippocampus (dHPC) and biological mechanisms typically used in developmental critical periods. The development of parvalbumin interneurons (PVIs) is an important mechanism of critical periods in sensory functions. Furthermore, episodic memory formation in adult mice involve PVIs in the dHPC. Whether PVIs in the dHPC play a role in the critical period of memory development remains unknown. We used Inhibitory Avoidance (IA) to model contextual episodic learning in infant rats at postnatal day 17 (PN17), and biochemical, immunohistochemical, pharmacological and genetic approaches to determine the regulation and functional role of PVIs in the dHPC during infantile memory formation. We found that training significantly increased the levels of parvalbumin 48 hours after training. Chemogenetic inhibition of PV+ cells in the dHPC disrupted infantile long-term memory. While a bilateral dHPC injection of the positive GABAAR modulator diazepam after training led to long-term memory expression, chemogenetic activation of PV+ cells in the dHPC did not prevent infantile forgetting. We conclude that infantile episodic learning regulates the expression of PV protein in the dHPC where the function of PVIs is necessary for the storage of latent infantile memory. Furthermore, although like for sensory functions, diazepam can accelerate functional maturation, i.e., memory expression, the activation of PVIs is not sufficient to accelerate this developmental maturation.

Disclosures: J. Miranda: None. **E. Cruz:** None. **B. Bessières:** None. **C.M. Alberini:** None.

Poster

P652. Threat/Fear: Acquisition

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P652.04

Topic: G.01. Fear and Aversive Learning and Memory

Support: MOST 110-2326-B-007-MY3

Title: Aberrant Orbitofrontal Cortical Activation Interfered the Encoding of Pavlovian Fear Conditioning

Authors: *C.-F. SUN, C.-H. CHANG;
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Abstract: The orbitofrontal cortex (OFC) can be subdivided into the lateral OFC (lOFC) and the medial OFC (mOFC). Previous studies have suggested that both subregions are involved in the modulation of negative emotions. However, the aberrant activities of the OFC on encoding the Pavlovian fear conditioning remain unknown. In Pavlovian fear conditioning, a neutral conditioned stimulus (CS), such as a tone, is associated with an aversive unconditioned stimulus (US), such as a footshock. After conditioning, the animals would demonstrate contextual fear to the conditioning chambers and cued fear to the tones. In this study, the mOFC or the lOFC was pharmacologically activated before fear conditioning on Day 1, followed by context test on Day 2 and tone test on Day 3 in Long-Evans rats. We found that animals under aberrant activation of either the mOFC or the lOFC showed normal within-session fear expression during conditioning. However, acquisition of contextual fear was impaired under mOFC activation, while the acquisition of cued fear was impaired under either the mOFC or the lOFC activation, that these animals showed lower freezing compared to controls during retrieval test. Together, our data revealed the importance of proper OFC activities in Pavlovian fear conditioning.

Disclosures: C. Sun: None. **C. Chang:** None.

Poster

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Program #/Poster #: P652.05

Topic: G.01. Fear and Aversive Learning and Memory

Support: MOST 110-2326-B-007-001-MY3

Title: Unilateral or Bilateral Activation of Lateral Orbitofrontal Cortex on Acquisition of Fear Extinction

Authors: *C.-C. SHIH[1], Y.-H. CHANG[1], R.-J. CHIOU[2], C.-H. CHANG[1];
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Abstract: Dysfunction of fear expression and extinction is the reason of many anxiety-related disorders. By repeatedly presenting an initially neutral conditioned stimulus (CS; a tone) with an aversive unconditioned stimulus (US; a footshock), animals learn the association between the CS and US in Pavlovian fear conditioning. Afterward, repeated presentation of the CSs alone results in decrement of the conditioned fear response to CS, a process called fear extinction. Researches have indicated that both the amygdala and the medial prefrontal cortex (mPFC) play critical roles in the fear extinction. In addition, aberrant activities of the lateral orbitofrontal cortex (lOFC) have been observed in patients with anxiety-related disorders. Given the role of the lOFC in modulating fear expression and extinction, we hypothesized that activation of the lOFC would change the balance of neuronal activities in the amygdala and the mPFC, which causes a learning deficit in fear extinction. First, we unilaterally or bilaterally activated the lOFC with N-methyl-D-aspartate receptor (NMDA) immediately before the fear extinction procedure. We found that left and bilateral lOFC activation would lead to an initially low then gradually increased freezing behavior in the extinction learning process, whereas activation of the right lOFC did not. In addition, all experimental groups showed impaired acquisition of fear extinction during retrieval test. Next, we quantified the neuronal activities using c-Fos as an index in related brain regions during extinction acquisition. Our data suggested that pharmacological activation of the lOFC caused higher levels of c-Fos expression in non-GABAergic neurons in the ipsilateral lateral/basolateral amygdala and the ipsilateral mPFC in experimental groups. Meanwhile, higher activation levels of GABAergic neurons in the ipsilateral lateral amygdala and the ipsilateral medial intercalated cells were also observed in experimental groups. There was no significant difference in c-Fos expression in the central amygdala between experimental groups and control groups. Together, our results indicated that pre-extinction activation of lOFC changed the balance in neuronal activity in numerous extinction-related brain regions, which may have impacts on the expression of conditioned fear and may lead to the impairment of fear extinction acquisition.

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Poster

P652. Threat/Fear: Acquisition

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Topic: G.01. Fear and Aversive Learning and Memory

Support: NCSU CVM Intramural Research Award

NIH Grant MH123768
BBRF NARSAD Young Investigator Award

Title: Sex-specific lateral septum circuits for threat memory acquisition

Authors: *K. ADCOCK BINION, A. N. YANKOGLU, S. BASU, E. K. LUCAS;
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Abstract: Dysregulation of emotional learning and memory is a core feature of anxiety disorders. Women disproportionately experience anxiety disorders, leading us to hypothesize that emotional memory circuits may differ between the sexes. Using cued threat conditioning as a model for emotional memory, we found robust lateral septum (LS) activation after training in females but not males. Since the LS does not receive input from primary sensory regions conveying the conditioned and unconditioned stimuli, we next questioned which upstream regions recruit the female LS during threat memory acquisition. We bilaterally infused the retrograde tracer cholera toxin subunit B (CTb) into the LS of male and female animals one week prior to cued threat conditioning. In ongoing analysis, we are quantifying regional expression of CTb+ as well as CTb/FOS+ neurons throughout the brain. Thusfar, we have not observed any sex differences in input to the LS. We found that the ventral hippocampus (vHC) is the primary source of input to the LS, but vHC→LS projections are robustly activated during training in both sexes. However, we found that LS-projecting neurons in the lateral orbital cortex are activated by training in males but not in females, whereas LS-projecting neurons in the infralimbic cortex are activated by training in females but not in males. These data suggest that hardwiring of LS circuitry does not differ between the sexes, but sex-specific activation of upstream regions may drive sex-specific LS activation during threat memory acquisition. Future experiments will determine the causal relationship of LS circuitry on threat memory formation.

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Poster

P652. Threat/Fear: Acquisition

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Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH Grant MH123768-01
Brain & Behavior Research Foundation Young Investigator Award
NCSU CVM Intramural Research Award

Title: The lateral septum drives sex-specific emotional memory encoding

Authors: *E. K. LUCAS[1], A. M. JEGARL[2], S. BASU[3];
[1]North Carolina State Univ., Raleigh, NC; [2]Yale Sch. of Med., New Haven, CT; [3]Cleveland Clin., Cleveland, OH

Abstract: The emotion of fear is a powerful trigger for memory storage, helping us avoid cues associated with dangerous outcomes. However, fear memories become maladaptive in common psychiatric diseases such as post-traumatic stress disorder, generalized anxiety disorder, panic disorder, and phobia. Women are twice as likely as men to experience these fear-based mental illnesses, suggesting that the neurobiological mechanisms underlying fear memory encoding may differ between the sexes. To explore this possibility, we quantified expression of the neural activity marker c-fos in select forebrain regions of mice subjected to auditory threat conditioning compared to naïve controls. While both sexes equally engaged canonical regions of cued threat memory circuit, such as basolateral and central amygdala, we found a robust female-specific increase of c-fos expression in the lateral septum (LS). We next employed chemogenetics for bidirectional modulation of LS activity during threat memory encoding and observed sex-specific effects on memory formation. In females, LS activation further enhanced threat memory encoding, whereas LS inhibition promoted threat memories that were more susceptible to degradation. In males, on the other hand, LS activation impaired, while LS inhibition enhanced, threat memory formation. To determine the downstream LS circuitry promoting threat memory, we used an activity-dependent tagging approach to express eYFP in neurons of the LS memory ensemble. Consistent with our c-fos data, significantly more tagged neurons were observed in trained females than trained males or naïve controls of either sex. Axons of LS memory ensemble neurons projected to canonical mediators of behavioral and physiological defensive responses, including the periaqueductal gray, lateral hypothalamus, and ventral hippocampus. Finally, we questioned whether LS neurons comprise a cued threat memory ensemble, and we found that LS neurons tagged during training were re-activated during cued memory recall. These data pinpoint the LS as a critical locus of sex-specific threat memory encoding that may promote women's susceptibility and men's resilience to fear-based psychiatric disease.

Disclosures: E.K. Lucas: None. **A.M. Jegarl:** None. **S. Basu:** None.

Poster

P652. Threat/Fear: Acquisition

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Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH Grant R01MH123768
NIH Grant T34GM131947
Brain & Behavior Research Foundation Young Investigator Grant

Title: Sex-specific neural networks of threat memory acquisition: a pilot study

Authors: *K. DU PLESSIS, E. K. LUCAS;
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Abstract: Women are twice as likely as men to be diagnosed with posttraumatic stress and anxiety disorders, and greater symptom severity leads to worse clinical outcomes in women than in men. Dysregulation of emotional memory is a core feature of these disorders, suggesting that emotional memory processing may differ between the sexes. While decades of elegant studies have established circuits for the acquisition and retrieval of cued threat memory,the most common preclinical model of emotional memory, most of these studies excluded female subjects. Thus, it is currently unknown whether threat conditioning engages similar circuits to the same extent in females versus males. To investigate sex-specific circuits for emotional memory, we conducted whole-brain quantification of c-fos in adult male and female mice after cued threat conditioning compared to naive controls (n=3/group). Two-way ANOVA revealed that trained females exhibited higher activation of the central amygdala, cortical amygdala, and primary somatosensory cortex than trained males. Interestingly, we also observed greater activation of the anterior hypothalamic nucleus, midbrain, and superior colliculus in females relative to males independent of threat conditioning. To further analyze these basal and learning-dependent sex differences in regional activity, we generated cross-correlation matrices for all groups. We observed patterns of highly synchronized interregional activity in both naive and trained males that were not observed in females. In trained males, co-activation patterns were particularly pronounced between thalamic subregions. In contrast, thalamic activity in trained females was consistently uncoordinated with activity throughout the rest of the brain. Overall, this pilot study provides preliminary evidence of sex differences in the individual circuit components and broader neural networks involved in threat memory acquisition. Subsequent studies will elaborate on these findings through estrous cycle monitoring to account for estrogen- and progesterone-mediated effects on threat circuitry. In addition, future analyses will identify hub regions that exert disproportionate influence over network activity. By expanding our understanding of sex-specificity in threat memory networks, we aim to identify potential targets for clinical interventions that are better equipped to accommodate sex differences in memory systems.

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Poster

P652. Threat/Fear: Acquisition

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Topic: G.01. Fear and Aversive Learning and Memory

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Title: Evidence for a prefrontal - bed nuclei of the stria terminalis circuit that constrains contextual fear generalization.

Authors: *R. T. LINGG[1], S. B. JOHNSON[1], T. D. SKOG[2], D. C. HINZ[1], M. LIZARAZU[1], S. A. ROMIG-MARTIN[1], S. ELIASEN[1], N. S. NARAYANAN[4], J. J. RADLEY[3]; [2]Psychological and Brain Sci., [3]Dept. of Psychological and Brain Sci., [1]Univ. of Iowa, Iowa City, IA; [4]Neurol., Univ. of Iowa Roy J and Lucille A Carver Col. of Med., Iowa City, IA

Abstract: Generalization of fear is an adaptive neurobiological process conserved across species. Nevertheless, dysregulation of the neural systems that mediate fear generalization has been implicated in the development of a variety of stress-related psychiatric illnesses, including PTSD. It is well documented that glucocorticoid stress hormones modulate memory consolidation and contribute to fear generalization. However, little is known about the stress-modulatory systems that regulate endogenous glucocorticoid activity and how these are engaged to mediate fear generalization. Our prior work combining optogenetic, behavioral, and neuroendocrine approaches highlight the anteroventral bed nuclei of the stria terminalis (avBST) as an important interface between HPA-related information processing and prefrontal cortical regions tasked with the regulation of emotional memory. Here we used intersectional viral approaches to examine the role of a rostral prelimbic cortex (PL)-avBST pathway in regulating contextual fear generalization. We applied optogenetic methods for manipulating neural activity immediately following training on a modified single-trial inhibitory avoidance procedure in rats, as this post-training period is critical for mediating the emergence of long-term fear generalization. First, post-training inactivation of the rostral PL-avBST pathway with *halorhodopsin 3.0* (eNpHR3.0) for 10 min exaggerated corticosterone output provoked by the footshock given in the training context as compared to the control group. Forty-eight hours later, retention latencies were tested in the training and a novel context. Whereas both eNpHR3.0 and control groups showed high retention latencies in the training context, only the eNpHR3.0 group showed a high degree of generalization, as indicated by a similar retention level in the novel as training context. Next, we pre-exposed rats to a neutral context prior to footshock training to bias them toward generalization and activated the rostral PL-avBST pathway with *channelrhodopsin 2* (ChR2) for 10 min immediately after training. When tested 48 h later, only controls showed high retention levels for both contexts, indicating generalization, whereas the ChR2 group showed comparable retention for shock but not neutral context. Interestingly, corticosterone increases in the post-training period were not affected by pathway activation with ChR2. Together, these data provide evidence for a novel circuit regulating consolidation processes that limit stress hormonal activation in the aftermath of a traumatic event and fear generalization thereafter.

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Poster

P652. Threat/Fear: Acquisition

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NIH Grant DK100685 to LR

Title: Gut-brain signaling about metabolic state alters passive avoidance behavior in male rats: Potential contribution of A2 noradrenergic neurons

Authors: *C. M. EDWARDS, T. DOLEZEL, L. M. RINAMAN;
Dept. of Psychology, Florida State Univ., Tallahassee, FL

Abstract: Competing motivational drives coordinate behaviors essential for survival. For example, negative energy balance suppresses innate anxiety-like behaviors in rats. In this set of experiments, we explored the impact of metabolic state on learned avoidance behavior, and whether the "hunger hormone" ghrelin plays a role. Male and female rats were trained in a passive avoidance task where they learn to avoid a context paired with a single mild footshock. When rats later were tested for avoidance of the same context, male rats tested in an overnight food-deprived state and female rats (regardless of feeding status) displayed significantly less avoidance compared to male rats that were fed *ad libitum* prior to testing. However, when the fasting-induced increase in ghrelin signaling was pharmacologically antagonized before the test, fasted male rats displayed more passive avoidance compared to fasted, saline-injected controls. Based on prior evidence that prolactin-releasing peptide (PrRP)+ A2 neurons and glucagon-like peptide 1 (GLP1)+ neurons within the caudal nucleus of the solitary tract (cNTS) are "silenced" by overnight fasting, we examined whether these neurons were activated in rats re-exposed to the shock-paired context in fed vs. fasted conditions. Compared to controls, context re-exposure in fed rats activated PrRP+ neurons, but not GLP1+ neurons, and also activated neurons within the anterior ventrolateral bed nucleus of the stria terminalis (vlBNST), which receives dense input from PrRP+ terminals. Food deprivation prior to context re-exposure reduced activation of PrRP+ A2 neurons in both sexes, and this effect was partially reversed by ghrelin receptor blockade. Our results support the view that PrRP+ neurons innervating the vlBNST contribute to expression of passive avoidance in a manner that is modulated by metabolic state, including a potential role for ghrelin signaling, in male rats.

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Poster

P860. Timing and Temporal Processing: Learning and Memory

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Nancy J. Newman, MD '78 & Valerie Biousse, MD Research Fund for Neuroscience

Title: Over- and under-segmentation of latent causes lead to different types of extinction failures

Authors: *I. AITSAHALIA[1], S. PISUPATI[1], A. LANGDON[1,2], Y. NIV[1,2];
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Abstract: Recent studies have proposed that individual differences in the failure to extinguish Pavlovian conditioned fear may emerge from differences in the inferred causal structure of the environment. Previous research has distinguished between individuals who attribute fear acquisition and extinction trials to a single cause and those who attribute these to different causes. Healthy participants in the latter group showed higher resurgence of fear than the former (Gershman and Hartley, 2015) while participants with PTSD were more likely to fall into the former category (Norbury et al. 2021). However, this delineation does not capture the full range of possible regimes that could contribute to differences in learning and fear extinction, especially in extinction tasks with both safe (CS-) and dangerous (CS+) stimuli. Here, we propose that inferred causal structures lie on a spectrum between too few and too many latent cause assignments, corresponding to "discrimination" failures (failures in distinguishing between safe and dangerous stimuli) and "over-accommodation" failures (failures in updating beliefs about stimuli that are no longer dangerous) respectively, with "good" extinction falling in between. Using simulations, we show that these extremes can be reached through changes in a parameter that controls beliefs about randomness in the world, captured in our model as a prior over observation probabilities. Starting out with a "deterministic" prior, i.e. a belief that probabilities are often all-or-nothing, leads to surprising observations being assigned to new causes. Creating too many latent causes can give the impression of rapid learning, but does not extinguish the original association, failing to generalize between acquisition and extinction trials. Such over-accommodating individuals could be more susceptible to spontaneous recovery of fear. On the other hand, a "stochastic" prior, i.e. a belief that probabilities are often close to chance, leads to attribution of disparate observations to a single cause and overgeneralization. The creation of too few latent causes leads to poor discrimination between stimuli and therefore poor extinction performance through the generalization of fear to safe stimuli. Our proposed delineation of latent cause regimes clarifies the mapping between erroneous inference of causal structure and different types of extinction failures. Moreover, we relate these regimes to a single parameter, the observation prior, that can reflect cognitive distortions or an individual's past experiences.

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Poster

P860. Timing and Temporal Processing: Learning and Memory

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Program #/Poster #: P860.02

Topic: H.08. Learning and Memory

Support: NIMH grant R01DA042065

Title: Learning a state-space map

Authors: *O. BEIN[1], N. W. SCHUCK[2], R. RAGHU[1], Y. NIV[1], A. J. LANGDON[1];
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Abstract: Goals drive how we represent the environment. If trying to cross the street, we might represent and base decisions on the speed of different cars, while when evaluating which car is more aesthetically pleasing, color might be more important. In the computational framework of reinforcement learning, the information relevant to a given decision is referred to as 'state', and a task can be described as a map of such states. This map is composed of a set of states, with transitions between them, and rewards and costs associated with some states and not others. Much research has been devoted to understanding how values are learned based on a given state representation. However, only recently researchers started to focus on understanding how the state representation itself is learned. Recent research has implicated the orbitofrontal cortex (OFC) in representing well-learned states (Schuck, Cai, Wilson & Niv, 2016), especially when relevant state information is partially unobservable (i.e., has to be recalled from memory rather than being perceptually available). Here, we used the same task to investigate the process of initially constructing the representation in the OFC: participants (N=27) were scanned using fMRI while they read the task instructions, practiced the task, and then performed it for hundreds of trials. The arbitrary nature of the task ensured that state representations had to be constructed de novo. In particular, participants observed stimuli composed of a superimposed house and a face, and made young/old judgements about either the face or the house. Knowing which category to judge, namely, what is the current 'state', required information from the previous trial, and therefore this aspect of the state was unobserved. Behaviorally, error rates and reaction times decreased with learning, indicating that participants learned and honed an effective state representation. Neural analyses include univariate activation, functional connectivity, and multivariate pattern analysis (MVPA classification and representational similarity analysis, RSA). Our analyses are focused on an OFC region of interest, as well as on the hippocampus and sensory brain regions, with the goal of understanding the circuit involved in learning a map of states to guide decisions.

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Poster

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Topic: H.08. Learning and Memory

Support: NIH Grant R01DA050647

Title: Changes in reward identity disrupt the reset of reward predictions and reveal model-based influences on reward prediction errors in dopamine neurons

Authors: S. K. HAROOTONIAN[1], Y. K. TAKAHASHI[2], T. A. STALNAKER[2], L. E. MUELLER[2], G. SCHOENBAUM[2], Y. NIV[1], A. LANGDON[1];
[1]Princeton Univ., Princeton, NJ; [2]Natl. Inst. on Drug Abuse Intramural Res. Program, Baltimore, MD

Abstract: The phasic activity of midbrain dopamine neurons is thought to encode the model-free prediction error signal posited by the temporal-difference (TD) reinforcement learning algorithm. In this interpretation, dopaminergic prediction errors are sensitive only to differences in the value of outcomes, irrespective of the form in which these outcomes are delivered. Recent findings, however, have challenged this idea by showing that unexpected reward identity—for example, a receiving a chocolate-milk reward rather than an equally valued vanilla-flavored reward—can elicit phasic increases in dopamine activity (Takahashi et al., Neuron, 2017). These results demonstrate that changes in reward identity alone are sufficient to produce significant prediction-error responses in dopamine neurons. Here, we present a model-based multi-prediction TD learning algorithm (Langdon et al. Curr. Opin. Neurobiol. 2018) to account for reward-identity-dependent responses of midbrain dopamine neurons in a task in which the identity and timing of rewards are systematically manipulated (Takahashi et al., SfN 2019). In particular, we focus on accounting for both the responses to changes in reward identity and the "reset" of reward predictions when a reward is obtained earlier than predicted (i.e., the fact that dopamine neurons do not show an omission signal at the original time of reward). Importantly, the data show that omission responses are restored when the identity of the now-early reward is also changed. In our model, we account for this identity-specific reset of reward predictions through model-based inference, in which only the specific prediction that is no longer relevant after the delivery of an early reward is reset, a mechanism that relies on the congruence between the expected and actual identity of the outcome. Our results suggest that dopamine neurons have access to complex, multi-dimensional reward expectations, in which multiple identity-specific predictions about reward outcomes can be sustained in parallel. Overall, our results provide compelling evidence

that prediction error responses in dopamine neurons do not correspond to simple model-free errors in scalar value expectations, but correspond instead to errors in model-based predictions about the specific reward outcomes expected in a task.

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Poster

P860. Timing and Temporal Processing: Learning and Memory

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Title: A unified framework for reversal learning based on latent cause inference

Authors: *S. PISUPATI[1], J. R. TAYLOR[3], Y. NIV[1,2], S. M. GROMAN[3,4];
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Abstract: Reversal-learning tasks have been extensively used to quantify cognitive flexibility across species by requiring subjects to adapt choice behavior to changing contingencies. Although reversal-learning tasks are traditionally thought to measure response inhibition, recent work suggests that this task may measure multiple interacting processes such as reward-based learning, change-point inference and rule/task-space learning (Izquierdo et al.'17, Groman et al.'19). Here, we present a model of reversal learning that unifies these processes based on the normative framework of latent-cause inference, in which individuals infer hidden or "latent" causes that dictate the relationship between cues, actions and outcomes in the task. Reversal learning in this model involves learning the structure of these causes, online inference about their change-point dynamics, and learning associations and response policies contingent on these causes.Our model accounts for several empirically observed features of reversal learning, such as abrupt rather than gradual changes in response policy, learning of new associations rather than unlearning of old ones, and a dissociation between abandoning previously rewarding actions and

learning to exploit newly rewarding ones. In addition, it predicts the existence of "interference" events where ambiguous evidence leads an agent to infer that an old cause has returned, and incorrectly perform the old response policy long after a new one has been learnt - a feature whose occurrence we confirm by fitting the model to a dataset of rodents performing a probabilistic reversal learning task (Groman et al. '19). Further, we use our model to understand the influence of prior experiences on reversal learning, and to interpret the effects of viral ablations of select projections to and from the orbitofrontal cortex, an area previously associated with latent cause inference. Our model offers a unified probabilistic view of multiple facets of reversal learning, and is of particular relevance to the application of reversal paradigms to psychiatric conditions involving the resurfacing of old behaviors after prolonged periods of time, such as relapse in addiction.

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Poster

P860. Timing and Temporal Processing: Learning and Memory

Location: Virtual Only

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Program #/Poster #: P860.05

Topic: H.08. Learning and Memory

Support: La Caixa ID 100010434. LCF/BQ/DI19/11730040

Title: Discovery of hierarchical representations in goal-directed behaviour

Authors: *M. SILVA, L. FUENTEMILLA, J. MARCO;
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Abstract: Goal-directed behavior relies on the ability to learn the relational structure of a complex environment. Prior research showed that humans can quickly acquire a map-like representation of multiple events that are associated by a uniform transition probability. Here, we examined how the acquisition of such topology structure can lead to the discovery of hierarchical representations hidden in a relational structure. We recorded scalp electroencephalography (EEG) while participants were asked to find out optimal paths that linked sequence of fractal pictures to a reward. Critically, picture sequences and paths coalesced around clusters or ‘communities’ of mutually predicting stimuli, thereby eliciting the emergence of hierarchical event memory representations for picture images, those that linked within and those that linked between (i.e., boundaries) communities. By implementing model-based analysis on behavioral and on EEG data, we demonstrate that the acquisition of memory representations containing such community structure precedes the discovery of the hierarchical topology hidden in the same structure. These findings provide important insights of the organizational principles that allow optimal goal-directed behavior via relational learning in humans.

Disclosures: M. Silva: None. **L. Fuentemilla:** None. **J. Marco:** None.

Poster

P860. Timing and Temporal Processing: Learning and Memory

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Program #/Poster #: P860.06

Topic: H.08. Learning and Memory

Support: R01 MH113071
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Title: Branch-specific dendritic plasticity in the retrosplenial cortex allows linking of memories across time

Authors: *M. SEHGAL[1], D. AMEIDA-FILHO[1], S. MARTIN[1], I. D. MEJIA[1], A. LAVI[2], A. PEKCAN[1], S. KIM[3], J. LEE[4], S. HUANG[5], W. HEO[4], J. T. TRACHTENBERG[6], A. J. SILVA[7]; [1]Univ. of California Los Angeles, Los Angeles, CA; [2]Dept. of Neurobiology, Psychiatry, Psychology and the Brain Res. Instit, Univ. of California, Los Angeles (UCLA), Los Angeles, CA; [3]IBS/KAIST, DAEJEON, Korea, Democratic People's Republic of; [4]IBS/KAIST, Daejeon, Korea, Democratic People's Republic of; [5]Neurobio., [6]UCLA, Los Angeles, CA; [7]Dept Neurobiol, UCLA Med. Ctr., Los Angeles, CA

Abstract: Memory formation is a dynamic process where single memories are stored, updated and integrated within the framework of other pre-existing memories to drive adaptive behavior. Previously our laboratory and others have shown that when two memories are acquired close in time, the recall of one leads to the recall of the other, a phenomenon referred to as memory linking. Memory linking is mediated by an overlap between the neuronal ensembles encoding these memories. However, little is known regarding the dendritic plasticity mechanisms that regulate memory linking and other memory integration phenomena. Here, we investigate the integration of two distinct contextual memories to study the role of somatic as well as dendritic plasticity within the retrosplenial cortex (RSC). We demonstrate that overlapping RSC neuronal ensembles are recruited to encode contextual memories acquired close in time (neuronal co-allocation). Moreover, individual neuronal firing as well as patterns of coactivity in RSC are more stable across context explorations within the same day vs 7days apart (conditions that drive contextual memory linking). We also show that this overlap in RSC neuronal ensembles is necessary and sufficient for linking contextual memories. Using structural as well as functional 2 photon imaging, we next investigated whether the exploration of a novel context causes branch-specific dendritic plasticity within RSC neurons such that the future dendritic plasticity is biased to the same dendritic segments (dendritic co-allocation). Longitudinal calcium imaging of RSC dendrites demonstrated that the same dendritic branches are activated when two contexts were

visited same day vs 7 days apart. We also found that structural plasticity in the form of spine addition and clustering was biased to the same dendritic branches under these conditions. These results demonstrate that contextual memory linking drives dendritic activation as well as plasticity within the same RSC dendritic branches. Finally, to further test the role of RSC dendritic co-allocation in contextual memory linking, we developed an optogenetic tool that allows the tagging as well as manipulation of previously activated dendrites. Using this system, we demonstrate that branch-specific reactivation of RSC dendrites alone was sufficient to link two contextual memories. These data demonstrate a key role for RSC dendritic co-allocation in contextual memory linking. We show that contextual learning results in branch-specific dendritic plasticity in RSC that biases future memory allocation to the same dendritic segments, and that this dendritic co-allocation is critical for memory linking.

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Poster

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Program #/Poster #: P860.07

Topic: H.08. Learning and Memory

Support: University of San Diego
Mount Holyoke College

Title: Medial entorhinal cortex lesions produce delay-dependent disruptions in memory for elapsed time in rats

Authors: A. VO[1], N. TABRIZI[1], T. HUNT[1], K. CAYANAN[1], S. CHITALE[2], L. ANDERSON[2], S. TENNEY[2], A. WHITE[2], M. SABARIEGO[2], *J. HALES[1];
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Abstract: Our awareness and experience of elapsed time is important for remembering and organizing events in a temporal context as well as for planning future actions. Several studies have supported the role of the hippocampus in temporal organization of episodic memory with the finding of hippocampal time cells that fire sequentially throughout a temporal gap. This sequential firing has recently been identified in the medial entorhinal cortex (MEC), an area that directly projects to the hippocampus, which suggests that the MEC may provide temporal input to the hippocampus that is integral for episodic memory formation and maintenance. The behavioral relevance of such firing remains unclear, however, as previous research has rarely used time as a meaningful variable or utilized tasks that depend on the MEC. In our study, we

investigated whether the MEC is necessary for rats to perform a time duration discrimination task (TDD), in which they were taught to distinguish between 10-s and 20-s delays. During every trial in the figure 8 maze, rats were held in a delay box at the end of the central arm for 10 or 20 seconds. At the end of the delay, rats were allowed to turn left or right, but they only were rewarded for making a left turn after a 10-s delay or a right turn after a 20-s delay. After reaching a 90% performance criterion in two out of three consecutive test days, rats were randomly assigned to receive an excitotoxic MEC-lesion or sham-lesion surgery. After a two-week recovery period, we found that MEC-lesioned rats were significantly impaired on the TDD task in comparison to sham-lesioned rats and did not return to performance criterion. This impairment was specific to the longer 20-s delays, suggesting an important role for the MEC in memory for elapsed time longer than 10 seconds possibly due to an increased reliance on long-term memory resources for processing prolonged time intervals.

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Poster

P860. Timing and Temporal Processing: Learning and Memory

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Title: Characterizing hippocampal CA1 network activity in the context of trace conditioning at multiple inter-stimulus intervals

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Abstract: The hippocampus plays a crucial role in associating discontiguous events and in the consolidation and encoding of memory. Recent research has shown that the hippocampus is functionally relevant for the creation and maintenance of different types of sequences. Of particular interest is the idea that some clusters of cells take up specific encoding tasks, termed ‘place cells’ and ‘time cells’. In previous work using trace eyeblink conditioning (TEC) protocol, where two events occur one after the other and are separated by an inter-stimulus interval (ISI) or

trace interval, we observed the emergence of time tuning in cells of the hippocampal CA1 pyramidal layer following learning of the trace association (Modi et al., eLife 2014). Time-cell remapping has been observed for learning tasks involving delay periods (W. Mau et al., Curr. Biol. 2018) but these take place over tens of seconds and do not address multiple simultaneous representations. Here, we seek to understand the time-cell formation, remapping, multiple representations, and extinction in the context of a series of trace timing tasks with multiple individuals and interleaved durations. Using TEC as the behavioral context, we investigate how the dorsal hippocampal CA1 network activity changes to encode new time scales of trace for a pair of a conditioned stimulus (CS: light) and an unconditioned stimulus (US:air-puff). We used head-fixed GCaMP6f mice (n=6 imaging, n=6 behavior) to train them on varying trace intervals (250 ms, 350 ms, 450 ms, and 550 ms). Each animal was first trained to criterion on the 250 ms trace over a few sessions on consecutive days. The trace duration was successively incremented once the animal reached the criterion. After learning the 550 ms trace, the animals went through 3-4 sessions of interleaved trials consisting of 6 blocks of 5 trials alternating between the 250 ms and 550 ms traces, for a total of 60 trials. Finally, the animals went through CS-only extinction sessions. We used an implanted cranial window to carry out in vivo 2-photon calcium imaging on head-fixed mice using a custom-built galvo-scan microscope. We recorded ~100 cells for each animal across ~25 sessions spanning the behavioral training, and also monitored locomotion using a rotary encoder on a treadmill. The behavior data suggests that the animals take longer to associate the first interval but are comparatively quicker to associate subsequently expanded trace intervals. We present our findings on the emergence and stability of time-cell sequences and how the sequential activity changes with the expansion of the trace intervals.

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Poster

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Program #/Poster #: P860.09

Topic: H.08. Learning and Memory

Support: HHMI

Title: Learning to anticipate: hazard rate updating in mice and monkeys

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Abstract: Anticipation of events allows us to infer cause/effect, test predictions, express and understand language and music. Its formal realization is the hazard rate, h(t): the probability of an event at t, given it has yet to occur. Monkeys and humans are known to produce faster reaction times (RT) to stochastic cues when h(t) is larger, and a neural representation of h(t) is observed in macaque parietal cortex (Janssen & Shadlen 2005). The mechanisms responsible for establishing these dynamical neural responses are unknown. We reason they might be tractable in the mouse. We therefore sought to determine whether mice exhibit anticipatory behavior consistent with hazard rate timing.

Mice were trained to wait in a central port for a "go" cue before moving to a second port to collect a reward. Initially, the wait time, τ, was uniformly distributed ($0.5 < \tau < 2.5$ s). After learning this unimodal distribution, $U(\tau)$, the mice were switched to a bimodal distribution, $B(\tau)$: $p=0.45$ that τ is from the 1st mode (0.5-0.75 s), $p=0.45$ that τ is from the 2nd mode (2.25-2.5 s), and $p=0.1$ that it is distributed uniformly between them. The hazard rate associated with $U(\tau)$ increases monotonically with waiting time; for $B(\tau)$ it is triphasic, rising for each mode and falling in between. One rhesus macaque was trained on a saccade task using $B(\tau)$ to facilitate comparison with past results.

We measured the RT of the mice after exposure to these schedules. For $U(\tau)$, RT decreased monotonically as a function of wait time. For $B(\tau)$, RT showed a triphasic pattern. These patterns are broadly consistent with anticipation mirroring the hazard rate. When $B(\tau)$ was introduced, RT decreased for trials from the first mode within the first session (rank sum $p<0.0001$), while RT in the second mode did not. The monkey, similarly, responded most quickly to trials from the first mode early in training and to trials from the second mode when fully trained. The effect of wait time on learning could also be seen in the slowing of RT in the sparsely sampled intermode epoch. RT slowed first for τ at the beginning of this epoch. Even late in training, mice exhibited changes in RT that were influenced by the wait time in the preceding trial. Thus they continue to adjust their behavior to potential changes in temporal milieu. For example, following a trial with τ drawn from the intermode epoch, mice responded faster than expected on the next trial.

These observations provide preliminary support for hazard rate timing by mice. They begin to constrain the class of models that can explain how temporal regularities are learned.

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Poster

P880. Learning and Value Processing in Disorders

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Title: The 'Expected Value' (not the surprising outcomes) of risky choices drives subjective pleasure of risky decision making in patients with Impulse Control Disorder

Authors: *B. LIEBENOW[1], A. JIANG[1], E. DIMARCO[1], L. P. SANDS[1], M. MOYA-MENDEZ[3], M. S. SIDDIQUI[2], I. HAQ[4], K. T. KISHIDA[1];
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Abstract: Dopaminergic signals have been implicated in heightened impulsive motivation and positive feelings initially associated with addictive stimuli. Impulse control disorder (ICD) is a behavioral addiction consisting of the sudden onset of increased risky decisions induced by dopaminergic therapies (DT) used to treat Parkinson's disease (PD). The connection between dopamine, ICD, and the experience of risky decision making is not understood. We hypothesized that patients with a history of ICD have increased motivation to take risks and that heightened dopamine-related signals increase the pleasure associated with those decision outcomes. We investigated the experience of risky decision making in patients with PD and a history of ICD (ICD, n = 18) compared to patients with PD and no history of ICD (non-ICD; n = 11), on and off their DT. All participants performed a 'Sure-bet vs. Gamble' Choice Task where they choose to either gamble or take a sure bet on a series of independent trials; on one-third of all trials, participants reported how they felt about the outcome. We fit each participant's behavior to Rutledge et al.'s 'happiness model' to predict subjective feelings about decision outcomes. Parameters in the model include: certain rewards (CR), expected value of the gamble (EV), and reward prediction errors over outcomes (RPE). No difference in the rate of gamble decisions or earnings were observed across groups; however, the ICD group showed a significant baseline bias towards subjective ratings of "very good" compared to non-ICD over all decisions and outcomes (on or off DT: $p < 0.001$). DT reduces positive feelings about these decisions in both ICD and non-ICD ($p < 0.001$). The EV of a gamble significantly increases positive feelings about gambles taken and related outcomes in ICD vs. non-ICD (on or off DT: $p < 0.001$). This effect is enhanced by DT in ICD ($p < 0.001$), but diminished by DT in non-ICD ($p < 0.001$). RPEs about decision-outcomes drive subjective ratings in both groups with no difference observed across ICD versus non-ICD off DT. However, DT appears to diminish the impact of RPEs in ICD ($p < 0.001$) and enhance the effect of RPEs in non-ICD ($p < 0.001$). Together, we show that enhanced positive experiences associated with risky decisions in patients with a history of ICD is driven by the anticipation of potential outcomes rather than experienced outcomes and that this effect is enhanced in the presence of DT. These results suggest that, in ICD, the reinforcing effects of dopamine may be most strongly associated with the onset of risky behavior and not on the outcome of these decisions, an effect that would lead to continued poor decision making in the face of adverse outcomes.

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Poster

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NCTIC Pilot Award (Wake Forest School of Medicine Neuroscience Clinical Trial and Innovation Center)

Title: Multi-shell diffusion-weighted magnetic resonance imaging for the study of white matter structural connectivity in treatment-resistant depression.

Authors: *C. K. JOHNSON[1], R. E. JONES[2], T. DURAN[2], H. E. DOUGLAS[1], P. GLIGOROVIC[1], K. T. KISHIDA[2];
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Abstract: Of the more than 300 million people diagnosed with depression worldwide, ~30% are classified as treatment-resistant (TRD). This occurs when a person fails to respond to conventional treatments, including a combination of psychotherapy with antidepressants and/or antipsychotics. The most effective intervention for TRD is electroconvulsive therapy (ECT), however, the mechanisms by which it alleviates symptoms of depression remain unknown. Several neuroimaging studies on depression, as well as those investigating effects of ECT, have demonstrated a difference in characteristics of white matter in the brains of depressed and non-depressed individuals and changes in white matter structure following ECT in individuals with

TRD, indicated by metrics such as fractional anisotropy (FA) and mean diffusivity (MD). However, results of these studies are inconsistent, primarily due to small sample sizes in some and imaging techniques that do not provide enough fine detail to reliably determine subtle differences in white matter structure, particularly where multiple fibers cross in a single voxel. Our approach uses multi-shell diffusion-weighted magnetic resonance imaging (DW-MRI), which includes multiple diffusion directions sampled along multiple spheres to provide high angular contrast necessary for distinguishing crossing fibers. Here, we conduct an observational study in which DW-MR images are collected from participants with no history of depression, those with mild/moderate depression, and those with severe TRD who are seeking ECT. Analysis of these images confirms that white matter structural integrity is diminished, as indicated by lower FA and higher MD, in cases of severe TRD (n = 8), and, to a lesser extent, in cases of mild/moderate depression (n = 11), when compared with images collected from individuals without depression (n = 25). Furthermore, our imaging approach allows us to detect significant differences in FA between participants with mild/moderate depression and those with TRD. Following ECT, we will use these techniques to identify with greater specificity than previously reported the white matter tracts most affected by this treatment for TRD and determine how they might track with functional changes in the brain and improvement of clinical symptoms. Finally, we will extend our work to include fitting penalized and cross-validated regression models to determine if it is possible to predict the effectiveness or side effects of ECT treatment. Together, our preliminary and ongoing work will further elucidate the mechanisms of ECT, thus allowing for better understanding of depression and, ultimately, higher quality of care for patients.

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Poster

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Wake Forest School of Medicine Neuroscience Clinical Trial and Innovation Center

Title: Slower learning from punishments and altered mapping of prediction errors to affective responses in patients with depression

Authors: *R. E. JONES[1], L. P. SANDS, III[1], A. JIANG[3], H. E. DOUGLAS[4], P. V. GLIGOROVIC[4], K. T. KISHIDA[2];
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Abstract: Major depressive disorder is characterized by persistent sadness and anhedonia, as well as deficits in cognitive function and decision-making. It is hypothesized that dopaminergic projections throughout mammalian reward circuitry encode temporal difference reward prediction error (TD RPE) signals, which quantify the difference between received and expected outcomes. Human functional magnetic resonance imaging (fMRI) studies have demonstrated reduced RPE responses to rewarding stimuli in the ventral striatum and ventral tegmental area in patients with depression; however, the neurocomputational components of punishment learning and how these may be altered in depression are less well understood. Standard computational reinforcement learning (RL) models equivocate rewards with punishments and thereby are unable to capture how humans, and especially patients with depression, may react non-linearly to these different classes of outcomes. We provide evidence suggesting that rewards and punishments are tracked by independent, valence-specific neural systems, though the bearings of such a valence-partitioned RL (VPRL) model on behavioral phenotypes in depression remains an open question. To address this, we conducted experiments in which healthy controls (HC; n=19), patients with depression (PwD; n=10), and patients with treatment-resistant depression (PwTRD; n=8) completed a probabilistic reward and punishment task while scanned with fMRI; participants reported how they felt about their outcomes on 33% of trials. We fit a standard TD learning model and a VPRL model to participant choice behavior and found that VPRL best captured how humans learn from rewards and punishments across cohorts. In this framework, PwD and PwTRD showed a specific decrease in their learning rate to punishment prediction errors (PPEs). We regressed RPEs, PPEs, and expected values (EVs) on participants' subjective ratings and determined that the three cohorts were best differentiated according to the influence of negative RPEs, positive PPEs, and the EV of the chosen and unchosen actions on subjective ratings. Our results demonstrate that how humans learn from rewards and punishments differentially impacts subjective feelings during decision-making, and that this mapping of learning signals to affective dynamics may be altered across depression severities. Our data collection and model-based fMRI analysis are ongoing. As our sample size increases we will investigate brain regions of interest that may track RPE and PPE signaling and how activity in these systems might be altered in depression.

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Poster

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Title: Time perception deficits are associated with individual differences in clinical presentation and comorbidities of Parkinson's disease

Authors: *E. DIMARCO[1], R. SADIBOLOVA[3], R. E. JONES[1], A. JIANG[1], B. LIEBENOW[1], I. U. HAQ[4], M. S. SIDDIQUI[2], D. B. TERHUNE[3], K. T. KISHIDA[1];
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Abstract: Striatal dopamine is commonly understood to mediate time perception and is the target of dopaminergic therapies (DT) prescribed to treat Parkinson's disease (PD); however, the role of dopamine in the intersection of timing and the non-motor symptomology of PD is incompletely understood. We hypothesize that specific deficits in interval timing will be associated with heterogeneity in PD presentation and comorbidities. To investigate, we recruited patients with PD (n = 19) to complete a temporal bisection task (interval durations: 500ms - 1100ms) while on and off their prescribed DT. We applied psychometric functions (PFs) to estimate temporal precision and accuracy for each participant in each medication state. Their timing performance was subsequently investigated in the context of different clinical features (e.g., age, PD duration, DT profile, comorbid depression diagnoses) and each patient's score on the Questionnaire for Impulsive-Compulsive Disorders in Parkinson's Disease–Rating Scale (QUIP-RS). We applied linear regression models with these clinical features as predictors and timing performance as outcome variables to examine how dopaminergic dysfunction related to interval timing performance. Although the PD patients on vs. off DT did not significantly differ in their timing ability ($p > 0.05$), we observed that longer PD duration of diagnosis (distribution = 1 – 8 years, $p < 0.01$) was associated with decreased temporal precision regardless of medication state. We additionally found that patients with suprathreshold QUIP-RS scores ($n = 11$, $p < 0.01$) showed increased temporal overestimation bias, and that patients with a depression diagnosis ($n = 4$, $p < 0.05$) showed increased bisection point error compared to euthymic subjects. This association between PD comorbidities and temporal deficits further highlights how PD symptom heterogeneity may be accounted for. Finally, patients who were prescribed poly-DT demonstrated an increase in proportion of correct responses and decreased bisection point error compared to patients prescribed Levodopa monotherapy ($n = 11$, $p < 0.01$). Together, these results demonstrate that non-motor PD symptoms are associated with specific changes in interval timing behavior consistent with changes in dopaminergic system function. This suggests that simple time perception tasks may be used to quantify and investigate specific mechanisms

underlying individual differences in the clinical presentation of PD; and, that investigating individual differences in time perception in PD patients may provide insights into the mechanisms of timing more generally in humans.

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Poster

P880. Learning and Value Processing in Disorders

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P880.05

Topic: H.10. Human Learning and Cognition

Support: MH114877
Philanthropic support

Title: Orbitofrontal beta-gamma rhythms influence reward processing and obsessive-compulsive behaviors

Authors: *S. GROVER, J. A. NGUYEN, V. VISWANATHAN, R. M. REINHART;
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Abstract: Many characteristic symptoms of various neuropsychiatric illnesses can be conceptualized as abnormalities in reward processing. This is particularly the case for disorders of compulsivity where computational models of decision-making have identified abnormalities in feedback-guided, goal-directed behavior. These abnormalities can be localized to a frontostriatal circuit consisting of the orbitofrontal cortex (OFC) and the striatum. Recent work in human electrophysiology has identified rhythmic activity in the high beta-low gamma (20-35 Hz) range as an index of reward processing. However, it is not conclusively known whether these rhythms originate from the OFC, and whether they selectively influence reward processing. In a randomized, double-blind, sham-controlled design, we entrained activity in the medial

orbitofrontal cortex (mOFC) using high-definition transcranial alternating current stimulation (HD-tACS) while human participants (N=60) performed a probabilistic reinforcement learning task. Personalized entrainment was performed at the peak beta-gamma frequency for every individual at which reward-related modulation could be observed in the electroencephalography (EEG). We found that personalized beta-gamma neuromodulation of the mOFC selectively modulated reward-guided behavior while leaving punishment-guided behavior intact. Our results provide direct evidence that beta-gamma rhythms are selectively associated with reward processing, and that the OFC is a likely source of these rhythms. Next, we examined the translational potential of beta-gamma rhythmic neuromodulation in the OFC. In a second randomized, double-blind, sham-controlled experiment (N=68), we examined the effect of chronic (over 5 days) mOFC neuromodulation on subclinical obsessive-compulsive symptoms. These symptoms were quantified at baseline, immediately after the fifth day of neuromodulation, and after 1 month, 2 months and 3 months post modulation. We observed a rapid reduction in obsessive-compulsive symptoms when mOFC was entrained at personalized reward-related beta-gamma frequencies. Critically, these improvements sustained over the next three months motivating the viability of this approach in clinical populations. Together, our results suggest that beta-gamma rhythms constitute the temporal code facilitating reward processing in the OFC, and that its chronic modulation can be used to reduce obsessive-compulsive symptoms. We speculate about the computational operations facilitated by beta-gamma rhythms, and examine the likelihood that beta-gamma rhythms facilitate connectivity between the OFC and the striatum.

Disclosures: S. Grover: None. **J.A. Nguyen:** None. **V. Viswanathan:** None. **R.M. Reinhart:** None.

Poster

P880. Learning and Value Processing in Disorders

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Program #/Poster #: P880.06

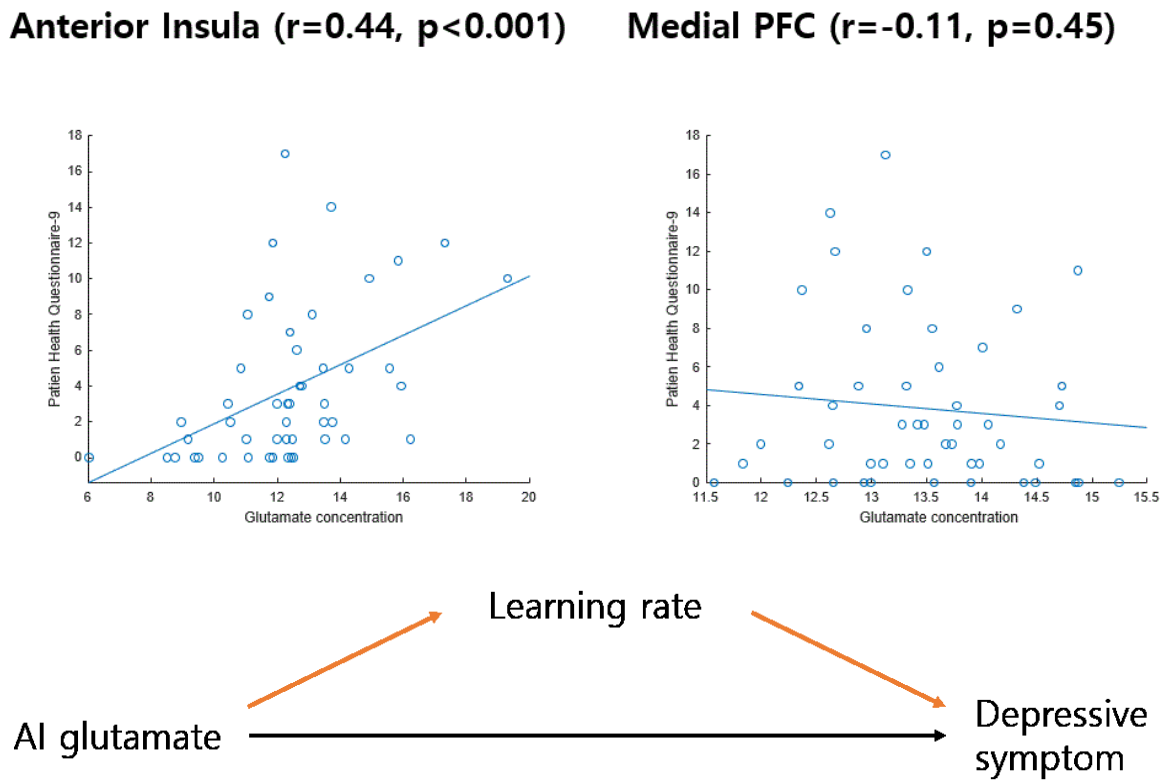
Topic: H.03. Decision Making

Title: Learning rate mediates effects of glutamate on depression

Authors: *H. PARK[1], S. KIM[1], D. YOON[1], J. KIM[2], B. JEONG[1];
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Abstract: Various learning paradigms have been used to show impairment in depression. However, neuro-computational mechanisms of abnormal decision-making in depression remain unclear. 52 subjects underwent 30 minutes of functional magnetic resonance spectroscopy (MRS) and performed both punishment-specific and reward-specific two-armed bandit decision-making tasks. Participants had to estimate the probability of some fractal stimuli to be associated

with reward or punishment. Dynamic accuracy and computational parameters of learning rate were compared with Patient Health Questionnaire-9 (PHQ9), which is depressive scale, using linear regression. Functional MRS focused on glutamate complex (Glx) in anterior insula (AI) and medial prefrontal cortex (mPFC) were performed to identify altered neurochemical change in depression. Time to reach threshold accuracy was negatively correlated with PHQ9 (r=-0.32, p=0.03). Model analysis consistently showed that a higher learning rate predicted higher PHQ9 performing linear regression (CI=[44.7 64.7]). Glx concentration in AI had a positive correlation with PHQ9 (r=0.44,p<0.001), but not with glutamate in mPFC (r=-0.11,p=0.45). Mediation analysis revealed that learning rate fully mediates effects of Glx on PHQ9 (p<0.05). Glutamate concentration abnormality in AI made depressive, which is mediated by higher learning rate.



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Poster

P889. Cognitive Development: Connectomics and Networks

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P889.01

Topic: G.02. Reward and Appetitive Learning and Memory

Support: National Science Foundation SBE Postdoctoral Research Fellowship Grant No. 1714321
National Science Foundation CAREER Award Grant No. 1654393
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Title: Reward-motivated memory processes and their underlying neural mechanisms change with age

Authors: *A. O. COHEN[1], M. M. GLOVER[3], X. SHEN[4], K. N. AVALLONE[1], C. V. PHANEUF[1], L. DAVACHI[5], C. A. HARTLEY[2];
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Abstract: Motivationally salient events are often prioritized in memory. In adults, memory for high-reward memoranda is related to increased connectivity involving mesolimbic dopamine systems, centered around the ventral tegmental area (VTA), the anterior hippocampus, and cortical areas both during and after encoding. More generally, dopamine-dependent plasticity in the hippocampus is critical for memory formation. Converging evidence from cross-species studies suggests that subcortical dopamine signaling is increased during adolescence, which may lead to stronger memory representations of rewarding events relative to mundane events. Additionally, given the distributed neural circuits that have been implicated in reward-motivated memory enhancements, it is possible that the underlying neural mechanisms change across age. To test these hypotheses, 89 participants ages 8 to 25 years-old completed a reward-motivated encoding fMRI paradigm with baseline and post-encoding active rest periods and returned 24-hours later for a behavioral memory retrieval test. We find that reward motivation enhances specific associative memory across age and leads to a boost in more general source memory during adolescence. Neuroimaging analyses suggest that reward-motivated encoding is supported by interactions between the prefrontal cortex and VTA across age, particularly in older participants. We also find that putative offline post-encoding consolidation mechanisms change with age such that experience-dependent increases in anterior hippocampus-VTA connectivity show a stronger relationship with reward-motivated memory benefits in younger participants. Taken together, our findings suggest that reward motivation enhances memory somewhat differently across age and that there may be age-related shifts from “offline” subcortical to “online” systems-level circuits supporting reward-motivated memory processes.

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Poster

P889. Cognitive Development: Connectomics and Networks

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P889.02

Topic: H.12. Aging and Development

Support: NHLBI R01-HL116585
CIHR MOP-81116
NICHD T32-HD098066

Title: Maternal education is associated with functional segregation of the fetal connectome.

Authors: K. M. COOK[1], **C. SITRIN**[2], *J. DE ASIS-CRUZ[1], S. D. BARNETT[1], D. KRISHNAMURTHY[1], C. LIMPEROPOULOS[1];
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Abstract: Parental education is consistently linked to children's cognitive and behavioral functioning including childhood IQ, executive functioning, and emotional processing as well as long-term outcomes of academic achievement, employment, and even prevalence of health concerns such as obesity. Despite this link and clear brain-behavior interactions, it is not known when in development education-driven differences become detectable in the brain. Moreover, the collinearity of socioeconomic status (SES) variables such as education and income, alongside the effect of greater neighborhood SES, makes it difficult to disentangle the effect of parental education later in development. To investigate these gaps, we utilized a cohort of fetuses from the DC/MD/VA area who underwent in utero, resting state fMRI scans. A total of 110 scans from 95 healthy fetuses between 19-40 weeks gestational age (GA; median: 34.93) were analyzed. Median education for participating pregnant mothers was 16 years, with 33% possessing a graduate degree. To assess neighborhood SES variables, we used participant reported zip code at the time of admission. Abstracted Census data suggest a median household income of $84,892, 12.8% live below the poverty line with an average 2021 SocioNeeds Index score of 21.3. Graph theory-based functional network metrics calculated from the fMRI data included clustering coefficient (*C*), characteristic path length (*L*), modularity (*Q*), global efficiency (*GE*), local efficiency (*LE*), normalized L (γ), normalized C (λ), and small-world-ness (σ). We utilized weighted Spearman Rank Correlation, controlling for GA at time of scan, to assess the relationship of education and SES indicators with brain network metrics. Statistically significant associations were observed for maternal education. Both *C* (r=-0.22) and *LE* (r=-0.24), metrics reflecting network segregation/specialization, were statistically significant for maternal education after controlling for gestation age, with both exhibiting a negative relationship. No connectivity metrics were significant for any of the Census data. Our findings show that relationships between emerging functional brain network structure and maternal education are detectable in utero, and that higher levels of education are associated with a diminished clustering coefficients and local efficiency - the latter of which we've observed decreases with fetal maturation in our prior work. Thus, even when controlling for GA, our findings suggest fetuses whose mothers have more education exhibit greater maturation (as measured by decreased network segregation) compared to those whose mothers have less education.

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Poster

P889. Cognitive Development: Connectomics and Networks

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P889.03

Topic: H.12. Aging and Development

Title: Structural and functional features of the adolescent brain in multiple cognitive domains

Authors: *C. HUMPHRIES[1], Y. LI[1], J. J. BERO[1], A. KUMAR[1], S. NAG[1], H. LEE[2], W.-Y. AHN[3,4], S. HAHN[3], H. KIM[5], D. LEE[6,2];
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Abstract: In recent years, a growing number of MRI studies have found associations between individual psychological variables on the one hand and structural or functional characteristics of specific brain areas on the other hand. For example, a study of emotional development might show a relationship between a metric of emotional maturity and the volume of the amygdala. One open question is whether the findings from these correlational studies probe unique associations of neuroanatomical and neurophysiological features with specific cognitive domains (e.g., memory, attention, sensory processing), or whether there are instead a few underlying sources of variation that appear across multiple domains.
To address this question, we selected a sample of 60 publications from the literature that investigated brain features associated with a wide variety of psychological variables, ranging from general cognitive and social skills to disorders, such as depression and anxiety. The modalities used in these papers were grouped into three broad categories: measurements of volume or surface-based structure, functional connectivity, and DTI metrics (e.g., FA values). The metrics from the MRI data used in these previous studies were recalculated using an independent MRI dataset collected from 459 adolescents aged 10 to 15 years, which included structural T1- and T2-weighted scans, a diffusion-weighted scan, and resting-state fMRI. For each of the previous studies, regions of interest (ROI) were chosen that matched the brain regions reported in the paper. Modality-specific metrics were then calculated for each ROI, and the resulting values were then normalized to z-scores. If a paper found an association between a psychological variable and multiple ROIs or modalities, then the resulting values were combined into a single measure using principal components analysis (PCA). In the end, we derived 74 metrics, each associated with a different psychological variable. The metrics were then analyzed using a hierarchical clustering analysis and PCA. The results showed three main clusters of

correlated metrics comprised of a variety of different psychological variables. In addition, the first 5 PCs of these metrics accounted for approximately 40% of their variance. These results suggest that multiple cognitive and emotional traits might be related to diverse functional and anatomical measures of the brain during adolescence.

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Poster

P889. Cognitive Development: Connectomics and Networks

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Program #/Poster #: P889.04

Topic: H.12. Aging and Development

Support: R21 DK118578
K99 HD100593
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R01 MH091351
SKIDI-Kids Program of the Academy of Finland 134950

Title: Maternal Pre-pregnancy Body Mass Index is Associated With Newborn Offspring Hypothalamic Mean Diffusivity: A Prospective Dual-Cohort Study

Authors: *J. RASMUSSEN[1], J. J. TUULARI[3], S. NOLVI[3], M. LAVONIUS[3], H. MERISAARI[3], L. KARLSSON[3], S. ENTRINGER[4], P. D. WADHWA[2], H. KARLSSON[3], C. BUSS[4];
[1]Univ. of California, Irvine, Long Beach, CA; [2]Univ. of California, Irvine, Irvine, CA; [3]Inst. of Clin. Med. Univ. of Turku, Turku, Finland; [4]Charité – Universitätsmedizin Berlin, Berlin, Germany

Abstract: Introduction Genomic, preclinical, and pharmacological studies all support the hypothalamus (HTH) as a critical regulator of energy (im)balance in the context of obesity. Further, a rich body of animal literature suggests that maternal obesity during pregnancy elicits structural changes to the developing offspring HTH, with implications for offspring obesity risk. Yet, the relationship between maternal BMI and the *newborn* human HTH remains unknown due to a lack of validated *in vivo* methods of measurement of HTH integrity. Recent evidence in adult humans supports HTH Mean Diffusivity (MD) as a replicable neuroimaging phenotype

associated with obesity. Here, using an international (Finland and California, USA) multi-site diffusion tensor imaging (DTI) dataset, we leverage this phenotype to test the hypothesis that maternal pre-pregnancy BMI is associated with newborn offspring HTH MD.
Methods HTH MD was independently quantified in two separate BMI-matched cohorts (up to Class I Obesity; BMI_{Range}=17-35) using a high-resolution atlas-based definition of HTH. A total of N=229 mother-child dyads were available for analysis ($N_{Site,1}$=152, Age at MRI=26.7+/-8.1 days, Gestational Age at Birth=39.9+/-1.2wks, $N_{M/F}$=82/70, BMI=24.9+/-4.3; $N_{Site,2}$=77, Age at MRI=25.6+/-12.5 days, Gestational Age at Birth=39.3+/-1.5wks, $N_{M/F}$=43/34, BMI=24.2+/-3.8). The association between maternal pre-pregnancy BMI and newborn offspring HTH MD was tested separately in each of these cohorts using linear regression controlling for gestational age at birth, postnatal age at scan, sex, whole white matter mean diffusivity, and quality control criteria. In post-hoc tests, additional potentially confounding factors including socioeconomic status, ethnicity, and obstetric risk, were adjusted for.
Results The distribution of maternal pre-pregnancy BMI was comparable across sites but differed in ethnicity and socioeconomic status in a geographically predictable manner. A positive linear association between maternal pre-pregnancy BMI and newborn offspring HTH MD was observed at both sites ($b_{Site,1}$ = 0.17, $p_{Site,1}$= 0.01; $b_{Site,2}$ = 0.22, $p_{Site,2}$=0.03) and remained significant after adjusting for cohort-relevant sociodemographic and clinical factors.
Discussion These findings demonstrate an association between maternal BMI and offspring hypothalamic microstructure consistent with animal literature. In addition to further replication and generalization, future studies aimed at identifying putative pathways in order to better inform primary intervention strategies are warranted.

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Poster

P889. Cognitive Development: Connectomics and Networks

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P889.05

Topic: H.12. Aging and Development

Support: NIGMS R01 GM134363-01

Title: Quantifying waveform shape of EEG alpha and mu oscillations across development

Authors: *A. S. BENDER[1], N. SCHAWORONKOW[2], B. VOYTEK[2,3,1,4];
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Abstract: Neural oscillations in the human brain are most prominent in the alpha band (8-13 Hz) as detected with EEG. While alpha oscillations in occipital cortex and mu oscillations in sensorimotor cortex occur at similar frequencies, it has been difficult to analyze mu oscillations due to their weaker amplitude relative to alpha oscillations. Because neural oscillations are commonly nonsinusoidal and the nonsinusoidal features have been shown to reflect underlying physiological and pathophysiological characteristics (Cole et al., 2017), quantifying the differences in waveform shape between alpha and mu oscillations may inform understanding of the physiological distinctions between these oscillations. In this study, we aim to quantify changes in waveform shape of both alpha and mu oscillations across development. We analyzed an open dataset of high density, resting-state EEG from 1205 participants (ages 5-21) of the Healthy Brain Network Biobank (Alexander et al., 2017). We used spatio-spectral decomposition to extract oscillatory components with enhanced signal-to-noise ratio in the alpha band. The spatial patterns for each of these components, which represent how each oscillatory source maps onto the electrodes, were matched to alpha and mu template patterns to identify whether each component represented an alpha or mu oscillation. We then used the bycycle package (Cole & Voytek, 2019) to quantify waveform shape features on a cycle-by-cycle basis. Using this approach, we found that the peak and trough of alpha waveforms were significantly less asymmetrical than those of mu waveforms ($t(1157)=12.9, p<0.001$), confirming that alpha and mu oscillations can be differentiated by their waveform shape. Additionally, we confirmed that alpha waveforms increase in frequency ($r=0.51, p<0.001$) and decrease in amplitude ($r=0.13, p=0.018$) across age, while there are no significant changes across age to waveform shape features. Mu waveforms, on the other hand, increase in frequency ($r=0.43, p<0.001$) and become more asymmetrical with age ($r=0.19, p<0.001$), but have no significant change in amplitude ($r=0.018, p=0.66$). The fact that alpha and mu oscillations undergo different changes during development suggests that the developmental modifications to physiological and pathophysiological characteristics underlying each of these oscillations are unique.

Disclosures: A.S. Bender: None. **N. Schaworonkow:** None. **B. Voytek:** None.

Poster

P889. Cognitive Development: Connectomics and Networks

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Program #/Poster #: P889.06

Topic: H.12. Aging and Development

Support: NIH Grant 1R21MH112019-01A1
Kavli Foundation Grant KSIP-2017-002
BrainsCAN at Western University

Title: Spatiotemporal dynamics in infant sleep spindles follow a longitudinal maturational pattern from 3 to 6 months of age

Authors: *A. PATHAK[1], S. E. PETERS[2], L. MULLER[1], A. A. BENASICH[3], T. J. SEJNOWSKI[4];
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Abstract: Spatiotemporal dynamics in infant sleep spindles follow a longitudinal maturational pattern from 3 to 6 months of age
Authors: A. PATHAK*, S. E. PETERS, L. MULLER, A. A. BENASICH, T. J. SEJNOWSKI
Sleep spindles are patterned bursts of oscillatory brain activity seen in the EEG, associated with NREM sleep, that are known to play a key role in information processing, neuroplasticity, and ongoing cortical development. Previous work (Muller et al.,eLife,2016) in our lab suggests that sleep spindles travel across the cortex as a neural traveling wave (nTW) rotating in the Temporal-Parietal-Frontal (TPF) direction via long and short-range cortical white matter tracts. Here we examine the same typically developing infants at 3 and 6 months using high resolution EEG acquired at each age during a daytime nap. We tested the hypothesis that traveling spindle waves are present and follow a maturational pattern. This developmental period is characterized by a high rate of active cortical myelination that follows a spatiotemporal trajectory from posterior to anterior and medial to lateral regions over the ages of ~3 to 10 months. We hypothesize that the developing topography and spatiotemporal dynamics of infant sleep spindles are associated with cortical myelinating fibers. Hemispheric differences in the rate of myelination in the developing cortex have been shown with delayed onset and a faster rate of myelination in the right hemisphere. The thin cranium at these ages allows EEG to capture neural activity with relatively high spatial resolution. By applying a phase-based method for detecting traveling waves in noisy multichannel data, we quantify the relative proportion of nTW patterns across these ages. Our computational algorithms for detecting rotating nTWs allowed us to characterize and classify the spatiotemporal patterns during thousands of episodes of spindle activity in several hours of sleep recordings at each age point. Our results show a significant increase in rotating waves from 3 to 6 months (n = 37, 21 females) in the TPF direction ($p<0.0001$). Interestingly, we observe the reverse trend in the opposite Temporal-Frontal-Parietal (TFP) direction across cortex, with a significant decrease in the wave activity from 3 to 6 months ($p<0.0001$). The nTW spindle analyses by hemisphere suggest greater individual variability in the nTW measure across ages in the left hemisphere as compared to the right, which is in line with a longer average myelination window in the left hemisphere. In this ongoing work, we will examine the hemispheric and sex differences at each age in a larger cross-sectional sample, along with behavioral data collected concurrently in this sample.

Disclosures: A. Pathak: None. **S.E. Peters:** None. **L. Muller:** None. **A.A. Benasich:** None. **T.J. Sejnowski:** None.

Poster

P889. Cognitive Development: Connectomics and Networks

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Program #/Poster #: P889.07

Topic: H.12. Aging and Development

Title: Learning-induced reorganization of neural representations in a biologically inspired neural network reveals developmental dynamics of numerosity representations

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Abstract: We implemented in-silico experiments to investigate plausible mechanisms of how learning induced numerosity representations emerge and evolve at a neuronal level over a key developmental period. We used a biologically inspired network (CORNet-S) developed for visual neuroscience whose architecture with 4 blocks (V1, V2, V4, inferior temporal cortex) maximized a composite Brain-score benchmark. A numerosity classification task was tested on this network pre-trained for object recognition. Subsequently, the network was trained on numerosity recognition and was probed at a neural level to measure various aspects of representational dynamics with learning. We show that as in some previous studies, spontaneous numerosity tuned neurons emerged in the pre-trained network. A key contribution of our study was to track the evolution of representational dynamics at a neuronal level with training. The spontaneously emergent neurons were not critical in the formation of subsequent representations and the improvement in accuracy was correlated to the dropping off of spontaneous neurons. Training reorganized the network with rapid representational drift in these neurons after even a single epoch of training. With training, a larger proportion of new neuronal groups became sensitive to specific numerosities, but continued to show representational drift with learning, manifested as an evolving mix of transient and persistent neurons. This continuous drift, possibly contributing to learning, was more pronounced at higher layers (V4 and IT). Using multidimensional scaling we found that the distributed representations in higher layers were predicated on two dimensions, one encoding magnitude, and the other encoding distance from the mid-point of the stimuli space (mid-point anchoring, similar to what has been demonstrated in numerical estimation tasks). The representational drift was smaller when measured at a distributed level compared to a neuronal level. Collectively this conveyed a learned cognitively plausible distributed representation almost independent of spontaneously emergent neurons. All of these aspects of drift were significantly correlated with classification accuracy. The biologically plausible network and cognitively plausible representations provided a unique environment for understanding how learning induced developmental changes may be manifested. This informs future considerations of training paradigms, and validates a platform to examine the neuronal basis for individual differences and mathematical learning difficulties, otherwise impossible to study at a neuronal level during this critical stage of development.

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Poster

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Topic: H.12. Aging and Development

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Title: Association of Wisconsin Card Sorting Test with Structural and Functional MRI in Healthy Adults

Authors: *I. CHI[1], A. YANG[1,2,3];
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Abstract: Background: The human brain is extremely abstract, and it is difficult to quantify the amount of consciousness and thought. Fortunately, the invention of scientific tools in the last decade has given us the opportunity to explore the world of thought and ideas without boundaries. In the theory of mind, thinking is subdivided into very different abilities, and cognitive functions are the most important of them. **Data and Methods:** The data used in this study were obtained from the Taiwan Aging and Mental Illness (TAMI) database. A total of 290 healthy adults aged 20 to 60 years were used. Each subject underwent a functional magnetic resonance imaging scan and cognitive function was assessed by the Wisconsin Card Sorting Test. The Pearson's product-moment correlation coefficient was used to identify significant brain areas. A corrected p-value was used to determine whether it was statistically significant, and stepwise analysis in multiple linear regression was used to develop a predictive model of cognitive function based on brain imaging metrics. **Results:** In the prediction model of cognitive function based on gray matter volume, the results showed that the important brain regions were the inferior frontal gyrus, the rectus gyrus, the right and left superior parietal gyrus, the angular

gyrus, and the caudate nucleus in the orbital region. In the predictive model of white matter bundle integrity, the right superior cerebellar peduncle was the most important, and the next most important areas were the medial lemniscus and the left posterior thalamic radiation. In terms of functional connectivity of the whole brain, the important brain regions are the right superior limbic gyrus, the pontine brain and the left and right brainstem.**Conclusion:** Using different structural and functional approaches, the resulting brain regions were found to be important areas that affect abstract thinking ability and participate in higher-level cognitive coordination, internal conceptualization and processing. **Key words:** cognitive function, Wisconsin Card Sorting Test, resting-state functional magnetic resonance imaging, diffusion tensor imaging, global functional connectivity

Disclosures: I. Chi: None. **A. Yang:** None.

Poster

P907. Cognition: Animal Models of Schizophrenia

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P907.01

Topic: H.13. Schizophrenia

Title: Suvn-5107002, a potent serotonin and weak dopamine modulator demonstrating antipsychotic effects with no motor side effects

Authors: *A. SHINDE, S. PETLU, N. NATH, A. RAYA, J. THENTU, P. ACHANTA, K. BOJJA, J. TADIPARTHI, V. GRANDHI, H. TALAKANTI, R. ABRAHAM, R. KALLEPALLI, R. NIROGI;
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Abstract: Recent interest in the role of serotonin (5-HT) in antipsychotic drug action is based mainly upon the fact that antipsychotic drugs are potent 5-HT_{2A} receptor antagonists and relatively weaker dopamine D_2 antagonists. These agents share in common low extrapyramidal side effects at clinically effective doses and greater efficacy to reduce negative symptoms. SUVN-5107002 is a lead compound with potent affinity at SERT transporter and 5-HT_{2A} receptor with weak affinity for dopamine receptors is being proposed for the treatment of psychosis. *In vitro* pharmacological properties of SUVN-5107002 were evaluated in radioligand binding assay and cell based reporter gene assay. The pharmacokinetic profile was evaluated in rats and dogs. Non- radiolabeled assay was used to assess the occupancy of test compound at SERT, 5-HT_{2A}, 5-HT_{1A} and D_2 receptors in rats. Brain microdialysis assay was used to assess the effects on the cortical levels of monoamine neurotransmitters. Functionality at SERT, 5-HT_{2A}, D_2 and 5-HT_{1A} receptor was evaluated by both *in vitro* and *in vivo* methods. SUVN-5107002 has potent affinity at SERT, 5-HT_{2A}, 5-HT_{1A} and weaker affinity at D_2 receptors. It showed antipsychotic and antidepressant effects in animal models. The efficacy doses were devoid of

motor impairment i.e., a clear separation between the doses which produced efficacy and side effects. From the current research we conclude that SUVN-5107002 has therapeutic potential for antipsychotic and antidepressant effects without inducing debilitating side effects associated with antipsychotics or antidepressants.

Disclosures: **A. Shinde:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **S. Petlu:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **N. Nath:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **A. Raya:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **J. Thentu:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **P. Achanta:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **K. Bojja:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **J. Tadiparthi:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **V. Grandhi:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **H. Talakanti:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **R. Abraham:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **R. Kallepalli:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **R. Nirogi:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd.

Poster

P907. Cognition: Animal Models of Schizophrenia

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Program #/Poster #: P907.02

Topic: H.13. Schizophrenia

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3503

Title: Activating $mGlu_3$metabotropic glutamate receptors rescues schizophrenia-like cognitive deficits through metaplastic adaptations within the hippocampus

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Abstract: Polymorphisms in *GRM3*, the gene encoding the $mGlu_3$ metabotropic glutamate receptor, are associated with impaired cognition and neuropsychiatric disorders such as schizophrenia. However, the mechanisms through which $mGlu_3$ receptors regulate synaptic plasticity and cognition are not known. Recent studies have provided evidence for a functional partnership between $mGlu_3$ and $mGlu_5$, prompting the current set of studies to investigate concerted actions between $mGlu_3$ and $mGlu_5$ in the enhancement of hippocampal synaptic plasticity and cognition. In this study, we performed a series of experiments in c57BL6 and cell-specific KOs of *Grm3* and *Grm5* to evaluate the role of $mGlu_3$ in hippocampal synaptic plasticity and cognition. We showed that $mGlu_3$ receptor activation enhanced trace fear conditioning, an associative learning task disrupted in patients with schizophrenia. Interestingly, it also reversed the deficits in cognition induced by subchronic administration of NMDA antagonist phencyclidine. To gain mechanistic insight of *in vivo* phenomenon, studies using selective pharmacological tools showed that activation of $mGlu_3$ receptor-induced metaplastic changes, biasing afferent stimulation to induce long-term potentiation at SC-CA1 synapse through a $mGlu_5$ receptor-dependent mechanism. Further, $mGlu_3$-mediated enhancement of LTP required CB1 receptor-dependent depression of inhibitory transmission onto pyramidal neurons. Selective deletion of either $mGlu_3$ or $mGlu_5$ from hippocampal pyramidal cells eliminated the effects of $mGlu_3$ activation on hippocampal LTP and cognitive enhancement reveal a novel mechanism by which $mGlu_3$ and $mGlu_5$ interact to enhance cognitive function. These studies suggest the potential utility of developing approaches to positively modulate $mGlu_3$ receptor function for treating cognitive disruption in schizophrenia and other psychiatric diseases.

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Poster

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Program #/Poster #: P907.03

Topic: H.13. Schizophrenia

Title: Roluperidone (MIN-101) Causes Brain Region-Specific Neurotransmitter Release, Increased Brain Activity and Functional Network Connectivity in the Rat Brain Related to Negative Symptoms in Schizophrenia

Authors: *R. HODGSON[1], N. ENGLISH[1], J. RUSSO[1], D. RADL[1], M. PAPAKOSTA[1], H. SUN[1], Z. CHERUVALLATH[1], J. MCLAUGHLIN[1], K. KOZMINSKI[1], J. SCHACHTER[1], A. SHATILLO[2], J. OKSMAN[3], R. KOZAK[1], H. MONENSCHEIN[1], H. SCHIFFER[1];

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Abstract: The lack of consistent and robust efficacy in treatments of negative symptoms in schizo-phrenia (SCZ) is a large burden for patients and represents an unmet need to develop novel therapies. Understanding polypharmacology, neurotransmitter release and brain circuit modulation of compounds demonstrating some efficacy in treating negative symptoms in SCZ patients will facilitate new drug development efforts. Roluperidone (MIN-101) is currently in clinical development by Minerva to treat negative symptoms in SCZ and previously demonstrated clinical efficacy in treating negative symptoms in SCZ in a phase 2b trial. MIN-101 shows a unique poly-pharmacology believed to drive its clinical efficacy: antagonizing serotonergic 5HT2A, sigma-2 and adrenergic alpha 1 receptor without direct binding to dopamine receptors. However, comprehensive preclinical data characterizing the MIN-101 mechanism of action are not publicly available. Studies focusing on structural and functional imaging and non-invasive electrophysiology implicate perturbations of the frontocortico-temporal circuits and disruption of cortico-striatal loops to negative symptoms in SCZ. We performed *in vivo* microdialysis and functional (f)MRI studies in naïve rats to obtain more insight into the neuropharmacological mechanism of MIN-101 and its impact on neurotransmitter chemistry and neurocircuitry. An acute oral dose of MIN-101 (1 mg/kg) significantly increased dopamine, norepinephrine and serotonin release in the hippocampus compared to vehicle dosing, but not in the striatum or prefrontal cortex. Additional less prominent changes in glutamate or acetylcholine release were observed and GABA release was unaffected. The brain activation pattern upon intravenous MIN-101 administration (1 mg/kg i.v.) was measured in rats using BOLD functional MRI (fMRI). In addition, the resting state functional connectivity changes induced by MIN-101 were studied. MIN-101 produced an acute and region-specific effect on the BOLD signal, showing strong activation of the medial prefrontal cortex (mPFC) and striatum. Interestingly, striatal activation was sustained during the whole 30 minutes measurement time in contrast to a sharp mPFC activation, which was followed by a signal decay in 10 min. No meaningful difference in BOLD signals was detected in hippocampus and thalamus. In resting state functional connectivity analysis, MIN101 significantly increased network connectivity over time. Our studies demonstrate that MIN-101 modulates hippocampal and cortical-striatal circuitry in naïve rats, which has been linked to negative symptoms in SCZ.

Disclosures: R. Hodgson: A. Employment/Salary (full or part-time):; Takeda. **N. English:** A. Employment/Salary (full or part-time):; Takeda. **J. Russo:** A. Employment/Salary (full or part-time):; Takeda. **D. Radl:** A. Employment/Salary (full or part-time):; Takeda. **M. Papakosta:** A. Employment/Salary (full or part-time):; Takeda. **H. Sun:** A. Employment/Salary (full or part-time):; Takeda. **Z. Cheruvallath:** A. Employment/Salary (full or part-time):; Takeda. **J. McLaughlin:** A. Employment/Salary (full or part-time):; Takeda. **K. Kozminski:** A. Employment/Salary (full or part-time):; Takeda. **J. Schachter:** A. Employment/Salary (full or part-time):; Takeda. **A. Shatillo:** None. **J. Oksman:** None. **R. Kozak:** A. Employment/Salary (full or part-time):; Takeda. **H. Monenschein:** A. Employment/Salary (full or part-time):; Takeda. **H. Schiffer:** A. Employment/Salary (full or part-time):; Takeda.

Poster

P907. Cognition: Animal Models of Schizophrenia

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P907.04

Topic: H.13. Schizophrenia

Title: Investigating the effects of acute and chronic exercise on cognitive dysfunction in a preclinical model of relevance to schizophrenia

Authors: *J. FLETCHER, T. WIGHT, J. GIGG, J. C. NEILL, B. GRAYSON, M. K. HARTE; Univ. of Manchester, Manchester, United Kingdom

Abstract: Cognitive impairment associated with schizophrenia remains an unmet clinical need. Increasing evidence indicates parvalbumin interneurons (PVI) and their associated perineuronal nets (PNN) play a key role in cognition. Reductions in PVI and PNN have been observed in schizophrenia patients and in vivo models of relevance to schizophrenia. Aerobic exercise has been correlated with both cognitive and PVI improvements in disease models, although the effect on PNN is unknown. This study aimed to investigate the behavioural and pathological outcomes in an acute and chronic model for exercise. 80 female lister hooded rats were dosed with sub-chronic phencyclidine (scPCP, 2mg/Kg, i.p.) or vehicle bi-daily for 7 days. Following a 1 week washout, baseline recognition memory was tested using the novel object recognition task (NOR). The difference between time spent at the novel and the familiar object was divided by total exploration to find the discrimination index (DI). Rats were subsequently exercised on individual running wheels for 1 hour/day. For the acute exercise cohort (AE), cognition was assessed immediately after exercise using NOR. The chronic exercise cohort (CE) were exercised 1 hour/day, 5 days/week. CE rats underwent NOR testing 24 hours after completing 3 and 6 weeks of exercise. After exercise, scPCP exercise (PE) and scPCP sedentary (PS) were tested with a continuous NOR (cNOR) protocol, comprising of 11 trials of NOR without intervention. CE brains were taken for immunohistochemistry (IHC), the prefrontal cortex was stained for parvalbumin (PV) and PNN. The baseline NOR test for AE and CE revealed scPCP treated rats had reduced DI compared to controls. In AE, all groups, apart from PS, spent significantly more time at the novel object, indicating acute exercise recovered scPCP induced cognitive impairment. This recovery was also seen in the PE group in the CE study at 6 weeks but not at 3 weeks, signifying separate mechanisms for AE and CE. In the first 4 trials of the cNOR, both PE and PS rats had intact recognition memory. However, in the final 4 trials, PS DI had decreased compared to PE, suggesting memory formation of PS rats is susceptible to distraction. IHC analysis revealed PV and PNN density decreased in scPCP treated rats relative to vehicle controls. Within treatments, exercise increased PV density and reduced PNN density. This study highlights the role of PVI and PNN in scPCP induced cognitive impairment and the ability of

aerobic exercise to reverse cognitive impairment and PVI pathology. Studies to investigate the wider mechanisms, including changes to PNN, are ongoing.

Disclosures: J. Fletcher: None. **T. Wight:** None. **J. Gigg:** None. **J.C. Neill:** None. **B. Grayson:** None. **M.K. Harte:** None.

Poster

P907. Cognition: Animal Models of Schizophrenia

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Program #/Poster #: P907.05

Topic: H.13. Schizophrenia

Title: Auditory steady-state response (40Hz) in rodents, a new tool for drug discovery in schizophrenia?

Authors: *J. VOLLE, C. HABERMACHER, V. DUVEAU, A. EVRARD, C. ROUCARD, Y. ROCHE;
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Abstract: Schizophrenia is a severe psychiatric disorder associated with persistent alterations of diverse neurocognitive functions, leading to lifelong psychosocial disabilities. Although schizophrenia has long been considered as a condition that specifically impairs the higher-order functions, recent research has demonstrated that basic sensory processing is also impaired, especially in the auditory modality. Neurophysiological approaches have provided evidence that most schizophrenic patients exhibit a wide range of clinically measurable dysfunctions in the processing of auditory stimulations, including the Auditory Steady-State Responses (ASSRs) which is one of the most consistent functional biomarker across schizophrenic patients. ASSRs consist in cortical electrophysiological oscillations entrained by the frequency and phase of a periodic auditory stimulus presented at a rhythm in the gamma range (that is, 30-80Hz). ASSRs are believed to reflect the interplay between cortical pyramidal neurons and parvalbuminergic interneurons. Consistent with the theory of imbalance between cortical excitation and inhibition in schizophrenia, patients show reduced power and phase locking, to stimuli presented at 40Hz. In recent years, we have developed the characterization of ASSRs in mice and rats to propose a translational solution for drug candidates. In this work, we particularly studied the 40Hz-ASSR modulations in a rat model of schizophrenia, induced by a glutamatergic antagonist MK-801. We will show in this poster how MK-801 (0.1, 0.15, 0.2mg/kg) induces a dose-dependent reduction of the 40Hz-ASSR phase locking in the cortex of rat and how antipsychotics (such as clozapine) modulate this pharmacological effect. We propose the 40Hz-ASSR phase locking as a specific translatable biomarker useful for the preclinical identification, selection and validation of new innovative therapeutics in psychiatric disorders.

Disclosures: **J. Volle:** A. Employment/Salary (full or part-time):; SynapCell SAS. **C. Habermacher:** A. Employment/Salary (full or part-time):; SynapCell SAS. **V. Duveau:** A. Employment/Salary (full or part-time):; SynapCell SAS. **A. Evrard:** A. Employment/Salary (full or part-time):; SynapCell SAS. **C. Roucard:** A. Employment/Salary (full or part-time):; SynapCell SAS. **Y. Roche:** A. Employment/Salary (full or part-time):; SynapCell SAS.

Poster

P907. Cognition: Animal Models of Schizophrenia

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Topic: H.13. Schizophrenia

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Title: Abnormal Perineuronal Nets and Gamma Band Oscillations in Mice with Vitamin D Deficiency

Authors: S. YU, *T. KIM;
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Abstract: Abnormal gamma-band oscillations (GBO) are possible mechanisms for cognitive dysfunction in patients with schizophrenia (SZ). In our brain oscillations, fast-spiking (PV) neurons are a pivotal component of GBO, and they are surrounded by perineuronal nets (PNNs), which are net-like extracellular matrix. Several studies showed that PNNs enable PV to make fast-spiking and GBO, and recent research suggested that a lack of vitamin D disrupts the PNNs integrity in the brain. However, the direct effect of vitamin D deficiency on GBO remains unknown. Here we hypothesized a decrease of PNNs by a vitamin D deficiency diet might change the power of GBO. First, we measured the evoked or spontaneous EEG power at the gamma range frequency in 4 to 6-month-old male PV-Cre mice. Then, the mice were grouped into control and vitamin D deficiency with normal and vitamin D depleted-diet for 6 weeks, respectively. To examine the impact of reduced PNNs on GBO, we injected chondroitinase ABC (chABC), an enzyme that digests PNNs, intracerebrally in the frontal cortex and measured optogenetically evoked GBO. Finally, we injected SB-3CT, an inhibitor of matrix metalloproteinase 9 (MMP-9), to rescue abnormal phenotypes in vitamin D deficiency. Immunohistochemistry was performed for PV neurons and PNNs, and analyzed with CellProfiler software. Our preliminary results showed that PNNs decreased in the vitamin D deficiency group, and the optogenetically evoked GBO decreased simultaneously. In contrast, spontaneous GBO increased in the vitamin D deficient group. Interestingly, after PNNs were reduced by chABC, the power of the evoked GBO decreased, and the spontaneous GBO increased

immediately. Vitamin D deficient mice treated with MMP-9 inhibitor demonstrated no changes in GBO and PNNs, Taken together, we suggest that the vitamin D deficiency could result in the decreased ability to generate GBO, especially around 40 Hz, by reducing PNNs. These results are reminiscent of the decreased auditory steady state response (ASSR) at 40Hz in schizophrenia. In addition, the relationship between vitamin D deficiency and schizophrenia has been suggested. Accordingly, the reduced PNN might be a necessary and sufficient condition for the reduced ASSR in schizophrenia.

Disclosures: S. Yu: None. **T. Kim:** None.

Poster

P907. Cognition: Animal Models of Schizophrenia

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Topic: H.13. Schizophrenia

Support: NIH R01 MH107491
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Title: Gamma band LFP activity in a cognitive control task is decreased after NMDAR blockade in primates

Authors: A. M. HUYNH[1], *D. A. CROWE[1], R. K. BLACKMAN[2], M. V. CHAFEE[2];
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Abstract: We measured local field potentials in the prefrontal and parietal cortices of monkeys to investigate the effects of NMDAR blockade (using phencyclidine; PCP) on gamma power during a cognitive control task. Hypofunction of NMDAR has been shown to be associated with schizophrenia-like symptoms. We utilized a task, the dot-pattern expectancy task (DPX), that isolates a specific deficit in patients with schizophrenia. Monkeys given PCP show the same pattern of errors on this task as patients with schizophrenia. In the DPX task two stimuli (the "cue", which could be either 'A' or 'B', and then the "probe", which could be either 'X' or 'Y') were displayed sequentially, separated by a delay period. The animal made a response based on the combination of these two stimuli. The four categories of 'Cue-Probe' pairs were 'AX', 'AY', 'BX', and 'BY'. 'AX' trials, 69% of the total, required a target response, the movement of a joystick to the left. The three other trials require a non-target response, a joystick movement to the right. Monkeys given PCP and patients with schizophrenia perform selectively worse on 'BX' trials. To study the effects of NMDAR blockade on neural signals related to this cognitive control task, we injected monkeys on alternate days with saline or 0.25 - 0.30 mg/kg PCP, and subsequently recorded local field potentials while the animals performed the DPX task. We converted the field potentials into power time courses (49 frequencies from 1-100 Hz) using a

wavelet analysis and then averaged across frequencies into five frequency bands. Our analysis here focuses on gamma power during the probe period of the task.In general, we observed that gamma power increased shortly after the display of the probe stimulus. In both the PCP and control conditions, gamma power increased faster after the display of 'X' probes than for 'Y' probes. Overall, gamma power was lower under the PCP condition during the end of the delay period in parietal but not prefrontal cortex. PCP administration reduced the stimulus-locked increase in gamma power in both brain areas, though it did not tend to alter the timing of such activity. Interestingly, probe-related differences in gamma power, as shown by a decoding analysis, were generally not affected by PCP.

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Poster

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Topic: H.13. Schizophrenia

Support: NIMH MH10748
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Title: Examining differential cancer rates in severe mental illness

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Abstract: Schizophrenia is a devastating psychiatric disorder that has a global prevalence of about 1%. Interestingly, overall cancer rates in schizophrenia are lower than the general population. Previous epidemiological studies of cancer rates in schizophrenia did not fully account for confounding variables such as smoking and substance abuse. We sought to confirm lower cancer rates in schizophrenia and explore mechanisms responsible for this unexpected observation.
Cell culture studies have shown dysregulation of kinase networks driven by antipsychotic administration with a focus on Dopamine 2 Receptor antagonism. Gene expression analysis of cancer cells treated with antipsychotic drugs has shown decreased expression of kinases involved in cell survival, proliferation, and drug resistance including AKT and PIM kinases. We hypothesize that typical antipsychotics are competitive inhibitors of AKT and PIM kinases. We postulate that inhibition of these and other kinases by antipsychotics accounts for reduced cancer incidence in patients with schizophrenia.

We applied a three-pronged approach to answer this question: 1) Epidemiological confirmation of reduced cancer incidence rates in patients with schizophrenia, 2) bioinformatics analyses of perturbagen structural moieties and gene expression data, and 3) in vitro kinase activity drug screening. Epidemiological data confirmed reduced risk of cancer incidence in patients with schizophrenia when accounting for age and smoking rates using the Healthcare Cost and Utilization Project (HCUP). Bioinformatics analyses of perturbagen structure and transcriptional profiles suggest a medication effect, as opposed to genetic factors (such as single nucleotide polymorphisms risk alleles for schizophrenia), as a main contributor to the observed decrease in cancer rates. In vitro kinase activity and protein expression will be measured in A549 cancer cells. In summary, we have identified AKT and PIM kinases as possible novel targets of typical phenothiazine antipsychotics. Taken together, our data suggest that phenothiazine antipsychotics act directly on protein kinases to lower cancer risk.

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Poster

P963. Computational and Analysis Tools II

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NIH grant number U24EB028998
NSF grant number 1904444-1042C

Title: Neuron 8.0: achievements of Neuron developer community to enable modern, performant and maintainable simulation environment

Authors: *J. G. KING[1], P. S. KUMBHAR[1], O. AWILE[1], F. PEREIRA[1], A. SAVULESCU[1], I. MAGKANARIS[1], O. LUPTON[1], N. CORNU[1], W. W. LYTTON[2], R. A. MCDOUGAL[3], S. DURA-BERNAL[5], A. NEWTON[6], M. L. HINES[4];
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Abstract: In order to build biologically ever-larger detailed reconstructions and simulations of large-scale neuronal circuits, the Blue Brain Project (BBP) and Yale University have added many new features to the NEURON simulator to improve efficiency and accessibility for massively parallel operation. Having been developed for the past 45 years, NEURON carries with it a substantial amount of legacy code which initially limited structural changes and prevented fast adoption of features from modern architectures. During the last two years, members from BBP, Yale university, and SUNY Downstate have organized regular discussion groups to consolidate ideas for advancing the state of NEURON source. As part of this developer community, we have made significant progress towards making NEURON into a modern, performant, portable and maintainable simulation environment including the following major developments: 1. A new build system using CMake in place of autotools; 2. Addition of documentation to nrn.readthedocs.io; 3. The introduction of an officially supported Python wheel for NEURON with automatic deployment on PyPi to enable installation of NEURON via pip; 4. Integration of CoreNEURON, an accelerated compute engine for NEURON providing more than 7-fold acceleration on NEURON simulations using CPUs; 5. Development of NMODL Framework, a source-to-source compiler for the domain-specific languages NMODL uses to describe biological models simulated in NEURON, allowing translation to C++, CUDA, OpenACC, ISPC, and LLVM IR backends to enable acceleration on a large variety of computing systems; 6. Addition of intracellular reaction-diffusion "chemophysiology" to electrophysiology; 7. Additional frontend development via GUIs and declarative language (NetPyNE).

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Poster

P963. Computational and Analysis Tools II

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Topic: I.06. Computation, Modeling, and Simulation

Support: MH116492-03S1

Title: Quantum machine learning applied to neuropsychiatric genomics datasets: proof-of-concept and biological interpretation

Authors: *P. S. EMANI, J. CHAPMAN, J. WARRELL, A. DONAGHEY, M. B. GERSTEIN; Mol. Biophysics and Biochemistry, Program in Computat. Biol. and Bioinformatics, Yale Univ., New Haven, CT

Abstract: Massively parallel classical computing continues to address computations on increasingly large datasets in genomics and neuroscience. However, the question remains whether alternate computing paradigms, such as quantum computing (QC), can capitalize on the promise shown on some algorithms to offer relative speedups or more efficient representations. Any gains need to be demonstrated on biologically and clinically relevant datasets, with complex interactions between many features. Accordingly, we explore functional genomics datasets from schizophrenia cases and controls from the PsychENCODE consortium, using quantum machine learning (QML) methods. Specifically, we aim to classify samples from the human prefrontal cortex according to neuropsychiatric disease status, based on sample gene expression. This is a complex classification problem, with a high-dimensional underlying structure. QML methods encompass many algorithms constructed as analogues to classical ML methods, including neural networks. We construct multiple versions of published quantum neural networks (QNNs), using the IBM Q library. Given that current QC hardware is limited in the number of quantum bits (qubits), we employ dimensionality reduction strategies: using a smaller set of genes known to be correlated with disease status; and using the top principal components of the full gene expression matrices. While the analysis was mostly done with QC simulators, we also run trained QNNs on a commercial quantum computer for validation. We further explore other aspects of QML: (1) representational differences relative to classical networks; and (2) biological interpretation. We compare the results of QNNs to classical NNs with a comparable number of parameters, using classification metrics. Additionally, we have designed a probabilistic classical NN whose architecture mimics that of the QNNs for a careful analysis of representational differences. Finally, we perform a saliency analysis to quantify the sensitivity of the best models to the input features, which are either genes or linear combinations thereof. We find (on PsychENCODE data) that QNNs can perform at an accuracy comparable to classical NNs, sometimes even improving upon the latter, with a certain degree of stochasticity in the model predictions. The representations are inherently different, and the saliency evaluation of features provides a more agnostic measure of shared biological information. We find differences in the saliency, with some of the QNN saliencies recapitulating the correlations of features with case/control status. Ongoing work is delving into the nature of these discrepancies.

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Poster

P963. Computational and Analysis Tools II

Location: Virtual Only

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Program #/Poster #: P963.03

Topic: I.06. Computation, Modeling, and Simulation

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Title: Spatiotemporal 3D image registration for mesoscale studies of brain development

Authors: *S. SHUVAEV[1], A. LAZUTKIN[2], R. KIRYANOV[3], K. ANOKHIN[4], G. ENIKOLOPOV[5], A. KOULAKOV[1];
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Abstract: Comparison of brain samples representing different developmental stages often necessitates registering the samples to common coordinates. Although the available software tools are successful in registering 3D images of adult brains, registration of perinatal brains remains challenging due to rapid growth-dependent morphological changes and variations in developmental pace between animals. To address these challenges, we propose a multi-step algorithm for the registration of perinatal brains. First, we optimized image preprocessing to increase the algorithm's sensitivity to mismatches in registered images. Second, we developed attention-gated simulated annealing (Monte Carlo) procedure capable of focusing on the differences between perinatal brains. Third, we applied classical multidimensional scaling (CMDS) to align ("synchronize") brain samples in time, accounting for individual development paces. We tested this multi-step algorithm on 28 samples of whole-mounted perinatal mouse brains (P0 - P9) and observed accurate registration results. Our computational pipeline offers a runtime of several minutes per brain on a personal computer and automates brain registration tasks including mapping brain data to atlases, comparison of averaged experimental groups, and monitoring brain development dynamics.

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Poster

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Topic: I.06. Computation, Modeling, and Simulation

Support: NSF Grant DBI2015317
NSF Grant DGE1745016
NSF Grant IOS1754869
NIH Grant R01 NS118606

Title: Modular neuromechanical model of Aplysia feeding behavior

Authors: *A. S. LIAO[1], R. SUKHNANDAN[1], Q. CHEN[2], B. KUNDU[3], J. P. GILL[2], G. SUTTON[3], P. J. THOMAS[2], V. A. WEBSTER-WOOD[1,4], H. J. CHIEL[2];
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Abstract: Animals exhibit behavioral flexibility in response to a dynamic environment. The coordination between the nervous system and periphery transforms sensory information into muscle activation to induce mechanical responses. The feeding behavior of the model organism *Aplysia californica* demonstrates this flexibility by switching between three behaviors - biting (an attempt to grasp an object), swallowing (grasping and ingesting food), rejection (egestion of inedible material) - based on mechanical and chemical stimuli. Understanding the neuromechanical principles underlying this behavioral flexibility is key to uncovering the mechanisms behind multifunctionality.

We developed an *Aplysia*-inspired neuromechanical model with a Boolean-like neural controller, first-order muscle models, and 1D biomechanical equations of motion. This model qualitatively demonstrated all three feeding behaviors and could flexibly switch between them depending on mechanical and chemical stimuli [1]. Furthermore, the simplified model components enabled the model to perform simulations faster than real time, but at the cost of physiological details in the components. Depending on the focus of the research application, components of the model may need to be altered to suit the level of complexity required.

Using a demand-driven complexity approach, we extended our original neuromechanical model to have three customizable modules: neural circuitry, muscles, and mechanics. This enabled us to qualitatively compare the simulation results between our original 1D mechanical model of the feeding apparatus and a 2D version as well as between our original first-order muscle model and a Hill-type muscle. With the new module versions, the model reproduced mechanical outputs that differed in the transient response but were qualitatively similar overall to that from the original model. However, the module replacement process was sensitive to model parameters and required hand tuning to match the original model's results.

Our modular modeling framework allows a demand-driven complexity approach to neuromechanical modeling of *Aplysia* feeding behavior. It is easily extensible, and each module can be replaced with alternative models that are more biologically realistic. We will explore the effect of incorporating more physiologically accurate neuron and muscle models on model output and quantitatively compare our modeled behavior to animal experimental data. Additionally, we are developing deep-neural-network-based algorithms for automatically tuning model parameters to match experimental data.

References: [1] Webster-Wood et al. *Biol Cybern*. 2020.

Disclosures: A.S. Liao: None. **R. Sukhnandan:** None. **Q. Chen:** None. **B. Kundu:** None. **J.P. Gill:** None. **G. Sutton:** None. **P.J. Thomas:** None. **V.A. Webster-Wood:** None. **H.J. Chiel:** None.

Poster

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Program #/Poster #: P963.05

Topic: I.06. Computation, Modeling, and Simulation

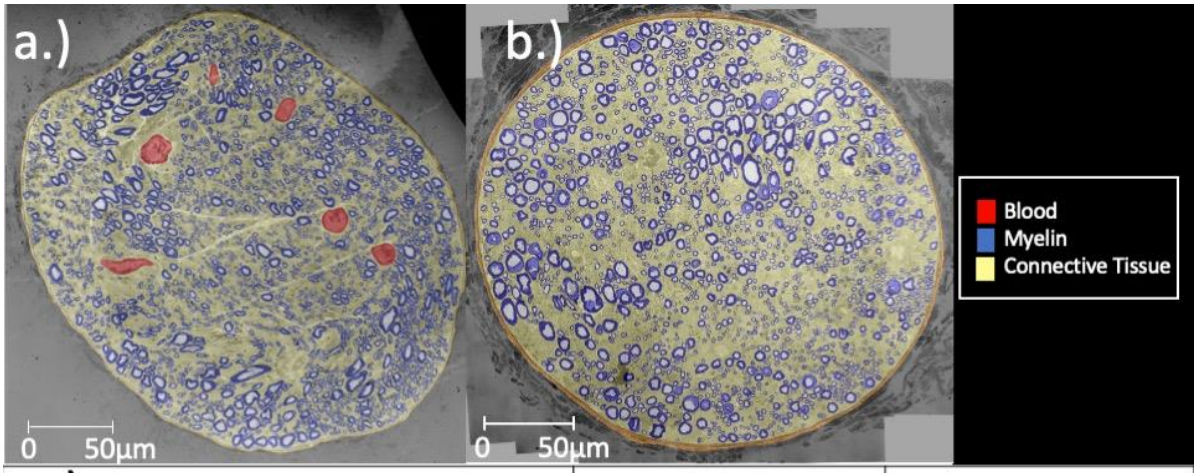
Support: NIH/NINDS grant R01NS120594

Title: Analysis of the Spatial Distribution of Myelinated Fibers in the Vagus Nerve for Spatially-Variant Targeted Neuromodulation

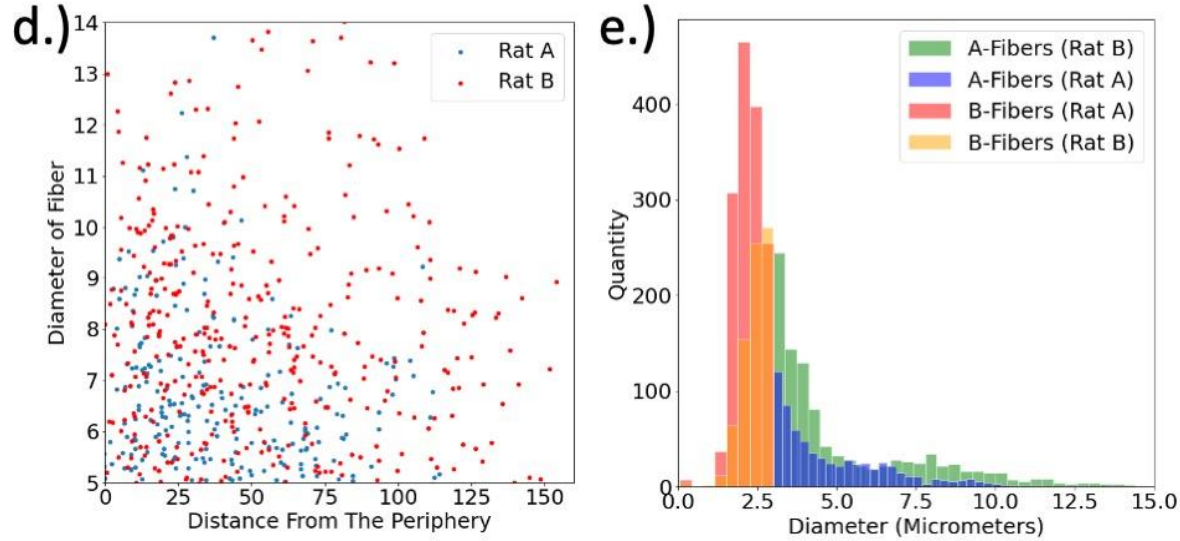
Authors: *A. CHO[1], H. JEONG[2], I. AY[3], G. BONMASSAR[4];
[1]Harvard Univ., Cambridge, MA; [2]Athinoula A. Martinos Center, MGH, Cambridge, MA; [3]Dept Radiol, Massachusetts Gen. Hosp., Boston, MA; [4]Harvard Med. Sch., Charlestown, MA

Abstract: Introduction: Vagus Nerve Stimulation (VNS) is an FDA-approved method for the therapeutic use of drug-resistant epilepsy, migraine, cluster headache, and depression. The vagus nerve contains both afferent and efferent fibers. Current VNS technologies are fiber non-selective: although the therapeutic effects of VNS are linked with the stimulation of the large afferent fibers, unintended stimulation of the efferent fibers results in adverse effects. This project aims to study the anatomy of the vagus nerve to develop new simulation technologies of the afferent A-fibers by modeling and characterizing the vagal fibers to minimize the current VNS side effects. **Methods:** Two cervical right vagus nerve samples of healthy adult male Wistar rats (350-450 g) were obtained and prepared on a Transmission Electron Microscopy (TEM) grid for imaging (HT7800, Hitachi, Japan). The images were preprocessed for noise removal, stitched, segmented, and morphed into a circle using MATLAB (MathWorks, MA, USA). The TEM images were adjusted for contrast for segmentation, masked based on B&W thresholds, partitioned, and manually cleaned up. ImageJ based on the color threshold was used to determine the diameters and locations of each fiber and blood vessel. A-fibers were defined as myelinated fibers with a diameter of 3 μm or greater, while B-fibers were determined to be myelinated fibers with a diameter of less than 3 μm **(1.c)**. Python and Prism8 (GraphPad, California, USA) software were used for quantitative distribution analysis. **Results and Discussion:** Rat 1's vagus nerve had an estimated diameter of 256.63 μm, whereas Rat 2's diameter was approximately 319.95 μm (Fig 1.c). 63.52% of Rat 1's A-fibers and 52.66% of Rat 2's A-fibers were located in the outer half of the nerve (Fig 1.d.e). **Conclusions:** The rat VNS segmentation allowed for the spatial characterization of the vagus nerve fibers and revealed a 2D

toroidal spatial distribution. This unique toroidal spatial distribution observed in our results dramatically simplifies the design of a focal and minimally invasive stimulator.



c.)	Rat A	Rat B
Nerve Area (µm²)	51,699.28	80358.72
Nerve diameter	256.63	319.95
Myelinated fiber number	2126	1795
Myelinated fiber density (fiber/mm²)	41122.43	22324.90
A-fiber number	658	1034
A-fiber density (fiber/mm²)	12727.45	12867.30



Disclosures: A. Cho: None. **H. Jeong:** None. **I. Ay:** None. **G. Bonmassar:** None.

Poster

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Program #/Poster #: P963.06

Topic: I.06. Computation, Modeling, and Simulation

Support: NIDILRR 90REGE0005-01

Title: Wearable Networks of Multijoint Elastic Elements for Upper-Extremity Neurorehabilitation

Authors: *P. RYALI[1], B. MALIZIA[1], Y. GHANI[1], D. MCDERMED[1], J. HE[1], C. CELIAN[2], J. L. PATTON[3];
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Abstract: Wearable robotic interfaces can not only make humans move better but can also help them train better -- offering an attractive modality for neurorehabilitation on post-stroke or spinal cord injured populations. Here, we present the advantages and demonstrate two neurorehabilitative applications of a passive orthosis, the ExoNET, which uses structural optimization with impedance (equilibrium point) control to tune moment arms and pretension properties of a network of elastic elements to satisfy desired upper-extremity torques.Tunable spring designs, such as the ExoNET, can not only provide assistive forces but also have the potential to mechanically amplify the errors made by the user. For example, destabilizing forces surrounding a reaching path can provide error augmentation, which has been shown to enhance motor recovery in brain-injured individuals. Another example is to reshape movement variability with an unstable limit push field that makes disturbance forces surrounding the boundary of a desired region. A crucial design variable is the number of impedance elements available at each joint, and there is a tradeoff where the system is more complex and bulky with more elements, but it is more accurate at approximation. Here we explore this variable. We measured the ExoNET's abilities to approximate upper extremity error augmentation and limit push forces for up to 5 elements at each joint. For error augmentation, errors averaged 4.120, 2.090, and 0.023 Nm for 1, 3, and 5 elements. For limit push, errors averaged 0.004, 0.002, and 0.001 Nm. Not only is it evident that accuracy increases with the number of elements, error augmentation fields require more elements, and they dramatically improved when 5 elements were used. Meanwhile, limit push may only require 1 element per joint. This work demonstrates the ExoNET's potential to perform complex and clinically significant tasks in assisting rehabilitation, and warrants further study.

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Poster

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Program #/Poster #: P963.07

Topic: I.06. Computation, Modeling, and Simulation

Support: NSF Grant 1552946
NIH Grant 1U19NS107464

Title: Sparse high-dimensional decomposition of non-primary cortical spectrotemporal receptive fields

Authors: *S. MUKHERJEE[1], B. BABADI[1,2], S. A. SHAMMA[1,2,3];
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Abstract: Characterizing neuronal responses to natural stimuli remains a central goal in sensory neuroscience. In auditory cortical neurons, the stimulus selectivity of elicited spiking activity is summarized by the neurons' spectrotemporal receptive fields (STRF). Canonically, the STRF can be inferred from spiking responses to a diverse range of auditory stimuli by reverse-correlation, assuming a linear relationship between spectrotemporal features of the stimuli and response. Alternatively, point process generalized linear models of stimulus-driven spiking can be applied to infer STRFs with greater predictive capacity.
Both approaches fundamentally seek to directly map the spectrotemporal features of auditory stimuli to neuronal responses. Though effective in characterizing primary auditory cortical responses, directly estimated STRFs of neurons in non-primary areas can be quite intricate, a reflection of mixed selectivity in neuronal responses. The complexity of non-primary STRFs poses challenges in interpreting the representation of spectrotemporal features in later stages of the auditory pathway.
To address this challenge using a point process model of neuronal spiking, we propose estimating receptive fields of non-primary auditory cortical neurons with respect to a high-dimensional model of the primary-cortical representation of auditory stimuli. Moreover, we employ a greedy estimation algorithm to capture the salient primary-cortical features that drive spiking responses. We additionally relate the resulting sparse cortical receptive fields to corresponding spectrotemporal features, and hence describe a biologically plausible hierarchical decomposition of non-primary STRFs. In application to simulated data, we validated that our proposed analysis can recover the cortical receptive field with statistical confidence. Additionally, in application to spiking activity of neurons in the ferret posterior ectosylvian gyrus, our proposed method yields highly predictive models of neuronal responses to both artificial and natural stimuli.

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Poster

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NSF DBI-2014862
Office of Naval Research N00014-20-1-2469
Luxembourg National Research Fund No.9984574

Title: Morphological principles of neuronal mitochondria

Authors: *G. C. GARCIA[1], R. MENDELSOHN[1], T. M. BARTOL, JR[1], C. LEE[2], P. KHANDELWAL[1], E. LIU[1], D. SPENCER[1], A. HUSAR[1], E. A. BUSHONG[3], S. PHAN[3], G. PERKINS[3], M. H. ELLISMAN[3], A. SKUPIN[4], T. J. SEJNOWSKI[1], P. RANGAMANI[2]; [1]Salk Inst., La Jolla, CA; [2]Mechanical and Aerospace Engin., [3]Ctr. for Res. in Biol. Systems, Univ. of California San Diego, La Jolla, CA; [4]Luxembourg Ctr. for Systems Biomedicine, Univ. of Luxembourg, Belval, Luxembourg

Abstract: In the dynamic metabolic landscape of a neuron, mitochondrial membrane morphology can provide critical insight into the energetic state of the cell. Theoretical calculations of functional outputs like ATP often represent mitochondria as idealized geometries and therefore can miscalculate the metabolic fluxes. To analyze mitochondrial architecture in neurons of mouse cerebellum neuropil, 3D reconstructions of synaptic and axonal mitochondria were generated using serial transmission electron microscope tomography images. We created high quality meshes from manual tracings of the membranes using new procedures in 3D computer graphics modeling software in order to produce minimal distortion from the original volumes with a granularity of 1.6 nanometer isotropic voxels. The resulting *in silico* representations were subsequently quantified by differential geometry methods in terms of the mean and Gaussian curvatures, surface areas, volumes, and membrane motifs, all of which can alter the metabolic output of the organelle. Finally, we identify structural motifs that are present across this population of mitochondria. These anatomical reconstructions are highly accurate and lay the foundation for future studies of the internal mitochondrial physiology and metabolism in neurons using Monte-Carlo techniques to simulate the biochemical interactions that takes place in the mitochondrial compartments.

Disclosures: G.C. Garcia: None. **R. Mendelsohn:** None. **T.M. Bartol:** None. **C. Lee:** None. **P. Khandelwal:** None. **E. Liu:** None. **D. Spencer:** None. **A. Husar:** None. **E.A. Bushong:**

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Poster

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P963.09

Topic: I.06. Computation, Modeling, and Simulation

Support: Department of Biotechnology (DBT), Ministry of Science and Technology, Government of India (BIO/17-18/303/DBTX/SRIN)

Title: A computational model to simulate vascular arborization based on neuron cytoarchitecture

Authors: *B. S. KUMAR, S. M. CHANDRAMOHANAN, S. G. RAJAPPA, V. S. CHAKRAVARTHY;
Indian Inst. of Technol. Madras, Chennai, India

Abstract: The electrical activity of the neurons in the brain is highly dependent upon timely and adequate blood supply from the cerebral vascular network. Since cerebral blood flow in turn is dependent on neural activity, it is understood that there is a bidirectional interaction between the neural and the cerebrovascular system. Therefore, there is an anticipation, supported by experimental evidence, that even the development of both neural and cerebrovascular systems should be interdependent. To understand the effect of activity and density distribution of the neurons on the development of vascular arborization, we propose a computational model known as the Vascular Arborization Model (VAM). We considered four segments of tissue volume from the murine cortex, wherein each segment has the dimensions: 100μm X 100μm X 800μm. The neurons and non-neural cells were distributed randomly in the volume, constrained by experimental data. The vascular network originates from a parent root node and grows to span the tissue volume based on the cytoarchitecture of the neural and non-neural cells of the cortex. There are two stages of vascular arborization: (i) Growth phase and (ii) Settling phase. In the growth phase, each terminal vessel grows closer to the neurons in its domain such that the mean squared distance (MSE) between the vascular terminal node and the neurons in its perfusion domain is minimized. When the MSE reaches a minimum, the vessel splits into two smaller vessels. The former perfusion domain is also split into two sub-domains, while the two child vessels proceed to perfuse the two newly formed subdomains. This process occurring iteratively results in vascular arborization. During the settling phase, the fully grown vascular network is rearranged to ensure optimal path length and wire length. The simulated vascular network grown from a single root node to a vast network could approximate the biological vascular network reasonably well. The microvascular volume density and the total vascular volume density estimated from the model were in close agreement with the experimental studies. The mean

Euclidean distance between vessels and neurons in the model was close to 14μm, the biologically observed value. The model also shows that the neural densities do not covary with the vascular density along the depth of the cortex, but predicted a high correlation between neural areal density and microvascular density when compared over a global scale (across animals and regions). Moreover, the range of estimated vessel radii that resulted in this vascular volume density agreed with the range of arteriolar diameter observed experimentally in rats and mice.

Disclosures: B.S. Kumar: None. **S.M. Chandramohanan:** None. **S.G. Rajappa:** None. **V.S. Chakravarthy:** None.

Poster

P963. Computational and Analysis Tools II

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Program #/Poster #: P963.10

Topic: I.06. Computation, Modeling, and Simulation

Support: NIH Grant R01-NS081118

Title: Quantitative methods to evaluate motor thalamic deep brain stimulation-induced gait ataxia with computer vision

Authors: *D. C. P. MARTICORENA, M. BLUMENFELD, A. BRINDA, B. REBECCA, J. D. KRIEG, L. SCHROCK, S. E. COOPER, J. MATSUMOTO, M. D. JOHNSON;
Univ. of Minnesota - Twin Cities, Minneapolis, MN

Abstract: Thalamic deep brain stimulation (DBS) is often used to decrease motor manifestations of Essential Tremor, but can be impeded by stimulation-induced gait ataxia. To further understand the neurophysiology of DBS-induced gait ataxia and avoid its emergence with DBS therapy, it is important to have methods that can quantify features of gait ataxia in a subject-specific manner with granularity beyond current clinical rating scales. In this study, we developed a markerless pose estimation approach to reconstruct a subject’s pose in real space for complex gait analysis during a DBS programming session. The apparatus includes a triad stereo camera system which captures heel-to-toe ambulation toward and away from the system. The collection methods involved activating the cameras (30 fps, 2160p) to collect ambulation videos for a given evaluation. Within the 2D videos taken from each camera, the subject's shoulders, elbows, wrists, hips, knees, and ankles were labeled using DeepLabCut, a markerless pose estimation tool. Given potential error derived from spurious labels attributable to loose clothing and occluded joints, a 10-sample median-difference-threshold filter was used to create a probability distribution of individual labels based on temporal consistency, identify low-probability labeled joints j (σj > 5), and eliminate them. Empty entries were refitted with a piecewise cubic Hermite interpolating polynomial. The resulting vectors were then smoothed

with a 3-point moving average filter. The camera intrinsics (focal point, FOV, etc.) and extrinsics (orientation with respect to one another) were calibrated on the basis of eight images of a distinctly-oriented checkerboard to minimize reprojection errors using linear least-squares, with average pixel reprojection error less than 0.4 pixels. To capture the real-world coordinates of the subject's joints, the filtered two-dimensional joint coordinate matrices were triangulated using a linear least-squares triangulation algorithm. These results serve as an initial estimate for the non-linear least squares optimization triangulation, which factors in camera extrinsics, joint depth, temporal consistency, and anthropomorphic constraints, which fits joint estimates onto a consistent model of the subject. This optimization greatly reduced error in pose reconstruction, and therefore increased the accuracy in related gait analysis. This low-cost, objective, and granular gait assessment method is poised to provide insight into subject-specific DBS optimization efforts to better elucidate the neural pathways involved in the induction of gait ataxia with DBS

Disclosures: D.C.P. Marticorena: None. **M. Blumenfeld:** None. **A. Brinda:** None. **B. Rebecca:** None. **J.D. Krieg:** None. **L. Schrock:** None. **S.E. Cooper:** None. **J. Matsumoto:** None. **M.D. Johnson:** None.

Poster

P979. Electrical Methods to Modulate Neural Activity III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P979.01

Topic: I.08. Methods to Modulate Neural Activity

Support: KAKENHI 19H04437
KAKENHI 20H04498
KAKENHI 20J22902

Title: Development and time interval learning of hippocampal-cortical co-culture model in vitro

Authors: *C. CHANG, K. SHIMBA, T. ASAHINA, T. FURUKAWA, K. KOTANI, Y. JIMBO;
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Abstract: Phenomenon and connection in brains have long been in interest of neuroscientists. In vitro cultural networks, consists of randomly connected neurons, have long been used to investigate basic properties of neuronal networks, and have provided insights about network level information processing and memory formation. In vitro studies have shown abilities of in vitro neuronal networks performing pattern separation, blind source separation, spatiotemporal memory and temporal interval learning by dissociated neuronal cultures. Dissociated neuronal cultures consisted of randomly connected neurons evenly spread over cultured area. Artificial

information can be given by electrical stimulation, optical stimulation or both. Studies have report modification of network activity for several seconds to minutes, shown by modification in network burst activities or spike events on recorded electrodes pairs. However, in brains, information and messages are transferred over layers of networks and processed across several structures. Considering above situation, in this study, we took advantage of microfluidic technologies and develop an in vitro model to separate neurons derived from separate brain structures, cortex and hippocampus. Specialized connection between cortical and hippocampal structures have been well-known for long-term memory formation, consolidation and memory recall. Cortical and hippocampal cells were plated into two different cell chambers to keep cell properties of the original structure. Communication between separated networks were performed by microstructures to allow axonal passage but restrain somatic entrance, forming a heterogenous network model to study memory formation by external stimulation in vitro. Hippocampal and cortical co-cultures showed communication between networks by sharing similar starting time of burst activities, on the other hand, hippocampal and cortical bursts showed different rhythms of local burst activities, suggesting localized network functions remained in each network. Memory formation by external stimulation was tested by time interval learning between cortical and hippocampus culture. Time interval training was designed as giving series of electrical stimuli first in cortical cultures, followed by another series of stimuli in hippocampal cultures. After training, direct evoked response by test stimulation in cortical culture showed significantly shortened duration of firing activity, and lasted for over 20 mins, suggesting the possibility of time interval learning to modify cortical-hippocampal connection by modifying network activity in cortical culture.

Disclosures: C. Chang: None. **K. Shimba:** None. **T. Asahina:** None. **T. Furukawa:** None. **K. Kotani:** None. **Y. Jimbo:** None.

Poster

P979. Electrical Methods to Modulate Neural Activity III

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P979.02

Topic: I.08. Methods to Modulate Neural Activity

Support: Howard Hughes Medical Institute (HHMI)-Wellcome International Scholar Awards from the Wellcome Trust (208468/Z/17/Z)
Basic Science Research Program through the National Research Foundation of Korea (NRF) (2020R1A2C2007285)
Samsung Science and Technology Foundation under Project Number SSTF-BA1602-11

Title: Stimulation-pattern dependence in the activation of Arc mRNA in live neurons

Authors: *D. KIM, H. MOON, B. LEE, J. SHIM, H. PARK;
Dept. of Physics and Astronomy, Seoul Natl. Univ., Seoul, Korea, Republic of

Abstract: Immediate early genes (IEGs) are genes that are rapidly induced by neural activities. The expression of IEGs such as Arc, c-fos, and Egr1 has been used to investigate the relation among external stimuli, neural activities, and memory formation. Inside cells, there exist signaling pathways where the chain reaction of enzymes leads to synaptic plasticity including gene expression and regulation. For electrically stimulated neurons, specific patterns are known to induce synaptic plasticity. But the relation between various stimulation patterns and gene expression in live cells is still in question. In this research, we investigated the effect of electrical stimulation on the expression of Arc mRNA in hippocampal neurons cultured from the Arc-PBS x PCP-GFP mice. We found that there are correlations among the pattern of the stimulation, calcium ion influx, phosphorylation of transcriptional factor CREB, and gene expression. Neurons stimulated by burst stimulation showed the highest Arc transcription rate and calcium influx at theta frequency. And Arc-positive neurons showed higher CREB phosphorylation levels than Arc-negative neurons.

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Poster

P979. Electrical Methods to Modulate Neural Activity III

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Program #/Poster #: P979.03

Topic: I.08. Methods to Modulate Neural Activity

Support: Loyola Research Program Support

Title: Neural inhibition with a miniature magnetic coil

Authors: *H. YE, L. BARRETT;
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Abstract: Numerous neurological dysfunctions are characterized by undesirable nerve activity. By providing reversible neural blockage, electric stimulation with implanted electrode holds promises in the treatment of these conditions. Externally applied electric currents have been used in inhibiting axon conductance and ganglion cell activities in clinical practices. However, there are significant limitations to its application, including poor bio-compatibility and decreased efficacy during chronic implantation. Magnetic coil with miniature size can mitigate these problems, by coating it with biocompatible material for chronic implantation. Via electromagnetic induction, the miniature magnetic coil could induce local electric field for neural stimulation. However, it is unknown if miniature coils could be effective in blocking neural

activity and if so, what are the underlying mechanisms. Here we report that a submillimeter magnetic coil could reversibly blocked action potentials in the unmyelinated axons from marine mollusk *Aplysia californica*. Biophysical modeling confirmed that the miniature coil induced a sufficient electric field in the vicinity of the targeted axon. Using a multi-compartment model of *Aplysia* axon, we found that the miniature coil caused a significant local depolarization in the axon, altered activation dynamics of the sodium channels, and prevented the traveling of the invading action potentials. We also report that the miniature coil could reversibly block ganglion cell activity, regardless the firing frequency of the neurons, or the presence of the ganglion sheath. The inhibitory effect was local to the soma, but was sufficient in blocking the neuron's functional output. Using a multi-compartment model of *Aplysia* ganglion neuron, we found that the high-frequency magnetic stimuli altered the ion channel dynamics that were essential for the sustained firing of action potentials in the soma. Results from this study produce several clinical insights to further develop the miniature coil technology for neural control by targeting axons or ganglion cells. With improved biocompatibility and capability of emitting high-frequency stimuli, micro coil provides an appearing alternative for electric blockage of neural activity.

Disclosures: H. Ye: None. **L. Barrett:** None.

Poster

P979. Electrical Methods to Modulate Neural Activity III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P979.04

Topic: I.08. Methods to Modulate Neural Activity

Support: NSF EAGER 1833288
NIBIB U01 EB025830
NIH 1R21EY028744

Title: On the design of electrical stimulus parameters for selective activation of retinal ganglion cells

Authors: *J. PAKNAHAD[1], M. HUMAYUN[2], G. LAZZI[3];
[2]Biomed. Engin. and Ophthalmology, [3]Electrical Engineering, Biomed. Engin. and Ophthalmology, [1]USC, Los Angeles, CA

Abstract: Retinal degenerative diseases such as retinitis pigmentosa (RP) and age-related macular degeneration (AMD) first affect the outermost layer of the retina, photoreceptors, and eventually lead to the damage of the innermost layer of the retina, consisting of retinal ganglion cells (RGCs). Retinal prosthetic systems have been developed with a tremendous influence on the life of patients suffering from the blindness for decades. Epiretinal implants aim at electrically stimulating targeted surviving retinal neurons and particularly RGCs with a relatively

longer functionality compared to outer retinal neurons such as bipolar cells (BCs) and photoreceptors. Several electrical stimulation strategies have been proposed and developed to augment the efficacy of current epiretinal prosthetic devices. However, there are still many challenges to be addressed such as simultaneous activations of various RGCs subtypes in response to electrical stimulation. The ability to selectively target subtypes of RGCs that carry particular visual information such as color and contrast would significantly enhance the performance of retinal prosthetic systems. In this study, we applied the 3D Admittance Method (AM)-NEURON computational platform to study the frequency response (up to 200 Hz) of the developed RGCs models, A2-monostratified and D1-bistratified, to a range of symmetric and asymmetric charge-balanced stimulus pulses. We further analyzed the response of RGCs as a function of modulations in charge amplitude over a range of stimulus pulses at 200 Hz. Our results indicate that D1 cells are more responsive to high stimulus frequency using symmetric, asymmetric short cathodic-first, and asymmetric long anodic-first biphasic pulses relative to A2 cells. Interestingly, asymmetric long cathodic-first stimulus pulses alter the excitability of RGCs such that A2 cells can better follow high rates of firing at high frequency compared to D1 cells. While the differential biophysical properties slightly changed the response of the cells, we found the soma size to be a predominant factor leading to this differential response of RGCs at high frequency across stimulus pulses. The differential sensitivity of stimulus strength-dependent response of RGCs at high frequency to various stimulus waveforms offers a great opportunity for selective activation of individual RGCs. This enables the possibility of encoding additional visual information such as color in future retinal prosthetics systems.

Disclosures: J. Paknahad: None. **M. Humayun:** None. **G. Lazzi:** None.

Poster

P979. Electrical Methods to Modulate Neural Activity III

Location: Virtual Only

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Program #/Poster #: P979.05

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH NEI 1R21EY028744-01

Title: Evaluating the Effect of Retinal Degeneration on Response of ON and OFF Cone Bipolar Cells to Extracellular Electrical Stimulation: A Computational Study

Authors: *S. FARZAD, J. PAKNAHAD, P. KOSTA, E. ISERI, G. LAZZI;
USC, Los Angeles, CA

Abstract: Bipolar cells (BCs) play critical roles in the visual signal processing through retinal layers by transmitting the signals from photoreceptors to ganglion cells (GCs). In retinal degenerative diseases, such as retinitis pigmentosa (RP), first photoreceptors start to degenerate,

followed by neuronal remodeling and topological alterations, which finally leads to loss of inner neurons such as BCs and GCs. Due to the loss of photoreceptors, the remaining cells go through intensive functional and morphological adjustments. Computational modeling of retinal cell response to electrical stimulation allows us to design stimulation strategies and improve the performance of current retinal prosthetic devices. Previous works either utilize single compartment-based simplified neuron models or use morphology of healthy retinal cells. With the aid of retinal connectomics databases of a healthy and an early stage degenerated retina, we can accurately model cells and compare their responses to electrical stimulation of varying stimulus parameters. Retinal connectomes are built by slicing the rabbit retina tissue in 70nm-thick slices, manually annotating different cell types, and generating 3D morphologies of retinal cells by connecting the annotations across all the slices. This way, the connectome databases provide an accurate and realistic of the cell morphologies, synapse information, and network topology details. Using the Admittance method (AM)-NEURON multi-scale modeling platform, we aim at analyzing the impact of retinal degeneration on neuronal activities induced by electrical stimulation. Leveraging this computational model, we compared the response generated in the healthy and degenerated ON-BCs morphologies due to electrical stimulation. Interestingly, we found that the degenerated ON-BCs have slower time constants compared to the healthy ON-BCs at the terminals. This may indicate the decreased average peak membrane potentials and therefore the less likelihood of neurotransmitter release at the presynaptic terminals of the degenerated cells relative to the healthy cells using short stimulation pulses. Characterizing the influence of retinal degeneration on BCs' response to electrical stimulation would allow us to effectively design stimulation strategies and enhance the efficacy of current retinal prosthetic systems.

Disclosures: S. Farzad: None. **J. Paknahad:** None. **P. Kosta:** None. **E. Iseri:** None. **G. Lazzi:** None.

Poster

P979. Electrical Methods to Modulate Neural Activity III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P979.06

Topic: B.06. Synaptic Transmission

Title: Optogenetic modulation of electrical coupling

Authors: *V. LERNER, M. LONDON, Y. YAROM;
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Abstract: Theoretical studies show that the number of neurons in an electrically coupled network can be experimentally estimated by comparing the membrane potential decay in a single neuron in two conditions: (1) following a brief stimulation to this neuron and (2) when the

stimulation is applied to all the electrically coupled neurons. The use of optogenetics may provide the experimental protocol to examine this prediction. Surprisingly, in cortical PV interneurons, we found that the voltage decay is identical in both conditions. We hypothesize that this results from temporary decoupling of the electrical connections between the neurons due to activation of Channelrhodopsin-2 (ChR2). We further examined this possibility on the electrically coupled network of the inferior olive, where subthreshold voltage oscillations occur when cells are electrically coupled. Indeed, during light activation of ChR2, expressed in inferior olive neurons, the subthreshold oscillations were eliminated, indicating a reduction in gap junction conductivity. Since it is well documented that gap-junction conductance is pH sensitive and that activation of ChR2 affects the pH, we suspect that this mechanism reduces coupling in the network. This finding may have implications on the interpretation of many experiments which use ChR2 to manipulate the activity of neurons that are electrically coupled.

Disclosures: V. Lerner: None. **M. London:** None. **Y. Yarom:** None.

Poster

P979. Electrical Methods to Modulate Neural Activity III

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Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P979.07

Topic: I.08. Methods to Modulate Neural Activity

Support: Interdisciplinary Pain Research Program

Title: Frequency dependent electrical stimulation of the prefrontal and anterior cingulate cortices provides pain modulation in rats

Authors: *H. F. XU, Y. LIU, B. VEMULAPALLI, H. J. JEE, Q. ZHANG, J. WANG;
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Abstract: As pain consists of both sensory and affective components, its treatment solely by pharmaceutical methods, such as opioids, remains difficult. While electrical stimulation is a promising alternative to pharmaceutical treatment, greater exploration into its targets and parameters of use, particularly its use in the brain, is required to support clinical application. We studied how electrical stimulation of the prelimbic-prefrontal cortex (PL-PFC) and anterior cingulate cortex (ACC) at different frequencies affects pain modulation within these two cortical regions in awake, freely-moving rats. In male Sprague-Dawley rats, we implanted stimulating electrodes in the PL-PFC, a region that provides top-down regulation of pain, and the ACC, a region that processes pain affect. To evaluate the effects of electrical stimulation frequency on prefrontal control of acute pain, we conducted a Hargreaves test by providing acute thermal stimulation to the plantar surface of the hind paw of a rat. We simultaneously administered electrical stimulation at a range of frequencies (0, 20, 40, 60, 80, 120 Hz) to the PL-PFC,

contralateral to the paw receiving thermal stimulation and recorded the paw withdrawal time as a measure of nociceptive pain response. We used the conditioned place aversion (CPA) assay in a standard two chamber setup to determine the rats' aversive response under stimulation of the PL-PFC and ACC at low (20 Hz) and high (120 Hz) frequencies. We found that lower frequency stimulations (20, 40, 60 Hz) of the PL-PFC prolong the withdrawal latency in the Hargreaves test, whereas high frequency stimulation (120 Hz) decreases withdrawal latency. Meanwhile, electrical stimulation of the PL-PFC decreased aversive response at low frequencies (20 Hz) and increased it at high frequencies (120 Hz). An opposite trend was observed in the ACC, as low-frequency stimulation increased aversive response, whereas high-frequency stimulation decreased aversive response. These results demonstrate that electrical stimulation can be used to modulate the sensory and aversive components of pain, and furthermore, such modulation is dependent on both brain targets and on stimulation frequencies.

Disclosures: H.F. Xu: None. **Y. Liu:** None. **B. Vemulapalli:** None. **H.J. Jee:** None. **Q. Zhang:** None. **J. Wang:** None.

Poster

P979. Electrical Methods to Modulate Neural Activity III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:15 AM - 10:15 AM

Program #/Poster #: P979.08

Topic: I.08. Methods to Modulate Neural Activity

Title: Treatment efficacy and technical advantages of ipsilateral temporary spinal nerve root stimulation for unilateral postherpetic neuralgia

Authors: M. HUANG[1], Q. CHEN[2], S. WU[1], J. HUANG[1], W. SUN[1], S. YANG[1], ***T. NGUYEN**[2], X. QIAN[2], L. XIAO[1];
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Abstract: Objectives: Post-herpetic neuralgia (PHN) is a common complication after herpes zoster infection (HZ). Whilst temporary spinal cord stimulation (tSCS) has been shown as an effective treatment for drug-resistant PHN, recent data suggests ipsilateral temporary spinal nerve root stimulation (tSNRS), a relatively newer technique, as a safe and viable alternative for treating PHN affecting unilateral dermatomes. However, there is no direct comparison on the treatment efficacy of tSNRS to the traditional tSCS. The current retrospective study aimed to describe the technical parameters and the therapeutic effects of tSNR for patients with unilateral PHN, and to compare them with those treated with tSCS.
Materials and Methods: 160 patients with unilateral PHN treated with tSCS (n=109) or tSNRS (n=51) were included in this single-center study. Technical factors between two groups, such as

procedure time, radiation dosage, number of electrodes used, and average cost were compared. Treatment efficacy, measured by analgesic coverage, pain visual analog scale (VAS), total analgesic agent consumption, Pittsburgh sleep quality index (PSQI), and physical and mental quality of life, was also compared between two groups at baseline, post-procedure, and 3 months after stimulation treatment.
Results: Patients who underwent tSNRS reported significant improvement in pain level, sleep quality, and overall quality of life immediately post-procedure and at the 3-month follow-up. This effect was comparable to the tSCS group. However, tSNRS was able to achieve this therapeutic effect with less stimulation sessions than tSCS. The overall cost, procedure time, number of implanted electrodes, and intraoperative radiation exposure were also significantly less in the tSNRS group than those received tSCS.
Conclusions: Whilst tSNRS provides similar therapeutic effect comparing to tSCS for patients with unilateral PHN, it offers several technical advantages. These advantages include shorter procedure time, less radiation exposure, less implanted electrodes, more effective stimulation, and lower overall cost.

Disclosures: M. Huang: None. **Q. Chen:** None. **S. Wu:** None. **J. Huang:** None. **W. Sun:** None. **S. Yang:** None. **T. Nguyen:** None. **X. Qian:** None. **L. Xiao:** None.

Poster

P979. Electrical Methods to Modulate Neural Activity III

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Program #/Poster #: P979.09

Topic: I.08. Methods to Modulate Neural Activity

Support: National Eye Institute Award 1R21EY030710-01
NSF Award DMR-1905575 and CBET-1752274
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Title: A Soft, Injectable Neuroelectronic Probe

Authors: *M. HILL[1], K. SEO[5], J. RYU[5], C. CHIANG[2], I. RACHINSKIY[1], Y. QIANG[6], M. TRUMPIS[3], C. WANG[4], J. VIVENTI[7], H. FANG[6];
[2]Biomed. Engin., [1]Duke Univ., Durham, NC; [3]Biomed. Engin., Duke Univ., Oakland, CA; [4]BME, Duke Univ., Durham, NC; [5]Thayer Sch. of Engineering, Dartmouth Col., Hanover, NH; [6]Northeastern Univ., Boston, MA; [7]Electrical and Computer Engin., Duke Univ. Dept. of Electrical and Computer Engin., Durham, NC

Abstract: Techniques to study brain activities have evolved dramatically, yet tremendous challenges remain in acquiring high-throughput electrophysiological recordings minimally

invasively. Here, we develop a soft neuroelectronic probe that is both filamentary and actively multiplexed. Specifically, onsite active multiplexing using miniaturized flexible silicon transistors achieves 256 neuroelectrodes on a filamentary substrate in only a 2.3×0.3 mm^2 area. A novel single-transistor multiplexing acquisition circuit further reduces noise from the electrodes. The filamentary form factor integrated with a rollable contact pad design allows the entire device to be also injectable through syringe injection, enabling minimally invasive probe delivery. Acute auditory experiments in rats successfully validated the ability of the array to record neural signals with high accuracy of tone decoding. Together, these results establish soft, injectable neuroelectronic probes as promising devices for neuroscience research and clinical applications.

Disclosures: M. Hill: None. **K. Seo:** None. **J. Ryu:** None. **C. Chiang:** None. **I. Rachinskiy:** None. **Y. Qiang:** None. **M. Trumpis:** None. **C. Wang:** None. **J. Viventi:** None. **H. Fang:** None.

Poster

P020. Regeneration: CNS

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P020.01

Topic: A.04. Transplantation and Regeneration

Support: NIH-NEI grant R00EY027467
Carthage College

Title: Learning to See Again: Using the optokinetic response to assess functional recovery after optic nerve injury

Authors: A. HERMANS, A. GORMAN, S. TAJNAI, ***S. HENLE**;
Carthage Col., Kenosha, WI

Abstract: The zebrafish (Danio rerio) eye has many anatomical similarities to the human eye but is unique in its ability to regenerate the optic nerve. Optic nerve regeneration is typically measured with histology. To better understand the functional regeneration of the optic nerve we investigated the optokinetic response (OKR) assays. The assay relies on the conserved ability to track movement across the whole field of vision in order to keep the visual acuity of the moving object. The response is unconditioned and reproducible, allowing it to be used for a visual assay. To perform an OKR assay, a fish is placed in a 3D printed drum with bars pulled by a motor controlled by an Arduino to elicit the OKR, a more cost-effective alternative to previous methods. Measuring changes in ocular angles recorded during the OKR assay, the zebrafish's visual function can be quantified. Previously, the ocular angles for adult zebrafish had to be manually measured in FIJI and then run through an R program to create a graph. We have also developed an automated imaging pipeline to analyze OKR in adult fish to speed up the process

and allowed for more fine-tuned OKR measurements over time. This will allow us to both better understand the process of optic nerve regeneration, and provide a new assay for measuring optic nerve regeneration that does not require sacrificing the animal.

Disclosures: A. Hermans: None. **A. Gorman:** None. **S. Tajnai:** None. **S. Henle:** None.

Poster

P020. Regeneration: CNS

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P020.02

Topic: A.04. Transplantation and Regeneration

Support: RPB-CDA
GRF-CFC3
NIH R01EY030138

Title: Osteopontin drives retinal ganglion cell resiliency in glaucomatous optic neuropathy

Authors: *K.-I. TOMA[1], M. ZHAO[1], B. KINDE[1], C. TAN[1], Y. HU[2], Y. HAN[1], X. DUAN[1]; [1]Univ. of California San Francisco, San Francisco, CA; [2]Dept. of Ophthalmology, Stanford Univ. Sch. of Med., Palo Alto, CA

Abstract: Retinal ganglion cells (RGCs) project from the retina to the central nervous system via the optic nerve, are representing the earliest step in visual processing. Damage to the RGCs or their axons in the optic nerve can lead to permanent vision loss. We and others have demonstrated that RGCs are not uniformly susceptible to injury and that certain RGCs appear to be more resistant to cellular death for acute injury. These studies showed that both the alpha-RGCs and the intrinsically photosensitive-RGCs expressing melanopsin (M1-RGCs) are more resistant in the ONC model and more chronic forms of injury. We further have previously demonstrated that alpha-RGCs preferentially express osteopontin (OPN, also named SPP1). It is critical for conveying increased resiliency and regeneration in combination with growth factors under ONC. However, the RGC subtype-specific survival has been explored to a lesser degree in mouse models of chronic injuries. Here, we explored RGC subtypes and subclasses with differential survival responses to elevated intraocular pressure (IOP). Furthermore, we assessed if similar mechanisms may protect certain RGC subtypes from chronic injury as occurs in glaucomatous disease states compared to acute injury in ONC models. For this, we employed two complementary models, the microbead occlusion model, and the silicone oil-induced ocular hypertension model, to induce and sustain elevated IOP to recapitulate the debilitating glaucomatous disease states that affect humans. We found that chronic ocular hypertension leads to significant loss of RGCs, with preferential survival of alpha-RGCs, and more specifically, survival of the alpha-ON-RGCs. We found that the OPN that is expressed in alpha-RGCs, is

critical for this resiliency as knockdown of OPN in alpha-RGCs abolishes this preferential survival, and over-expression of OPN in non-alpha-RGCs induces survival. We provide mechanistic insight into the molecular players that specifically drive alpha-RGC survival in the context of chronic glaucomatous injury. Finally, we demonstrated that OPN is expressed, and its expression is correlated with disease severity in the human retina. Our study demonstrated the novel role of OPN in promoting RGC resiliency in a mouse experimental glaucoma model. This work highlights how intrinsic properties of different RGC subsets equip them with differential survival abilities, which may be exploited to prevent premature and permanent vision loss in glaucoma.

Disclosures: K. Toma: None. **M. Zhao:** None. **B. Kinde:** None. **C. Tan:** None. **Y. Hu:** None. **Y. Han:** None. **X. Duan:** None.

Poster

P020. Regeneration: CNS

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P020.03

Topic: A.04. Transplantation and Regeneration

Support: NSF #1652312

Title: The planar cell polarity pathway is required to terminate adult neurogenesis in-vivo

Authors: *S. J. HACK, W. S. BEANE;
Biol. Sci., Western Michigan Univ., Kalamazoo, MI

Abstract: Understanding how to initiate the regrowth of axons and other neural structures following injury is a central goal of regenerative medicine. However, successful implementation of translational therapies will require more than how to initiate neurogenesis *in-vivo*. Understanding how neuronal growth self-terminates will be equally critical, as small alterations in growth regulation can easily result in carcinogenesis. To date, few studies have investigated the elusive process by which adult neurogenesis ceases. One of the few model systems where this is being investigated is planarians, which are flatworms capable of regenerating their entire CNS (including brain). Our previous work shows the planar cell polarity (PCP) pathway plays a key role in restricting or stopping neurogenesis during planarian regeneration. Specifically, loss of PCP signaling results in the formation of ectopic photoreceptor neurons and hyperplasia throughout the entire nervous system, concurrent with elevated levels of mitosis weeks after new growth is normally considered finished. In this study, we sought to determine which neural sub-populations and adult stem cell (ASC) subtypes require PCP signaling to terminate their growth. The PCP pathway has long been implicated in regulating cell size, proliferation, cell migration, and tissue patterning, and is a known human tumor suppressor gene associated with several

cancers. Our data suggest that PCP loss has variable effects on different neuronal subtypes, with some populations not being affected, some increased, and some decreased. For example, pan-neural and photoreceptor neuron markers reveal significant increases in the number of both neurons and neural synapses, while markers specific for chemosensory and GABAergic neurons show no increase and dopaminergic neuron numbers are decreased. Ongoing investigations are characterizing the requirement for PCP in specific neural populations and neural ASCs. Because very little is known about the mechanisms governing termination of neurogenesis, these investigations will give insights into how tissues 'know' when they have been properly patterned and scaled. Furthermore, understanding how neuronal growth stops will be crucial for translating future *in-vivo* therapies. This work was supported by NSF grant #1652312.

Disclosures: S.J. Hack: None. **W.S. Beane:** None.

Poster

P020. Regeneration: CNS

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Program #/Poster #: P020.04

Topic: A.04. Transplantation and Regeneration

Support: R01 EY022030
U01 EY027267
T32 NS105864

Title: Inhibition of NFkB signaling promotes neuron regeneration in the mammalian retina

Authors: *I. PALAZZO[1], L. TODD[2], S. BLACKSHAW[3], A. J. FISCHER[1];
[1]Neurosci., The Ohio State Univ. Neurosci. Grad. Program, Columbus, OH; [2]Biol. Structure, Univ. of Washington, Seattle, WA; [3]Johns Hopkins Univ, Sch. Med., Baltimore, MD

Abstract: Loss of vision is caused by irreversible death of retinal neurons. Müller glia (MG) are the primary support cell of the retina and are capable of reprogramming into progenitor cells with potential to regenerate neurons following retinal damage. The capacity for MG mediated retinal neuron regeneration varies widely across species. In lower vertebrate species, MG undergo a neurogenic reprogramming event that results in regeneration of functional neurons to restore loss of vision. However, in mammalian systems, MG undergo a reactive "gliotic" response associated with activation of inflammatory gene networks, and do not regenerate neurons. NFkB signaling is a master regulator of a pro-inflammatory response in the nervous system. The goal of this study was to investigate the role that NFkB signaling plays in regulating the neurogenic vs. gliotic response in MG after damage in the mammalian retina. We investigated patterns of NFkB activation in the mammalian retina after NMDA-mediated excitotoxic damage using an NFkB-eGFP reporter mouse line, as well as probing for NFkB

signaling components in single cell RNA-seq (scRNAseq) libraries from damaged mouse retinas (Hoang et al. 2020). Additionally, we utilized an Ascl1 overexpressing mouse line (GlastER-cre/LNL-tTA/tetO-Ascl1-GFP) (Jorstad et al., 2017) to investigate neuron regeneration after NMDA damage. We performed intraocular injections of NMDA and a pharmacological inhibitor of NFkB signaling (PGJ2) followed by immunohistochemistry or scRNAseq library preparation to identify retinal neuron regeneration and transcriptomic networks underlying the response, respectively. We find that NFkB signaling is rapidly activated in Müller glia cells after NMDA damage, and that this activation is dependent on the presence of microglia in the retina. Additionally, inhibition of NFkB signaling decreases macrophage populations within the retina. We find that inhibition of NFkB signaling promotes Ascl1-mediated retinal neuron regeneration by 30%. Further, inhibition of NFkB signaling repressed damage induced upregulation of reactivity gene networks as well as gene networks that drive reactive Müller glia back to a quiescent state, which underlies the increase in neurogenesis from these cells. In conclusion, these data indicate that inhibition of NFkB signaling suppresses a pro-inflammatory response and promotes neurogenic reprogramming of Müller glia in the damaged mammalian retina.

Disclosures: I. Palazzo: None. **L. Todd:** None. **S. Blackshaw:** None. **A.J. Fischer:** None.

Poster

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Marie Skłodowska-Curie Innovative Training Networks (ASCTN Training Network, grant agreement #813851)

Title: Purification and characterization hPSC-derived striatal progenitor subpopulations for transplantation in Huntington's Disease

Authors: *F. J. MOLINA-RUIZ, P. SANDERS, C. GOMIS, G. BOMBAU, M. GALOFRÉ, S. ARTIGAS, C. INTRONA, V. MONFORTE, J. M. CANALS;
Biomed. Sci., Univ. of Barcelona, Barcelona, Spain

Abstract: Huntington's disease (HD) is a movement disorder primarily characterized by the degeneration of striatal medium spiny neurons (MSNs). Cell replacement therapy to substitute degenerated neurons is a promising strategy to cure this devastating disease. The efficient differentiation of human pluripotent stem cells (hPSCs) to MSNs is a critical step in the implementation of cell therapy strategies for treating HD. Current *in vitro* striatal differentiation protocols are inefficient and generate heterogeneous cell products, which have a risk of undesired results following transplantation. Here we show a robust immunomagnetic cell sorting pipeline that uses a novel cell surface marker to select striatal neuroblast (NB) subtypes from a heterogeneous cell population differentiated in vitro from hPSCs. Our findings demonstrate that the use of this cell surface marker is a consistent method to isolate NBs which rapidly differentiate into mature striatal neurons expressing Dopamine Receptor (DR)-D1 and DR-D2. In addition, cell sorting of the heterogeneous differentiated hPSCs under different conditions produces specific NB subtypes related to striatal MSN sub-populations. Furthermore, following transplantation of these sorted committed NBs into adult mouse striatum we show evidence of graft survival and integration into the host tissue without graft overgrowth. In conclusion, we demonstrate that the cell selection process results in high-yield enrichment of different striatal NB subtypes under different conditions, and a reduction of cell product heterogeneity and batch-to-batch variation in different hPSC lines. We anticipate that the use of this pipeline for striatal NB selection prior to transplantation could accelerate the implementation of safe and reproducible cell products as part of future clinical cell replacement strategies for the restoration of functional deficits in HD.

Disclosures: F.J. Molina-Ruiz: None. **P. Sanders:** None. **C. Gomis:** None. **G. Bombau:** None. **M. Galofré:** None. **S. Artigas:** None. **C. Introna:** None. **V. Monforte:** None. **J.M. Canals:** None.

Poster

P020. Regeneration: CNS

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Program #/Poster #: P020.06

Topic: A.04. Transplantation and Regeneration

Support: 5R01EY027347
5R01EY030883
5R01EY031779

Title: Using multi-omics prediction and in vivo screen to explore transcriptional regulation of optic nerve regeneration

Authors: *C. QIAN[1], C. QI[1], Y. XIN[2], F.-Q. ZHOU[3];
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Abstract: Weak inherent axonal regrowth in the adult mammalian CNS hampers circuitry reconnection and functional recovery after various types of injuries or axonopathies. Emerging evidence showed that developmentally downregulated transcriptomic and epigenomic regulators, including Klf6/7, Sox11, Jun, Myc, Uhrf1 and Lin28, functioned to promote CNS axon regeneration sufficiently. Some developmentally upregulated factors such as Klf4/9 and Rest functioned to inhibit CNS axon regeneration. We thus hypothesize that manipulating developmentally regulated transcriptional factors (TFs) in retinal ganglion cells (RGCs) may act to promote optic nerve regeneration. An advanced integrative bioinformatics analysis approach was established to explore a mouse retinal embryonic and postnatal developmental deep-sequencing dataset (GSE87064) including RNA-seq, ChIP-seqs of histone markers and ATAC-seq. The RNA-seq was used to sort out differential TFs across development. By characterizing both the chromatin accessibility transition and active enhancer regions, a motif-based footprinting analysis (notches in the ATAC peaks) was utilized to predict TF-promoter as well as TF-enhancer-gene regulations. We then used a scoring algorithm to identify a group potential candidate TFs that might be manipulated to enhance optic nerve regeneration. Among all the candidate TFs, several novel targets were predicted with prioritized score in interacting with the regeneration-related transcriptome via either promoter or enhancer regions. Besides, some well-known regeneration promoting TFs, such as Sox11, Myc, Ascl1, Fos and Id2, also received high scores. Next, we are going to perform the in vivo screen with intermediate throughput of candidates in optic nerve axon regeneration. We will use CRISPRa and traditional ORF overexpression to explore the positive targets. Meanwhile, we will use CRISPRi, Cas13 and traditional shRNA interference through AAV2 infection to reveal the role played by suppressive targets in CNS regeneration.

Disclosures: C. Qian: None. **C. Qi:** None. **Y. Xin:** None. **F. Zhou:** None.

Poster

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Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P020.07

Topic: A.04. Transplantation and Regeneration

Support: NIH Grant EY018132
NIH Grant EY027310
Gilbert Family Foundation Vision restoration Initiative

Title: Hey1 and Id2b regulate Müller glia's injury-response threshold and the proliferation of Müller glia-derived progenitors in the zebrafish retina

Authors: *A. SAHU, S. DEVI, J. JUI, D. GOLDMAN;
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Abstract: The zebrafish and mammalian retina share structure and function; however, only zebrafish can regenerate a damaged retina. Key to this regenerative response is Müller glia (MG), which responds to retinal injury by reprogramming and dividing to produce multipotent retinal progenitors that regenerate all major neuron types. In the uninjured retina, Notch signaling is restricted to quiescent MG. However, following retinal injury, it is suppressed in injury-responsive MG and this suppression is necessary for retina regeneration. Studies suggest that MG engages in a regenerative response when exposed to injury-derived signals that exceed a certain threshold (MG's injury-response threshold). This threshold appears to be controlled by Notch signaling since pan-retinal Notch inhibition recruits normally quiescent MG near the injury site to participate in the regenerative response. Notch signaling inhibition is also associated with the proliferation of MG-derived progenitors. Using a combination of ATAC-seq and RNA-seq, we show that Notch controls MG's injury-response threshold by regulating the expression and chromatin accessibility of certain regeneration-associated genes. Intersection of RNA-seq gene lists obtained from injured MG and Notch inhibited uninjured MG identified 89 regeneration-associated genes that are regulated by Notch signaling in the uninjured retina. Importantly, 2 Notch target genes, *hey1 and id2b*, were found to mediate the effects of Notch signaling were also found to regulate MG's regenerative response. Using a combination of gene inactivation and transgenic overexpression studies, we show that *hey1* regulates MG's injury-response threshold, while *id2b* regulates the proliferation of MG-derived progenitors. Furthermore, using morpholino and CRISPR mediated gene suppression, transgenic overexpression studies, along with epistasis experiments, we show that notch ligands, Dll4 and Dlb, act through Notch3 receptor to regulate *hey1* and *id2b* expression. Importantly, these studies show that Hey1 suppression is sufficient to lower MG's injury-response threshold, while Id2b regulates the proliferation of MG-derived progenitors.

Disclosures: A. Sahu: None. **S. Devi:** None. **J. Jui:** None. **D. Goldman:** None.

Poster

P020. Regeneration: CNS

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P020.08

Topic: A.04. Transplantation and Regeneration

Title: Evaluation of functional repair process in iPSC-derived in vitro model of damaged neuronal network

Authors: *S. SHIOMOTO[1], W. LIN[1], K. MURAMATSU[1], T. HOSOYA[1], K. SAWADA[2]; [1]Biomed. Business Ctr., Ricoh Company, Ltd., Kawasaki-shi, Japan; [2]Grad. Sch. of Pharmaceut. Sci., The Univ. of Tokyo, Tokyo, Japan

Abstract: Neurodegenerative diseases cause abnormalities at different scales of the nervous tissue, from neuron subcellular structures to neuronal network functions. The repair of damaged neurons and their networks has been found to be difficult in the mammalian central nervous system (CNS). Therefore, there is a great need for new treatment methods that promote recovery from neuronal damage. However, the mechanisms for neuronal repair is poorly understood and replicating the human CNS microenvironment remains a challenge. Here, we combined human induced pluripotent stem cell (hiPSC)-derived neurons with microengineering to assess the functional connectivity of neuronal networks and their repair after disconnection.
Using hiPSC-derived neurons, two spatially separated clusters of cells were placed on a microelectrode-array. Neurites were observed to elongate between the co-cultured clusters, and the establishment of functional connections was evaluated by analyzing synchronized bursts within and between the two distinct networks. Cutting the neurites between the two clusters induced a desynchronization of the bursting activity, indicating that the neuronal networks had been damaged. Further culture of the damaged networks for a week induced the neurites to extend again beyond the severed area. Synchronous bursts reappeared between the two clusters, thus demonstrating that functional connection could be repaired over time.
Our results provide a method that can be used to evaluate the process and outcome of disrupting and repairing functional connections between two populations of neurons. This approach is expected to be useful in developing screening assays to evaluate the efficacy of drugs that reduce damage or promote functional recovery, and in understanding the mechanisms of brain plasticity and regeneration.

Disclosures: S. Shiomoto: A. Employment/Salary (full or part-time):; Ricoh Company, Ltd. **W. Lin:** A. Employment/Salary (full or part-time):; Ricoh Company, Ltd. **K. Muramatsu:** A. Employment/Salary (full or part-time):; Ricoh Company, Ltd. **T. Hosoya:** A. Employment/Salary (full or part-time):; Ricoh Company, Ltd. **K. Sawada:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Ricoh Company, Ltd..

Poster

P020. Regeneration: CNS

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P020.09

Topic: A.04. Transplantation and Regeneration

Support: DOD Grant WHX81-16-0715
DOD Grant W81XWH-15-0079
NIH Grant R01EY026546
NIH Grant U01 EY027257
NIH Grant EY14801

Title: Optic Nerve Regeneration Lipidomics: A Comparison of Four Phospholipid Profiles

Authors: *S. MEEHAN[1], E. NEAG[2], S. K. BHATTACHARYA[3];
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Abstract: Purpose As the quantity of discovered regenerative factors increases, we can evaluate regenerative processes from multiple perspectives. Successful optic nerve regeneration requires axon membrane expansion. The neuronal membrane contains more than 50% of phospholipids and is a likely source of significant shifts in regeneration. A comparison of phospholipid lipid profiles may provide insight into the essential lipid species and properties for regeneration.
Methods We evaluated four regenerated lipid profiles at 7 days and 2 weeks of regeneration: 2 pharmacological (Zymosan and Wnt3a), 1 biophysical (optogenetics - transgenic channel rhodopsin (ChR)), and a Phosphatase and Tensin homolog (PTEN) Cre-Lox knockout. Raw mass spectrometry files were obtained from Metabolomics Workbench and analyzed using LipidSearch. Significant lipid species and properties were determined using Metaboanalyst and Lipid Ontology (LION).
Results Of the four lipid profiles, the optogenetic transgenic mouse showed no significant specie or ontology shift between ChR groups and was omitted from the comparisons. Phospholipid classes were compared between regeneration, vehicle, and control groups. Zymosan group had 3/12 and 0/12 significant classes at 7 and 14 days post crush, respectively. Wnt3a had one significant class (LPC) at 15 days. PTEN KO had no significant phospholipid classes. At the species level, Zymosan group had 24/762 and 117/796 significant species at 7 and 14 days post crush, respectively. Wnt3a group had 1/286 and 15/286 significant species at 7 and 15 days, respectively. PTEN KO had 19/561 and 2/561 significant species at 7 and 14 days, respectively. Comparing all three lipid profiles showed no matched significant species. However, Wnt3a and Zymosan had two matched crush-specific lipids: PG(22:6/22:6) and PG(18:1/22:6). Zymosan and PTEN had one matched lipid specie: PI(17:0/20:4) with opposite shifts. Comparing all three lipid ontology profiles, at 7 days post crush, there is a consensus of upregulated lysophospholipids (LPE/LPC) and decreased alkyl- and acyl-phosphoethanolamines. At ~2 weeks, both Zymosan and PTEN KO groups had decreased lysophospholipids while Wnt3a maintained increased lysophospholipids.
Conclusions Based on this approach of optic nerve regeneration phospholipid profile comparisons, total phospholipid classes and single species may not provide insight into regeneration. Single phospholipid species may be associated with injury-specific shifts. The lipid ontology comparison provided the most insight into axon regeneration. Lysophospholipid and phosphoethanolamine regulation may be a clue in regenerative capacities.

Disclosures: S. Meehan: None. **E. Neag:** None. **S.K. Bhattacharya:** None.

Poster

P020. Regeneration: CNS

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P020.10

Topic: A.04. Transplantation and Regeneration

Support: NYS/DOH/SCIRB/ Grant C32247GG

Title: Neuroregenerative neurotrophic effect mediated by agents that promote cytidine diphosphate diacylglycerol biosynthesis

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Abstract: CDP-diacylglycerol (cytidine disphosphate diacylglycerol) is a critical intermediate in the biosynthesis of phosphatidylinositols for various cell signaling and structural functions. Receptors utilizing phosphatidylinositol-4,5-bisphosphate (PIP2) in signal transduction may couple through phospholipase C to induce PIP2 hydrolysis with release of inositol phosphate and diacylglycerol second messengers, or the receptors may couple through phosphatidylinositol-3-kinase (PI3K) to convert PIP2 into PIP3 which activates downstream intracellular signaling mediators. Neurotrophic factor receptors, especially the brain-derived neurotrophic factor (BDNF)-sensitive TrkB receptor, depend on both of these PIP2 cascades for normal function. We previously showed that a number of chemically diverse pharmacological agents could increase the biosynthesis of CDP-diacylglycerol with onward conversion of the nucleolipid into enhanced production of phosphatidylinositides in neural tissues (Undie, 1999; Tyeryar et al, 2007; 2009). Here, we surmised that a strategy to promote cellular PIP2 availability from increasing CDP-diacylglycerol could boost PIP2-associated receptor signaling events with physiologic consequences. To test this, cortical primary neurons obtained from neonatal rat pups and grown in culture for 7 days were mechanically injured using a commercial neuronal injury device. Injured neuronal populations were immediately exposed to predetermined concentrations of chemical compounds known to promote the synthesis of CDP-diacylglycerol. After 3 days, the cultures were immunostained with *Tub3* antibodies to mark neuronal soma and processes, followed by quantitative assessment of neuronal regrowth into the region of injury. As observed, multiple CDP-diacylglycerol-promoting compounds, including some dopaminergic agents and some clinical antidepressants, boosted the regenerative regrowth of injured rat cortical neurons in culture. For comparison, the regenerative effect of SKF38393 (1-phenyl-7,8-dihydroxy-2,3,4,5-tetrahydro-1H-3-benzazepine HCl) at 30 uM concentration was comparable to the effect of BDNF 40 ng/ml. Hence, in addition to direct stimulation of neurotrophin receptors, boosting PIP2 substrate availability could lead to neuroregenerative neurotrophic effects in neural tissue. The direct role of TrkB receptor signaling in these drug effects is being investigated.

Disclosures: W. Kang: None. **A.D.S. Tchamna:** None. **A.S. Undieh:** None.

Poster

P020. Regeneration: CNS

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P020.11

Topic: A.04. Transplantation and Regeneration

Title: The role of MMP2 in zebrafish retinal regeneration

Authors: L. BATIS[1], J. WILKINSON[1], P. GARRETT[2], K. FRIEDEL[1], J. SAAVEDRA[2], K. PAYNE[3], B. TONKS[4], *E. SANDQUIST[1];
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Abstract: Cell transplantation therapy has the potential to remedy presently incurable neurodegenerative diseases and injuries of the retina. However, low cell survival and retention is observed following transplantation to the retina. Disrupted environmental architecture and the presence of growth inhibitory molecules can reduce the efficiency of transplanted cells. Replicating regenerative stem cell niches such as those observed in zebrafish may improve cell survival and retention following transplant. Further, the use of developing zebrafish allows for live visualization of cell migration, an important step for stem cell integration following transplant. Matrix metalloproteinase-2 (MMP2) is promotes cell migration during development, with some evidence of involvement in regeneration. The objective of this study was to confirm whether MMP2 plays a role in retinal repair. Following photolesion to the zebrafish retina at three days postfertilization, the expression of MMP2 was measured at 1, 2, 3, 5, and 7 days post lesion. Results demonstrated that the gene was upregulated in the eye compared to controls. Significant changes in MMP2 expression indicates that further investigation of MMP2 in retinal regeneration is warranted. A second goal was to explore the validity of larval, rather than adult, zebrafish as a model for retinal regeneration. Immunohistochemistry to detect proliferating cell nuclear antigen (PCNA) was used to measure cell proliferation in the retina following injury. To identify proliferative Müller glial cells, *Tg(alpha-1 tubulin:EGFP)* zebrafish were used. This can determine whether Müller glia are the source of new cells, as is seen in adults, or if the cells originate in the ciliary marginal zone, which is responsible for normal retinal development. Future experiments will use live imaging to characterize the migration patterns of retinal stem cells during regeneration, as well as the effects of loss of MMP2 on this process. The ability to live image retinal stem cell migration *in vivo* will be an additional tool available to scientists as they investigate the mechanisms of regeneration. A better understanding of the manner in which stem cells migrate within their environment during repair, such as with matrix metalloproteinases, will lead to optimized cell delivery methods and improved prognosis for patients with retinal disease.

Disclosures: L. Batis: None. **J. Wilkinson:** None. **P. Garrett:** None. **K. Friedel:** None. **J. Saavedra:** None. **K. Payne:** None. **B. Tonks:** None. **E. Sandquist:** None.

Poster

P022. Axon Growth and Guidance: Axonal Transport and Trafficking

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P022.01

Topic: A.05. Axon and Dendrite Development

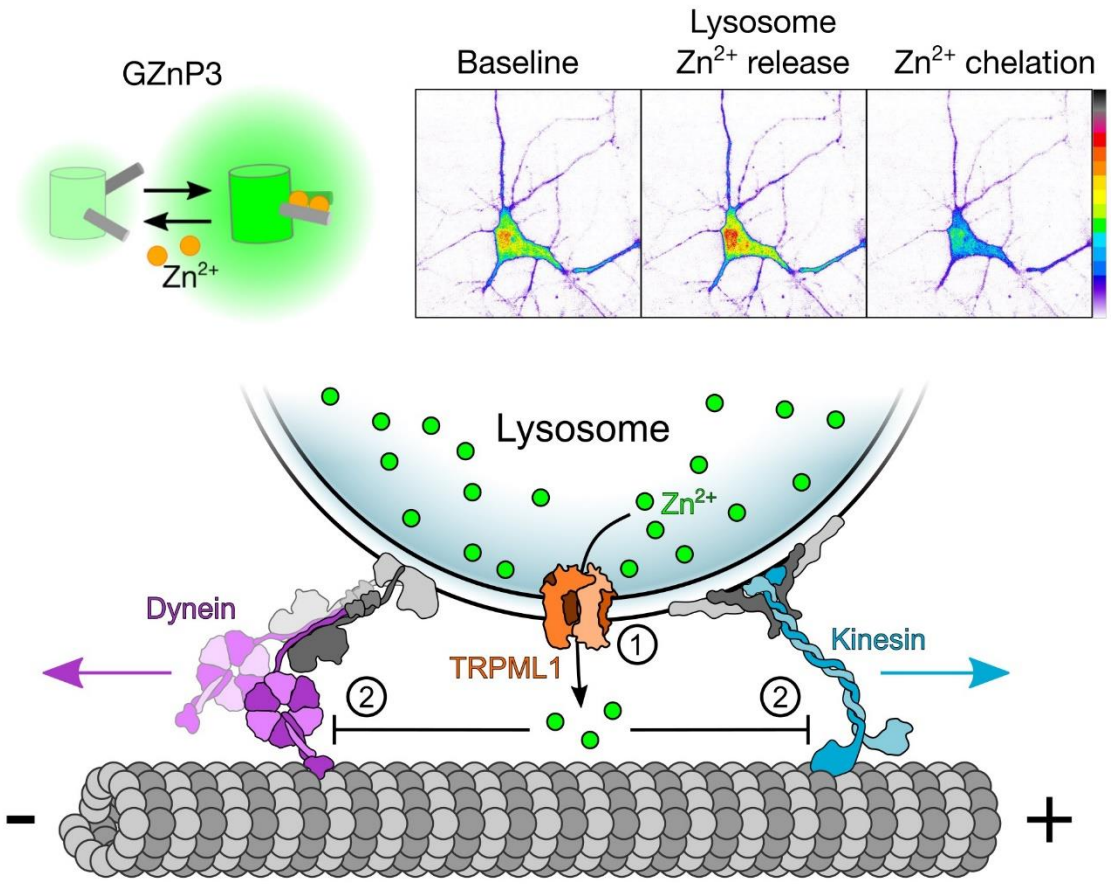
Support: NIH Grant R01NS110590
NIH Grant R01GM118492
NIH Grant R35GM139483
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NIH Grant R35GM131744

Title: Lysosomes self-regulate their axonal transport through TRPML1-mediated zinc release, directly inhibiting motor protein motility

Authors: *T. F. MINCKLEY[1], A. M. DISCHLER[1], D. H. FUDGE[2], E. GHAZVINI ZADEH[3], W.-H. LI[3], K. VERHEY[4], S. M. MARKUS[5], Y. QIN[1];
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Abstract: Accurate delivery of cargo over long distances through axonal transport requires precise spatiotemporal regulation in neurons, and in this work, we identify a novel role of zinc (Zn^{2+}) as a signaling molecule in axonal transport. First, we developed a novel, sub-nanomolar sensitive genetically encoded Zn^{2+} sensor, GZnP3, that has 13-fold dynamic range, and is able to detect cytosolic fluctuations in Zn^{2+} between 100 pM and 100 nM, characterized both *in situ* and *in vitro*. Using this sensor, both cytosolic and subcellularly-targeted, we established via confocal microscopy that the lysosomal ion channel, transient receptor potential mucolipin 1 (TRPML1), was permeable to physiological levels of Zn^{2+}. We found that primary rat hippocampal neurons have lysosomes with high concentrations of Zn^{2+} and pharmacological activation of TRPML1 can release this intracellular pool of Zn^{2+} into the cytosol. In addition, TRPML1-mediated Ca^{2+} signals were much smaller and shorter-lived in neurons than in HeLa cells. Conversely TRPML1-mediated Zn^{2+} signals were completely absent in HeLa cells, leading us to believe that TRPML1-mediated Zn^{2+} release from lysosomes may play an important role in neuronal health. Further we found that TRPML1-mediated Zn^{2+} release was more robust in neurites as compared to the soma. This led us to investigate the role of TRPML1 activation on axonal transport and we found that TRPML1-mediated Zn^{2+} release inhibited bidirectional axonal transport of lysosomes.

We were able to replicate this inhibition of lysosomal axonal transport by depolarization-induced Zn^{2+} influx. *In vitro* ATPase assays using purified motor protein and preformed microtubules were used to determine that both dynein and kinesin were inhibited by physiological increases in Zn^{2+}. We therefore propose a novel model for Zn^{2+} as a brake signal on axonal transport, wherein lysosomes can self-regulate their transport through TRPML1-mediated Zn^{2+} release.



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Poster

P022. Axon Growth and Guidance: Axonal Transport and Trafficking

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P022.02

Topic: A.05. Axon and Dendrite Development

Support: NIH Grant R01NS114253

Chan-Zuckerberg Initiative - Ben Barres ECA Award

Title: Muscleblind-like proteins selectively associate with kinesin motors through their zinc-finger domains to regulate RNA transport

Authors: *R. HILDEBRANDT[1], K. MOSS[2], L. KNUDSON[2], A. JANUSZ-KAMINSKA[2], K. LIN[2], T. SAXENA[1], L. DENES[1], G. J. BASSELL[2], E. T. WANG[1];
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Abstract: Localization of mRNAs to axons and dendrites supports efficient control of local protein synthesis and maintains proper neuronal function. RNA-binding proteins (RBPs) and cytoskeletal motors are important for mediating this process. One such family of RBPs are the Muscleblind-like proteins (MBNLs), splicing factors with cytoplasmic functions, including localization of RNA targets to various subcellular locations. Previous studies have shown that depletion of MBNLs by expanded CTG repeats in myotonic dystrophy may cause RNA mis-localization; however, mechanisms and domains responsible for MBNL-mediated RNA localization remain uncharacterized. Through live imaging of primary mouse cortical neurons, we observed MBNL-containing granules moving at speeds consistent with microtubule transport. Thus, we hypothesized that MBNL associates with a kinesin motor to mediate RNA localization. To elucidate these potential MBNL-kinesin associations, we utilized assays designed to identify novel kinesin-cargo interactions. We found that tail domains of Kif1bα and Kif1c selectively associate with MBNL and, surprisingly, these interactions depend on MBNL C_3H zinc-finger (ZnF) domains in a non-RNA binding dependent manner. Subsequent experiments with other C_3H ZnF-containing proteins suggest a common mechanism for kinesin association. MBNLs have been shown to localize RNAs to membranes, indicating a potential role for MBNL in facilitating not just transport, but terminal anchoring. Indeed, we found that MBNL mutants lacking unstructured domains (exon 3 and C-term) no longer co-fractionate with membranes and do not efficiently form cytoplasmic granules. Finally, we found that dominant negative expression of kinesin tails or functional depletion of MBNL by CTG repeat over-expression in a neuronal cell model causes mis-localization of nucleolin (*Ncl*) RNA, revealing a functional target that may be perturbed in myotonic dystrophy. Overall, these findings advance our understanding of kinesin-RBP interactions and how RNA transport is mediated in neuronal systems.

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Poster

P022. Axon Growth and Guidance: Axonal Transport and Trafficking

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Program #/Poster #: P022.03

Topic: A.05. Axon and Dendrite Development

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NRF Grant 2020M3E5E2039894
NRF Grant 2017R1A5A1015366
NRF Grant 2021R1A2C3010639

Title: The role of schizophrenia-associated MAD1L1(Mitotic Arrest Deficient-1 like 1) in neuronal development

Authors: B. GOO, D. MUN, S. KIM, T. THI MY NHUNG, B. SUH, Y. WOO, S. KIM, *S. PARK;
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Abstract: The neurodevelopmental hypothesis of schizophrenia is evidenced by the alteration of ventricle sizes and cortical volumes in patients. Until now, novel schizophrenia risk genes have been revealed via multiple genome-wide association studies (GWAS). Although GWAS has identified an association between *MAD1L1* (*Mitotic Arrest Deficient-1 like 1*) and the predisposition of schizophrenia, the molecular mechanisms underlying this association remain unclear. Here, we report the novel functions of *MAD1L1* in neuronal development. First, we found that MAD1 (gene product of *MAD1L1*) is an important regulator of neuronal polarity in migrating neurons. Our data revealed that polarity defect caused by *MAD1L1*-deficiency affects neuronal migration and neurite outgrowth. Positioning of Golgi apparatus, which determines the cellular polarization, was notably changed by *MAD1L1*-deficiency in migrating neurons. Second, we observed that endogenous MAD1 is enriched at the Golgi apparatus in neurons. When we induced knockdown of MAD1, significant changes in morphology and trafficking functionality of Golgi apparatus were observed. Finally, we identified kinesin-like protein KIFC3 as a novel protein-protein interaction partner of MAD1 by yeast-two-hybrid screening. Our data also show that MAD1-KIFC3 collaborates to maintain the integrity of the Golgi apparatus in developing neurons, critical for proper migration and differentiation. Collectively, our findings suggest that *MAD1L1* has a novel function in neuronal development, which might be further relevant to schizophrenia pathophysiology.

Disclosures: B. Goo: None. **D. Mun:** None. **S. Kim:** None. **T. Thi My Nhung:** None. **B. Suh:** None. **Y. Woo:** None. **S. Kim:** None. **S. Park:** None.

Poster

P022. Axon Growth and Guidance: Axonal Transport and Trafficking

Location: Virtual Only

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Program #/Poster #: P022.04

Topic: A.05. Axon and Dendrite Development

Support: R01 DC005798 to GS
CoBRE Grant P30 GM103503 to GS
P41 GM103412 to ME
Howard Hughes Medical Institute
Gordon and Betty Moore Foundation

Title: Rapid terminal growth dynamics within local territories during neural circuit formation

Authors: *D. JACKSON[1], S. SCHELLHASE[1], R. NICHOL, IV[1], J. HEDDLESTON[2], T.-L. CHEW[2], S. M. YOUNG, JR[3], M. H. ELLISMAN[4], G. A. SPIROU[1];
[1]Med. Engin., Univ. of South Florida, Tampa, FL; [2]Advanced Imaging Ctr., Janelia Res. Campus, Ashburn, VA; [3]Dept. of Anat. and Cell Biol., Univ. of Iowa, Iowa City, IA; [4]Dept Neurosci, UCSD BSB 1000, La Jolla, CA

Abstract: The largest mammalian CNS nerve terminal, the calyx of Held (CH), provides strong and temporally precise excitation of target principal cells of the medial nucleus of the trapezoid body (MNTB). The CH exhibits hallmark developmental features of strengthening and pruning necessary to yield monoinnervation of MNTB neurons. Using serial block-face scanning electron microscopy (SBEM) of tissue collected from neonatal littermate mice (24-48 hr sampling), we previously found that between postnatal (P) days 2 and 4, most of the 10-20 small pioneering inputs on each cell are pruned while 1-3 terminals grow at rates of 200 μm^2 per day. By P6, 75% of principal cells are monoinnervated. During CH formation, collateral processes of variable length extend from the edges of the developing terminal and are known sites of Ca^{2+} entry. Although we found no evidence of direct interaction between CHs along the principal cell soma, collaterals largely occupy shared territories. However, little is known about the function of the calyceal collaterals. Our lab is interested in the direct interaction among developing inputs that lead to the emergence of a dominant CH. Here, we sought to examine the temporal dynamics of synaptic organization in the MNTB as it relates to the ultrastructure revealed with SBEM. We employed lattice light-sheet (LLS) microscopy which offers rapid and high-resolution image acquisition with minimal bleaching. Acute coronal brainstem slices (300-600 μm thickness) were collected in neonatal mice ranging from P0-14. Following the 4D image acquisition, data was imported into syGlass ™, a custom software package designed in-house, for immersive virtual reality (VR) aided-analysis. Processes were manually tracked in syGlass™ to reveal the growth dynamics of CHs and their associated processes, including filopodia- and growth cone-tipped terminal arbors. We found these collaterals form a fluid field around each CH as well as exhibit an age-dependent relationship in their motility that rivals or exceeds rates of axonal extension described elsewhere in the CNS. Growth cones, perhaps the most dynamic structure in this system, extend fastest during the ages of CH expansion yet slow after monoinnervation is established. Moreover, live imaging and ultrastructural analysis presented here establishes that CH arbors repeatedly form transient associations between developing terminals and such contacts may serve a communicative role in circuit formation. These data are effective in

monitoring the navigation patterns of assembling neural circuits in an intact system and reveal the dynamic nature of growth and retraction of developing neurite reorganization.

Disclosures: D. Jackson: None. **S. Schellhase:** None. **R. Nichol:** None. **J. Heddleston:** None. **T. Chew:** None. **S.M. Young:** None. **M.H. Ellisman:** None. **G.A. Spirou:** A. Employment/Salary (full or part-time):; syGlass.

Poster

P022. Axon Growth and Guidance: Axonal Transport and Trafficking

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P022.05

Topic: B.11. Glial Mechanisms

Support: Canadian Institutes of Health Research/MOP-142231
MS Society of Canada
CAPES/CNPq
International Society for Neurochemistry

Title: Schwann cells provide iron to axonal mitochondria and its role in nerve regeneration

Authors: *B. S. MIETTO[1], P. JHELUM[2], K. SCHULZ[3], S. DAVID[4];
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Abstract: Iron is an essential co-factor for several metabolic processes, including the generation of adenosine triphosphate (ATP) in mitochondria, which is required for axonal function and regeneration. However, it is not known how mitochondria in long axons, such as those in sciatic nerves, acquire iron *in vivo*. Due to their close proximity to axons, Schwann cells (SCs) are a likely source of iron for axonal mitochondria in the peripheral nervous system. Here we demonstrate the critical role of iron in promoting neurite growth *in vitro* using iron chelation. We also show that SCs express the molecular machinery to release iron, namely, the iron exporter, ferroportin (Fpn) and the ferroxidase ceruloplasmin (Cp). In *Cp* knockout (KO) mice, SCs accumulate iron, because Fpn requires to partner with Cp to export iron. Axons and SCs also express the iron importer transferrin receptor 1 (TfR1), indicating their ability for iron uptake. In teased nerve fibers, Fpn and TfR1 are predominantly localized at the nodes of Ranvier (NR) and Schmidt-Lanterman incisures (SLIs), axonal sites that are in close contact with SC cytoplasm. We also show that lack of iron export from SCs in *Cp* KO mice reduces mitochondrial iron in axons as detected by reduction in mitochondrial ferritin, affects localization of axonal mitochondria at the NR and SLIs, and impairs axonal regeneration following sciatic nerve injury.

These finding suggest that SCs contribute to the delivery of iron to axonal mitochondria, required for proper nerve repair.

Disclosures: B.S. Mietto: None. **P. Jhelum:** None. **K. Schulz:** None. **S. David:** None.

Poster

P022. Axon Growth and Guidance: Axonal Transport and Trafficking

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P022.06

Topic: A.05. Axon and Dendrite Development

Support: NSF Grant #1748523
NSF Grant #1826871
NSF Grant #1832282

Title: Mutations in UNC-33, a microtubule associated protein, causes defects in transport of autophagic vacuoles in neurites of C. elegan's neurons: impact of microtubule organization on axonal autophagic flux

Authors: *R. JAUREGUI, **A. HOLGADO**;
Dept. of Biol. Sci., St. Edward's Univ., Austin, TX

Abstract: Autophagy is an important process for maintaining homeostasis wherein cells degrade intracellular components to enable nutrients to be utilized in other ways. Neurons are postmitotic cells that rely on this process to ensure that dysfunctional proteins, organelles, and aggregates are safely degraded. Neurodegenerative diseases such as Alzheimer’s disease display characteristic intracellular aggregates that make autophagy a target of research. Neurites are elongated, polarized projections that enable communication between neurons. Collapsin Response Mediator Protein-2 (CRMP-2), along with it's ortholog UNC-33 in *C. elegans*, are responsible for proper microtubule assembly and neurite elongation. UNC-33 has been shown to form a complex with UNC-44 and UNC-119 to secure microtubules to the cortex of neurons. Proper microtubule organization enables the functioning of the autophagic pathway and ensures that synaptic dysfunctional proteins are safely degraded and recycled in the cell. Our study aims to understand the role that UNC-33 has in the autophagic process. In vivo confocal imaging of the *C. elegans'* AIY neuron with the aid of a tandem LGG-1 marker was used to count and track individual autophagic vacuoles (AV) and their flux through the autophagy pathway. This tandem marker contains a pH sensitive, two color fluorescent marker that, when fused with degradative cellular compartments such as lysosomes and endosomes, quenches the green signal and shows a single red puncta. Images of red and green AV’s were randomized and blindly scored for location, number, and maturity. Each control or experimental group analyzed contained n>27 animals. Statistical examinations of the results demonstrated that two *unc-33* mutants alleles *unc-33(e204)*

and *unc-33(mn407)* are found to have a significantly higher number of LGG-1 positive puncta in their AIY neurites. The *unc-33* mutants also possess a higher number of mature autolysosomes in the AIY neurites when compared to wild type. *unc-44(hrt-2)* mutants show a similarly higher number of AV in the AIY neurite. This study further validates the role that *unc-33* plays in maintaining homeostasis by enabling transport of autophagosomes through neurites. A better understanding of how autophagosomes are able to clear dysfunctional proteins has important implications on how scientists may be able to remedy the aggregates seen in neurodegenerative diseases such as Alzheimer's Disease.

Disclosures: R. Jauregui: None. **A. Holgado:** None.

Poster

P022. Axon Growth and Guidance: Axonal Transport and Trafficking

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P022.07

Topic: A.05. Axon and Dendrite Development

Support: NSF Grant #1748523
NSF Grant #1826871
NSF Grant #1832282

Title: Defects in autophagy flux related to microtubule associated protein UNC-33 mediate changes in basal and starvation-induced autophagy

Authors: *E. CARRILLO, E. HOLECHEK, A. OTT, A. HOLGADO;
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Abstract: Autophagy is the intracellular degradative process responsible for the recycling of inefficient proteins or organelles. Regulation of autophagy is essential for axonal development and maintenance via CRMP-2/UNC-33, a microtubule associated protein (MAP) that contributes to neuronal development and transport of autophagosomes. Together with UNC-119 and membrane-associated Ankyrin (UNC-44), UNC-33 forms a ternary complex that anchors microtubule bundles to the cortex and inhibits microtubule sliding, herein referred to as the ternary microtubule associated cortical complex (MACC). Deficiencies in cargo degradation and autophagy flux produced by *unc-33* mutants have the potential to contribute to the development of neurodegenerative diseases, such as Alzheimer's. Our laboratory studies autophagic flux via the cleavage of the double fluorescently tagged autophagosomal protein LGG-1/LC3 in *C. elegans unc-33 mutants*. We predict that lack of UNC-33 will result in defects in autophagy flux.To test this hypothesis we monitored autophagosome maturation and pathway flux using western blotting techniques and measured the levels of the double fluorescently tagged autophagosomal protein CERULEAN-VENUS-LGG-1(dFP)(MW 75) versus the monomeric

fluorescent protein (mFP)(25 kDa). We compared induced autophagy via starvation against basal autophagy. The cleavage of dFP into mFP upon arrival at the lysosome was used to construct a ratio of 25 kDa/75 kDa herein defined as the index of autophagy flux. Analysis of westernblot densitometry showed that *unc-33* mutants undergoing induced autophagy have an increase in the index of autophagy flux. In basal autophagy, there was a decrease in the index of autophagy flux. Additionally, *unc-104(e1265)* mutants appeared to have similar indices to wildtype. Together, these results suggest that UNC-33 and the ternary MACC may play a notable role in autophagy flux and clearance of cargo compared to the kinesin motor protein UNC-104. These results are indicative of the role UNC-33 plays towards neuronal autophagy and neurodegenerative disorders.

Disclosures: E. Carrillo: None. **E. Holechek:** None. **A. Ott:** None. **A. Holgado:** None.

Poster

P022. Axon Growth and Guidance: Axonal Transport and Trafficking

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Program #/Poster #: P022.08

Topic: A.05. Axon and Dendrite Development

Support: NSF Grant #1748523
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NSF Grant #1832282

Title: Investigating the effects of infection of Pseudomonas aeruginosa on the regulation of the expression of neuronal GFP driven by the unc-33 promoter in C. elegans

Authors: *A. CARRASCO-PENA[1], S. AVANT[1], E. TORRES[1], K. SALAZAR[1], A. HOLGADO[2];
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Abstract: Schizophrenia’s familial transmission and heritability have been known for several decades, although the relationship between the disorder and genetic components have been made evident, it has been difficult to identify the particular causative molecular players involved. It has been hypothesized that environmental and genetic factors have contributed to the development of Schizophrenia particularly through the disruption of the mammalian target of rapamycin (mTOR) pathway. *Pseudomonas* is an aerobic and gram-negative bacteria that belongs to the *Pseudomonadaceae* family which is often found in soil and water. *Pseudomonas aeruginosa* is commonly described as an antibiotic resistant and opportunistic pathogen that causes infection in plants and animals with the inclusion of humans. *Pseudomonas aeruginosa* has been shown to be pathogenic to *C. elegans* nematodes. Genes, including *daf-16*,activate the nematode’s immunity upon the pathogen's presence. It is proposed that infection with *Pseudomonas aeruginosa*

activates the DAF-2 signaling pathway as well as induces autophagy. Our previous findings support the hypothesis that external factors, such as temperature, play a role in the regulation of the expression of GFP driven by the *unc-33* promoter. In our studies, we incubated *C. elegans* at 16°C and 25°C and found that those developing at a high temperature had a lower expression of neuronal GFP as an indicator of the activity of the *unc-33* promoter. In contrast, nematodes that develop at 16°C show a high expression of GFP in the nerve ring. Herein we will study the effects of environmental stressors such as *Pseudomonas aeruginosa* infection on the expression of GFP driven by the *unc-33* promoter. To this end, *C. elegans* hermaphrodites were incubated at 16°C in a plate seeded with OP50 or *Pseudomonas aeruginosa* until they reached the L3/L4 stage. Confocal microscopy was then utilized and the head of the *C. elegans* hermaphrodites were imaged to determine GFP expression driven by the *unc-33* promoter in the nerve ring and glial cells. The analysis of the quantification of twenty five worms per condition has shown that *C. elegans* infected since embryo with *Pseudomonas aeruginosa* have decreased neuronal GFP driven by the *unc-33* promoter. While neuronal expression is reduced due to infection, GFP glial expression is increased. This study provides insight into the role that infection has on regulating the expression of GFP driven by the *unc-33* promoter, shedding some light into external factors controlling one of the molecular players (UNC-33/CRMP2/DPYSL2) shown to be repressed by polymorphic variations in patients suffering from schizophrenia.

Disclosures: A. Carrasco-Pena: None. **S. Avant:** None. **E. Torres:** None. **K. Salazar:** None. **A. Holgado:** None.

Poster

P042. Animal Models of Autism

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Program #/Poster #: P042.01

Topic: A.07. Developmental Disorders

Support: NIH Grant 1R01MH121433-01

Title: Quantifying the developmental trajectory of autism associated brain overgrowth using magnetic resonance and 3D cellular resolution imaging

Authors: *F. A. KYERE[1], C. M. MCCORMICK[2], I. CURTIN[2], O. KRUPA[2], M. R. GLASS[1], T. M. FARAH[2], C. F. ESCOBAR-TOMLIENOVICH[2], Q. HE[3], T.-W. WANG[3], A. RUMPLE[4], J. A. JIMENEZ[5], M. S. ITANO[6], M. J. ZYLKA[6], Y.-Y. I. SHIH[3], G. WU[4], J. L. STEIN[7]; [1]Dept. of Cell Biol. and Physiology, Neurosci. Curriculum, UNC Neurosci. Ctr., [2]Dept. of Genetics, UNC Neurosci. center, [3]Departments of Neurology, Biomed. Engineering, and Biomed. Res. Imaging Ctr., [4]Dept. of Psychiatry, Neuro Image Res. and Analysis, [5]Curriculum in Toxicology & Envrn. Med., [6]Dept. of Cell Biol. and Physiology, UNC Neurosci. center,

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Abstract: Disruptions in embryonic brain development can manifest as altered postnatal brain structure and function, leading to neuropsychiatric illness. One such disruption is mutation in Chromodomain helicase DNA binding protein 8 (*Chd8*); a gene that encodes an ATP-dependent chromatin remodeler. Heterozygous *Chd8* loss of function mutations result in macrocephaly in both humans and mouse models, and autism in humans. However, how *Chd8* haploinsufficiency impacts brain structure across cortical development is unknown. Importantly, previous work demonstrated that increases in brain volume are observed across several brain regions including the cortex, requiring a whole brain imaging approach to study the mutation's effects. Here, we employ magnetic resonance imaging (MRI), tissue clearing technology, and light-sheet microscopy to acquire MR and 3D cellular resolution images in intact whole brains from a mouse model with a single heterozygous point mutation in *Chd8* leading to truncated protein (*Chd8*$^{+/-}$). Our goal is to quantify neural progenitors and neuronal cell-types within annotated areas of the developing neocortex in *Chd8*$^{+/-}$ mice and their sex-matched wild-type (WT) littermate controls to determine the cell types that drive this ASD-associated cortical hyper-expansion. At present, we have generated 2 littermate pair brain MR images at postnatal day 4 (P4) (60µmx60µmx60µm). We found increased cortical volume in *Chd8*$^{+/-}$ mice(7%) relative to WT littermate controls in both males and females, replicating previous work. Additionally, we have generated 3D cellular high resolution (0.75µmx0.75µmx4µm) images from 5 littermate pairs at P4 labeled with upper- (*Brn2*) and lower-(*Ctip2*) layer neuronal markers. Using NuMorph, a suite of image analysis tools developed in our lab, we have registered one of these images to the Allen Developing Brain Atlas and quantified all the nuclei in the cortex. We detected about 13.8 million cortical nuclei comprised of 21% each of upper- and lower-layer neurons. These results demonstrate cortical volume increases in *Chd8*$^{+/-}$ mouse brains. Moving forward, we aim to perform similar analyses for 9 biological replicates per sex and per genotype at P4 and other early developmental timepoints to elucidate the cellular basis and spatial localization of brain overgrowth in our mouse model, leading to a better understanding of how genetic variation can affect cortical development and alter risk for neurodevelopmental disorders.

Disclosures: F.A. Kyere: None. **C.M. McCormick:** None. **I. Curtin:** None. **O. Krupa:** None. **M.R. Glass:** None. **T.M. Farah:** None. **C.F. Escobar-Tomlienovich:** None. **Q. He:** None. **T. Wang:** None. **A. Rumple:** None. **J.A. Jimenez:** None. **M.S. Itano:** None. **M.J. Zylka:** None. **Y.I. Shih:** None. **G. Wu:** None. **J.L. Stein:** None.

Poster

P042. Animal Models of Autism

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Topic: A.07. Developmental Disorders

Support: NIH Grant 1T32NS115753-01
ARCS Foundation Fellowship
UCLA Neuroscience Interdepartmental Program
NIH Grant R01HD054453
NIH Grant R01NS117597

Title: Deficits in social touch in genetic and environmental mouse models of autism

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Abstract: Social withdrawal/social anxiety in individuals with autism spectrum disorders (ASD) may result from hypersensitivity to social touch, such as hugging or shaking hands. Social touch deficits have been relatively understudied in rodent models of ASD, and the circuits involved have not been identified. We designed a novel assay to investigate the behavioral responses associated with social touch and differences between ASD mice and healthy controls. A *test* mouse, head-fixed but free to run on an air-suspended polystyrene ball, is allowed to voluntarily (whisker-whisker) or forced (snout-snout) to interact with an inanimate object or with a *visitor* mouse (familiar or stranger) head-fixed on a moving platform. Each social interaction type (order is randomized) occurs in several bouts to mimic typical mouse to mouse interactions. We use high speed videos (50 FPS) to analyze avoidance behaviors and aversive facial expressions with custom MATLAB routines and DeepLabCut (Mathis et al. 2018). We tested two ASD models, the genetic model of Fragile X Syndrome (*Fmr1*$^{-/-}$ mice) and the environmental maternal immune activation (MIA) model (progeny of dams injected with Poly I:C at E12.5). We find that ASD mice in both models spent more time running in the direction away from the stranger mouse during forced and voluntary social touch interactions compared to 'no-contact' or voluntary object interactions. Healthy control mice (*Fmr1*$^{+/+}$ mice or progeny of dams injected with phosphate buffered saline) showed significantly less avoidance during social touch than with object touch (and less avoidance compared to ASD mice). Furthermore, we observed a significant increase in aversive facial expressions (orbital tightening, prolonged whisker protraction) during forced social touch compared to both voluntary social and object touch in both *Fmr1*$^{-/-}$ and MIA mice, while there was no difference in healthy controls across conditions. We also observed that autism model mice demonstrate gaze avoidance during object and social touch, a behavioral phenomenon also observed in ASD individuals. We conclude that our novel social touch assay for mice shows a strong applicability for investigating maladaptive behaviors to social touch in models of autism. In the future, we intend to use Neuropixels silicon probes to record the activity of behaviorally relevant neuronal populations in the barrel field of primary somatosensory cortex (S1BF) and the basolateral amygdala (BLA) during social touch to correlate neuronal activity with behavioral changes in autism models.

Disclosures: T. Chari: None. **A. Hernandez:** None. **C. Portera-Cailliau:** None.

Poster

P042. Animal Models of Autism

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P042.03

Topic: A.07. Developmental Disorders

Support: Startup account

Title: Human infant gut microbiomes may influence social behavior in mice

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Abstract: Background: The gut microbiome has pleotropic effects on our body with recent research suggesting individual variation in gut microbiome composition may influence brain development and increase risk for some psychiatric and neurodevelopmental disorders. This includes work by our group showing that certain features of the infant gut microbiome are associated with fear reactivity. Given the observational nature of our human infant studies, it remains unclear whether the human infant gut microbiome causes individual differences in fear reactivity. To address this gap, the present study was undertaken to investigate whether transplant of gut microbiota from human infants into germ-free pregnant mice altered anxiety-related behaviors, social behaviors, and cognition in their offspring.
Methods: Pregnant Swiss-Webster germ-free mice were divided into four groups. Group 1 (BIF group) was inoculated with a mixed human fecal slurry derived from infants with relatively high levels of *Bifidobacterium*. Group 2 (BAC group), was inoculated with a mixed human fecal slurry derived from infants with relatively high levels of *Bacteroides*. Group 3 (SPF group) received fecal slurry from specific pathogen free mice (SPF group) and group 4 (GF group) received autoclaved fecal slurry from SPF mice. Offspring were assessed in the elevated plus maze (EPM), novel object recognition, and three-chamber sociability tests between 6-8 weeks of age. Both males and females were studied (N = 13, 6, 4, 7, 4, 4, 6, and 5 for BIF males, BIF females, BAC males, BAC females, SPF males, SPF females, GF males, and GF females, respectively).
Results: No statistically significant group differences were observed when males and females were combined. In sex stratified analyses, SPF male mice spent more time with the familiar mouse in phase 2 of the social interaction tests than BIF male mice (*$p<0.05$) or BAC male mice (**$p<0.01$), but all 4 groups spent a similar amount of time with the novel mouse. Germ free males appeared more similar to the SPF males, but did not differ significantly from any other group. Sex stratified analyses of the EPM and novel object recognition test did not indicate and significant group differences.
Conclusion: Results suggest that male mice carrying humanised infant gut microbiomes may

differ in their social behaviour compared with SPF and GF mice. Our next step will be to examine histopathological and imaging data from these mice to determine if behavioural changes are accompanied by changes in brain volumes, resting-state connectivity, myelination, and dendritic morphology.

Disclosures: H. Dubey: None. **A. White:** None. **S. Liu:** None. **R. Knickmeyer:** None.

Poster

P042. Animal Models of Autism

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P042.04

Topic: A.07. Developmental Disorders

Title: Toward Understanding the Molecular Basis of Prenatal Stress as an Environmental Risk Factor for Autism Spectrum Disorder

Authors: J. T. DUNN[1,2], **A. GUIDOTTI**[2], *D. R. GRAYSON[2];
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Abstract: It has long been posited that symptoms of autism spectrum disorder (ASD) may be the consequence of an imbalance of excitation and inhibition (E/I) in the brain. However, the E/I profile of neural substrates for ASD-relevant behavioral endophenotypes remains largely undefined on the molecular level with respect to the etiological risk factor of prenatal stress. We examined the behavioral and molecular endophenotypes of adult offspring of C57BL/6J dams subjected to prenatal stress during gestation. We chose to examine this genetic background as it is commonly used in the generation of preclinical models for the study of ASD. Pregnant C57BL/6J dams underwent three 45-minute restraint stress sessions per day from gestational day 9 to parturition. Male and female offspring of the stressed dams (PRS mice) and non-stressed dams (NS) underwent ASD-relevant behavioral testing in adulthood. The differential expression of genes related to excitatory and inhibitory neurotransmission was evaluated in the medial prefrontal cortex, amygdala, hippocampus, nucleus accumbens (NAc) and dorsal striatum of male PRS mice using qRT-PCR. Adult PRS mice demonstrated reduced sociability along with increased marble burying behavior relative to NS control mice. The PRS mouse behavioral phenotype coincided with aberrant expression of glutamate and GABA marker genes (e.g. Grin1, Grin2b, Gls, Gat1, Reln) in brain regions with putative involvement in social behavior. We tested drugs with known epigenetic properties (e.g. clozapine (5 mg/kg) + 18-hr washout) and observed the successful pharmacological rescue of the male PRS mouse sociability deficit. This result is consistent with the hypothesis that genes that govern sociability are epigenetically regulated. Analysis of the expression levels of genes relevant to epigenetic regulation using NanoString fluorescent barcoding indicated that those genes which encode enzymes that alter histone

methylation and acetylation revealed alterations in clozapine-treated PRS NAc samples compared to the NAc of vehicle-treated PRS mice. The identification of etiology-specific mechanisms underlying clinically relevant behavioral endophenotypes provides alternative targets for the development of novel pharmacotherapies for treating ASD.

Disclosures: J.T. Dunn: None. **A. Guidotti:** None. **D.R. Grayson:** None.

Poster

P042. Animal Models of Autism

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P042.05

Topic: A.07. Developmental Disorders

Title: Development of the midbrain and social behavior in theMonodelphis domesticaanimal model

Authors: *I. PEREZ[1], E. I. ALANIZ[1], A. MENDIOLA[2], A. MAR[1], J. L. VANDEBERG[3], M. GIL[4];
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Abstract: The Brazilian short-tailed opossum (*Monodelphis Domestica)* is an understudied animal model, compared to the *Mus musculus,* and has been identified as an ideal candidate for studying neurodevelopment (VandeBerg, & Robinson, 1997). *Monodelphis* is an ideal animal model to study neurodevelopment because the embryo develops outside the pouch of the mother providing easy noninvasive access to track changes across different developmental stages (Mate et al., 1994). Having this noninvasive access provides an opportunity to study natural midbrain development, which has implications for typical behavior and neurodevelopmental disorders. The objective of the present study is to compare the area and volume of the *Monodelphis* midbrain across four different developmental stages as well as describe any changes in behavior. We utilized Zen and ImageJ software to perform area analyses of different stages of brain development (Mironov, 2017). As expected, based on our preliminary results, the embryonic day 14 specimen had the smallest area in the midbrain, area= 27260.36 μm ^2, m= 381.376. At postnatal day 1 (~24 hrs after birth) the midbrain area increased to 49917.28 μm^2, m= 404.12; increases continued on postnatal days 6 and 27 to 81866.66 μm^2, m=166.016 and 98442 μm^2, m= 227.734, respectively. Dissections were also performed from the midbrain at two developmental time points, PND0 (at birth) and PND1 (24 h post-birth). Tissue was then lysed and protein extracted for spectrophotometry and western immunoblotting (WB), and ImageJ software was used to quantify band densities. We have determined that extraction of the midbrain from a neonate (PND1-5) yields approximately 1,000,000 cells. After adjusting with a

loading control, the WB procedure yielded a brighter band for a target protein tyrosine hydroxylase (TH) at PND0 (band density = 0.97) compared to PND1 (band density = 0.19). Thus, our data suggest that TH protein levels are highest at birth and then decrease over subsequent developmental time points. Regarding behavior, aggression appeared to increase as post-weaning pups matured to adolescence and young adulthood. Interestingly, in litters with fewer animals, there was decreased expression of aggressive behavior in later stages in comparison to the litter with the most animals. There was no significant difference in the means for general motor activity and social behavior. Our study provides support for the use of the *Monodelphis Domestica* for the study of early neurodevelopment and neurodevelopmental disorders. A future goal is to analyze glia and neuronal number across different early neurodevelopmental stages using stereological techniques.

Disclosures: I. Perez: None. **E.I. Alaniz:** None. **A. Mendiola:** None. **A. Mar:** None. **J.L. Vandeberg:** None. **M. Gil:** None.

Poster

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Title: Early Development of Social Attention Brain Circuits in Infant Primates: A Combined Behavioral and Structural MRI Study.

Authors: *Z. KOVACS-BALINT[1], C. POOLE[1], S. SAAVEDRA[1], T. JONESTELLER[1], L. LI[2,5], M. STYNER[6], M. ALVARADO[1], J. BACHEVALIER[1,3], M. SANCHEZ[1,4];
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Abstract: Non-human primate (NHP) models are critical for understanding typical development and alterations in social attention in neurodevelopmental disorders, such as Autism Spectrum Disorder (ASD). The goal of this study was to characterize the developmental trajectories of

social attention neurocircuits in infant rhesus macaques and their behavioral correlates. Longitudinal brain structural MRI scans were acquired at 2, 4, 8, 12 and 24 wks in 31 male infant macaques (equivalent to 2-24 mos in human infants), and at 16 and 20 wks in 21 of the subjects, all living in social groups. T1- and T2-MRI images were acquired using a 3T MRI scanner and analyzed for volumetric changes in the ventral and dorsal attention networks (VAN: V1,V3, lateral intraparietal area -LIP-, frontal eye field -FEF-; DAN: V1, V3, temporoparietal junction -TPJ-, ventrolateral prefrontal cortex -vlPFC-), and other regions involved in social attention (i.e. caudal temporal parieto-occipital area -TPOc-; area PG associated region in the superior temporal sulcus -STS-). Measures of social visual attention were collected from a subset of subjects (n=11), using: (1) focal observations (ethogram), (2) rating scales of atypical social behaviors (juvenile macaque Social Responsiveness Scale -jmSRS-, adapted from the human SRS, widely used for ASD diagnosis), (3) measures of visual orientation / following / attention with the Schneider Neonatal Assessment for Primates (SNAP), and (4) eye-tracking measures of visual attention to social vs. nonsocial stimuli. Our results showed increased social ($F_{(5,14.695)}$=10.802; p=1.66x10^{-4}) and solitary play ($F_{(5,25.714)}$=10.792; p=1.10x10^{-5}) behaviors by 12 wks, when infants increase independence, exploration and social interactions. These changes were paralleled by region-specific changes in developmental trajectories of social attention neurocircuits, although the strongest volumetric growth was observed in most brain regions between 2 and 8-12 weeks. Our findings suggest that during the first 8-12 wks, social attention brain networks undergo rapid and robust structural changes that parallel important social behavior milestones (increased play and independence from mother) in infant macaques. The neural changes likely support the infant's adjustment to independence and refined attention to relevant social visual cues for appropriate social interactions in complex social environments. Understanding how normative social attention and underlying brain networks develop in a NHP model can help elucidate the roots of brain-behavior pathogenesis of human neurodevelopmental social derailments, such as those in ASD.

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Poster

P042. Animal Models of Autism

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Program #/Poster #: P042.07

Topic: A.07. Developmental Disorders

Support: Simons Foundation Autism Research Initiative Grant 345034
NIMH R01 MH123661

Title: Delay discounting but not probability discounting promotes choice inflexibility in males in a mouse model of 16p11.2 hemideletion

Authors: *G. R. ROJAS, L. S. CURRY-POCHY, A. T. HELLER, N. M. GRISSOM;
Univ. of Minnesota, Minneapolis, MN

Abstract: People with neurodevelopmental disorders have previously been demonstrated to have an impaired ability to associate actions with their outcomes (Sinha et al., 2014). 16p11.2 hemideletion is a copy number variant which is associated with the diagnosis of multiple neurodevelopmental diagnoses, including autism spectrum disorder, language disorder, and attention deficit hyperactivity disorder. Mice modeling hemideletion of the syntenic 16p11.2 region display altered reward-seeking behavior and concomitant changes in striatal neurobiology. However, it is unknown whether altered reward-seeking behavior reflect slower or more uncertain value updating, versus other factors such as impulsivity. We asked whether 16p11.2 hemideletion impacts the ability of mice to learn about choice contingencies in a series of discounting tasks, and how animals reduce uncertainty when introduced with different tasks. We had mice modelling 16p11.2 hemideletion undergo delay and probability discounting in order to observe how mice adapt to different types of incrementing costs. Our design of delay and probability discounting presented animals with two choices: a small immediate reward or a large delayed/risky reward. Animals learned that blocks of trials either started with no delay/risk but increased delay/risk as mice progressed through a session ("Worsening") or started with a large delay/risk but decreased delay as mice progressed throughout a session ("Improving"). We gave mice amphetamine during the last rounds of probability discounting in order to examine if mice shift choice according to the order of the task (St Onge et al., 2009). In our current results, we found male 16p11.2 hemideletion mice, but not female 16p11.2 hemideletion mice, preferentially choose to experience large delays to receive more reward, suggestive of slower value updating, but not choice impulsivity per se. As training continued, male 16p11.2 hemideletion mice adjusted to delays with increased training, indicating long-term adaptation to shifting schedule demands. We are currently examining if animals exhibit differences in how they explore the chamber during different choice epochs.

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Poster

P042. Animal Models of Autism

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Topic: A.07. Developmental Disorders

Title: Murine influenza A virus disrupts immune profiles in the maternal gut and placenta but not the fetal brain

Authors: *A. M. OTERO[1], A. M. ANTONSON[2];
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Abstract: Maternal immune activation (MIA) during pregnancy is linked to neurodevelopmental disorders, such as schizophrenia and autism, in offspring. The majority of animal models initiate MIA using pathogen mimetics, which induce a controlled innate immune response. In this study, we use a mouse model of live H3N2 influenza A virus (IAV), which is moderately pathogenic and activates both innate and adaptive immune responses. As respiratory IAV infection is known to cause downstream immune dysregulation in the intestine, we hypothesized that this would contribute to inflammation in the fetus. Small intestine, placenta, and fetal brains were collected from C57BL/6NTac mice on gestational day 17, seven days post inoculation (dpi) with IAV (n=17) or saline (n=15), across four identical replicates. Despite no detectable virus in the small intestine, respiratory IAV infection upregulated the gene encoding RORγt, the main transcription factor for T_H17 and ILC3 cells of type III immunity. RORγt$^+$ type III immune cells release pro-inflammatory IL-17 cytokines, which are implicated in MIA-driven developmental abnormalities. However, transcription of *Il17a*, *Il17f*, and *Il17ra* was unaltered in maternal intestinal tissue regardless of treatment, as was production of IL-17A protein. To investigate if IAV-induced MIA impacted the fetus, we looked at immune profiles in the placenta and fetal brain. The placenta, which acts as a physical and immune barrier between dam and fetus, demonstrated altered immune profiles in infected dams. However, neuroinflammatory genes in the fetal brain were not significantly different between groups. Our results demonstrate that a moderately pathogenic IAV infection upregulates type III immune cells in the maternal gut at seven dpi with no significant difference in IL-17 production. Furthermore, changes in the placental immune response, but not fetal, suggest the placenta remains a resilient barrier during moderate maternal infection. Our data confirm the necessary use of live pathogens in MIA modeling to evaluate the complete immune response and improve translation to the clinic.

Disclosures: A.M. Otero: None. **A.M. Antonson:** None.

Poster

P042. Animal Models of Autism

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P042.09

Topic: A.07. Developmental Disorders

Support: NINDS T32MH020068

Title: Effect of genetic background in a mouse model of autism due to ASH1L mutation

Authors: *C. M. PAPENDORP[1], D. A. HAQUE[1], L. A. GOICOURIA[1], J. S. LIU[2];
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Abstract: Autism spectrum disorder (ASD) is a heterogenous condition with a broad spectrum of severity. While most studies of ASD use targeted disruption of a single gene, in affected humans, multiple genetic interactions are likely at play. Absent, small, or homeotic-like (ASH1L) is a highly penetrant ASD-associated gene. Humans with ASH1L mutations have variable phenotypes: while some may have mild ASD/intellectual disability (ID) and few other symptoms, others have severe ID, no verbal communication, intractable seizures, and multiple congenital anomalies. To study how loss of ASH1L leads to ASD, I use mice with a gene-trap (gt) mutation in ASH1L, rendering the mice hypomorphic for ASH1L. Previous studies have reported that homozygous loss of ASH1L is lethal at birth. I find that on an inbred C57BL/6J background, homozygous ASH1L$^{gt/gt}$ die around weaning age ($p = 0.0025$ at P21, Chi squared test for expected versus observed allelic frequencies, n = 61 mice from 10 litters). This lethality reflects the fact that humans are heterozygous for ASH1L mutations. However, when backcrossed three times to the outbred CD1 strain, I discovered that ASH1L$^{gt/gt}$ mice survive to adulthood at normal Mendelian ratios (no difference by Chi squared test at P21, n = 68 mice from 9 litters). While previous studies on a C57BL/6J background had reported that ASH1L is required for fertility in mice, on a CD1 background, ASH1L$^{gt/gt}$ mice are fertile and bear offspring. Motivated by the association of ASH1L with ASD, I examined neuroanatomical phenotypes caused by loss of ASH1L. While BL/6 ASH1L$^{gt/gt}$ mice are microcephalic, CD1 ASH1L$^{gt/gt}$ may be microcephalic or macrocephalic. To see if changes in brain size are accompanied by differences in neuronal morphology, I performed Golgi staining in 2-month-old mice. While ASH1L$^{gt/+}$ mice on a BL/6 background have normal morphology of layer V pyramidal cells compared to BL/6 wild type, ASH1L$^{gt/gt}$ mice on a CD1 background have decreased soma size and fewer intersections by Sholl analysis compared to CD1 wild type (n = 6 mice per genotype/background, 3 or more cells per mouse). Because about 30% of people with ASH1L mutations have seizures, I also examined differences in seizure frequency and baseline EEG power spectra between ASH1L mutant and wild type mice on the BL/6 and CD1 backgrounds (n = 6 mice per genotype/background). Overall, I find striking effects of genetic background on lethality, fertility, and brain structure and function in a mouse model of ASD due to ASH1L mutations. This study underscores the importance of considering genetic background in mouse studies of ASD and provides clues about the wide spectrum of severity in ASD due to ASH1L mutations.

Disclosures: C.M. Papendorp: None. **D.A. Haque:** None. **L.A. Goicouria:** None. **J.S. Liu:** None.

Poster

P165. Neuroinflammation: Microglia I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P165.01

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant 1R21NS087472-01A1
NIH Grant 1R01NS095937-01A1
NIH Grant1R01NS094306-01A1
DoD Grant W81XWH-14-1-0543

Title: Neuro-immune signatures in chronic low back pain subtypes

Authors: *Z. ALSHELH[1], L. BRUSAFERRI[1], A. SAHA[1], E. MORRISSEY[1], P. KNIGHT[1], M. KIM[1], Y. ZHANG[2], J. HOOKER[1], D. ALBRECHT[1], A. TORRADO-CARVAJAL[1], M. PLACZEK[1], O. AKEJU[2], J. PRICE[1], R. EDWARDS[3], J. LEE[1], R. SCLOCCO[1], C. CATANA[1], V. NAPADOW[1], M. L. LOGGIA[1];
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Abstract: We recently showed that patients with different chronic pain conditions demonstrated elevated brain and/or spinal cord levels of the glial marker 18kDa translocator protein, which suggests that neuroinflammation might be a pervasive phenomenon observable across multiple etiologically heterogeneous pain disorders. Interestingly, the spatial distribution of this neuroinflammatory signal appears to exhibit a degree of disease specificity, suggesting that different pain conditions may exhibit distinct “neuroinflammatory signatures”. To explore this hypothesis, we tested whether neuroinflammatory signal can characterize putative etiological subtypes of chronic low back pain patients based on clinical presentation. Specifically, we explored neuroinflammation in patients whose chronic low back pain either did or did not radiate to the leg (i.e., “radicular” vs. “axial” back pain). Fifty-four chronic low back pain patients, twenty-six with axial back pain (43.7±16.6 y.o. [mean±SD]) and twenty-eight with radicular back pain (48.3±13.2 y.o.), underwent PET/MRI with [^{11}C]PBR28, a second-generation radioligand for the 18kDa translocator protein. [^{11}C]PBR28 signal was quantified using standardized uptake values ratio. Functional MRI data were collected simultaneously to the [^{11}C]PBR28 data 1) to functionally localize the primary somatosensory cortex back and leg subregions and 2) to perform functional connectivity analyses. PET and functional MRI measures were compared across groups, cross-correlated with one another and with the severity of “fibromyalgianess”. Furthermore, statistical mediation models were employed to explore possible causal relationships between these three variables. For the primary somatosensory cortex representation of back/leg, [^{11}C]PBR28 PET signal and functional connectivity to the thalamus were: 1) higher in radicular compared to axial back pain patients, 2) positively correlated with each other and 3) positively correlated with fibromyalgianess scores, across groups. Finally, 4) fibromyalgianess mediated the association between [^{11}C]PBR28 PET signal and primary somatosensory cortex-thalamus connectivity across groups.Our findings support the

existence of “neuroinflammatory signatures” that are accompanied by neurophysiological changes, and correlate with clinical presentation in chronic pain patients. These signatures may contribute to the subtyping of distinct pain syndromes and provide information about inter-individual variability in neuro-immune brain signals, within diagnostic groups, that could eventually serve as targets for mechanism-based precision medicine approaches.

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Poster

P165. Neuroinflammation: Microglia I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P165.02

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: KAKENHI [JP19K07119]
Smoking Research Foundation

Title: Panaxytriol inhibits lipopolysaccharide induced microglia activation

Authors: G. HIRAMATSU[1], *D. UTA[1], K. MIHARA[2], T. KUME[1];
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Abstract: Introduction: In neurodegenerative diseases, brain inflammation is triggered, and activated microglia produce inflammatory mediators that damage surrounding neurons. Therefore, suppression of activated microglia can be an effective therapeutic target in neurodegenerative diseases. Then, we focused on panaxytriol, an Nrf2 pathway activator contained in *Panax ginseng* extracts, as an inhibitor of activated microglia. Since the effect of panaxytriol on microglia activation has not been revealed so far, we investigated its effects of the lipopolysaccharide (LPS)-induced microglia activation both *in vivo* and *in vitro*. Methods: C57BL/6Ncr mice intracerebroventricularly injected LPS were used as a brain inflammation mouse model. Panaxytriol was simultaneously administrated with LPS. We performed immunostaining and measured spontaneous behavior activity. BV-2 cell was utilized *in vitro* study and nitric oxide (NO) production was detected as an index of activation by griess assay. The expression levels of the inflammatory cytokine mRNA and protein were detected by real-time PCR and Western blotting, respectively, and translocation of NF-κB to the nucleus was detected by immunocytochemistry. Results: In a brain inflammation mouse model induced by

LPS, Iba1 positive cells were increased and morphological change of Iba1 positive cells were observed. GFAP positive fluorescence intensity was increased. Spontaneous behavior was decreased. Simultaneous intraventricular injection of panaxytriol with LPS suppressed the increase of Iba1 positive cells, morphological change, and the increase of GFAP positive fluorescence intensity. Similar to the response to LPS, microglia were more sensitive to panaxytriol than astrocytes. Therefore, we investigated the mechanism of inhibitory effect of panaxytriol on microglia activation. In BV-2 cell, LPS increased NO production and iNOS expression. Panaxytriol suppressed the increase of NO production and iNOS expression by simultaneous treatment with LPS. In addition, panaxytriol suppressed the increase of proinflammatory cytokine mRNA expression induced by LPS. To investigate the mechanism of inhibitory effect of panaxytriol on microglia activation, we investigated the involvement of the MAPK pathway. Panaxytriol treatment had no effect on MAPK pathway. Panaxytriol suppressed the translocation of NF-κB into nucleus induced by LPS. Conclusion: These results suggested that panaxytriol suppressed microglia activation by suppressing the NF-κB signaling pathway. Taken together, panaxytriol can inhibit brain inflammation and may be useful as a therapeutic agent against neurodegenerative diseases.

Disclosures: G. Hiramatsu: None. **D. Uta:** None. **K. Mihara:** None. **T. Kume:** None.

Poster

P165. Neuroinflammation: Microglia I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P165.03

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant R25-GM07174514A1
NIH Grant 5R01DA047743-02

Title: Impact of neonatal opioid exposure and withdrawal on the adult innate immune response in female mice

Authors: *C. LEMCHI[1], C. J. BROWN[2], J. R. FERRANTE[1], J. E. MENGAZIOL[1], A. D. DUNN[1], J. A. BLENDY[1];
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Abstract: Every year, thousands of pregnant women are prescribed prescription pain relievers, resulting in prenatal exposure to opioids. Babies born to an opioid-dependent mother experience neonatal opioid withdrawal syndrome (NOWS); a combination of somatic withdrawal symptoms including high pitched crying, sleeplessness, irritability, gastrointestinal distress, and in the worst cases, seizures. The long-term effects of chronic morphine treatment during early life are not

well characterized, particularly the effect on microglia activation and neuroinflammation. Determining the impact of neonatal opioid exposure and withdrawal on the innate immune response in adulthood will lead to new understanding and therapy of NOWS. We developed a novel morphine exposure and withdrawal paradigm to test if opioid exposure, over the human equivalent of all three trimesters, dysregulates the neuroimmune response to opioids, either at birth or later in adulthood. Female mice (n=34) were exposed to either morphine or saline during embryonic development (E0-E21) and treatment was continued via injections upon birth to postnatal day 14. On PND 15, 24 h after the final morphine or saline injection, pups showed significant signs of withdrawal including somatic signs, increases in ultrasonic vocalizations and thermosensitivity. In addition, several immune-related mediators are elevated in cortex, including complement component 1q (C1q), a protein complex which is part of the innate immune system. C1q elevation in newborn mice might have functional relevance for the development of long-term consequences of NOWS due to its role in synaptic pruning, however whether this elevation persists in adulthood is not known. Further, elevation of C1q has not been reported in adults after exposure to opioids. We found that elevation of C1q is not maintained in adulthood, following chronic morphine exposure (PND 90-595), but we see a significant downregulation of expression in cortex following an exposure paradigm of escalating doses of morphine (20-100 mg/kg for 5 days; which induces dependence as evidenced by withdrawal symptoms in adults. These finding reveal disordered neuroimmune regulation in female mice exposed to NOWS, suggesting susceptibility to autoimmune disorders or reoccurring infections later in life, and aid in providing important targets for the development of rational treatment approaches for NOWS.

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Poster

P165. Neuroinflammation: Microglia I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P165.04

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIGMS P20 GM103429

Title: Cultured microglia exhibit time-dependent recovery after ethanol exposure

Authors: *B. BISHOP, C. KEISLING, A. HARMSWORTH, D. DONLEY;
Harding, Searcy, AR

Abstract: Microglia are central nervous system-resident immune cells that activate in response to dyshomeostasis. Upregulation of phagocytosis and the release of inflammatory cytokines are

two markers of activation. Ethanol has been identified as a cause of microglial activation. Therefore, an examination of the microglial responses to ethanol contributes to a better understanding of microglia function during insult. Published research demonstrates that microglial cells exhibit decreased phagocytosis and increased inflammatory cytokine production in response to ethanol exposure. While an increase in cytokine production is consistent with activation, decreased phagocytosis is indicative of microglia damage. Ethanol exposure is typically transient as compared to other activating signals such as misfolded proteins in neurological disorders. The long-term effects of ethanol exposure and their ability to recover after simulation is not well understood. To model the transient nature of ethanol exposure, we treated immortalized mouse microglial cells with 25 mM or 100 mM ethanol for two hours in the same plate as control cells. To measure activation, phagocytosis was quantified by engulfment of fluorescent latex beads, tumor necrosis factor-alpha (TNF-a) production was measured by ELISA, and metabolism was measured using a Seahorse XF-HS metabolic analyzer. Data was analyzed using an ANOVA. We found that microglial phagocytosis is impaired, metabolic capacity is altered, and TNF-a is increased by two-hour ethanol exposure. To address the ability of cells to recover after ethanol exposure, we changed the culture media after the two-hour stimulation and allowed cells a recovery period of 0, 12, or 24 hours. Our data suggests partial, time-dependent recovery upon removal of ethanol marked by a recovery of phagocytosis. We found that markers of inflammation including TNF-alpha production and glycolytic flux indicate a low-level activation that resolves over time, but in the absence of changes in cell proliferation. Together, these data suggest that cells are recovering and not being replaced. Future studies aim to explore the mechanism(s) of recovery and will contribute to the understanding of the dynamic responses of microglia during dyshomeostasis.

Disclosures: B. Bishop: None. **C. Keisling:** None. **A. Harmsworth:** None. **D. Donley:** None.

Poster

P165. Neuroinflammation: Microglia I

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P165.05

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant P20GM103429
NSF Grant 1827066

Title: Myeloperoxidase is a pro-oxidant enzyme in microglial cells that shapes the response to extrinsic stimuli

Authors: *S. EWING[1], D. DONLEY[2], A. WILCOX[1], B. BISHOP[1];
[2]Biol., [1]Harding Univ., Searcy, AR

Abstract: Microglial cells are the resident immune cells of the central nervous system. Reactive oxygen species (ROS) are highly reactive molecules that result from cellular metabolism and participate in cell signaling. In microglia, excess ROS causes oxidative stress, which is hypothesized to promote neurodegenerative diseases. The pro-inflammatory enzyme myeloperoxidase (MPO) is a known pro-oxidant enzyme in immune cells such as neutrophils but has not been widely studied in microglial cells. In addition, buildup of the Alzheimer's Disease-associated protein, beta-amyloid (Abeta) is associated with oxidative stress on microglia. However, the link mechanism(s)of how oxidative stress associated with Abeta develops in microglia are still not fully understood. Abeta is predominantly found in 2 forms: AB-40 and AB-42. AB-42has been found to be more toxic than AB-40. The goal of this study was to determine the impact of myeloperoxidase on the development of microglial oxidative stress in response to extracellular Abeta. To measure ROS, cultured microglial cells were stained with the fluorogenic dye, DCFDA, that detects ROS. Following staining, the cells were either treated with HBSS (as a control) or treated with hydrogen peroxide (as an ROS generator). After treatments, myeloperoxidase inhibitor-I was added to half of the samples. Data was continuously collected for two hours following initial treatment. Results show that the MPO inhibitor decreases the production of reactive oxygen species in hydrogen peroxide-treated microglial cells when compared to controls. These data indicate that myeloperoxidase could account for a major portion of the oxidative stress in microglial cells during neurological diseases. When microglia are exposed to toxic substances, such as AB 40 or 42, they produce inflammatory cytokines, such as Tumor necrosis factor alpha (TNFa). An ELISA assay was used to measure cytokine production. Inhibiting myeloperoxidase decreased cytokine production significantly across all samples indicating that myeloperoxidase could account for the majority portion of TNF-a production in microglial cells in response to pro-inflammatory stimuli. Given the connection between ROS and metabolism, we also measured the impact of MPO inhibition on the metabolic activity of microglia. Direct injection of a MPO inhibitor into control and AB 40or 42 treated cells increased glycolysis and decreased oxidative phosphorylation. However, MPO inhibition had a larger effect on control cells than cells stimulated with Abeta. Together these data show that myeloperoxidase impacts the oxidative state of the cell and participates in their response to external stimuli.

Disclosures: S. Ewing: None. **D. Donley:** None. **A. Wilcox:** None. **B. Bishop:** None.

Poster

P165. Neuroinflammation: Microglia I

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Program #/Poster #: P165.06

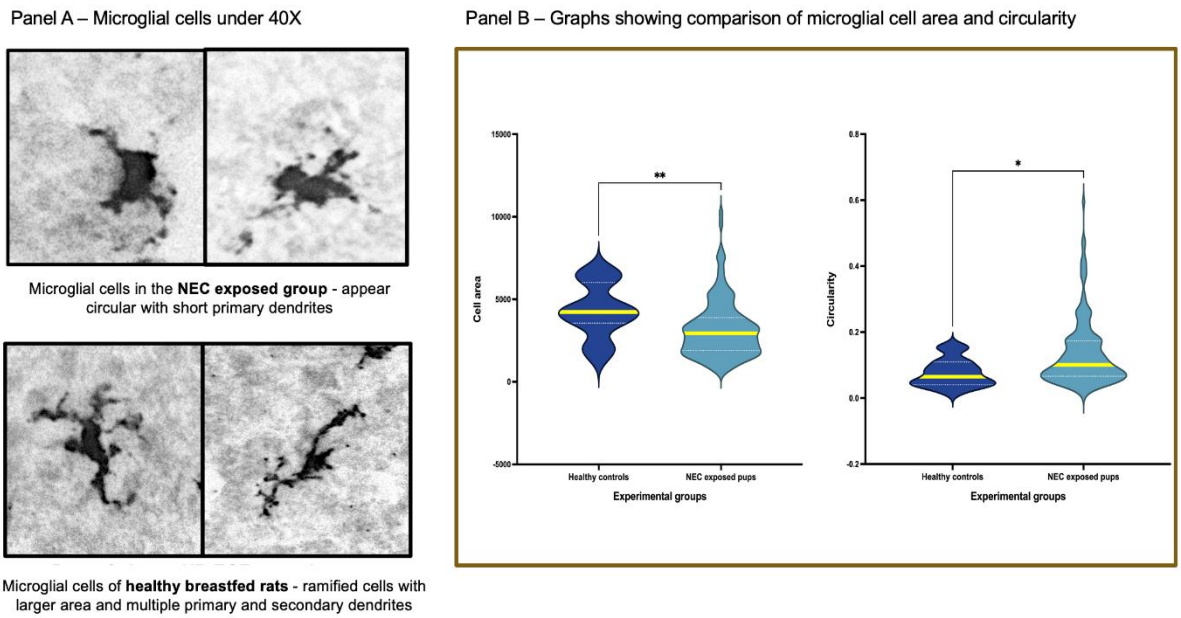
Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: Loma Linda University GCAT grant 2190400

Title: Changes to microglial cell morphology in response to early post-natal environment as seen in a rodent model of Necrotizing enterocolitis (NEC)

Authors: V. CHAWLA[1], L. C. SIEBOLD[2], *C. G. WILSON[3];
[1]Neonatal-Perinatal Med., [3]Lawrence D. Longo MD Ctr. for perinatal biology, [2]Loma Linda Univ., Loma Linda, CA

Abstract: NEC presents with high mortality and neuro-developmental impairment in neonates. Gut-brain interactions may be key to NEC-related neuroinflammation and microglial cells in the CNS are key elements of neuroinflammation. Microglial morphology has a phenotypical spectrum, ranging from ramified to ameboid forms determined by micro-environment and patho-physiologic state. Microglia transition from ramified to more ameboid shapes in acute inflammatory states. Morphometric differences have the potential to differentiate and measure inflammation/restorative states when studying the effects of NEC on the brain. We hypothesized that microglial cells in the brainstem of rat pups with NEC have a compact, pro-inflammatory phenotype, compared to larger, ramified microglial cells in healthy rat pups.We compared two groups, 1) Healthy full-term breastfed rat pups housed with the dam and 2) NEC exposed pups delivered prematurely on embryonic day 21 via C-section and exposed to a NEC protocol that included repetitive hypothermia, hypoxia, lipopolysaccharide and hyperosmolar feeds. Intestinal injury was graded based on villous damage. Brainstems were removed, stained for IBA1, and analyzed using *ImageJ*. NEC incidence in the exposure group was 45% (n=38). Exposed rat pups had smaller brains compared to breastfed pups ($p<0.05$, n=50). Microglia in brainstems of breastfed pups had larger hull area, span, perimeter, bounding circles, radii, and increased "roughness" suggesting a resting (non-inflammatory) state compared to denser, compact (pro-inflammatory) microglia in NEC-exposed pups ($p<0.05$, n=170). We saw no correlation between brain size and microglia size suggesting that microglial cell parameters are independent of brain weight. Exposure to early postnatal risk factors was associated with smaller brains—emphasizing the importance of early developmental windows in long-term neurological outcomes. NEC-exposed microglia showed significant reduction in cell ramification and increased circularity indicating brainstem inflammation begins early postnatally.



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Poster

P165. Neuroinflammation: Microglia I

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Program #/Poster #: P165.07

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIAAA

Title: Cortical microglia gene and protein expression changes in human alcohol use disorder suggest shifts in microglial phenotype

Authors: *L. QIN, R. P. VETRENO, F. T. CREWS;
Bowles Ctr. Alcohol Studies, Sch. Med, Univ. North Carolina, Chapel Hill, NC

Abstract: Microglia are essential for CNS homeostasis, synapse regulation, and innate immune signaling that could contribute to neuroplasticity and neurodegeneration. Microglial genes and proteins were determined in orbital frontal cortex (OFC) of individuals with alcohol use disorder (AUD) and age-matched moderate drinking controls using RT-PCR and immunohistochemistry (IHC). In AUD OFC, we found significant increases in mRNA levels of microglial markers CD11b (207%) and CX3CR1 (337%). However, Iba1, CD68, and P2RY12 mRNA were not changed. Microglial marker protein assessment using IHC revealed increases in Iba1+ (13-fold), CD68+ (4-fold), CD11b+ (Mac1: 1.4-fold; OX42: 2-fold), CX3CR1+ (3-fold), and P2RY12+ (2-fold) immunoreactivity in the AUD OFC, compared to control subjects. Microglia in alcoholic

brain showed varied morphologies, including enlarged cell bodies, intensified staining, and activated morphology. In contrast, the microglial marker TMEM119, which is widely used in transgenic mice, was reduced in AUD OFC. TMEM119 mRNA and protein (IHC) were reduced by 47% and 51% in the AUD OFC, relative to controls. TMEM119 microglial morphology is ramified, similar to morphology associated with resting-trophic microglia. This differs from most other markers. Control TMEM119+IR cells show a ramified morphology that in AUD is faint with fewer cells. The six other microglial markers (Iba1, CD68, Mac1, OX42, CR3CR1, and P2RY12) show dark staining rounded microglial morphology associated with activation. These data are consistent with shifts in microglial phenotype from a resting state to an activated state with associated changes in gene expression, including loss of TMEM119 expression and morphological phenotype. AUD altered microglia may contribute to changes in synapses and neurocircuitry associated with AUD-induced changes in brain function.

Disclosures: L. Qin: None. **R.P. Vetreno:** None. **F.T. Crews:** None.

Poster

P165. Neuroinflammation: Microglia I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P165.08

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: Dr. Miriam and Sheldon Adelson Medical Research Foundation (LIB)
NIH (5 R01 EY027881-02, LIB)
NIH (NICHD IDDRC HD018655, LIB)
NIH (NEI MBED T32 EY007145, SLP)
Medical Technology Enterprise Consortium (LIB)

Title: Complement proteins and myeloid cells support optic nerve regeneration via myelin clearance

Authors: *S. L. PETERSON, K. A. WONG, C. SUN, A. CATACORA, L. I. BENOWITZ; Boston Children's Hosp., Boston, MA

Abstract: Axon regenerative failure in the mature CNS contributes to functional deficits following traumatic injuries, ischemic injuries, and neurodegenerative diseases. Although neuro-immune interactions modulate many CNS functions, potential contributions of the classical complement cascade and myeloid cells to CNS axon regeneration (or its failure) are largely unknown. We recently reported (SfN 2017) that genetic deletion of complement C1q, C3, or CD11b (CR3) attenuates retinal ganglion cell (RGC) axon regeneration following experimental mouse optic nerve injury (ONI), suggesting that complement proteins and $CR3^{+}$ myeloid cells are each required for axon regeneration. However, the underlying mechanisms for this

regeneration effect remained unclear. The classical complement cascade is initiated by C1q, and C3b is a downstream opsonin that binds to CR3 receptor on myeloid cells (resident microglia, infiltrating monocytes, neutrophils) to induce phagocytosis. In the current study, we characterized the complement and myeloid cell response to ONI, and tested the hypothesis that complement and phagocytic myeloid cells support RGC axon regeneration by altering the local environment of the injured optic nerve. To achieve this, we performed protein and molecular analyses (markers: C1qa, C3, CD11b, Iba1/Aif1, CD68, CD206, CD45, P2RY12, MBP) on optic nerves and retinas from mice with and without ONI (uninjured, 1d, 3d, 5d, 7d, 14d), and with and without complement manipulation ($CR3^{-/-}$, $CR3^{+/+}$, WT, anti-C1q, IgG). Our data show that classical complement proteins C1q and C3 and $CR3^{+}$ myeloid cells each increase markedly near the ONI site. Further, axon regeneration results from local injections of C1q function-blocking antibody experiments confirmed that complement acts primarily within the optic nerve to support axon regeneration. In the optic nerve, we observed an MBP-negative area that expanded from the injury site over time and closely corresponded to an area that was densely populated with $CR3^{+}$ and $CD68^{+}$ myeloid cells, including some that contained internalized MBP. Interestingly, this region also contained an increased density of regenerating axons. Finally, our data indicate inefficient myelin clearance (increased myelin debris remaining near the ONI site) in $CR3^{-/-}$ mice compared to $CR3^{+/+}$ controls. Collectively, these results suggest that classical complement proteins and myeloid cells act within the optic nerve to support RGC axon regeneration through phagocytic removal of growth-inhibitory myelin debris. These results emphasize the importance of the axonal compartment and highlight a beneficial role for the innate immune response in CNS repair.

Disclosures: S.L. Peterson: None. **K.A. Wong:** None. **C. Sun:** None. **A. Catacora:** None. **L.I. Benowitz:** None.

Poster

P165. Neuroinflammation: Microglia I

Location: Virtual Only

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Program #/Poster #: P165.09

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: Jones Foundation

Title: Transcriptome & Mitochondrial Analysis of ASD Children Compared to Healthy Controls

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Abstract: Autism has been described as a gastrointestinal and neurological condition that is represented by impairments in language and social situations. Recently reported increases in the occurrence of Autism Spectrum Disorder (ASD) have resulted in increased research into the environmental differences between areas with a high occurrence, as in the “Western” diet populations, and a more locally derived diet population, wherein the rate appears to be significantly less. Thirty autistic children (USA) ages 6 to 21, were sampled at three different intervals: prior to any intervention, post-xylitol intervention, and post-probiotic intervention. The sampling consisted of buccal swabs for Mitoswab testing, and saliva for metatranscriptomics to determine the entire range of microorganisms and the metabolome. In addition, a “Blue Zone” subject component that included 30 Colombian children, ages 6-16, and who were healthy and within normal behavior standards, also had buccal swabbing and salivary sampling performed for comparison. Their saliva samples were analyzed metatranscriptomically and artificial intelligence was utilized to discern important differences in the microbiome that included bacteria, archaea, fungi, and viruses, and microbial functions. Significant differences were noted, and also seen in the temporal data, demonstrating shifts that may have been precipitated by the interventions, with both the probiotics and the polyols. Particular bacterial strains were significantly more prevalent in the autism subject group including a strain that reduces neurotransmitters by enzymatic degradation. This provided supporting evidence to the hypothesis that the occurrence of autism may also be influenced by low Gamma Amino Butyric Acid (GABA) levels during development. Also, of interest was the un-expected finding that verbal skills dramatically increased in 6 of the 30 ASD subjects following xylitol supplementation.

Disclosures: M. Cannon: None. **S. Ganeshan:** A. Employment/Salary (full or part-time):; Religen Lab. **V. Alvarez:** None. **M. Vuyisich:** A. Employment/Salary (full or part-time):; VIOME. **G. Banavar:** A. Employment/Salary (full or part-time):; VIOME.

Poster

P173. Neuroinflammation: Animal Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P173.01

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: Kentucky Science & Engineering Foundation (KSEF-3937-RDE-020, JC)
NIEHS NIH (P30ES030283, JC for pilot grant).

Title: Genetic susceptibility and high fat diet-induced obesity underlie changes in neurobehavior, white matter microstructure, and hippocampal metabolites in C57Bl/6 mice

Authors: W. ZENG[1], T. CHU[1], J. GAO[1], E. GOZAL[1], R. JAGADAPILLAI[1], G. N. BARNES[2], ***J. CAI**[1];
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Abstract: Introduction: Accumulating evidence indicates that obesity-related cognitive deficits and mood disorders are associated with white matter (WM) impairment. Increased energy intake and reduced high-fiber food consumption (*e.g.* high fat diet, HFD), as well as genetic susceptibility, are major driving forces for the epidemic of obesity. It remains unclear whether HFD or genetic susceptibility plays a significant role in obesity-relevant brain complications. C57BL/6J mice, but not C57BL/6NJ mice, develop impaired glucose homeostasis control and reduced insulin secretion due to a natural deletion of nicotinamide nucleotide transhydrogenase exons, thus may be particularly proned to HFD-induced effects. Therefore, we adopted C57BL/6J and C57BL/6NJ mice to evaluate the gene-environment interaction on neurofunction, WM integrity, and brain metabolites. We hypothesized that diet and genetic susceptibility exert diverse effects on impairments of neurofunction, WM integrity, and brain metabolism. Methods: 3-week-old male C57BL/6J and C57BL/6NJ mice were fed a normal-fat diet or a HFD for 12 weeks or 24 weeks. Body weight (BW) and non-fasting blood glucose were monitored weekly. Fasting glucose and intraperitoneal glucose tolerance test were performed before sacrifice. Behavioral tests, including elevated plus maze, open field, Y-maze, object recognition test, and object location tasks, were conducted. WM microstructure and hippocampal metabolites were assessed by diffusion tensor imaging and magnetic resonance spectroscopy. All data are presented as mean ± SD and analyzed by two-way ANOVA with diet-by-strain interaction. Significance was set at $p<0.05$. Results: HFD exposure increased BW but reduced glucose intolerance in both strains, suggesting that the decrease in glucose intolerance was independent from their genetic differences. Short-term HFD exposure induced anxiety-like behavior only in C57BL/6J mice; while long-term HFD exposure caused cognitive decline in both strains. Decreased fractional anisotropy and increased radial diffusivity were observed in genu corpus callosum for both strains after long-term HFD exposure, suggesting a compromised WM integrity elicited by HFD. Both short- and long-term HFD exposures significantly elevated the myo-inositol and choline to creatine ratios in hippocampus for both strains, indicating glial activation and inflammation in the brain. Conclusions: HFD causes impairments in glucose intolerance, cognition, callosal WM integrity, and hippocampal metabolism. Diet-by-strain interaction exerts adverse effect on anxiety-like behavior.

Disclosures: W. Zeng: None. **T. Chu:** None. **J. Gao:** None. **E. Gozal:** None. **R. Jagadapillai:** None. **G.N. Barnes:** None. **J. Cai:** None.

Poster

P173. Neuroinflammation: Animal Models I

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Program #/Poster #: P173.02

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant T32HD094671 (AAE)
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NIH Grant 1R01NS114578-01 (EJB)
Jean and George Brumley Jr. Neonatal Perinatal Research Institute

Title: Neonatal sepsis leads to ependymal cell injury and a loss of subventricular zone neurogenesis

Authors: *A. A. EPSTEIN[1], B. RIKARD[2], V. JAIN[3], A. S. CHAO[1], K. ABDI[2], N. YOUNGE[1], K. PEGRAM[1], S. GREGORY[3], E. J. BENNER[1];
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Abstract: Preterm neonates are at high risk of brain injury leading to neurodevelopmental impairments. Prematurity-specific mechanisms of neonatal neurological disease are not well-defined and no therapies are available at this time. The subventricular zone (SVZ) is part of the germinal matrix critical for postnatal brain development. The SVZ includes multiciliated ependymal cells forming a barrier between the cerebral spinal fluid and neural stem cell niche. Notably, high rates of neurodevelopmental impairments are associated with complications of premature birth, including necrotizing enterocolitis and bowel perforations. Our laboratory has developed a clinically relevant rodent model where experimental bowel perforation on postnatal day 5 (P5) lead to neonatal sepsis that replicates brain injuries seen among preterm-born infants. These injuries include white matter injury as well as motor deficits. In this study we aim to describe neural stem cell niche responses to neonatal sepsis in the animal model. We employed single cell transcriptomics to demonstrate broad shifts in SVZ ependymal, glial, and neural progenitors in septic mice compared to littermate controls. qPCR was utilized to measure localized inflammatory cytokine production. Immunohistology (IH) of SVZ whole mount and primary ependymal cell cultures were used to validate observations from transcriptomic data. Single cell transcriptomics identified changes in the SVZ with increased frequency of inflammatory cells and decreased progenitor populations and ependymal cells following 2 days of sepsis. IH confirms sepsis-induced localized periventricular microglial activation. IH of SVZ whole mounts confirms ependymal cell degeneration using FoxJ1-EGFP tg-mice expressing green fluorescence protein in ependymal cells, as well as broad loss of cilia visualized using electron microscopy. Sepsis leads to increased mRNA expression of IL-1 and TNFalpha in SVZ compared to control littermates. NFκB-specific signal transduction and cytokine expression follows LPS stimulation of ependymal cells in culture, thus replicating ependymal cell activation in vitro. Our animal data suggests that inflammation and sepsis lead to an injury impacting the neural stem cell niche during a critical time of neurodevelopment. This previously undescribed injury may be an important molecular mechanism linking sepsis and poor neurodevelopmental outcomes in human infants.

Disclosures: A.A. Epstein: None. **B. Rikard:** None. **V. Jain:** None. **A.S. Chao:** None. **N. Younge:** None. **K. Pegram:** None. **S. Gregory:** None. **E.J. Benner:** None.

Poster

P173. Neuroinflammation: Animal Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P173.03

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Title: Alteration of glycosphingolipid levels and its implications on neuroinflammation

Authors: *A. CHEONG, S. RYAN, L. LEE, D. OFENGEIM, J. PROTO;
Rare and Neurologic Dis. Res., Sanofi, FRAMINGHAM, MA

Abstract: Glycosphingolipids (GSLs) are one of the most abundant lipid molecules in the CNS. Synthesized within the Golgi, the conversion of ceramide to glucosylceramide (GL1), the major precursor of complex GSLs, is the rate-limiting step of the GSL biosynthesis pathway. GSLs can modulate a variety of cellular functions including cell proliferation, signal transduction, and glial cell activation and GSL dysregulation is implicated in numerous human inflammatory and metabolic diseases. While murine models of CNS disease have been used to understand various aspects of lipid biology and nervous system homeostasis, human-cell based systems are critical for advancing therapeutics. In particular, a better understanding GSL biology in the context of neuroinflammation is critical for generating precise therapeutics for neuroinflammatory disorders. In this study, we employed an *in vitro* iPSC-derived triculture system composed of astrocytes, neurons, and microglia to examine the effect of acute inhibition of GL1 synthesis during homeostatic and inflammatory conditions. We performed RNA sequencing analysis to examine the impact of GSL modulation in both an acute setting and after immune activation by endotoxin. Following immune activation of the tri-culture system, the data show global transcriptional changes including the induction of interferon-related (*ISG15*, *SOCS1*) and HLA class II (*HLA-DRB1, HLA-DQB1*) genes. Multiple synaptogenesis related (*VAMP2*, *SYN1*, and *GRM2*) gene expression levels were also altered indicating an impact on neuronal functions. In the absence of inflammatory stimuli, acute reduction of GL1 synthesis had only modest effects on gene expression compared to vehicle alone. However, GL1 inhibition subsequent to endotoxin stimulation resulted in profound gene expression changes. This included marked downregulation of genes related to synaptogenesis and cytoskeletal signaling. Taken together, this data supports the use of human iPSC-derived triculture systems as models for interrogating cell-cell interactions under healthy (homeostatic) and disease (inflammatory) conditions. Furthermore, this study underscores the importance of including neuro-immune perturbations in these types of models. Acute reduction of GL1 reflected the importance of achieving balanced glycosphingolipid levels when exposed to inflammatory stimulus and further signified the importance of glycosphingolipids in maintaining a balanced intracellular metabolic homeostasis in the CNS. A better understanding of this relationship will give rise to new opportunities for developing precise therapeutics towards inflammatory and metabolic diseases.

Disclosures: A. Cheong: None. **S. Ryan:** None. **L. Lee:** None. **D. Ofengeim:** None. **J. Proto:** None.

Poster

P173. Neuroinflammation: Animal Models I

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Program #/Poster #: P173.04

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: CIHR
MS Society of Canada Doctoral Studentship

Title: Fibrosis of PDGFRβ+ cells following spinal cord demyelination is exacerbated with age

Authors: *B. LOZINSKI, Y. DONG, E. JELINEK, W. YONG;
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Abstract: The process of remyelination is the endogenous response in demyelinating diseases such as multiple sclerosis (MS). However, in people with MS remyelination is heterogeneous and often insufficient for functional recovery. Remyelination failure is known to be a result of aging and of the inhibitory lesion environment. Fibrosis of the CNS following immune mediated injury has recently been shown to effect the glial cell response. Fibroblasts are a possible source of CNS fibrosis and produce collagen that is capable of impairing oligodendrocyte progenitor cell migration. Other stromal cells found in the CNS, such as pericytes and smooth muscle cells, have also been suggested to both respond to demyelination and produce various other extracellular matrix (ECM) molecules including laminins and proteoglycans. However, it is unclear how fibrosis affects remyelination, and if fibrosis impairment of remyelination worsens with age. We hypothesize that PDGFRβ+ cells responding to demyelination form a fibrotic scar that is inhibitory to oligodendrocyte lineage cells and this is exacerbated with age. To test this hypothesis, we used the lysolecithin model to demyelinate the ventral white matter of the spinal cord in young (2-3 months) and middle-aged (12 months) mice. PDGFRβ+ cells formed a scar area by 7 days post induction (dpi); the scar area persisted throughout the lesion site up to 21 dpi. Cells of the oligodendrocyte lineage were scarce in areas occupied by PDGFRβ+ cells at all time points. In middle-aged mice, the scar area was larger than in young mice and corresponded with reduced oligodendrocyte repopulation. Oligodendrocyte lineage cells were determined by Olig2+PDGFRα+ for OPCs, and Olig2+CC1+ for mature oligodendrocytes. Collectively, age was found as a significant factor that influenced the response of PDGFRβ+ fibrotic cells following demyelination and the scar area they formed were less hospitable to oligodendrocyte lineage cells. Further studies are needed to determine how this response impairs remyelination and if it can be targeted for therapeutic intervention.

Disclosures: B. Lozinski: None. **Y. Dong:** None. **E. Jelinek:** None. **W. Yong:** None.

Poster

P173. Neuroinflammation: Animal Models I

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Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: The Gilbert Family Foundation
Dr. Miriam and Sheldon G. Adelson Medical Research Foundation

Title: Spontaneous Remyelination in the regenerating optic nerve

Authors: *S. DE LIMA[1], T. ZHANG[1], N. P. BISCOLA[2], N. ZHANG[4], L. A. HAVTON[3], P. A. ROSENBERG[5], L. I. BENOWITZ[6];
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Abstract: In the past 10 years, several groups have achieved appreciable optic nerve regeneration by activating the growth potential of mature retinal ganglion cells (RGCs). We previously showed that, with appropriate stimulation, some RGCs can extend axons from the eye to subcortical visual target areas (de Lima et al., PNAS, 2012), and that some regenerating axons become myelinated and re-establish the electrically excitable domains required for action potential propagation (Marin, de Lima et al., J.Neurosci, 2016). A more recent study reported an absence of spontaneous remyelination of regenerating axons in the optic nerve, and that stimulating myelination requires modulating GPR17 and microglia (Wang et al., 2020). However, we report here that after inducing regeneration (by combining PTEN deletion in RGCs with the growth factor oncomodulin + cAMP elevation), myelination of regenerating axons occurs spontaneously in the vicinity of the injury site and to a lesser extent distally. A longitudinal study using immuno-electron microscopy reveals that: 1) regenerating axons (identified by GAP-43 immunostaining or anterograde tracing with cholera toxin subunit B, CTB) show wraps of myelin as early as 2 weeks post-injury; 2) all regenerating axons reach the threshold diameter (≥ 300 nm) to become myelinated by 9 weeks, although not all axons that attain this diameter become remyelinated; and 3)regenerating axons that become myelinated exhibit a normal g ratio. We also investigated the dynamics of the oligodendroglial population from 3 days post-injury(dpi) to 42 dpi using as a marker Olig2, a transcriptional factor expressed by oligodendrocytes regardless of their state of differentiation. At the injury site, there is a75% drop in the number of Olig2+ cells 3 dpi, though at 42 dpi the number of Olig 2+cells is somewhat closer to normal. These results show that areas close to the injury site are more efficiently remyelinated than in distal areas, that some regenerating axons that reach the

appropriate threshold become myelinated, that remyelinated axons can reach a normal g ratio, and that the remyelination process observed in our study occurs in parallel with an increase in the total number of oligodendroglia.

Disclosures: S. de Lima: None. **T. Zhang:** None. **N.P. Biscola:** None. **N. Zhang:** None. **L.A. Havton:** None. **P.A. Rosenberg:** None. **L.I. Benowitz:** None.

Poster

P173. Neuroinflammation: Animal Models I

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Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIAA Grant P60AA011605
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Title: Pdgfr inhibitor regulates both ng2 glia and microglia: role of ng2 glia in ethanol-induced brain dysfunction

Authors: *J. ZOU[1], L. G. COLEMAN, JR[2], F. T. CREWS[3];
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Abstract: NG2 glia are a distinct glial cell population distributed throughout brain. They are identified by the expression of specific markers such as chondroitin sulfate proteoglycan 4 (CSPG4) and platelet-derived growth factor alpha (PdgfRα). However, the role of NG2 glia in ethanol-related brain dysfunction remains unknown. The present study used a PDGFR inhibitor CP673451 to dissect NG2 glia in contributing to ethanol-induced neuroimmune signaling and neuronal gene expression in a hippocampal-entorhinal cortex (HEC) slice culture. The concentration-dependent studies show that treatment of CP673451 at lower concentration (100nM) for 4 days induced specific loss of NG2 glia without disrupting the other types of cell signatures, for example: CSPG4 (Control:100±1; CP100nM4D:34±3); PdgfRα (Control:100±2; CP100nM4D:21±1); Iba-1 (Control:100±4; CP100nM4D:109±3); Sall1 (Control:100±2; CP100nM4D:96±0.3); GFAP (Control:100±3; CP100nM4D:145±1) and NeuN (Control 100±1; CP100nM4D:138±4). However, PDGFR inhibitor CP673451 at higher concentration (1000nM) depleted both NG2 and microglia markers and cell numbers as indicated by mRNA levels and IHC staining. To uncover the functional role of NG2 glia in ethanol-induced brain dysfunction, we treated the slices with lower concentration of CP673451 (100nM) for 4 days to induce the selective loss of NG2 glia, either copresence of ethanol (100mM) or pretreatment for 4 days followed by 4-day ethanol exposure. The results indicated that ethanol alone significantly

increased the expression of NG2 glia gene CSPG4 and PdgfRα. The loss of NG2 glia (both copresence and pretreatment) significantly reduced ethanol-induced neuroimmune signaling, for example (from an experiment with 4-day pretreatment of CP673451 followed by 4-day ethanol without CP673451): IL-1β (Control:100±5; CP100nMW4D:145±14; EtOH4D:187±8; E+CP100nMW4D:55±3, $p<0.001$). The loss of NG2 glia also significantly blocked ethanol-induced expression of neuronal genes (NR2A, NR2B and PSD95). Interestingly, myelin-associated protein genes such as PLP and MBP were markedly potentiated after NG2 ablation in response to 4-day ethanol. Together, these results reveal the unique role of NG2 glia in ethanol-induced neuroimmune signaling and neuronal activity. Our results also suggest that PDGFR inhibitors may offer a unique tool to selectively explore the physiological and pathological role of NG2 glia during ethanol exposure.

Disclosures: J. Zou: None. **L.G. Coleman:** None. **F.T. Crews:** None.

Poster

P173. Neuroinflammation: Animal Models I

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Program #/Poster #: P173.07

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NHGRI R35 HG010718

Title: Tet1 isoform-mediated regulation of immune response pathways in neural cells.

Authors: E. JASHIM[1], G. LUPICA-TONDO[2], C. B. GREER[3], E. GAMAZON[4], ***G. A. KAAS**[4];
[1]Belmont Univ., Nashville, TN; [3]Pharmacol., [4]Genet. Med., [2]Vanderbilt Univ., Nashville, TN

Abstract: Ten eleven translocation (TET) dioxygenase enzymes have emerged as key regulators of inflammation—both under normal and pathological conditions. Recently, we reported that two isoforms of *Tet1* are co-expressed in the adult mammalian brain, and their disruption results in opposite effects on physiology and cognition (Greer *et al*. 2021). In addition, we find that suppression of the full length, canonical isoform ($Tet1^{FL}$) in neurons induces a robust immune response, suggesting it might normally function to suppress neuroinflammation in the brain. To investigate these findings further, and in a more tractable system, we have now generated *Tet1* isoform mutant Neuro2A (N2a) cell lines using CRISPR-Cas9 editing. We find that loss of $Tet1^{FL}$ expression in N2as is sufficient to upregulate many immune-related genes, including *Nfkb1, Nfkb2, Tnfa, and IL-7.* Surprisingly, we also find that *pan-Tet1* loss reduces the expression of these same genes, whereas loss of the opposite isoform, *Tet1 Short* ($Tet1^{S}$), only mildly changes their transcript levels. Together, our results reveal that each isoform serves a unique, oppositional role in the regulation of inflammation in neural cells. Moreover, given that

many immune pathways contribute to neural physiology, these data suggest that *Tet1* isoforms may influence brain function, in part, through the epigenetic regulation of immune response genes.

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Poster

P173. Neuroinflammation: Animal Models I

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Program #/Poster #: P173.08

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

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Title: SRI-42127, a novel small molecule inhibitor of the RNA Regulator HuR, potently attenuates glial activation in a model of neuroinflammation

Authors: *R. CHELLAPPAN, A. GUHA, Y. SI, T. KWAN, B. NABORS, N. FILIPPOVA, X. YANG, A. S. MYNENI, S. MEESALA, A. S. HARMS, P. H. KING;
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Abstract: Glial activation with production of pro-inflammatory mediators is a major driver of disease progression in neurological conditions ranging from acute traumatic injury to chronic neurodegenerative disease. Our prior work has shown that the RNA regulator HuR plays a major positive role in regulating glial production of pro-inflammatory mediators through its interaction with adenine- and uridine-rich sequences (ARE) in the 3' untranslated region (3' UTR) of the mRNA. Upon binding to the ARE, HuR promotes RNA stabilization and increases translational efficiency of the transcript. We have developed a small molecule inhibitor of HuR, SRI-42127, that blocks its ability to dimerize and translocate to the cytoplasm, two steps necessary for proper function. Here, we test the ability of this inhibitor to attenuate glial activation and neuroinflammatory responses using lipopolysaccharide (LPS) as a model of neuroinflammation. Methods: Cultured murine microglial and astroglial cells were stimulated with LPS and treated with SRI-42127. Cellular localization of HuR was determined by single-cell flow cytometry and immunohistochemistry (IHC). Cytokine production was measured by qPCR and ELISA. RNA kinetics were measured in LPS-stimulated BV2 microglial cells after actinomycin D treatment. A luciferase-based reporter assay was used to assess promoter function of select cytokine genes. Glial migration was assessed with a transwell assay. For in vivo assessment, mice were injected with LPS intraperitoneally (i.p.) followed by SRI-42127 (10 mg/kg). Brains were assessed for

cytokine mRNA expression by qPCR, neutrophil and monocyte infiltration by flow cytometry, and microglial activation by Iba1 fluorescence intensity with IHC. Results: SRI-42127 blocked HuR cytoplasmic translocation in glial cells and the concomitant production of pro-inflammatory cytokines including IL-1β, IL-6, TNF-α, iNOS, CXCL1, and CCL2. There was marked suppression of TNF-α and IL-1β promoter activity and a mixed effect on RNA kinetics of select cytokines. Glial cell migration was suppressed. In vivo, SRI-42127 blocked induction of cytokine mRNAs and the infiltration of neutrophils and monocytes in all regions of the brain. Microglial activation was attenuated. Conclusion: SRI-42127 suppresses neuroinflammatory responses by activated glia through its inhibitory effect on HuR function. Systemic administration of SRI-42127 suppresses CNS neuroinflammatory responses including the recruitment of peripheral immune cells into the CNS. These findings open up a new avenue of therapeutic development for the many neurological diseases driven by neuroinflammation.

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Poster

P173. Neuroinflammation: Animal Models I

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Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

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Duke Microbiome Center Grant
Duke University Anesthesiology Research funds

Title: Sting signaling in peripheral sensory neurons confers protective immunity against listeria monocytogenes infection

Authors: *A. S. ANDRIESSEN[1], S. BANG[1], A. MCGINNIS[1], R.-R. JI[1,2,3], C. R. DONNELLY[1,4];
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Abstract: STING signaling in peripheral sensory neurons confers protective immunity against *Listeria monocytogenes* infection
Amanda S. Andriessen, Sangsu Bang, Aidan McGinnis, Ru-Rong Ji, Christopher R. Donnelly
Listeria monocytogenes (*L.m.*) is a bacterial pathogen responsible for the foodborne illness listeriosis, which is among the most frequent causes of death and is responsible for the highest

hospitalization rate among all foodborne illnesses. Importantly, *L.m.* is among the most common causes of bacterial meningitis and/or meningoencephalitis among newborns, and once *L.m.* crosses into the central nervous system (CNS), lethality exceeds 50%, even with aggressive medical treatment. Notably, the mechanism used by *L.m.* to bypass the blood-brain barrier to enter the CNS is poorly understood. STING is a sensor of host-derived double-stranded DNA and bacterially derived cyclic dinucleotides. *L.m.*, an intracellular pathogen, potently activates STING in immune cells, which initiates or amplifies a protective immune response. Interestingly, we recently demonstrated that STING is highly expressed by pain-sensing peripheral sensory neurons (nociceptors). Given that the peripheral afferents of nociceptors are present within virtually all barrier tissues and are frequently used by viral pathogens as a conduit to enter the CNS, we hypothesized that *L.m.* may infect nociceptors, and STING signaling in nociceptors may confer protective immunity against *L.m.* To test this hypothesis, we co-cultured sensory neurons with fluorescently labeled *L.m.* and performed live-cell imaging, which revealed robust infection of nociceptors by *L.m. in vitro*. Additionally, administration of *L.m.* in mice leads to robust infection of nociceptors *in vivo*. To evaluate whether STING signaling in peripheral sensory neurons confers protective immunity against *L.m. in vivo*, we performed a *L.m.* challenge experiment in signaling-deficient STING global knockout mice (STING-goldenticket mice, or STING-gKO) and mice lacking STING selectively in peripheral sensory neurons ($STING^{fx/fx}$; $Na_v1.8$-Cre; STING-cKO), as well as their wildtype (WT) littermates. Interestingly, both STING-gKO and STING-cKO exhibited significantly worse survival compared to WT controls, but not relative to one another. Collectively, these data suggest that STING signaling in peripheral sensory neurons confers protective immunity against *L.m.*, and ongoing work is aimed at uncovering the mechanism responsible for these effects.

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Poster

P173. Neuroinflammation: Animal Models I

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Program #/Poster #: P173.10

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

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Korea Health Technology R&D Project (HI21C0572) through the Korea Health Industry Development Institute (KHIDI), funded by the Ministry of Health & Welfare, Republic of Korea

Title: Effect of neurokinin 1 (NK 1) receptor antagonist on chronic osteoarthritic pain in MIA-induced OA rats

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Abstract: Osteoarthritis (OA) is a progressively degenerative joint disease, causing structural degradation of articular cartilage and adjacent connective tissue elements. This eventually induces pain and loss of joint function. Despite the considerable medical need, treatment options for the pain are insufficient. Substance P (SP) is well known to involve the induction of neurogenic inflammation and pain transmission in arthritis through activation of neurokinin 1 (NK1) receptor. The aim of this study is to investigate and compare the effect of two different neurokinin 1 (NK1) receptor antagonist, GR82334 and L-732,138 on arthritic pain in rats with MIA-induced OA.
Monosodium iodoacetate (MIA 4 mg/50 μl) was injected into the right knee joint cavity of Sprague-Dawley rats (180-200g) to induce OA. Two different NK1 receptor antagonist, GR82334 (10 μM diluted with 30 μl of saline) and L-732,138 (10 μM diluted with 30 μl of saline), was injected into the ipsilateral knee joint cavity using 30-gauge insulin syringe at 7 and 14days after MIA injection. Before and after NK1 antagonist administration, paw withdrawal threshold (PWT), knee joint diameter, knee bending test, and weight bearing were measured for determining hyperalgesia, edema, movement evoked pain and functional difference of hind limbs. Maximal Possible Effect (MPE) from the value of PWT and Area Under the Curve (AUC) of MPE were calculated to compare the effect of NK1 receptor antagonist between the groups. PWT significantly increased after administration in both GR 82334 group and L-732,138 group, while GR82334 group showed more rapid and dramatic analgesic effect compared to that of L-732,138 group in MPE and AUC graph. However, both GR82334 and L-732,138 did not reversed the reduction of weight load of inflamed leg and increased knee joint score on the ipsilateral hind-limb after MIA injection.
The present data showed that two different NK1 receptor antagonist, GR82334 and L-732,138 alleviate mechanical hyperalgesia in hind paw of rats with MIA induced OA through mediating the activity of the peripheral nociceptor, decreasing the excitability of the spinal cord. The finding suggests NK1 receptor antagonist as a potential mediator and drug target in OA, but each NK1 receptor antagonist has its characteristics and results in different effects under same condition, despite the same function.

Disclosures: H. Yoo: None. **M. Kwon:** None. **J. Kim:** None.

Poster

P173. Neuroinflammation: Animal Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P173.11

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH NINDS R01 1R01NS089688
R01NS062019

Title: The effects of daily melatonin administration on the tissue reactions to neural implants

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Abstract: Intracortical microelectrode arrays that record neural activities are key component of brain computer interface and neurophysiological studies. Despite their potential, recording quality often suffers from low yield and signal degradation. Host immune cells respond to the implant, encapsulating the foreign material and generating reactive oxygen and nitrogen species (RONS). The RONS serve as signals to other cells, amplifying the response, while also damaging both the device and native neurons. Pharmacologic methods of improving the long-term electrode performance by controlling the extent of RONS in the tissues have been investigated. One particularly interesting antioxidant is melatonin, a natural hormonal antioxidant which can modulate cellular responses through native receptors, while also having the ability to directly scavenge free radicals. Daily melatonin administration before and after electrode insertion resulted in enhanced electrode performance over 16 weeks. However, the short-term effects of melatonin have not been investigated. In this works, we use a combination of approaches including two photon microscopy, post mortem histology, and gene sequencing were employed to provide a better mechanistic understanding how melatonin affects the host tissues after short term implantations. In the two photon experiments, mice expressing green fluorescent protein in their microglia were given daily melatonin injections while the reactions of the microglia to the implant were visualized in real time. Characteristically, microglia attempted to encapsulate the implant. However, the microglial responses were slowed when melatonin was administered, with probe coverage decreased from 51.8% in control animals to 25.1% in treated animals at four days post implant. Postmortem histology and gene sequencing were used to examine the responses of the brain to the implant after one week. Staining of the tissues shows reduced astrocytic activation, decreased apoptosis, and elevated neural counts near the electrodes when the animals were treated with melatonin. We are currently underway in our attempts to determine the gene expression levels for implanted mice treated with melatonin or vehicle.

Disclosures: K. Woeppel: None. **Q. Yang:** None. **D. Krahe:** None. **T. Cui:** None.

Poster

P178. Neuro-Oncology: Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P178.01

Topic: B.13. Neuro-Oncology

Support: Rowan University's Camden Health Research Initiative

Title: Hypoxic regulation of gene expression in T98 glioblastoma multiforme cells by RNA sequencing identifies IRE1 as a target for therapy

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Abstract: Glioblastoma multiforme (GBM) is the most common primary brain cancer and carries a very poor prognosis. Intra-tumoral hypoxia is associated with dysregulation of various genes, including those involved in carbon metabolism, angiogenesis, and immunosuppression. These alterations characterize a subpopulation of cells that is particularly aggressive and resistant to multimodal therapy. IRE1, an endoplasmic reticulum stress sensor involved in the unfolded protein response, has been implicated in the increased angiogenic activity demonstrated in other cancer cell lines subject to hypoxia, including U87 GBM cells (Drogat et al., 2007). In our study, human T98G cell lines were cultured in a humidified incubator at 37° C and 5% CO_2 and were grown in normoxia (21% O_2) or hypoxia (95% N_2, 5% CO_2) for 72 hours. Three independent cultures (n=3) were used for each condition. Total RNA was harvested, and global gene expression was evaluated via RNA sequencing. RNA-seq alignment files were generated using the Illumina DRAGEN Bio-IT Platform (version 3.3.7), and the FASTQ alignment files were processed through Rsubread in Bioconductor package (version 2.0.0) to generate feature counts per sample. These counts were then processed through both DESeq2 (version 1.26.0) for differential expression analysis. *P* values were calculated using gene set enrichment analysis (GSEA) with Bonferroni correction. Gene Ontology and KEGG analyses were performed to identify processes and pathways enriched in hypoxia. The DrugBank database was queried to identify molecules targeting gene products upregulated in hypoxia. Hypoxia in T98G cells led to upregulation of genes implicated in multiple biological processes including canonical glycolysis, focal adhesion, and extracellular matrix reorganization. We document 690 genes and 11 associated KEGG pathways that demonstrated significant enrichment ($p \leq 0.01$) in hypoxia, including the IRE1-mediated unfolded protein response. DrugBank database analysis identified five molecules targeting gene products upregulated in hypoxic T98G cells: tenecteplase (5

targets), succinic acid (7 targets), artenimol (13 targets), copper (22 targets), and fostamatinib, a tyrosine kinase inhibitor targeting the IRE1 protein encoded by *ERN1*. In conclusion, this gene discovery study showed hypoxic upregulation of genes involved in multiple pathways associated with neoplastic behavior. The IRE1 protein showed enrichment in hypoxic T98G cells, corroborating results in another GBM cell line and suggesting a potential therapeutic target. The IRE1 inhibitor fostamatinib may hold therapeutic potential and warrants further preclinical study.

Disclosures: B. White: None. **Y. Liu:** None. **H. Hakonarson:** None. **R.J. Buono:** None.

Poster

P178. Neuro-Oncology: Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P178.02

Topic: B.13. Neuro-Oncology

Support: PRODEP UDG

Title: G protein-coupled estrogen receptor blockade decreases cell viability and increases apoptosis of glioblastoma cells

Authors: *C. E. GUTIERREZ-ALMEIDA[1], J. M. DUEÑAS-JIMENEZ[2], S. H. DUENAS-JIMENEZ[1], A. SANTERRE[3];
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Abstract: Background: Glioblastoma remains one of the most difficult to treat and worst prognosis tumors. Patients who undergo the standard treatment of surgical resection, chemotherapy, and radiation have 16 to 18 months of survival and a high recurrence rate. Different reports have shown that female patients have a survival advantage over males who have a higher glioblastoma incidence. The reason for these sex differences is believed to be associated with estrogens. G protein-coupled estrogen receptor (GPER) is a seven-transmembrane-domain receptor. This receptor participates in the estrogen rapid non-genomic signaling such as the activation of mitogen-activated protein kinase/extracellular signal-regulated kinase (MAPK/ERK), phospholipase C (PLC), and phosphatidylinositol 3-kinase (PI3 K) pathways, crucial pathways for cancer development. Rat tumor Leydig cells (R2C) express GPER and suffer apoptosis after GPER agonist G1 treatment, in vitro, and in vivo. GPER is also expressed in human squamous oral carcinoma cell lines (SCC4 and HSC-3), and the antagonist G15 induces apoptosis through caspases activation. Nevertheless, it is presently unclear the role of GPER in Glioblastoma. Objective: This study aimed to evaluate the role of GPER in C6 glioblastoma cells viability and apoptosis after the administration of GPER agonist (G1) and its antagonist (G15). Methods: C6 rat glioblastoma cell line was treated with a GPER agonist (G1,

10nM, for 48 hours) and antagonist (G15, 10μM, for 48 hours). Cell viability was evaluated using an MTT assay. To determine apoptosis, we evaluated caspase-3 expression by immunofluorescence and ELISA. Results: MTT assay showed C6 cell viability decreased after treatment with G1 in combination with G15 compared to control. Furthermore, G1, in combination with G15, significantly increased the percentage of C6 caspase-3 immunostaining cells and G15 (10μM) alone compared to control. Also, caspase-3 protein expression increased significantly in C6 cells treated with G1 and G15 compared to control. Conclusion: The GPER antagonist (G15) decreases cell viability while increasing caspase-3 protein levels. Our results demonstrate that GPER blockade promotes C6 cells apoptosis, thus showing the role of this receptor in cell death in glioblastoma cells.

Disclosures: C.E. Gutierrez-Almeida: None. **J.M. Dueñas-Jimenez:** None. **S.H. Duenas-Jimenez:** None. **A. Santerre:** None.

Poster

P178. Neuro-Oncology: Molecular Mechanisms I

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Program #/Poster #: P178.03

Topic: B.13. Neuro-Oncology

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Margaret Q. Landenberger Research Foundation
De Drie Lichten

Title: Knocking out miR-21 cripples glioma

Authors: *L. NIELAND[1], T. S. VAN SOLINGE[1], P. S. CHEAH[5], L. M. MORSETT[2], J. E. KHOURY[2], J. I. RISSMAN[3], B. P. KLEINSTIVER[3], M. L. D. BROEKMAN[6], X. O. BREAKEFIELD[4], E. R. ABELS[7];
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Abstract: Non-coding RNAs, including microRNAs, support the progression of glioma. miR-21 is a small, non-coding transcript, involved in regulating gene expression in multiple cellular

pathways, including the regulation of proliferation. High expression of miR-21 has been shown to be a major driver of glioma growth. Manipulating the expression of microRNAs is a novel strategy in the development of therapeutics in cancer. In this study we used CRISPR genome editing technology to disrupt the miR-21 coding sequence in glioma cells. We found that targeted deletion of miR21 resulted in the upregulation of many downstream target mRNAs involved in proliferation. Phenotypically, miR21 deficient glioma cells showed reduced migration, invasion and proliferation *in vitro.* In an immunocompetent mouse model, miR-21 knockout tumors showed reduced growth resulting in an increased overall survival. In summary, we show that by knocking out a key miRNA in glioma, these cells have decreased proliferation capacity both *in vitro* and *in vivo*. Overall, we identified miR-21 as a potential target for CRISPR-based therapeutics in glioma.

Disclosures: L. Nieland: None. **T.S. van Solinge:** None. **P.S. Cheah:** None. **L.M. Morsett:** None. **J.E. Khoury:** None. **J.I. Rissman:** None. **B.P. Kleinstiver:** None. **M.L.D. Broekman:** None. **X.O. Breakefield:** None. **E.R. Abels:** None.

Poster

P178. Neuro-Oncology: Molecular Mechanisms I

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Program #/Poster #: P178.04

Topic: B.13. Neuro-Oncology

Support: IMMUN-HUB “Sviluppo di nuove molecole di seconda generazione per immunoterapia oncologica”, CUP E51B19000550007 – Call HUB Ricerca e Innovazione
co-funded by POR FESR 2014-2020 (Regional Operational Programme, European Regional Development Fund)

Title: Selective HDAC6 inhibitor ITF3756 down-regulates PD-L1 in Gli36R cells, boosting the PBMCs-mediated immune response

Authors: *L. TAIAROL[1], M. KRAVICZ[1], M. RIZZUTO[2], L. BARBIERI[2], M. COLOMBO[2], C. STEINKÜHLER[3], E. MESSUTI[1], A. BENTIVEGNA[1], F. RE[1];
[1]Med. and surgery, [2]Biotechnologies and biosciences, Univ. of Milan-Bicocca, Milan, Italy; [3]Italfarmaco S.p.A, Cinisello Balsamo, Italy

Abstract: Glioblastoma multiforme (GBM) is a severe IV grade astrocytoma with high degree of recurrence attributable to GBM stem cells (GSCs) and to the immunosuppressive microenvironment. Immunotherapy gained a lot of attention due to its ability to prevent metastatization and relapse better than conventional therapies. Immune checkpoint (ICs) such as Protein Death-1 and its ligand (PD-L1) are negative modulators of T-cells and Natural killer cells

(NKs) immune surveillance, being able to switch off the immune response eliciting immune escape of GBM. Histone deacetylases inhibitors (HDACis) are promising epigenetic modulators emerged as a therapy for various malignancies by modulating the expression of ICs. Here, we have characterized a nanoformulation of ITF3756, a selective HDAC6 inhibitor, on two *in vitro* models of GBM: Gli36ΔEGFR-2 (Gli36R) cells, which are resistant to chemotherapy and carry an EGFRvIII variant mutation present in 27-54% of GBMs, and PZG7, GSCs isolated from human biopsy. Results were compared with normal human astrocytes (NHA) as controls. ITF3756, dissolved in 7.5% PBS/DMSO, was formulated in liposomes (LIP-ITF) composed of cholesterol and sphingomyelin 1:1 added with 1.5 mol% of 1,2-dimyrystoil-snglycero-3-phosphate and prepared by extrusion procedure. LIP-ITF had a diameter of 125.3±1.5 nm with a PDI <0.2 and a surface charge -22.02±3.06 mV which make them stable and homogeneous drug vehicles. The cytotoxic effect of LIP-ITF was evaluated by MTT assay. Results showed that after 24 h both PZG7 and NHA were quite tolerant to LIP-ITF until the dose of 25 μM, while Gli36R started to be sensitive from 10 μM. Since HDACs relate to ICs expression, we have investigated the effect of LIP-ITF on PD-L1 in Gli36R at the doses of 25 and 50 μM using Tubastatine A, a known HDAC6 inhibitor, as a positive control. The mean reduction after 24 h of PD-L1 was about 70%, indicating that this drug might be an excellent candidate for immunotherapy. To confirm this hypothesis, we performed a live-imaging experiment using the Operetta System that involved PBMCs (T-cells and other mononuclear immune cells isolated from healthy donors' peripheral blood) co-cultured with Gli36R cells treated with 25 μM LIP-ITF for 24 h. Intriguingly, the treatment amplified the immune response of PBMCs against cancer cells in respect to untreated cells and we can speculate this effect was mainly due to the decrease of PD-L1 expression. Taken together, these unprecedented results lay the foundations for further studies aimed to better define the potential of ITF3756 for immunotherapy of GBM and its formulation in multifunctional LIP to boost blood-brain barrier crossing and tumor targeting.

Disclosures: **L. Taiarol:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Italfarmaco S.p.A. **M. Kravicz:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Italfarmaco S.p.A. **M. Rizzuto:** None. **L. Barbieri:** None. **M. Colombo:** None. **C. Steinkühler:** A. Employment/Salary (full or part-time):; Italfarmaco S.p.A. **E. Messuti:** None. **A. Bentivegna:** None. **F. Re:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Italfarmaco S.p.A.

Poster

P178. Neuro-Oncology: Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P178.05

Topic: B.13. Neuro-Oncology

Title: Cytotoxicity effects of mesoporous SiO_2 nanoparticles functionalized with folic acid and loaded with temozolomide in glioma cell lines.

Authors: *C. RODRÍGUEZ-PÉREZ[1], M. MANRÍQUEZ-RAMÍREZ[2], A. GONZALEZ-AGUILAR[1], E. ORTIZ-ISLAS[1];
[1]Natl. Inst. of Neurol. and Neurosurg., MEXICO CITY, Mexico; [2]ISIQIE, Natl. Polytechnic Inst., Mexico City, Mexico

Abstract: Glioblastoma multiforme is known as the most frequent primary brain tumor, which develops and originates in the non-neuronal glial cells of the brain. Despite the advances in the treatment, the median survival of patients is of 15 months. Currently, temozolomide is a first-line chemotherapy drug concomitantly with radiotherapy. Temozolomide, is a derivative of imidazotetrazine, it can cross the blood-brain barrier and at physiological pH, is converted by hydrolysis to the active form and then it is converted to 5-aminoimidazole-4-carboxamide and methyl diazonium ions. The formation of these methyl diazonium ions causes methylation at the guanine sites of the N7 and O6 atoms and the adenine site of the N3 atoms, leading to DNA double helix breakage, cell cycle interruption, and consequently cell death caused by cytotoxic DNA damage. The amount of temozolomide reaching the brain tumor is low (2%) relative to the total dose of drug circulating in the blood and the half-life is noticeably short (approximately 1.8 h). An alternative approach to improve the treatment of gliomas is use nanomaterials as drug carriers. The surface modification of nanomaterials has allowed them to preferentially accumulate in tumors and release drugs, making them interesting candidates for treat glioblastoma. In this study, we evaluated the cytotoxicity *in vitro* of temozolomide release of SiO_2 nanoparticles functionalized with folic acid in different human cell lines of gliomas (LN18, U87 and M059K) using MTT assay. The aim of functionalizing the silica nanoparticles with folic acid is that the nanoparticles can differentiate between cancer cells and healthy cells. The cytotoxic effect of temozolomide-SiO_2 nanoparticles was dose-dependent and clearly increased the cytotoxic effect of the drug alone. These results, suggest that these silica nanoparticles functionalized could have an important role in glioblastoma cancer treatment avoiding probably many side effects of chemotherapeutic drugs.

Disclosures: C. Rodríguez-Pérez: None. **M. Manríquez-Ramírez:** None. **A. Gonzalez-Aguilar:** None. **E. Ortiz-Islas:** None.

Poster

P178. Neuro-Oncology: Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P178.06

Topic: B.13. Neuro-Oncology

Support: PRIN 2017-2017XJ38A4_003

Title: Tumor-associated glial cells, macrophages, and extracellular matrix. Morpho-molecular dynamics in the mouse GL261-C57/Bl6J model of glioblastoma

Authors: A. VIRTUOSO, I. VISCOVO, R. CIRILLO, G. CIRILLO, ***M. PAPA**, C. DE LUCA; Dept. of Mental, Physical Hlth. and Preventive Med., Univ. della Campania Luigi Vanvitelli, Napoli, Italy

Abstract: Glioblastoma multiforme (GBM) is the most common primary brain tumor, associated with poor prognosis: scarce chances of long-term survival and rapid clinical worsening with state-of-the-art treatments. The microenvironment in the GBM comprehends as key cellular components microglia, macrophages, and astrocytes. Recent studies showed their involvement in nurturing tumor cells and grant GBM invasiveness. In the present study, we aim to characterize the time-related changes in the GBM landscape, using a syngeneic mouse model of primary GBM. GL261 glioma cells were injected in the right striatum of immuno-competent C57Bl6J mice and animals were sacrificed after 7, 14, and 21 days (7D, 14D, 21D). The tumor development was assessed through 3D tomographic imaging.Brain tissues were processed using immunohistochemistry, immunofluorescence, and western blotting. Our results showed how tumor progression temporally changes the morphology of astrocytes and microglia/macrophages. In the early stages, the proliferating tumor (Ki67+) appeared with a speckled distribution. The proliferating elements triggered astrocyte activation (GFAP+, glial fibrillary acidic protein) surrounding the GBM cells. The phagocytic cells, microglia, and macrophages (Iba1+, Ionized calcium-binding adaptor molecule 1), on the other hand, were scarcely represented and the brain architecture was conserved. A dense multilayer of reactive astrocytes with a morphological gradient of activation towards the well-defined tumor bulk is evident at 14D. At the same timepoint, we observed the increase of the phagocytic elements in the peritumoral area. The cellular changes may prompt the release of extracellular matrix (ECM) proteins and proteases to favor the GBM invasiveness locally and across the contralateral hemisphere. Metalloproteinase 9, fibulin, and tenascin C expression were affected at this stage. Accordingly, CD133+ glioma stem cells migrated out of the bulk at 21D, while the necrotic primary lesion was being infiltrated by macrophages. Protein expression of the specific microglial marker TMEM-119 (transmembrane protein 119) was downregulated, suggesting diminished activity of tumor-associated-resident microglia and increased contribution of circulating invading macrophages. The present study emphasizes the role of functional changes in the microenvironment during the GBM progression, fostering the studies on novel multi-targeted, time-dependent therapies in an experimental model similar to the human disease.

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Poster

P178. Neuro-Oncology: Molecular Mechanisms I

Location: Virtual Only

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Program #/Poster #: P178.07

Topic: B.13. Neuro-Oncology

Support: UCSF School of Medicine Summer Explore Grant
NIH Grant 5K08NS110919-03

Title: Tsp-1 mediated glioma-network integration in high grade glioma

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Abstract: Glioma exists in the complex neural circuitry of the brain, making the interface between neoplastic and healthy neurons and glia potentially damaging to long range neural networks and stimulatory to tumor growth. Thrombospondin-1 (TSP-1), an astrocyte derived neurogenic factor expressed by glia of the normal brain, has been found to be upregulated in intratumoral regions with high network functional connectivity (HFC). This modified cell signaling represents cancer cell hijacking of normalphysiology with direct impact on tumor biology. There is emerging evidence that neuronal activity influences glioma proliferation and gliomas promote neuronal hyperexcitability. In humans, we have recently shown that bidirectional cellular interactions between gliomas and neurons alter cognitive circuit dynamics and ultimately patient survival. Previously, a subpopulation of human high-grade glioma cells which are enriched for tumor cells with synaptogenic potential were identified (HFCIDHwtGBM). We studied the mechanisms of TSP1 signaling in three different established glioma models (1) HFC-IDHwtGBM hippocampal neuron co-culture, (2) HFC-IDHwtGBM + induced neuron organoids, (3) patient derived xenografts (PDX) for in vivo GCaMP calcium imaging. This project aims to test the hypothesis that increased TSP-1 secretion from HFC-IHDwtGBM cells plays a central role in the maintenance of an invasive and proliferative tumor phenotype when compared with control conditions. Our results show that TSP-1 silencing using a lenti shRNA knockdown construct decrease the proliferative and invasive phenotype of HFC-IHDwtGBM cells evidenced by the reduced expression of Ki67 and MET markers, respectively in hippocampal neuron co-culture and PDX tissue specimens. Further, the HFC-IDHwtGBM cells lacking TSP-1 exhibit a less synaptogenic and integrative potential tested via hippocampal neuron co-culture and induced neuron organoid models. We hope our study guides future work focused on preventing the infiltration of tumor cells into healthy brain tissues.

Disclosures: M. Negussie: None. **S. Krishna:** None. **S. Hervey-Jumper:** None.

Poster

P178. Neuro-Oncology: Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P178.08

Topic: A.03. Stem Cells and Reprogramming

Title: Primary brain tumour cells live morphological analysis and association with cancer stem cell biomarker SOX2 and CD133

Authors: *Z. HUMA[1,3], I. ABBAS[4,5], I. KHAN[4], D. HUSSEIN[6], S. WAZIR[2], S. AYUB[7]; [1]IBMS, [2]Khyber Med. Univ., Peshawar, Pakistan; [3]Anat., Rehman Med. Col., Peshawar, Pakistan; [4]Khyber medical university, Peshawar, Pakistan; [5]Anat., Northwest Sch. of Med., Peshawar, Pakistan; [6]King Abdulaziz Univ., Jeddah, Saudi Arabia; [7]Hayatabad Med. Complex, Peshawar, Pakistan

Abstract: ABSTRACT: Primary Brain Tumour Cells Live Morphological Analysis and association with cancer stem cell biomarker 'SOX2' and CD133**Background:** Despite recent advancements mortality rates remain high for brain tumour patients in Pakistan which being a developing nation lacks proper healthcare facilities. Majority of brain tumour patients suffer greatly and eventually die despite standard methods of treatment used. To understand brain tumour complexities and develop effective strategies for prevention and treatment proper documentation and research in this field is imperative. Primary cultures developed from patient derived tissues are indispensable tools for studying the biology of disease and for carrying out pre-clinical testing. Developing primary brain tumour cell lines from Peshawar based patients to study the biology of brain tumours will set the framework for research in the field of neuro-oncology for our people as there is paucity of research in this field in our country.**Methods:** Primary brain tumour cell lines were established with tumour samples from Hayatabad medical complex and Northwest general hospital. The tissue samples were processed in the cell culture laboratory and dissociated tumour cells were cultured in growth media and incubated at 37°C in 5% CO2.The attached monolayer cells were analyzed for various morphologies. Immunofluorescence was performed on the cultured cells as well as original tumour tissue for detection of stem cell biomarker SOX2.**Results:** Cell lines were developed from three samples of primary brain tumours which included two gliomas and one meningioma. The cell counts over a four-week period revealed that glial cell type was the dominant morphology at end of 4th week. SOX2 expression was found in all cells and tissues but higher in cells with a mean percentage of 45% as compared to tissue having mean percentage expression of 11.3%.**Conclusion:** Primary cultures from brain tumour developed. Morphological analysis of the primary brain tumour cultures demonstrated the presence of different cell types with glial type being dominant in our 3 samples. SOX2 was detected in cultured cells and original tumour tissue in all samples. No apparent correlation was found between dominant morphology and SOX2 expression with tumour aggressiveness.Keywords Primary brain tumours, Cell culture, Brain tumour stem cells, SOX2.

Disclosures: Z. Huma: A. Employment/Salary (full or part-time):; Rehman Medical college. **I. Abbas:** None. **I. Khan:** A. Employment/Salary (full or part-time):; khyber medical university. **D. Hussein:** A. Employment/Salary (full or part-time):; King Abdulaziz university. **S. Wazir:** A. Employment/Salary (full or part-time):; khyber medical university. **S. Ayub:** A. Employment/Salary (full or part-time):; Hayatabad medical complex.

Poster

P198. Alzheimer's Disease: Genetics, Genomics, and Other Omics Approaches VII

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P198.01

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH EY04067
VA Merit Review

Title: Neurodegeneration of retinal ganglion cells in a mouse model of Alzheimer's Disease

Authors: L. PÉREZ DE SEVILLA MÜLLER[1], A. M. CHEN[1], *N. BRECHA[1,2];
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Abstract: Introduction: Visual alterations are common in AD patients with a substantial impact on the quality of life. Retinal abnormalities in AD include loss of retinal ganglion cells (RGCs), including the intrinsically photosensitive RGCs (ipRGCs). We have tested if there is a loss of RGCs, including ipRGCs in the AD mouse model, APPswe/PS1DE9.
Methods: Double transgenic mice on the genetic background of C57BL/6J and C3H harboring the chimeric mouse/human APP (APPSWE) and mutant human presenilin 1 (PS1ΔE9) genes, and their aged-matched WT littermates, were purchased from the Jackson Laboratories (Bar Harbor, ME, strain #004462). Both APPSWE and PS1ΔE9 mutations are associated with early-onset Alzheimer's disease. The "humanized" APPSWE and PS1ΔE9 mutations mediate expression of the human Aβ peptide and presenilin 1. Retinas from mice at 10 and 12 months of age were used. Non transgenic female littermates and C57bl/6J were used as controls. To label and quantify the entire RGC population we have used RBPMS antibodies and for ipRGCs, specific ipRGC antibodies.
Results: Our data show a reduction in the number of RGCs in the ganglion cell layer and in the inner nuclear layer, and significant loss of RGCs in the superior and inferior retinal regions. These findings are in agreement with reports of RGC degeneration in the superior quadrants of the retina in AD patients. The near-complete loss of ipRGC dendrites in a 10-month-old APP_{swe}/PS1ΔE9 transgenic mouse model is consistent with postmortem studies in human AD retinas.
Conclusion: Symptoms referable to the visual system may be the earliest signs of AD and dementia. Damage or loss of ipRGCs in the retina might account for many AD-related visual dysfunctions, including impaired ocular motility, a reduction in amplitude of the PLR, and circadian alterations of melatonin. With ipRGC dysfunction occurring in the pre-symptomatic AD stages that precede cognitive decline, *in vivo* measurements of ipRGC function with chromatic pupillometry may be a potential biomarker for early diagnosis and progression of AD.

Its use in AD screening may provide information on the state of the CNS, sampled via ipRGCs, and become a promising tool for clinical evaluation for the onset, control, and progression of AD.

Disclosures: L. Pérez de Sevilla Müller: None. **A.M. Chen:** None. **N. Brecha:** None.

Poster

P198. Alzheimer's Disease: Genetics, Genomics, and Other Omics Approaches VII

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P198.02

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: U54 AG054345

Title: Unraveling disease mechanisms in a novel, humanized mouse model of late-onset Alzheimer’s

Authors: *K. KOTREDES[1], A. OBLAK[2], C. PREUSS[1], R. PANDEY[1], P. R. TERRITO[3], S. J. SUKOFF RIZZO[4], G. CARTER[1], M. SASNER[1], G. R. HOWELL[5], B. T. LAMB[6];
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Abstract: OBJECTIVE:Late-onset Alzheimer’s disease (LOAD) is the most common form of dementia without an approved therapy. Current animal models do not fully recapitulate LOAD and thus are not ideal for therapy development. Hence, the Model Organism Development and Evaluation for Late-onset AD (MODEL-AD) Center is developing and distributing novel mouse models carrying human-relevant risk factors. Characterization of aging mice that better phenocopy human disease will reveal more appropriate molecular targets useful in the treatment of LOAD. METHODS:Two risk factors of LOAD, humanized *APOEε4* and *Trem2*R47H*, were incorporated into C57BL/6J mice to produce the LOAD1 mouse strain. The effects of expressing an additional genetic factor, humanized amyloid-beta (Aβ) allele (to form the LOAD2 strain), was investigated. Cohorts of mice were aged on multiple sites to 4-, 12-, 18-, and 24-month timepoints. In half, high-fat diet replaced normal mouse chow. A phenotyping pipeline designed to qualify neurodegenerative disease states observed in humans was implemented to measure behavior, cognition, and wellness. *In vivo* imaging of brain perfusion and metabolism, was also performed, as was post-mortem analyses of blood chemistry, neuropathology, transcriptomics, metabolomics, proteomics, spatial profiling, and electrophysiology. RESULTS:LOAD2 mice show increased CSF neurofilament-light in 12-month animals and KC/GRO plasma KC/GRO in 18-month animals, however no overt hippocampal pathology nor differences in long-term

potentiation were observed. Expression of human amyloid did not yield plaque formation in 18-month mice. High-fat diet increased frailty scores and weight in all cohorts irrespective of genotype. In 18-month-old LOAD2 mice, high-fat diet reduced activity and exploratory drive. Correlation of transcriptional profiling with human AMP-AD modules determined individual and synergistic effects between genetic and environmental risk factors. Continuing efforts dissect the downstream effects of sex, age, genotype, and diet via RNAseq, neuropathology, and *in vivo* imaging of brain tissue are in progress, as are proteomic and metabolomic assays on brain homogenates and plasma. CONCLUSIONS:The MODEL-AD consortium has established the LOAD2 mouse model to study the effects of genetic and environmental risk factors of LOAD. This strain serves as a platform for the incorporation of additional AD risk factors (both genetic and environmental) to more closely align phenotypes in the mouse to outcomes observed in the clinic. Here we show the evidence of emerging disease pathology in an animal model exposed to genetic and environmental risk factors.

Disclosures: K. Kotredes: None. **A. Oblak:** None. **C. Preuss:** None. **R. Pandey:** None. **P.R. Territo:** None. **S.J. Sukoff Rizzo:** None. **G. Carter:** None. **M. Sasner:** None. **G.R. Howell:** None. **B.T. Lamb:** None.

Poster

P198. Alzheimer's Disease: Genetics, Genomics, and Other Omics Approaches VII

Location: Virtual Only

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Program #/Poster #: P198.03

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH R56 AG049870 (JIL, PRH)
NIH R01 AG059028 (JIL)
NIH P01 AG005138 and P30 AG066514 (PRH)

Title: Neuronal vulnerability in prefrontal and visual cortex in Alzheimer's disease analyzed by multiplexed immunofluorescence

Authors: *M. VARGHESE[1], S. S. IN[1], W. CHANG[2], S. KRITZER[1], C. M. FREIRE-COBO[1], B. WICINSKI[1], E. MCDONOUGH[3], L. LOWERY[3], M. MEDALLA[2], J. I. LUEBKE[2], D. E. MEYER[3], P. R. HOF[1,4];
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Abstract: Neurons in the prefrontal cortex (PFC) are more susceptible to degenerative changes in aging and Alzheimer's disease (AD) compared to neurons in the primary visual cortex (V1).

Whether interaction of neurons with other cells in their vicinity or protein expression in specific neuronal and non-neuronal cell populations contribute to differential vulnerability of neurons remain open questions. Postmortem brain samples from the PFC and V1 of human subjects with AD (clinical dementia rating CDR 3, Thal stage 3-4, Braak stage V-VI), mild cognitive impairment (CDR 0.5, Thal stage 1-3, Braak stage I-V) and age-matched healthy controls (CDR 0, Thal stage 0-1, Braak stage I-II) were used to prepare tissue microarrays. These tissue microarrays were subject to multiplexed immunofluorescence detection (MxIF), with markers of cellular identity, neuropathology, oxidative stress and inflammation. The images were processed using machine learning-based segmentation and classification algorithms, to detect and classify cells based on their expression of various markers. Tissue retention on the slides across the 13 rounds of MxIF was well-preserved for most of the samples in the study. Qualitative assessment of MxIF images showed robust expression of cellular markers Hu-antigen D for neurons, glial fibrillary acidic protein and aldehyde dehydrogenase 1 family member L1 for astrocytes, ionized calcium-binding adapter molecule 1 for microglia, and collagen IV for vasculature. Cellular stress markers (3-nitrotyrosine, cleaved caspase-3) and pathological markers (amyloid beta, phospho-Ser202/Thr205 tau and TAR DNA-binding protein 43) were also detected. Further analysis is underway to determine whether neuron density is changed, which specific subtypes of neurons are affected in AD, and if pathological or oxidative stress markers are altered in specific neurons, glia, or vasculature in AD. We will also investigate whether previously reported cortical layer-specific susceptibility can be replicated using this method. The outcomes from this study will provide information on *in-situ* cellular interactions and protein changes that contribute to neuronal vulnerability in aging and AD.

Disclosures: M. Varghese: None. **S.S. In:** None. **W. Chang:** None. **S. Kritzer:** None. **C.M. Freire-Cobo:** None. **B. Wicinski:** None. **E. McDonough:** A. Employment/Salary (full or part-time):; General Electric Company. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); General Electric Company. **L. Lowery:** A. Employment/Salary (full or part-time):; General Electric Company. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); General Electric Company. **M. Medalla:** None. **J.I. Luebke:** None. **D.E. Meyer:** A. Employment/Salary (full or part-time):; General Electric Company. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); General Electric Company. **P.R. Hof:** None.

Poster

P198. Alzheimer's Disease: Genetics, Genomics, and Other Omics Approaches VII

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P198.04

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIA Grant 1R43AG065039-01

Title: Transcriptomics of Alzheimer's Disease determined from dried whole blood on filter paper

Authors: *M. BABIC, A. DIAZ, K. SCULLY, J. YEAKLEY, B. SELIGMANN;
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Abstract: Blood contains a complex mixture of cells that are exposed to local signals and metabolic states in all parts of the organism. As such, the overall transcriptomic state of such cells can reflect disease states in various parts of the body, and individual biomarker signals that correlate to specific conditions can be teased out. This is true even for complex conditions such as Alzheimer's Disease (AD), for which several transcriptomic signatures have been published. However, determination of such signatures presents significant technical challenges and costs: blood samples have to be collected, nucleated cells separated, RNA extracted, reverse-transcribed, RNA-seq normalizations performed before sequencing, and the final dataset has to be analyzed by expert bioinformaticists. We have developed a novel approach in which a targeted transcriptomic assay, TempO-Seq®, is applied to small samples of dried blood spots on filter paper, collected noninvasively by simple finger sticks. Rich transcriptomic data, up to the whole transcriptome, can be generated from a <5 mm^2 sample of a blood spot, which is added directly into the assay with no extraction or reverse transcription. Dried blood spots are stable at room temperature, allowing easy handling and shipping, properties that are especially important in poor or medically underserved areas. The assay is simple and inexpensive, with a single technician easily processing >400 samples in parallel in <4 hours of hands-on time. Since only targeted probes are sequenced, bioinformatics is fully automated; the final product is a table with read counts corresponding with levels of each RNA, without any need to analyze sequences directly at any point. The assay also possesses single-base specificity, which allows detection of expressed SNPs, mutations, splice variants, etc. A version of the assay can be utilized to measure DNA as well, finding mutations, unexpressed SNPs, and other alleles of interest. In this report, we show whole blood transcriptomic signatures of AD, comparing to controls, and to Parkinson's Disease patients. Preliminary data indicates that both AD and mild cognitive impairment induce changes in expression level of the non-receptor tyrosine kinase BMX, as well as changes in beta-catenin and FGFR signaling pathways. We also demonstrate detection of alleles of interest from the same dried spots using a DNA genotyping assay.

Disclosures: M. Babic: A. Employment/Salary (full or part-time):; BioSpyder Technologies, Inc. **A. Diaz:** A. Employment/Salary (full or part-time):; BioSpyder Technologies, Inc. **K. Scully:** A. Employment/Salary (full or part-time):; BioSpyder Technologies, Inc. **J. Yeakley:** A. Employment/Salary (full or part-time):; BioSpyder Technologies, Inc. **B. Seligmann:** A. Employment/Salary (full or part-time):; BioSpyder Technologies, Inc..

Poster

P198. Alzheimer's Disease: Genetics, Genomics, and Other Omics Approaches VII

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P198.05

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIAMS R01 AR041398 (PI Kiel)
NIGMS P20GM121301 (PI Liaw)

Title: Transcriptomic analysis of alpha synuclein in blood and bone to explore ADRD peripheral effects

Authors: *A. NAGARAJAN[1], M. GERETY[2], S. REPPE[3], K. GAUTVIK[3], S. SESHADRI[4], A. L. DESTEFANO[4], D. A. BENNETT[5], C. A. FIGUEROA[2], C. J. ROSEN[2], D. P. KIEL[6], C. W. LARY[2];
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Abstract: Association of Alzheimer's Disease and Related Dementias (ADRD) with fractures and low bone mineral density (BMD) are often observed prior to cognitive decline. A prior network analysis of ovariectomy-induced bone loss in mice highlighted the involvement of SNCA, and other literature supports its role in ADRD peripheral effects including insulin resistance and reduced gut motility. We hypothesized that SNCA would have a role in the ADRD-bone association. We conducted a transcriptomic analysis of SNCA using blood gene expression data from the Framingham Heart Study (FHS) Offspring cohort at exam 8 (N = 2,311, average age = 66, 54% female) for 17,873 transcripts, and whole blood (N = 104, average age = 65) and bone biopsy (N=84, average age = 65) gene expression data for 28,867 transcripts from a study of postmenopausal women in Norway. We performed co-expression analysis with FDR correction and pathway and network analysis of significantly co-expressed genes. In the FHS cohort, we also performed a WGCNA (Weighted gene co-expression network analysis) in a subset of participants with two bone assessments available (N = 1,350) for 4,220 transcripts, as their variance was in the top quartile of transcript variability. We found a strong enrichment for mitophagy and ubiquitination among co-expressed genes across all data sets, with top correlated candidates including FBXO7, ADIPOR1, and PINK1. WGCNA of the FHS blood data revealed a module containing SNCA, FBXO7, and ADIPOR1 that showed a significant inverse correlation between the intramodular connectivity, a statistic which measures gene connectivity in the same module, and change in femoral neck BMD. Additionally, SNCA was itself highly connected (intramodular connectivity above the 75th percentile for the module) and significantly associated with change in femoral neck BMD with inverse correlation. All three genes have been implicated in neurodegenerative disease pathogenesis, and our results further link SNCA to mitochondrial and protein degradation along with fat homeostasis. This analysis highlights a potential role of SNCA in ADRD interactions with peripheral tissue, given similar co-expression patterns in bone and peripheral blood that will be further explored using the SNCA interactome in brain transcriptomic data sets in the future.

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Poster

P198. Alzheimer's Disease: Genetics, Genomics, and Other Omics Approaches VII

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P198.06

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant 1RF1AG071683

Title: Single-nucleus chromatin accessibility and transcriptomic characterization of Alzheimer's disease

Authors: *S. MORABITO[1], E. E. MIYOSHI[2], N. MICHAEL[2], S. SHAHIN[2], A. C. MARTINI[2], E. HEAD[2], J. SILVA[2], K. LEAVY[2], M. PEREZ-ROSENDAHL[2], V. SWARUP[2]; [1]Mathematical, Computat. and Systems Biol., [2]Neurobio. & Behavior, Univ. of California, Irvine, Irvine, CA

Abstract: The gene-regulatory landscape of the brain is highly dynamic in health and disease, coordinating a menagerie of biological processes across distinct cell types. The genetic, molecular, and cellular underpinnings of many complex multifactorial neurological diseases such as Alzheimer's Disease (AD) are not fully understood, hindering our ability to design effective therapeutics. Here, we present a multi-omic single-nucleus study of 191,890 nuclei from the prefrontal cortex in late-stage AD, profiling chromatin accessibility and gene expression in the same biological samples and uncovering vast cellular heterogeneity. We independently identified neuronal and glial cell types using two orthogonal genomics techniques and performed an integrated analysis to clarify the relationship between the epigenome and the transcriptome at cellular resolution. We found that the cellular composition of the cortex was significantly altered in AD in both neuronal and glial populations, highlighting a significant decrease of homeostatic microglia. We identified cell-type-specific, disease-associated candidate cis-regulatory elements and their candidate target genes, including an oligodendrocyte-associated regulatory module containing links to AD-associated genetic risk genes including *APOE* and *CLU*. Furthermore, we characterize the cis-regulatory architecture in specific cell types at a subset of AD genetic risk loci defined by genome-wide association studies, demonstrating the utility of this multi-omic single-nucleus approach for contributing to our understanding of AD genetics. We performed integrated pseudotime trajectory analysis in oligodendrocyte, microglia, and astrocyte populations, revealing disease-relevant transcription factors, such as SREBF1, and their regulatory targets throughout disease-associated activated glial responses. Finally, we introduce single-nucleus consensus weighted gene co-expression analysis, a co-expression network

analysis strategy robust to sparse single-cell data and perform a systems-level analysis of the AD transcriptome.

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Poster

P198. Alzheimer's Disease: Genetics, Genomics, and Other Omics Approaches VII

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P198.07

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant AG060144

Title: Neuronal cell cycle re-entry mice and their relevance to late-onset Alzheimer's disease

Authors: *K. PARK[1], R. PANDEY[2], G. CARTER[2], K. STANGIS[1];
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Abstract: Alzheimer's disease is the most prevalent form of dementia, affecting 6 million individuals in US alone. There is an urgency for identifying novel druggable targets. There is no treatment available for modifying the disease progression and numerous therapeutic strategies targeting the removal of Aβ have been unsuccessful. The majority (90-95%) of the AD cases are sporadic, yet much of the current research utilizes familial AD mouse models arising from amyloid precursor protein (APP) and presenilin gene mutations that represent 1-2% of AD cases (Jankowsky and Zheng, 2017). Aberrant neuronal cell cycle re-entry is an alternative pathogenic mechanism in AD that has gained substantial support in the literature. We have demonstrated that ectopic neuronal cell cycle re-entry (NCCR) in wildtype mice generates many of the late-stage AD-related pathologies (Park et al., 2007; Park and Barrett, 2020). We recently combined our NCCR mice with App^{NLF} KI FAD mice, which possess humanized Aβ domain and FAD-linked Swedish and Iberian mutations (Saito et al., 2014), to generate NCCR-App^{NLF} mice. These newly generated NCCR-App^{NLF} mice demonstrate that neuronal cell cycle re-entry can induce and enhance many AD-related pathological features such as hyperploidy, neuroinflammation, brain leukocyte infiltration, PHF-1 phospho-tau lesions, neurodegeneration, and cortical atrophy compared to App^{NLF} mice (Barrett et al., 2021). As a follow-up study, we evaluated the effect of neuronal cell cycle re-entry in App^{NL} KI mice. App^{NL} mice possess humanized Aβ domain and FAD-linked Swedish mutation. However these mice predominantly produce $Aβ_{40}$, and do not deposit Aβ plaques or show neuroinflammation (Saito et al., 2014; Salas et al., 2018). Here we present gene expression profiling data for evaluating the late-onset AD (LOAD) relevance of neuronal cell cycle re-entry mice. NanoString nCounter Mouse AD panel has been developed to

correlate human disease processes and pathways with mouse brain transcripts as a means for evaluating the LOAD relevance of mouse models (Preuss et al., 2020). Our neuropathological and NanoString nCounter AD panel gene expression profile data from NCCR-*App*^{NL} mice provide additional evidence for the LOAD relevance of the NCCR mice.

Disclosures: K. Park: None. **R. Pandey:** None. **G. Carter:** None. **K. Stangis:** None.

Poster

P198. Alzheimer's Disease: Genetics, Genomics, and Other Omics Approaches VII

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Program #/Poster #: P198.08

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NHMRC grant 211255168

Title: Tmem106b increases with physiological ageing in the human hippocampus

Authors: *J. LEE[1], D. J. HARNEY[2], M. LARANCE[2], A. S. DON[1,3];
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Abstract: Ageing is the greatest risk factor for major forms of dementia, including Alzheimer's disease (AD), frontotemporal dementia (FTD) and dementia with Lewy bodies (DLB). These diseases are categorised neuropathologically by protein aggregates that develop with ageing and usually precede clinical dementia. As such, brain ageing is hypothesized to form a prodromal state to neurodegenerative dementias. Allelic variants of *APOE,* encoding apolipoprotein E, are the greatest genetic risk factor for AD, while also regulating risk for DLB and age at onset for FTD. This study employed proteomic analysis of human hippocampus samples to investigate how ageing and *APOE* genotype modulate the risk of developing dementia. Post-mortem tissue samples from the hippocampus (CA1 region) of 76 cognitively- and neuropathologically-normal donors, aged 64 to 104, were used for data-independent mass spectrometry to acquire proteomic data. The dataset was inquired for age- and *APOE* genotype-driven changes at the systems level using Weighted Gene Co-expression Network Analysis (WGCNA). Multiple regression models adjusted for post-mortem interval and sex were applied to determine the effects of age and *APOE* genotype on individual proteins of the hippocampus. All p values were adjusted for false discovery rate (FDR) using the Benjamini-Hochberg correction. As typically observed in the ageing brain, networks of co-expressed proteins enriched with proteostatic, mitochondrial, and astrocytic/endothelial cytoskeletal proteins were associated with age. Seven proteins were significantly associated with age: TMEM106B, MAOB, TBCA, ANXA1, ANXA2, CFL1, and METTL7A. Lysosomal transmembrane protein TMEM106B was most significant ($\beta = 3.10$,

FDR = 2.27 x 10^{-3}, R^2 = 0.25). TMEM106B is a key regulator of lysosomal function and myelination, and its genetic variants modulate the risk for AD, FTD, PD, hippocampal sclerosis, and hypomyelinating leukodystrophy. TMEM106B levels were associated with proteins involved in lysosomal acidification and lipid catabolism (ATP6AP1, ASAH1), myelination (QKI), axon guidance (PLXNB1) and lipid metabolism (ERLIN2, DHRS7). Our results provide the first evidence that TMEM106B protein abundance is increased with normal ageing in the human hippocampus. The association of TMEM106B levels with lysosomal proteins and positive association with a protein critical for myelination (QKI) support literature that have implicated TMEM106B in lysosomal transport and myelination. We speculate that the age-driven increase in TMEM106B levels may offer neuroprotection in the human hippocampus by maintaining lysosomal function and myelination.

Disclosures: J. Lee: None. **D.J. Harney:** None. **M. Larance:** None. **A.S. Don:** None.

Poster

P214. Function and Processing, and Activity of Amyloid

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P214.01

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant AG 44486
NIH Grant AG 15379
BrightFocus Foundation grant A2019056F

Title: Limited substrate specificity of PS/g-secretase is supported by novel multiplexed FRET analysis in live cells

Authors: *M. HOUSER, Y. TURCHYNA, F. PERRIN, O. BEREZOVKSA, M. MAESAKO; Massachusetts Gen. Hosp., Charlestown, MA

Abstract: Presenilin (PS)/γ-secretase plays a pivotal role in essential cellular events as well as in numerous diseases via proteolytic processing of transmembrane proteins, including APP and Notch 1. To better understand the spatiotemporal regulation of PS/γ-secretase activity, we have engineered Förster resonance energy transfer (FRET)-based APP C99 YPet-mTurquoise-GL (C99 Y-T) and Notch1 N100 Y-T biosensors that allow for the first-time recording the activity of endogenous PS/γ-secretase activity overtime, on a cell-by-cell basis, in live cells (Maesako et al. 2020). Using these new biosensors, we uncovered that PS/γ-secretase activity is heterogeneously regulated among cells. However, whether PS/γ-secretase preferentially processes its different substrates (e.g., APP vs. Notch1) in live cells is still poorly understood. Recent structural studies of PS/γ-secretase enfolding a fragment of APP or Notch1 showed that these two key substrates engage the protease in broadly similar ways, suggesting the limited substrate specificity of PS/γ-

secretase. Here, we report the development of a new near-infrared (NIR) C99-720-670 biosensor that incorporates an immediate PS/γ-secretase substrate APP C99 with miRFP670 and miRFP720 as the donor and acceptor fluorescent proteins, respectively (Houser et al. 2020). Our validation demonstrates that the C99 720-670 biosensor permits quantitative monitoring of endogenous PS/γ-secretase activity and is spectrally compatible with our previously developed visible range C99 Y-T biosensor. We then utilized this multiplex imaging platform to monitor how PS/γ-secretase processes two different substrates in the same cell. Overall, we found that PS/γ-secretase similarly cleaves Notch1 N100, wild-type APP C99, and familial Alzheimer's disease (FAD)-linked APP C99 mutants in Chinese hamster ovary (CHO) cells (Houser et al. 2021). These findings support limited PS/γ-secretase substrate processing specificity. Our new multiplexed FRET assay could help better understand the consequences of changes in PS/γ-secretase activity and variability in the processing of multiple substrates in normal and disease conditions.

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Poster

P214. Function and Processing, and Activity of Amyloid

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P214.02

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH-NIA Grant 15379
NIH-NIA Grant 44486

Title: Glutamate transporter GLT-1 and PS1/γ-secretase are cross-regulated in term of function and activity in Alzheimer's disease

Authors: *F. PERRIN[1], L. C. ANDERSON[2], Y. TURCHYNA[1], M. MAESAKO[2], M. C. Q. HOUSER[1], O. BEREZOVSKA[1];
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Abstract: Rational: Mutations in Presenilin 1 (PS1), the catalytic sub-unit of γ-secretase, cause early-onset familial Alzheimer's disease (fAD) by inducing a "closed" structural conformation in PS1 and production of longer Aβ species. Interestingly, clinical data revealed that patients with presenilin mutation carriers have significantly higher incidence of epileptiform activity, suggesting that fAD PS1 mutations (Aβ-related or not) could contribute to hyperactivity. We have recently discovered an interaction between PS1 and the major CNS glutamate transporter GLT-1. Here we aim to investigate; i) if mutations in PS1 modulate interactions between PS1 and GLT-1, ii) whether PS1 wt or PS1 fAD impact GLT-1 function in term of localization and

activity, iii) does GLT-1 and PS1 binding affect APP processing. **Methods:** To test whether sporadic AD (sAD) effects the PS1/GLT-1 interaction we examined brain tissue from control, sAD, and frontotemporal dementia (FTD) cases. To examine if the PS1/GLT-1 interaction is disrupted by PS1 pathogenic mutations we use fluorescence lifetime imaging microscopy assay (FLIM) in intact cells *in vitro*. Next, we sought to determine if changes in the PS1/GLT-1 interaction caused by fAD PS1 mutations result in functional changes in GLT-1 cell surface expression by flow cytometry and cell surface biotinylation. We also tested in native condition GLT-1 multimerization profile (GLT-1 dimer and trimer) on different cell lines. In addition, we used glutamate uptake assay to measure the PS1 effects on GLT-1 function. Finally, we monitored the production of different Aβ species in cell lines and neurons expressing PS1/GLT-1 with a multiplex assay. **Results:** We showed that the PS1/GLT-1 interaction occurs in the human brain and this interaction is reduced in sporadic AD brains. We found that the presence of PS1 wt increased GLT-1 cell surface expression and GLT-1 multimerization. Importantly, our preliminary data indicate that GLT-1/mediated glutamate uptake was increased in the presence of PS1. In terms of APP processing, GLT-1 impacts PS1/g-secretase cleavage favoring shorter Aβ(38) species production. **Conclusions:** The change in the PS1/GLT-1 interaction induces functional changes in GLT-1 and PS1. These results could explain the strong connection between fAD PS1 mutations and epileptiform activity in fAD.

Disclosures: F. Perrin: None. **L.C. Anderson:** None. **Y. Turchyna:** None. **M. Maesako:** None. **M.C.Q. Houser:** None. **O. Berezovska:** None.

Poster

P214. Function and Processing, and Activity of Amyloid

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P214.03

Topic:

Support: NIH-NIA Grant 15379
NIH-NIA Grant 44486

Title: Glutamate transporter GLT-1 impacts PS/ɣ-secretase activity and conformation

Authors: *Y. TURCHYNA, M. MAESAKO, F. PERRIN, M. C. Q. HOUSER, O. BEREZOVSKA;
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Abstract: Recently GLT-1 was identified by our group as a novel PS1-interacting protein, which allowed us to explore the link between two major hallmarks of Alzheimer's disease - abnormal Aβ production/accumulation and hyperexcitability of neuronal networks. Current study

investigates impact of GLT1 on Presenilin/ɣ-secretase to explore functional outcomes of their interaction. First, Aβ42 and 40 concentrations were measured with ELISA in conditioned medium of CHO PS70 cells transiently transfected with GLT-1 DNA or empty vector. Results revealed that extracellular concentration of both Aβ species was significantly decreased in the presence of GLT-1, compared to control. Among other possibilities, it suggests that activity of PS/ɣ-secretase may be altered. Therefore, using genetically encoded FRET-based biosensor C99 YT and confocal microscopy we monitored endogenous PS/ɣ-secretase activity and discovered that it was significantly decreased due to GLT-1 overexpression. Interestingly, when GLT-1 function was inhibited by 1mM DHK, the activity of PS/ɣ-secretase was not significantly different from control level. This suggests that not only GLT-1 expression, but its functional state is important for modulation of the PS/ɣ-secretase function. Abovementioned changes in extracellular Aβ42 and 40 concentrations measured with ELISA might also be a result of abnormal Aβ production. Since it was previously established that changes in Aβ production may be linked to changes in conformation of PS1, we employed FRET-based FLIM assay and GFP-PS1-RFP genetically encoded probe to determine if GLT-1 expression impacts PS1 conformation. We found that PS1 conformation is shifted to more “open” state in cells expressing GLT-1. Together our results suggest that GLT-1 has powerful impact on PS1 functioning and Aβ generation. GLT-1/PS1 interaction as a potential new target for developing drugs aimed at treating Alzheimer`s disease will be discussed.

Disclosures: Y. Turchyna: None. **M. Maesako:** None. **F. Perrin:** None. **M.C.Q. Houser:** None. **O. Berezovska:** None.

Poster

P214. Function and Processing, and Activity of Amyloid

Location: Virtual Only

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Program #/Poster #: P214.04

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIA Grant AG060238
NIA Grant AG062251
NIA Grant AG033570
AHA 836269

Title: Depletion of Caveolin-1 in Type 2 Diabetes model induces Alzheimer’s disease pathology

Authors: *A. SHETTI[1], J. BONDS[3], A. RAMAKRISHNAN[1], K. VO[1], R. MINSHALL[2], O. LAZAROV[1];
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Abstract: Type 2 Diabetes is a major risk factor for the development Sporadic Alzheimer Disease. Vascular dysfunction and impaired insulin signaling are thought to be involved in the development of dementia and Alzheimer's disease (AD) in T2DM but a mechanism connecting these pathologies is currently unknown. Here we show that levels of Caveolin-1 (Cav-1), an endothelial cell enriched protein and its downstream signaling component eNOS were reduced in the brains of T2DM patients compared to healthy ageing individuals. The levels of Cav-1 and eNOS were inversely correlated with β-amyloid (Aβ) in these patients. Depletion of Cav-1 was recapitulated in the brains of T2DM mouse models and correlated with upregulated levels of amyloid precursor protein (APP), β-secretase-1 (BACE-1) and hyperphosphorylated tau (p-tau) species. In addition, the T2DM mice exhibited impairment in recognition memory. Importantly, restoration of Cav-1 in the hippocampus of T2DM mice rescued recognition memory and reduced AD pathology i.e. Aβ, APP, BACE-1 and p-tau levels. Together, our results suggest that reduced levels of Cav-1 in the brain in T2DM accelerate the development of AD hallmarks. Restoring Cav-1 levels may attenuate or prevent AD pathology and cognitive decline in T2DM.

Disclosures: A. Shetti: None. **J. Bonds:** None. **A. Ramakrishnan:** None. **K. Vo:** None. **R. Minshall:** None. **O. Lazarov:** None.

Poster

P214. Function and Processing, and Activity of Amyloid

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Program #/Poster #: P214.05

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Hawaii Community Foundation #19ADVC-95450
NIH Grant P20GM103457

Title: The exocyst complex is an insulin-sensitive regulator of amyloid precursor protein (APP) trafficking and beta-amyloid generation in neurons

Authors: G. Y. PATWARDHAN[1], R. K. SACHS[1], M. A. ORTEGA[3], A. J. LEE[4], K. M. ORMSBEE[2], M. K. WILLIAMS[1], B. E. HEW[1], J. B. OWENS[1], N. G. JAMES[2], R. A. NICHOLS[2], *B. C. FOGELGREN[1];
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Abstract: Alzheimer's disease (AD) is a progressive neurodegenerative disorder characterized by the loss of cognitive function, language, and memory. Despite decades of AD research, there are still no effective therapies and the pathogenic mechanisms are not fully understood. One prominent histopathological hallmark of AD is the accumulation of amyloid beta (Aβ) peptide

plaques. Neuronal generation of the Aβ peptide arises from proteolytic processing of the amyloid precursor protein (APP) by β- and γ-secretases, and although not fully understood, regulation of APP intracellular trafficking plays a key role in the balance of amyloidogenic APP processing. The exocyst is a highly-conserved eight-protein complex that acts as a Rab GTPase effector to guide subsets of intracellular transport vesicles to their destination for vesicle fusion. Assembly and targeting of the exocyst is controlled by a variety of protein kinases and small GTPases through mechanisms that are often cell type-specific. For example, adipocytes and muscle cells use the exocyst for dynamic exocytosis of GLUT4-containing vesicles in response to insulin signaling. We hypothesize that the exocyst plays a key regulatory role in the neuronal intracellular trafficking of APP. We show in human SH-SY5Y neuroblastoma cells differentiated into neurons, and in primary mouse hippocampal neurons, exocyst subunits were co-localized with APP on intracellular vesicles via high-resolution imaging. We confirmed these findings using proximity ligation assays (PLAs), a novel technique to measure protein-protein interactions within the cell, showing that APP and Exoc5 protein co-localized within 40nm. However, after 15 mins of insulin treatment, this APP-Exoc5 interaction was greatly reduced, while the association of Exoc5 with GLUT4 was increased. Using a stable SH-SY5Y cell line that we generated to express APP containing several familial AD mutations and which secretes high levels of Aβ, RNAi silencing of each exocyst subunit led to dramatic decreases in sAPP and Aβ secretion, with intracellular accumulation of full-length APP. In addition, we observed the same results with RNAi silencing of Exoc3L2, an uncharacterized exocyst gene homolog linked to late onset AD through GWAS. Using live-cell TIRF microscopy of fluorescently-labeled APP and exocyst proteins in differentiated SH-SY5Y neurons, we observed highly correlated movement of APP and Exoc4/7, with the Exoc1 protein often marking the site of APP exocytosis. These experiments show for the first time that the exocyst plays a key role in APP trafficking and Aβ secretion in neurons, and this activity is directly regulated by insulin signaling.

Disclosures: G.Y. Patwardhan: None. **R.K. Sachs:** None. **M.A. Ortega:** None. **A.J. Lee:** None. **K.M. Ormsbee:** None. **M.K. Williams:** None. **B.E. Hew:** None. **J.B. Owens:** None. **N.G. James:** None. **R.A. Nichols:** None. **B.C. Fogelgren:** None.

Poster

P214. Function and Processing, and Activity of Amyloid

Location: Virtual Only

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Program #/Poster #: P214.06

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: the University Grants Committee of Hong Kong GRF14123718

Title: A potential crosstalk between YKL-40 and PTEN in amyloid-beta induced signalling and biological effects in Alzheimer’s disease

Authors: *X. ZENG, K. CHEUNG, P.-Y. OR, Z. LI, T. LIU, A. CHAN;
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Abstract: YKL-40 has been established to be a diagnostic biomarker of Alzheimer's disease (AD), and its increased immunoreactivity has been observed in astrocytes in the frontal cortex of AD patients. However, the functional role of YKL-40 in the process of AD is still not known. In our previous study, we have confirmed a cell-type specific relationship between YKL-40 and PTEN, which has been implicated in long term depression and synaptic plasticity. Here, we found significantly increased YKL-40 expression level in cerebral spinal fluid (CSF) of 5xFAD mice. By confocal imaging, we found an astrocyte-specific expression pattern of YKL-40. Similarly, in primary cultured astrocytes, we observed increased YKL-40 expression in conditioned medium (CM) following amyloid beta (Aβ) treatment. Interestingly, we also observed activated PTEN-related AKT/mTOR and JAK2/STAT3 pathway in Aβ-treated astrocytes. Since YKL-40 is a secreted protein, we exposed primary neuronal cultures with YKL-40 and Aβ simultaneously. Immunofluorescence staining showed reduced synaptic puncta following Aβ and YKL-40 treatment. Also, by microelectrode array (MEA) recording, we observed significant reduction in the amplitude of spontaneous action potential. In particular, Western blotting analysis revealed increased expression of PTEN, YKL-40 and Caspase-3, and decreased expression of PSD-95 and synaptophysin (SYP), as well as activated AKT/mTOR pathway following YKL-40 and Aβ treatment. All these observations implicate YKL-40 in mediating the neurotoxic effects of Aβ in neuronal cells. In conclusion, our study demonstrates that Aβ increased YKL-40 secretion and this activates the AKT/mTOR pathway, leading to impaired synaptic plasticity in primary cortical neurons. Our results provide impetus in developing anti-YKL-40 therapy as a strategy to alleviate neuronal attrition in AD brains.

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Poster

P214. Function and Processing, and Activity of Amyloid

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Program #/Poster #: P214.07

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH/NIA Grant R01AG067048
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Cure Alzheimer's Fund
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Owens Family Foundation

Title: A Tau-mTORC1-SOD1 pathway couples nutrient availability to mitochondrial DNA synthesis and its disruption by Amyloid-beta oligomers

Authors: *A. NORAMBUENA[1], X. SUN[1], H. WALLRABE[1], R. CAO[1], N. SUN[2], E. PARDO[1], N. SHIVANGE[1], D. BIGLER[1], L. A. POST[1], H. A. FERRIS[1], S. HU[2], A. PERIASAMY[1], G. S. BLOOM[1];
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Abstract: Alzheimer's disease (AD) features accumulation of amyloid-β (Aβ), tau phosphorylation, insulin resistance and mitochondrial dysfunction, but molecular mechanisms connecting these phenomena remain unclear. We recently discovered nutrient-induced mitochondrial activity (NiMA), an inter-organelle signaling pathway whereby activation of mechanistic target of rapamycin complex 1 (mTORC1) on lysosomes by insulin or amino acids (herein, nutrients) stimulates mitochondrial oxidative metabolism (OXPHOS) and suppresses mitochondrial DNA (mtDNA) replication in cultured neurons. Aβ oligomers (AβOs), which activate plasma membrane, but not lysosomal, mTORC1 by a tau-dependent mechanism (Norambuena et al, 2017. *Alzheimer's & Dementia* 13: 152-167), were found to counteract the effects of nutrients on both OXPHOS and mtDNA replication (Norambuena, et al. 2018. *EMBO J* 37: e100241). These observations stimulated further investigation of the mechanism by which tau regulates mtDNA replication and how such regulation may be compromised in AD. Using EdU incorporation and two-photon fluorescence lifetime imaging of NAD(P)H in live cultured neurons, and tracking oxygen metabolism in the live mouse brain with multi-parametric photoacoustic microscopy, we found that mTORC1-mediated phosphorylation of superoxide dismutase 1 (SOD1) at threonine 40 regulates mtDNA synthesis through the NiMA pathway. Mechanistically, we found that in neurons tau enables nutrient-induced activation of mTORC1 at lysosomes, which in turn suppresses SOD1 activity and mtDNA synthesis. In contrast, AβO-induced insulin resistance in neurons increases the lysosomal content of the tuberous sclerosis complex (TSC), an inhibitor of mTORC1 kinase activity, and by extension upregulates SOD1 activity and increases mtDNA synthesis. Dysregulated mtDNA synthesis was also observed in fibroblasts from two patients with tuberous sclerosis, a developmental disorder caused by reduced TSC activity and mTORC1 hyperactivity. Finally, SOD1 activity was found to be elevated in human AD brain. Thus, tau and SOD1 are part of a nutrient-sensing machinery that functionally couples nutrient availability to mtDNA replication and function under normal conditions, and mechanistically links mitochondrial dysfunction to AD.

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Poster

P214. Function and Processing, and Activity of Amyloid

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P214.08

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH (NINDS): 2R01NS045860-16A1 (RET)

Title: Axonal Generation of Amyloid-beta from Palmitoylated APP (palAPP) is Regulated by Mitochondria-associated Endoplasmic Reticulum Membranes (MAMs)

Authors: *R. BHATTACHARYYA[1], S. E. BLACK[3], M. S. LOTLIKAR[3], R. H. FENN[3], M. B. TARANTINO[3], M. JORFI[3], D. M. KOVACS[4], R. E. TANZI[2];
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Abstract: The subcellular localization of amyloid-β (Aβ) generation and its downstream effects on neurons are key events in Alzheimer's disease (AD) neuropathology. At least 40% of total neurotoxic Aβ is generated via processing of β-amyloid precursor protein (APP) in neuronal processes and axons. The cellular and the molecular mechanisms involved in the axonal generation of the AD-associated Aβ remain largely elusive. We have previously reported that palmitoylated APP (*pal*APP) serves as a preferred substrate for β-secretase (BACE1) in lipid rafts leading to Aβ generation. Mitochondria-associated Endoplasmic Reticulum (ER) Membranes (MAMs) are cholesterol rich lipid domains that bear remarkable similarity to lipid rafts. Recent studies have shown that MAMs are upregulated in AD. APP, BACE1 and γ-secretase components are all found in MAMs, prompting detailed study of the role of MAMs in Aβ generation in AD. In addition, other proteins enriched in MAMs, including inositol-1,4,5-trisphosphate receptor subunit 3 (IP3R3), sigma-1 receptor (S1R) and acyl-coenzyme A: cholesterol acyltransferase (ACAT), have emerged as potential drug targets for AD. Here, we have investigated the role of MAMs in the processing and trafficking of *pal*APP and specifically in the axonal generation of Aβ. For this purpose, we primarily employed human neuronal progenitor cells expressing APP with familial AD (FAD) mutations (FAD hNPCs). These cells differentiated into neurons in a three-dimensional (3D) matrix exquisitely recapitulate AD neuropathology. First, we show that *pal*APP is enriched in MAMs, both in FAD hNPCs and in mouse brain. Next, we show that downregulating MAM assembly by either siRNA-mediated silencing or pharmacological modulation of the MAM-resident sigma1 receptor (S1R) leads to attenuated β-secretase cleavage of *pal*APP. Upregulation of MAMs has promoted trafficking of *pal*APP to the cell surface and has increased β-secretase cleavage. We also show that modulation of MAMs via S1R regulates *pal*APP stabilization. Using a novel microfluidic device, we show that modulation of MAM levels regulates Aβ generation specifically in neuronal processes and axons, but not in cell bodies. Confocal imaging studies have indicated that *pal*APP partitions into axonal MAMs, while total or de-palmitoylated APP resides in somal MAMs. These findings provide a better understanding of the cellular and molecular mechanisms underlying Aβ

generation in axons, and carry significant implications for alleviating Aβ pathology by targeting MAM-associated *pal*APP in axons.

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Poster

P214. Function and Processing, and Activity of Amyloid

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P214.09

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Fondecyt 1200908
Conicyt 21141247

Title: Impact of Transglutaminase type 2 transamidating activity on Amyloid beta aggregation and mitochondrial function in Alzheimer disease models

Authors: *J. PANES-FERNANDEZ, J. A. GAVILAN, P. A. GODOY RIVERA, O. RAMIREZ-MOLINA, A. M. MARILEO, C. F. BURGOS, C. MUNOZ-MONTESINO, G. E. YEVENES, G. MORAGA, P. A. CASTRO, L. GUZMAN, J. FUENTEALBA;
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Abstract: Alzheimer's disease (AD) is a neurodegenerative disorder characterized by cognitive impairment that increasingly afflicts the elderly population. The main features of AD have been related with cellular and molecular events like neuronal loss, mitochondrial dysfunction, and aberrant aggregation of Amyloid beta peptide (Aβ) in specific areas of the brain; however, the molecular events associated with Aβ pathology are not well understood. The transglutaminase type 2 (TG2) is a ubiquitous enzyme that have as main role the Ca^{2+}-dependent transamidation of proteins. This enzymatic activity has been closely related with several steps of neurotoxicity in AD pathogenesis, including alterations on mitochondrial O_2 consumption, as well as Aβ crosslinking. In this work, we studied the contribution of TG2 in Aβ aggregation process, and its impact on mitochondrial function in neuronal model of Aβ-pathology as well as J20 mice model. First, we studied the toxic effects of Aβ soluble oligomers (AβOs, 0.5 μM, 24 h) on mitochondrial function parameters as well as on key proteins that regulate mitochondrial biogenesis in hippocampal neurons treated with TG2 inhibitor (Z-DON, 30nM). Using fluorescence approaches, we found that mitochondrial membrane potential (ΔΨm) decrease induced by AβOs was partially prevented by a Z-DON (AβOs 56% vs AβOs + Z-DON = 73%). In parallel, TG2 inhibition could prevent the decrease of PGC-1α levels induced by AβOs (AβOs: 66%, AβOs + Z-DON = 75%). Later, we tested if Aβ peptide is a substrate of endogenous TG2 in an *ex vivo* model of hippocampal slices. We observed a concentration-

dependent raise on TG2 transamidating activity treated Aβ peptide (1-5 μM). Furthermore, hippocampal slices from J20 mice, shown a substantial increase on TG2 transamidating activity (330%), compared with control samples. Finally, we performed an in silico analysis to study the potential TG2-Aβ complex (docking score: -138.3, ΔG_{bind}:-13.2 kcal/mol). Thioflavin-T (ThT) fluorescence assays shown that the recombinant human TG2 (rhTG2) was able to potentiate the Aβ aggregation kinetics, promoting the oligomeric or fibrillar-like structures. These findings reinforce our previous works and lead us to raise the comprehension about TG2 transamidating activity at the onset of Aβ pathology and mitochondrial alterations in AD.

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Poster

P234. Therapeutic Strategies: Preclinical Animal Models and Small Molecule Therapeutics IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P234.01

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: SATT AXLR /UM 210384

Title: Fluoroethylnormemantine (FENM) used in combination with other therapeutic drugs offers new opportunities in Alzheimer's disease

Authors: *A. FREYSSIN[1,2], A. CARLES[1], S. GUEHAIRIA[1], G. RUBINSTENN[2], *T. MAURICE[1];
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Abstract: Memantine, a non-competitive N-methyl-D-aspartate (NMDA) receptor antagonist, has been approved for use in Alzheimer's disease (AD). Memantine works by blocking NMDA receptors in the brain and may help improve the symptoms of dementia. However, memantine can cause mild or serious side effects include drowsiness, dizziness, headache, or confusion. Fluoroethylnormemantine (FENM) is a memantine derivative, recently shown to be a superior neuroprotectant than its parent molecule, in the $Aß_{25-35}$ mouse model of AD [Couly et al., Int J Neuropsychopharmacol 2020]. FENM is considered as an anti-excitotoxic drug that may be adequately used in combination for a greater therapeutic efficacy. We therefore tested several FENM-based combinations with drugs acting on different targets of interest in AD: a selective sigma-1 receptor agonist, an acetylcholinesterase inhibitor, an antioxidant, an agonist of GABA-

A receptor and a nonsteroidal anti-inflammatory drug. Memory deficits were induced in mice by intracerebroventricular injection of oligomeric $A\beta_{25-35}$ peptide. Two behavioral tests, namely spontaneous alternation in the Y-maze and passive avoidance response, were used in parallel and protective and symptomatic effects were examined. After determination of the minimal active doses for each compound, the combinations were tested, and combination index (CI) were calculated according to the isobologram representation of drug combination. Results confirmed that FENM presents a different neuroprotective profile than its parent molecule memantine and which extend to the way FENM is active when used in combination with combination with other pharmacological agents. Particularly, outstanding results for combination potentially useful in the treatment of AD will be exposed and discussed.

Disclosures: **A. Freyssin:** A. Employment/Salary (full or part-time):; ReST employment. **A. Carles:** None. **S. Guehairia:** None. **G. Rubinstenn:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); stock, patent holder,CEO ReST. **T. Maurice:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; SATT AxLR. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Patent holder.

Poster

P234. Therapeutic Strategies: Preclinical Animal Models and Small Molecule Therapeutics IV

Location: Virtual Only

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Program #/Poster #: P234.02

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH/NIA R21AG056947
NIH/NIA T32AG00115
NIH/NIA P01AG
NIH/NIGMS T32GM008541
NIH/NINDS F31NS068219

Title: Does Modulation of Tonic Inhibition Control Sharp Wave Ripples and Hippocampal Place Cell Dynamics?

Authors: M. H. RATNER[1], S. S. DOWNING[1], O. GUO[3], K. ODAMAH[1], T. M. STEWART[2], V. KUMARESAN[1], R. ROBITSEK[1], *D. H. FARB[4];
[1]Dept. of Pharmacol. and Exptl. Therapeut., [2]Boston Univ. Sch. of Med., Boston, MA; [3]Dept. of Biol., Boston Univ., Boston, MA; [4]Dept. of Pharmacol. and Exptl. Therapeut., Boston Univ. Sch. Med., Boston, MA

Abstract: Hippocampal hyperactivity is a hallmark of amnestic mild cognitive impairment (aMCI) in aging and memory dysfunction in Alzheimer's disease, leading to the hypothesis that hyperactivity in the hippocampal trisynaptic circuit impairs memory. As a test of this hypothesis, we sought to recapitulate hyperactivity in young adults by attenuating tonic inhibition in the hippocampal trisynaptic circuit. We used chronically implanted arrays of 96 electrodes arranged as tetrodes and 32 channel silicon probes to record place cell activity within and across the CA1 subregion of young adult rats either foraging in familiar and novel environments or resting in a familiar environment (without foraging) while awake but immobile for ripple band recordings. We find that the nootropic drug α5IA, a selective negative modulator of tonically active predominantly extrasynaptic α5 GABA receptors and established enhancer of episodic memory, enhances CA1 place cell firing rates. Yet properties of place cells believed to reflect memory processing such as spatial information content and pattern separation did not reflect nootropic drug action. This conundrum is resolved by the finding that α5IA substantially augments sharp wave ripple (SPW-R) amplitude, which is known to correlate with improved remembering. These observations strongly suggest that the nootropic actions of α5IA may result from induction of CA1 place cell hyperactivity and enhanced CA1 SPW-R amplitude. Surprisingly, spatial mapping characteristics of place cell firing rates with location were unaffected. We used three strains of rats (Long Evans, Fisher 344, and Sprague Dawley) and found that the outcomes were not strain specific. These results suggest that tonic inhibition may be a key control point in modulating SPW-R amplitude during awake immobility, perhaps leading to enhanced consolidation of encoded information. We posit that modulation of ripple dynamics may be controlled via alterations in extrasynaptic tonic inhibition mediated in part by α5 GABA-A receptors.

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Poster

P234. Therapeutic Strategies: Preclinical Animal Models and Small Molecule Therapeutics IV

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Program #/Poster #: P234.03

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: 1R01NS096225-01A1 NIH/NINDS
19TPA34850047 AHA
Magale Foundation
Louisiana State University Research Council

Title: Prmt 4 causes cerebral blood flow derangements and cognitive dysfunction in dementia

Authors: *G. CLEMONS[1], A. DO COUTO E SILVA[1], C. H. ACOSTA[1], C. Y.-C. WU[2], C. T. CITADIN[1], R. H.-C. LEE[2], H. LIN[2];
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Abstract: Alzheimer's disease (AD) is a leading cause of mortality, disability, and long-term care burden in the United States. Women are more likely to develop a rapid progression of dementia than men with a greater risk of developing vascular dementia (VaD) compared to males when predictive factors such as obesity, lifespan, and enhanced stroke severity are considered. While AD-related dementia is associated with tau and amyloid beta accumulation, concurrent derangements in cerebral blood flow (hypoperfusion) have been observed alongside these proteinopathies in humans and mice, a phenomenon known as vascular dementia. Restoring or maintaining cerebral blood flow can improve prognosis and slow dementia progression, making vascular dementia a critical point of investigation in aging and neurodegeneration. Few therapies exist to treat vascular dementia, and there is an urgent need for the discovery of novel targets that treat disease progression and improve patient prognosis. As L-arginine is a crucial driver of nitric oxide synthesis, an ideal target would be involved in this process, and may exist in protein arginine methyltransferases (PRMTs). PRMTs are a novel class of enzymes which can exhibit vasoactivity via the nitric oxide synthase signaling pathway by methylating arginine residues, this methylation reduces the availability of the canonic NOS ligand, l-arginine. This makes PRMTs a relevant target as PRMT products can act on NOS upstream of nitric oxide production. We used aged (9-12 month) 3xTg female mice to recapitulate dementia. We measured PRMT4 protein levels using ProteinSimple capillary-phoresis, and our data suggests that 3xTg female mice have increased levels of PRMT4 (180.0% ± .37% in hippocampus, 5.0% ± .13% in cortex). We assessed peroxynitrite levels using an ELISA kit and our findings suggest that peroxynitrite was increased (40.8% ± 14.0% in hippocampus, 126.8% ± 1.0% in cortex). We used laser speckle contrast imaging to measure regional cortical cerebral blood flow; we found that in aged 3xTg female mice, CBF was reduced (26.0% ± 5.0%). Additionally, administration (30mg/kg/day, 1 week, IP) of specific PRMT4 inhibitor TP-064 improved cerebral blood flow by 162.0% ± 40.4% compared to untreated age/sex matched 3xTg mice. To observe differences in learning and memory, we utilized the modified T-maze; we found that female 3xTg mice score lower (51%), compared to C57 age/sex-matched controls. Overall, our data suggests that enhanced levels of PRMT4 in AD brain results in NOS uncoupling which contributes to peroxynitrite accumulation, hypoperfusion, and functional deficits.

Disclosures: G. Clemons: None. **A. Do Couto E Silva:** None. **C.H. Acosta:** None. **C.Y. Wu:** None. **C.T. Citadin:** None. **R.H. Lee:** None. **H. Lin:** None.

Poster

P234. Therapeutic Strategies: Preclinical Animal Models and Small Molecule Therapeutics IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P234.04

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: KHIDI grant HI18C1801
Genuv Inc.

Title: Mek1/2 inhibition rescues neurodegeneration by activation of autophagic lysosomal function in alzheimer's disease

Authors: *Y. CHUN[1], M.-Y. KIM[1], S.-Y. LEE[1], M. KIM[1], H. KIM[1], T.-I. KAM[2,3], S. HAN[1]; [1]Genuv Inc., Seoul, Korea, Republic of; [2]Neuroregeneration and Stem Cell Programs, Inst. for Cell Engin., [3]Dept. of Neurol., Johns Hopkins Univ. Sch. of Med., Baltimore, MD

Abstract: Alzheimer's Disease (AD) is a progressive neurodegenerative disorder, which is characterized by cognitive deficit due to synaptic loss and neuronal death. Extracellular amyloid β plaques are one of pathological hallmarks of AD. MEK/ERK signaling pathway is activated in the brain of AD patient and AD animal models, and in primary neurons cultured with Aβ oligomers. Here, we showed that inhibition of MEK/ERK signaling using a clinically available MEK1/2 inhibitor, trametinib (GSK1120212, SNR1611) protected neurons in AD-model mice (5XFAD). The 5-month-old 5XFAD mice were administered either vehicle or 0.1 mg/kg of trametinib for 10 weeks by oral gavage once a day. Administration of trametinib recovered impaired neuronal structures, synaptic dysfunction, and cognitive deficits. Trametinib also protected from apoptotic neuronal death. RNA-sequencing analysis of the brain cortex revealed upregulation of autophagic lysosomal genes by trametinib administration. In addition, trametinib elevated autophagic lysosomal activity in 5XFAD cortex and primary cortical neurons cultured with Aβ oligomers. From these observations, we concluded that MEK inhibition provides neuronal protection by increasing autophagic lysosomal activity, and thus MEK inhibition might be effective therapeutic strategy for AD.

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Poster

P234. Therapeutic Strategies: Preclinical Animal Models and Small Molecule Therapeutics IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P234.05

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: 7,8-dihydroxyflavone attenuates cognitive deficit in icv-stz rat model of alzheimer's disease reversing oxidative stress, mitochondrial dysfunction, and insulin resistance

Authors: *A. AKHTAR, S. P. SAH;
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Abstract: Objectives: Sporadic Alzheimer's disease (SAD) is the most commonly prevalent dementia and progressive neurodegenerative disease. A polyphenolic flavonoid, 7,8-dihydroxyflavone (7,8-DHF) was used to reverse the cognitive deficit in a rat SAD model by reversing oxidative imbalance, mitochondrial enzyme dysfunction, and insulin resistance. **Methods:** For the SAD model, streptozotocin (STZ-3 mg/kg) was injected intracerebroventricularly (ICV) in male Wistar rats to induce cognitive dysfunction. Cognitive functions were evaluated by Morris water maze (MWM) and novel object recognition (NOR) tests, while locomotor activity was determined in actophtometer. 7, 8-DHF was given orally in doses of 5 mg/kg, 10 mg/kg, and 20 mg/kg, and reference standard drug rivastigmine in a dose of 2 mg/kg. Antioxidant enzymes, mitochondrial enzyme complexes were determined biochemically, insulin-degrading enzyme (IDE) and p-tau by ELISA, and histopathology by H&E staining. **Results:** 7,8-DHF attenuated cognitive deficit induced by ICV-STZ in MWM and NOR. Moreover, in the cortex and hippocampus regions of the brain, levels of reduced glutathione, catalase, superoxide dismutase, and mitochondrial complex enzymes and increased lipid peroxidation, protein carbonylation, and nitrite levels were subsequently reversed by 7,8-DHF and rivastigmine. IDE and p-tau protein were found to be altered. Histopathological examination revealed halted neurodegeneration. **Conclusions:** Conclusively, 7,8-DHF was found to be neuroprotective in the ICV-STZ rat model of SAD by ameliorating oxidative stress, mitochondrial dysfunction, and insulin resistance, thereby improving cognitive functions evident with the behavioral results. Hence, these results can further have clinical relevance in human AD.

Disclosures: A. Akhtar: None. **S.P. Sah:** None.

Poster

P234. Therapeutic Strategies: Preclinical Animal Models and Small Molecule Therapeutics IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P234.06

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: John G. Kulhavi Professorship in Neuroscience at CMU
The Field Neurosciences Institute
CMU Summer Scholar Grant

Title: Therapeutic Effect of Progesterone to Reduce Cognitive Deficits in the 3xTg Mouse Model of Alzheimer's Disease

Authors: *E. LAUZON[1,2,3,4], S. KONERU[1,2,3], N. WEDSTER[1,2,3], P. OTERO SEQUEIROS[1,2,3,5], E. IDZIOR[1,2,4], C. BUENO ALVAREZ[1,2,3], J. WASSELL[1,2,3], B. KEMMERLING[1,2,3], R. SCHALAU[1,2,3], K. BELGYA[1,2,3,6], J. ROSSIGNOL[1,2,3,5], G. L. DUNBAR[1,2,3,7,8];
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Abstract: Alzheimer's Disease (AD) is a neurodegenerative disease that first affects the entorhinal cortex and hippocampus regions of the brain. It is characterized by the aggregation of amyloid-beta (Aβ) protein in the form of plaques in the extracellular space and inclusions in the blood vessels, and hyperphosphorylation of the protein tau, in the form of intraneuronal aggregates of neurofibrillary tangles (NFTs). Clinical manifestations of AD include progressive memory loss, psychological and emotional disturbances, attention deficits, and visuospatial deterioration. Currently, no treatment has been shown to effectively modify the disease progression in AD. Most of the current treatments focus on specific aspects of AD and provide, at best, palliative effects on the targeted symptoms. We chose to test progesterone to treat memory dysfunction in the 3xTg mouse model of AD because of its pleiotropic effects. Progesterone is a major gonadal and neurosteroid hormone that has been shown to exert neuroprotective effects in *in vitro* and *in vivo* studies of brain injury and disease, including some rodent models of AD. Progesterone influences neuronal function by modulating gene transcription and cellular activity by binding to intracellular receptors abundant in the central nervous system. The mouse model used in the current study, 3xTg-AD shows both amyloid-beta plaques and neurofibrillary tangles, as well as the synaptic dysfunction that mimics what is observed in AD patients. The 3xTg-AD mice display behavioral impairments such as severe memory loss, as well as the emotionality and psychological changes that are analogous to those seen in AD patients. As the primary goal of this project was to determine if progesterone is an effective treatment for reducing the memory deficits in the 3xTg model of AD, we utilized a spatial reversal task using a water-T-maze. Daily oral-gavage injections of progesterone (5 mg/kg) or equivalent volumes of the vehicle was administered for 40 days. Our results indicated that progesterone-treated C57 wild-type male mice made fewer errors than vehicle-treated control and 3xTg mice, with the progesterone-treated 3xTg male group showing marginal improvement in their performance on the water-T-maze tests. Although further data analysis is required of additional behavioral tests, our results suggest that progesterone may provide benefits in reducing AD-induced cognitive deficits.

Disclosures: E. Lauzon: None. **S. Koneru:** None. **N. Wedster:** None. **P. Otero Sequeiros:** None. **E. Idzior:** None. **C. Bueno Alvarez:** None. **J. Wassell:** None. **B. Kemmerling:** None. **R. Schalau:** None. **K. Belgya:** None. **J. Rossignol:** None. **G.L. Dunbar:** None.

Poster

P234. Therapeutic Strategies: Preclinical Animal Models and Small Molecule Therapeutics IV

Location: Virtual Only

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Program #/Poster #: P234.07

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R56AG05755501 (to L.X. (PI), M.E.F.-P., P.R., P.S.)
Hunter RISE Grant 2R25GM060665-21

Title: The phosphodiesterase inhibitor ibudilast alleviates Alzheimerr's disease pathology and improves cognition in a transgenic rat model

Authors: *G. OLIVEROS[1], L. XIE[2], P. ROCKWELL[1], P. A. SERRANO[3], M. E. FIGUEIREDO-PEREIRA[1];
[1]Biol. Sci., [2]Computer Sci., [3]Psychology, CUNY Hunter Col., New York, NY

Abstract: Alzheimer's disease (AD) is a multifactorial disease characterized by the development of amyloid-beta (Aβ) plaques, neurofibrillary tangles, neuroinflammation, neuronal loss in the brain, and cognitive deficits. Therefore, treatment efficacy is likely dependent on a multi-target drug approach. Using a multi-scale predictive modeling framework, we identified Ibudilast (IBU), a phosphodiesterase (PDE) inhibitor and Toll-like receptor 4 (TLR4) antagonist, as a potential drug candidate. We predicted that IBU inhibits the interleukin-1 receptor associated kinase 1 (IRAK1) as an off-target drug effect. The promise IBU has shown in multiple sclerosis (MS) clinical trials and its efficacy in treating asthma and stroke by reducing neuroinflammation, makes IBU a favorable candidate for treating AD. The Fisher transgenic 344-AD rat model, which expresses human mutant "Swedish" amyloid-precursor protein (APPsw) and Δ exon 9 presenilin-1 (PS1ΔE9), was used to test the efficacy of IBU. TgF-344-AD rats exhibit age-dependent Aβplaques, tau paired-helical filaments, neuronal loss, microgliosis, and cognitive deficits, mimicking the progression of AD in humans. IBU-treatment started at 5 month of age. Following a 6 month IBU-treatment, we assessed spatial learning and memory in transgenic (Tg) and wild-type (WT) male and female rats compared to age-matched non-treated controls using the Active Place Avoidance Task (aPAT). At 11 months of age, untreated Tg rats exhibited deficits in acquisition and retention compared to their untreated WT counterparts. IBU-treatment mitigated these spatial learning deficits and improved memory retention in the Tg rats compared to untreated Tg rats. IBU-treatment also reduced hippocampal (hilar region) Aβ plaque and tau paired-helical filament, and reduced microgliosis. RNA sequencing analysis of Tg hippocampal tissue showed that IBU affects the expression of genes involved in the TLR and ubiquitin/proteasome pathways in a sex-dependent manner. With respect to the TLR pathway, IBU increased the expression of IRAK3, a negative regulator of IRAK1 activity, and altered the expression levels of TRAF6 and other TLR-related ubiquitin ligases and conjugases. Our results

support IBU as a promising drug candidate for AD treatment via its multi-target effects on the TLR and ubiquitin/proteasome pathways to alleviate AD symptoms including cognitive deficits.

Disclosures: G. Oliveros: None. **L. Xie:** None. **P. Rockwell:** None. **P.A. Serrano:** None. **M.E. Figueiredo-Pereira:** None.

Poster

P234. Therapeutic Strategies: Preclinical Animal Models and Small Molecule Therapeutics IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P234.08

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Weston Brain Institute
Ontario Veterinary College

Title: Neurometabolite 5α-androstane-3α,17β-diol enhances object recognition memory and hippocampal dendritic morphology in 3xtg-ad mice

Authors: *N. KUWAHARA[1], S. BHULLAR[1], K. TANG[1], L. K. ISAACS[1], K. C. NICHOLSON[1], K. JARDINE[2], B. D. WINTERS[2], N. J. MACLUSKY[1];
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Abstract: Women have an increased risk of developing Alzheimer's disease (AD) and experience worsened cognitive deterioration at the same stage of the disease, compared to men. This may be due, in part, to the sex differences in age-related decline of circulating gonadal steroid hormone levels. In animal studies, testosterone has been shown to exert neuroprotective and neurotrophic effects in the brain, while men with AD tend to have reduced free levels of circulating testosterone. Recent studies have demonstrated that the steroid hormone-mediated neuroprotection may be augmented through the conversion of primary gonadal steroid hormones into metabolites that may have distinct biological properties from those of their parent hormones. Previously, we showed that the 5α-reduced metabolite of testosterone, 5α-androstane-3α,17β-diol (3α-diol), may play a role in protecting neurons against the neurotoxic effects of amyloid beta exposure and hence contribute to the observed sex differences in AD. Inhibiting the synthesis of testosterone-derived neurosteroids impaired object recognition memory (ORM) and exacerbated AD-like neuropathology in 6-month-old male 3xTg-AD mice (expressing mutations associated with human familial AD: APP Swedish, MAPT P301L, and PSEN1 M146V). However, the possible protective effects of increasing 3α-diol levels *in vivo* remains unknown. To determine whether supplementation with 3α-diol might confer protection against the development of AD-like pathology, male and female 3xTg-AD mice were implanted s.c. with Silastic capsules releasing 3α-diol at 3 months of age (n=15-16 per treatment group). After 6

months of treatment, 3α-diol increased hippocampal CA1 dendritic branching and apical dendritic spine density in females compared to males, and improved ORM in 3xTg-AD females, eliminating the ORM deficit normally observed in female 3xTg animals at this age. These findings suggest that long-term treatment with 3α-diol may induce neuroprotective effects, counteracting the preferential loss of cognitive function otherwise observed in females. Ongoing experiments will determine the effects of 3α-diol on cognitive performance and AD-like neuropathology as the disease progresses beyond 9 months of age.

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Poster

P234. Therapeutic Strategies: Preclinical Animal Models and Small Molecule Therapeutics IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P234.09

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: ReST/UM 201446
SATT AxLR/UM 210324

Title: Neuroprotective effects of Fluoroethylnormemantine (FENM) after chronic infusion by Alzet pumps in the Aß$_{25-35}$ mouse model of Alzheimer's disease

Authors: *A. CARLES[1], A. FREYSSIN[1,2], S. GUEHAIRIA[1], G. RUBINSTENN[2], T. MAURICE[1];
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Abstract: Alzheimer's disease (AD) is a devastating neurodegenerative disease and available treatments are mainly symptomatic. In particular, memantine (Ebixa) is a non-competitive NMDA receptor antagonist which mechanism of action suggested promising symptomatic and neuroprotective effects. Fluroethylnormemantine (FENM) is a structural analogue of memantine originally synthesized as a positron emission tomography radioligand, with a substitution of the Fluor atom with 18F, to label NMDA receptors in vivo [Salabert et al., Nucl Med Biol 2015, 2018]. Recently, we showed that FENM, administered intraperitoneally, prevented the toxicity, neuroinflammation and learning deficits in Swiss mice intracerebroventricularly injected with oligomerized Aβ$_{25-35}$ peptide. The drug showed a superior efficacy as compared with Memantine and an absence of direct amnesic effect at higher doses [Couly et al., Int J Neuropsychopharmacol 2021]. In the present study, we compared two modes of administrations of FENM and memantine, namely chronic subcutaneous infusion with Alzet pump and bolus

intraperitoneal injections. C57Bl/6 mice were treated intracerebroventricularly with oligomerized $A\beta_{25-35}$ peptide and after different timepoints behavioral, biochemical and physiological parameters were analyzed. Different memory tests were used including spontaneous alternation in the Y-maze and object recognition test. Then, toxicity induced in the mouse hippocampus or cortex was analyzed biochemically or morphologically, using markers of apoptosis, neuroinflammation and oxidative stress. (De)regulations of NMDA receptor subunits were also analyzed in the hippocampus by western blots. Interestingly, data showed that infused FENM is as effective after infusion than observed after bolus administration, with effective doses in the 0.1-0.3 mg/kg/day range. FENM therefore appeared as a potent neuroprotective drug using two administration modes in the pharmacological model of AD. These results open the possibility of using the compound either as a patch or tablets in human therapy.

Disclosures: A. Carles: None. **A. Freyssin:** A. Employment/Salary (full or part-time):; ReST employment. **S. Guehairia:** None. **G. Rubinstenn:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); stock, patent holder, CEO ReST. **T. Maurice:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; SATT AxLR.

Poster

P243. Biomarkers in Neurodegenerative Diseases I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P243.01

Topic:

Support: University of Florida AI Initiative

Title: Percolation Theory Modeling and Biomarker Identification in an Alzheimer's Disease Mouse Model

Authors: *P. KOTLARZ[1], L. DA SILVA[1], K. XHINDI[1], M. HAGSTROM[1], L. SOSA[1], M. FEBO[2], J. NINO[1];
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Abstract: Alzheimer’s disease (AD) is a neurodegenerative disease that can lead to severe neurocognitive deficits. Due to the degenerative nature of AD, percolation theory, which examines how networks respond to damage, can be used as a potential model. Our objective was to utilize connectomic analysis in conjunction with percolation theory to understand network changes in AD to identify connectivity-biomarkers via fMRI. Effects of Aβ and inflammatory interleukin-6 (IL6), alone or in combination, on in vivo functional network connectivity in the

TgCRND8 mouse model for Alzheimer's-type Aβ deposition were examined using high-field functional magnetic resonance imaging (fMRI). Using a single shot echo planar imaging sequence at 11.1 Tesla, we scanned the brains of 8 months of age mice expressing brain-targeted IL6 or enhanced glial fibrillary protein (EGFP controls). Connectivity matrices were obtained via pre-processing the fMRI images. The matrices were averaged as a group, normalized, and thresholded at 10% graph density, or normalized and thresholded individually. Both global and nodal quantifiers were analyzed. Statistical significance of the quantifiers' difference between AD and control mice were evaluated using the Wilcoxon-rank sum test via nonparametrics. Differences between groups were further elucidated using network-based statistics employing t-tests comparing nodes and edges. Finally, networks and their response to targeted attacks based on specific quantifiers were analyzed. Significant differences were found in four global quantifiers (characteristic path length, global efficiency, network diameter, average strength) as well as specific nodal regions, notably the left ventral tier of the thalamus and left mesencephalic reticular formation. Examining network connectivity indicated reduced connectivity in anterior regions of the AD brain compared to control. This disconnected anterior cluster was also modeled utilizing iterative betweenness centrality and collective influence attack schemes. Percolation theory demonstrated an increase in vulnerability in AD brains in response to network attacks both in terms of overall network composition and sub-regions. While these results point toward significant differences in terms of vulnerability and connectivity between AD and control brains, employing percolation theory modeling can potentially identify preclinical AD via fMRI through examining anterior-to-posterior connectivity, vulnerability via slope analysis, and nodal baselines.

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Poster

P243. Biomarkers in Neurodegenerative Diseases I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P243.02

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Graph signal processing tools reveal links between brain regions and serum NfL levels in healthy subjects

Authors: *S. SIHAG[1], S. NAZE[2], F. TAGHDIRI[3], M. GUMUS[3], C. H. TATOR[4], R. GREEN[3], B. COLELLA[3], K. BLENNOW[5], H. ZETTERBERG[5], L. G. DOMINGUEZ[6], R. WENNBERG[3], D. MIKULIS[3], M. C. TARTAGLIA[6], J. R. KOZLOSKI[7];
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Toronto, ON, Canada; [5]The Sahlgrenska Acad. at the Univ. of Gothenburg, Mölndal, Sweden; [6]Univ. Hlth. Network, Toronto, ON, Canada; [7]IBM - TJ Watson Res. Ctr., Yorktown Heights, NY

Abstract: Neuro-axonal brain damage releases neurofilament light chain (NfL) proteins, which enter the blood. Serum NfL has recently emerged as a promising biomarker for neuro-axonal injury, monitoring treatment responses, and predicting prognosis in neurological diseases. Importantly, serum NfL level also increases with normal aging, so the interpretation of serum NfL levels in neurological diseases is confounded because a reliable model for age-related variation in serum NfL level in healthy subjects is currently lacking. Graph signal processing (GSP) has emerged as a powerful analytical tool to produce measures of relationships of functional dynamics of brain activity with the underlying structural white matter anatomy. Here, we leveraged a set of low and high graph frequency features extracted from the blood oxygen level dependent (BOLD) signals and structural connectome (Desikan-Killiany atlas, 66 cortical brain regions) using tools from GSP to test their association with serum NfL levels collected from healthy subjects with no history of neurological or developmental disorders (n = 20, mean age = 49.38 years). Age was significantly correlated with serum NfL in this cohort (Pearson's correlation = 0.71, p-value = 4.1e-4). A low and a high graph frequency feature were extracted from the BOLD data for each brain region, resulting in 132 total GSP features per subject. The low or high graph frequency features for a brain region were the metrics of increasing or decreasing conformity of its corresponding functional activity with the underlying structural anatomy. Using a linear model based univariate feature selection analysis, we found the low graph frequency features from isthmus cingulate (F-value = 20.175, false discovery rate (FDR) corrected p-value = 3e-4) and parahippocampus (F-value = 9.68, FDR corrected p-value = 6e-3) in the right hemisphere to be the most significantly related to serum NfL. Furthermore, using linear regression analysis, we observed that the low graph frequency feature from the isthmus cingulate explained 52% of the variance in serum NfL in this cohort. Additionally, mediation analysis revealed that the low graph frequency feature from isthmus cingulate had a significant partial mediation effect (p-val = 0.0056) on the association between age and serum NfL. Therefore, our results suggest that, in addition to age, these GSP-based neuroimaging features may be included in models of baseline variance when evaluating NfL as a biomarker in different contexts. Furthermore, our results raise new questions about the causal association between serum NfL levels and GSP features from certain brain regions during aging.

Disclosures: S. Sihag: None. **S. Naze:** None. **F. Taghdiri:** None. **M. Gumus:** None. **C.H. Tator:** None. **R. Green:** None. **B. Colella:** None. **K. Blennow:** None. **H. Zetterberg:** None. **L.G. Dominguez:** None. **R. Wennberg:** None. **D. Mikulis:** None. **M.C. Tartaglia:** None. **J.R. Kozloski:** None.

Poster

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Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P243.03

Topic:

Support: NIH Grant P01-AG066597

Title: De-differentiation of functional networks in corticobasal syndrome

Authors: C. T. MCMILLAN[1], **S. SIHAG**[1], J. S. PHILLIPS[1], P. A. COOK[2], M. GROSSMAN[1], J. C. GEE[2], D. IRWIN[1], T. SATTERTHWAITE[3], L. M. MASSIMO[4];
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Abstract: There is an increasing acceptance that neurodegenerative diseases manifest as network disorders. In particular, we have established that corticobasal syndrome (CBS) is characterized by structural network degeneration involving grey matter and white matter. Moreover, graph-theoretic analyses of diffusion magnetic resonance imaging (dMRI) have related efficiency of intrinsic brain networks to neuropathological features of CBS. However, little is known about the mechanisms of functional network degeneration in CBS. In this study, we draw upon evidence in the developmental literature that intrinsic networks segregate in adolescence to facilitate functional specialization of cognition and hypothesize that these networks become de-differentiated in neurodegeneration. We analyzed resting state functional MRI in clinically diagnosed CBS patients (23 subjects, 13 males, 10 females, mean age = 68.69 y) and Healthy Controls (12 subjects, 8 males, 4 females, mean age = 57.7 y). We focused our analyses on 5 network parcellations from the 200 parcel Schaefer Atlas (somatomotor, salience, limbic, frontoparietal and default mode network) and quantified de-differentiation using participation coefficient (PC) where higher PC corresponds to decreased network segregation, within-module connectivity for 5 modules, and their respective inter-modular connectivity. We additionally used ordinal regression to relate network segregation features to global clinical dementia rating scale (CDR) for CBS. All analyses were adjusted for age, sex, and average root-mean-square of head displacement, and their statistical significance was evaluated after false discovery rate (FDR) correction for multiple comparisons. The comparisons of segregation measures between CBS and healthy controls revealed that the within-module connectivity in somatomotor network was significantly lower in CBS and subsequently, PC in this network was higher in CBS cohort, consistent with de-differentiation of somatomotor network in CBS. Ordinal regression models suggested significant decreasing trends in within-module connectivity of frontoparietal network (β = -23.37, FDR corrected p-value = 0.04) and inter-modular connectivity between limbic and salience subnetworks (β = -37.21, FDR corrected p-value = 0.04) with worsening disease severity on CDR. These results are consistent with the hypothesis that de-differentiation of critical intrinsic networks is a feature of CBS-related neurodegeneration that increases with disease severity.

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Translational Bioinformatics, LLC. He also receives an honorarium as Associate Editor of NeuroImage: Clinical.. **S. Sihag:** None. **J.S. Phillips:** None. **P.A. Cook:** None. **M. Grossman:** None. **J.C. Gee:** None. **D. Irwin:** None. **T. Satterthwaite:** None. **L.M. Massimo:** None.

Poster

P243. Biomarkers in Neurodegenerative Diseases I

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P243.04

Topic: C.02. Alzheimer's Disease and Other Dementias

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Guangdong Provincial Fund for Basic and Applied Basic Research (2019B1515130004)

Title: A high-performance biomarker panel for Alzheimer's disease screening and staging identified by large-scale plasma proteomic profiling

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Abstract: Blood proteins are emerging candidate biomarkers for Alzheimer's disease (AD). A comprehensive investigation of the AD blood proteome will help identify additional biomarkers to delineate the disease's pathways and define specific AD stages. We quantified 1,160 plasma proteins in a Hong Kong Chinese cohort (n = 106 AD patients, n = 74 healthy controls) by high-

throughput proximity extension assay to identify AD-associated plasma proteins. Specifically, we showed that plasma proteins involved in diverse biological processes were dysregulated in AD. A subset of plasma proteins was selected to represent the AD plasma protein profile, which formed the basis of a scoring system that can accurately classify AD and associated endophenotypes. In addition, we showed that certain plasma proteins and biological processes exhibit stage-specific dysregulation in AD, thus adding biological annotations to AD stages. This study comprehensively profiled the AD plasma proteome and serves as a foundation for a high-performance, blood-based test for clinical AD screening and staging.

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Poster

P243. Biomarkers in Neurodegenerative Diseases I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P243.05

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: DGAPA/PAPIIT/UNAM IA210620
CONACYT Postdoctoral Fellowship
CONACYT PhD Student Fellowship

Title: Identification of potential new biomarkers for the diagnosis of Alzheimer's disease, through bioinformatic analysis.

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Abstract: Introduction. Alzheimer's disease (AD) is the most common cause of dementia, where substantial neuronal loss and neuropathological lesions can damage many brain regions, symptoms of the disease begin with mild memory difficulties and evolve towards cognitive impairment. The discovery of biomarkers that confer high confidence AD diagnosis would be a valuable tool to study the etiology of the disease, find risk factors, discover more treatments, and develop new drugs. Bioinformatics have made possible to analyze large amounts of transcriptomics data bases with greater efficiency to identify genes whose relationship with diseases such as AD was previously unknown. **Methods.** We used the total RNA sequencing

databases microarrays of tissue from the medial temporal lobe (MTL) and peripheral blood (PB) of patients with AD, which were processed by TrueSeq RNA-seq. The values were normalized with the Limma package of the statistical programming language. Subsequently, with the data obtained, we performed a computational analysis of the genetic differential expression using t-statistic-Limma. The data were corrected with the Benjamini and Hochberg approach and the genes with p values equal to or less than 0.05 were considered statistically significant. The direction of the change in gene expression was determined by its variation in the "log2-fold change" between controls and patients. A functional enrichment analysis of genetic ontology was performed using goana and topGO-Limma. The results were compared and confirmed with clusterProfilerR, we also applied the Fry-Limma analysis. **Results.** With MTL database we obtained 359 genes down regulated (DR) and 324 genes up regulated (UR), the altered pathways UR were: transcription, mitochondrial function and hydrolase activity, while DR: gene expression and regulation of cell proliferation, nuclear alterations, transmembrane receptor signaling. KEGG Analysis showed that metabolic pathways are altered UR and cytokine pathways and their receptors are altered DR. The analysis of PB samples showed 383 genes DR and 257 genes UR. KEGG analysis showed alterations in the Retinol and metabolism pathways. **Conclusion.** The intersection of the differentially expressed genes in the two databases exhibited 34 genes shared between the brain and blood arrangement: UR: SCG2, CA14, AHSP, HAPLN2, RALY-AS1, PLEKHG4, PRSS35, LINC01116; DR: DBH, NME8, CXCL6, SERPINA7, OR10H3, NYNRIN, MOS, C3, IL10RB-DT, CDKN2A-DT, CCDC60, CPB2, RBPMS6 CRST, T4ENT5D, DGKG. TRIM58 matched in both databases, DR in patients with AD. Search for altered genes in AD brain tissue will allow us to validate them in in blood samples, as a possible diagnostic tool.

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Poster

P243. Biomarkers in Neurodegenerative Diseases I

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Program #/Poster #: P243.06

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Artificial Intelligence Catalyst Research Fund

Title: Brain cortical differences in functional connectome strength in subjects with subjective memory complaints, mild cognitive impairment, and Alzheimer's disease

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Abstract: The early behavioral signs of Alzheimer's disease (AD), which include cognitive difficulty and mood changes, begin years-to-decades before the clinical symptoms of mild-to-severe cognitive impairment and dementia. The objective of the present study was to determine how functional connectivity (FC) between nodes located in distributed brain regions is modified at distinct stages of cognitive decline and AD. To gain insight into strength of connectivity and overall network topology, we applied graph theory analysis to graphs generated from functional connectivity data. We investigated functional connectivity between 300 nodes embedded within 17 distinct networks in control elderly participants (n=51), and participants classified as having subjective memory complaints (SMC, n = 18), early mild cognitive impaired (EMCI, n=34), MCI (n=11), late MCI (LMCI, n=13), or AD (n=7). Subjects were part of the ADNI study (ages: 61-96; n=134, 50% female) and the data were from their initial visit. All resting state functional MRI scans were collected on a Siemens Prisma Fit 3-Tesla scanner with the following parameters: EPI with 197 image repetitions, TR=3seconds, TE=30ms, 3.4mm isotropic voxel resolution. Graph theory-based calculations were applied to weighted undirected matrices constructed from 44,550 pairwise correlations between fMRI signals from 300 regions (Yeo parcellation). We identified a cluster of nodes in the somatomotor and dorsal attention networks that showed a gradual decline in node strength from controls to MCI and AD. A separate cluster of nodes located in default/dorsal attention network showed a transient increase in node strength. Pairwise permutation tests between each group vs controls indicated that differences in node strength varied rostral-caudally across the cortex from SMC to MCI and AD. In AD, significant differences in node strength were observed in occipital, temporal lobe, and superior parietal areas. Large-scale regional changes in node strength across pre- and postcentral gyri, superior parietal and occipital lobe appear to track differences from SMC to MCI and AD. Although the link to underlying synaptic function is unclear, the results suggest that the strength of communication across the cortex could vary over the course of progression of cognitive decline. While applying these network analysis techniques to AD is novel, our aim is to give an overview of our technique and the possible future application of connectomics as biomarkers of cognitive decline.

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Poster

P243. Biomarkers in Neurodegenerative Diseases I

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Program #/Poster #: P243.07

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: R01AG051086
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P30AG010133
P01AG014449
P30AG053760

Title: Role of novel microRNAs in neurodegenerative diseases

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Abstract: Neurodegenerative diseases, such as Alzheimer's disease (AD), are incurable and debilitating conditions that result in progressive degeneration and/or death of nerve cells. In particular, AD results from alterations of key biochemical pathways that lead to classic AD pathology of brain extracellular neuritic plaques generated from aggregates of amyloid-β peptide (Aβ), which is cleaved from Aβ precursor protein (APP), and intraneuronal fibrillary tangles of the hyperphosphorylated microtubule-associated protein tau. More importantly, this is accompanied by progressively debilitating dementia. Dysregulation of proteins involved in Aβ production, such as APP and β-secretase (or BACE1), and/or Aβ degradation, such as membrane metallo-endopeptidase (MME or neprilysin), contribute to excess Aβ deposition. Excess Aβ deposition contributes to tau hyperphosphorylation. MicroRNAs (miRNAs) are non-coding small RNAs that typically downregulate mRNA translation via mRNA 3'-untranslated region (UTR) sequences, and, consequently, many cellular processes. Notably, miRNA expression is often dysregulated in sporadic human disorders, including AD. We tested human specific miRNA species regulating human genes involved in AD. We used human cell cultures and brain tissues from control and AD subjects. Notably, we observed regulation of APP by miR-20b, miR-101 and miR-153; BACE1 by miR-9 and miR-339-5p; and MME by miR-181d. These miRNAs regulate protein expression via the respective target's mRNA-3'UTR (see an exception below). Interestingly we observed that overexpression of miR-298 reduces levels of APP, BACE1 and specific tau protein moieties. Further, we found that miR-346 uniquely *upregulates* APP levels via the APP mRNA *5'UTR* in human cultures and that this activity was part of iron (Fe) homeostasis. Here we report a novel human microRNA (miR20b) that alters AD risk probability and neuronal function. A specific polymorphism close to the MIR20B gene alters levels of CSF Aβ and structure of a critical brain region. Further, we have identified specific miRNAs in brain tissues with altered levels in AD patients vs. controls. Together, these multiple unique regulatory interactions may serve as novel therapeutic targets and enable the development of treatment strategies beneficial against human diseases, including AD. Complexity of interaction effects with other miRNAs may shed further light on brain health, AD and other neurodegenerative disorders.

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Poster

P243. Biomarkers in Neurodegenerative Diseases I

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Program #/Poster #: P243.08

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: R01AG051086
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P30 AG053760

Title: Alteration of a Specific MicroRNA Regulating Membrane Metalloendopeptidase Expression in Alzheimer's Disease

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Abstract: Alzheimer's disease (AD) is characterized by accumulation of amyloid β (Aβ) plaques and aggregates of hyperphosphorylated tau protein. Aβ biogenesis and degradation play critical roles in Aβ accumulation. Membrane metallo-endopeptidase (MME, neprilysin) is an Aβ-clearing enzyme. microRNA (miRNA), a group of small non-coding RNAs, regulates protein translation from mRNA. Disruption of miRNA regulation of MME could contribute to AD. Bioinformatics predicted miRNA binding sites on the MME mRNA 3'-untranslated region (3'-UTR), including the miR-181-5p family. We tested miR-181 activity with recombinant fusion reporter gene- MME 3'-UTR constructs. All four miR-181-5p (miR-181a, miR-181b, miR-181c, miR-181d) sequences downregulated reporter signal. We quantified miR-181-5p in human brain tissue specimens from non-cognitive impairment (NCI), mild cognitive impairment (MCI), and AD subjects. Of these, only miR-181d-5p significantly associated with disease status. Human neuronally differentiated cells were transfected with miR-181d-5p mimics. miR-181d-5p treatment produced significant reduction of MME mRNA and protein. Dysregulation of MME by (presumably excess) miR-181d could contribute to AD pathogenesis.

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Poster

P243. Biomarkers in Neurodegenerative Diseases I

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Topic: C.02. Alzheimer's Disease and Other Dementias

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Title: Lysosomal, glial and neuronal health biomarkers are altered in CSF and plasma from sporadic frontotemporal dementia (FTD) patients and FTD patients with GRN, MAPT or C9ORF72 mutations

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Abstract: Frontotemporal dementia (FTD) is a common cause of early-onset dementia. FTD is highly heritable, and 10-20% of all cases are caused by mutations in three genes: MAPT, GRN, and C9ORF72. As therapeutics are being developed for FTD, it is essential to identify disease-associated biomarkers in clinically accessible biofluids that may be used as pharmacodynamic biomarkers for dose selection and pathway engagement. It is also important to determine the levels of such biomarkers longitudinally along the course of disease progression.
In collaboration with ALLFTD/NCRAD, we took a discovery and validation approach for patient biomarker analysis. We used a pilot set of CSF and plasma samples from FTD patients with or without GRN mutations for biomarker discovery, and a large longitudinal set of samples including GRN, MAPT and C9ORF72 mutations for biomarker confirmation, examination of genotype effect and longitudinal stability. We used a multidisciplinary approach including targeted mass spectrometry analysis for lipids/metabolites, quantitative immunoassays for microglial and neurodegeneration biomarkers, and untargeted and targeted proteomics for synaptic and lysosomal proteins.
We found plasma glucosylsphingosine (GlcSph), a lysosomal biomarker proximal to progranulin, was elevated in GRN-FTD patients compared to non-GRN clinically normal controls, indicating GlcSph could be a useful peripheral pathway biomarker in clinical studies. CSF bis(monoacylglycerol)phosphate (BMP), a lysosomal phospholipid, showed decreases in sporadic FTD and MAPT-FTD. Glial biomarkers, YKL-40 and GFAP, were elevated in all FTD patients for YKL-40, and in sporadic-FTD, GRN-FTD & MAPT-FTD patients for GFAP. Consistent with previously reported findings, neuronal health related biomarkers, including

neurofilament light chain (Nf-L) and UCH-L1, were elevated for all FTD patients. Synaptic biomarkers NPTXR/1/2 were reduced in CSF of all FTD patients. Interestingly, Nf-L and NPTX2 levels were correlated with disease severity (FTLDCDR-SB) in GRN-FTD & MAPT-FTD patients. Nf-L levels were relatively constant during disease progression, positioning it as a disease related biomarker with potential for monitoring treatment effect.
Taken together, we have identified a panel of progranulin pathway and disease related biomarkers in CSF and plasma from FTD patients, which could serve as useful markers for monitoring treatment effects in the clinic.

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Poster

P243. Biomarkers in Neurodegenerative Diseases I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P243.10

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: AG046200

AG062378
Alzheimer Association IIRG 10-173180
MUSC interprofessional

Title: Dietary Methionine restriction modifies gut microbiota and improves memory in an Alzheimer Disease model

Authors: *K. SAMBAMURTI[1], E. J. AMELLA[2], I. CROMWELL[3], S. ZHANG[3], M. A. PAPPOLLA[5], C. VASU[4], D. K. LAHIRI[6], N. H. GREIG[7], P. VASUDEVARAJU[3];
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Abstract: High serum homocysteine is a risk factor for Alzheimer's Disease (AD). It has been reported to influence the accumulation of its characteristic lesions - senile plaques (SP) and neurofibrillary tangles (NFT) - containing amyloid β protein (Abeta) and microtubule-associated protein Tau (MAPT), respectively. It is also a known risk factor for retinal degeneration and typically increases in the aging population. We hypothesize that metabolic failure with aging might drive conditions that increase homocysteine and favor SP formation. Reduced food or calorie intake or branch chain amino acids are already known to benefit AD model mice. Restriction of Met, an essential aa, contributes to an increase in longevity without calorie restriction. We treated APP/PS1 transgenic mice with a 75% Met restricted (MR) diet and compared it with an isocaloric isonitrogenous complete diet (CD) for up to 12 months. MR diets significantly alter gut microbiota. As expected, the bacteria that metabolize Met and Cys are reduced. Furthermore, several major species of bacteria were reduced significantly, and the overall diversity of microbiota appears to be increased. Our data show that the treatment significantly drops brain soluble Abeta at 3 and 6 months. Abeta levels are very high and presumably saturated in older mice, explaining the lack of statistically significant differences between the different diets. Nevertheless, older mice show substantial improvement in behavior in a novel object recognition task after MR treatment. MR treatment is known to be senolytic, and therefore, we are currently examining related processes, such as preservation of lysosomal capacity, changes in metabolome, mRNA, and miRNA profiles. Senolysis and improvements in AD pathology may also be mediated by reduced inflammation due to the increased diversification of the microbiome. Thus, such diets may play an important role in strategies to prevent AD, retinal degeneration, and other age-associated disorders.

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Poster

P243. Biomarkers in Neurodegenerative Diseases I

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Program #/Poster #: P243.11

Topic: C.02. Alzheimer's Disease and Other Dementias

Title: Differential atrophy along the longitudinal hippocampal axis in Alzheimer's disease

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Abstract: Alzheimer’s disease (AD) is a progressive neurodegenerative disorder that primarily affects the hippocampus. Since hippocampal studies have highlighted a differential subregional regulation along its longitudinal axis, a more detailed analysis addressing subregional changes along the longitudinal hippocampal axis has the potential to provide new relevant biomarkers. This study analysed structural brain MRI data of 600 participants (aged 55-92 years-old) from the Alzheimer’s Disease Neuroimaging Initiative (ADNI). Hippocampi of cognitively normal (CN) subjects (n=200), mild cognitively impaired (MCI) subjects (n=200) and AD patients (n=200) were segmented with Freesurfer v6.0 and sub-segmented along its longitudinal axis in anterior, intermediate and posterior sub-divisions using an in-house algorithm.Total hippocampal volume normalized to intracranial volume (ICV) negatively correlated with age in all groups. Standardized residuals from the linear regression of normalized hippocampal volume by age were then calculated, showing that the mean relative atrophy corrected for the aging effect was higher in CN than in MCI, and lower in AD, with significant differences between all groups. Both anterior, intermediate and posterior hippocampus negatively correlate with age in all groups, with no significant differences between them. However, when normalizing to total hippocampus, only CN subjects show a significant positive correlation between anterior sub-division and age, and only CN and MCI posterior hippocampus negatively correlate with age. The longitudinal ratio of hippocampal atrophy (anterior sub-division divided by the posterior one) shows a significant increase with age in CN and MCI but not in AD.Normal aging affects the hippocampus differently along its longitudinal axis: its posterior aspect shows more atrophy and the anterior one becomes relatively more prominent. In AD, this antero-posterior gradient is disrupted early on, and hippocampal atrophy occurs in a similar manner along its longitudinal axis from late-adulthood to older ages.

Disclosures: T. Oliveira: None. **R. Morais-Ribeiro:** None. **A. Coelho:** None.

Poster

P265. Therapeutic Strategy: Preclinical: Small Molecule

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P265.02

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: DoD Grant W81XWH1810374 to SC
VA Merit Grant I01 CX001923-01 to SC

Title: Nexinhib-20 and CX546 ameliorates gut-derived extracellular HMGB1 induced BBB dysfunction and neuroinflammation in an experimental model of Gulf War Illness

Authors: *R. K. SETH[1], D. BOSE[1], P. SAHA[1], P. A. JANULEWICZ[2], J. O'CALLAGHAN[3], L. P. REAGAN[4], A. K. SHETTY[5], K. SULLIVAN[2], S. CHATTERJEE[1,6];
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Abstract: The persistence of inflammatory bowel disease, chronic fatigue, neuroinflammation, and cognitive impairment, are the common characteristics of Gulf War Illness (GWI). These symptoms predominantly involve the gut and brain and are associated with exposure to pesticides (permethrin, Per), pyridostigmine bromide (PB), multiple vaccinations, stress, etc., in the war theatre. In the current study, we aimed to examine how permethrin and PB exposure induced gut-derived extracellular high mobility group box-1 (HMGB1) led to blood-brain-barrier (BBB) dysfunction and neuroinflammation in a mouse model of GWI. The study also tested extracellular vesicles (EV) biogenesis inhibitor Nexinhib-20 (Rab27 inhibitor) and glutamate activator ampakines (CX546) in attenuating such neurological ailments. While using an established mouse model of GWI, the C57BL/6 mice (n=6 for each group) were exposed to GW agents (PB+Per) along with restraint stress for one week. Other groups of mice were co-exposed with GW agents and Nexinhib-20 or CX546. Results showed that GW agent exposure and stress increased EV biogenesis and HMGB1 expression in the small intestine and concurrently increased circulatory EVs loaded with HMGB1. Data also showed a decreased Zonal Occludin-1 (ZO1) and Claudin-1 mRNA expression and serum albumin accumulation in the frontal cortex, thus suggesting blood-brain barrier (BBB) integrity loss that likely enabled the entry of circulatory HMGB1 in the brain. Increased HMGB1 in both the frontal cortex and hippocampus activated microglia (M1 phenotype), thus inducing an inflammatory response via TLR4 signaling. We observed that HMGB1 induced neuroinflammation suppressed brain-derived neurotropic factor (BDNF) in the hippocampus and thus might modulate neuroplasticity. The GW agents exposed mice co-treated with EV inhibitor (Nexinhib20) or glutamate activator (ampakine, CX546) showed better BBB integrity and neuroplasticity, and reduced neuroinflammation. In summary, our findings suggested that the gut-derived extracellular HMGB1 played a crucial role in GWI pathogenesis, and Nexinhib 20 and CX546 might be promising therapeutics for GWI.

Disclosures: R.K. Seth: None. **D. Bose:** None. **P. Saha:** None. **P.A. Janulewicz:** None. **J. O'Callaghan:** None. **L.P. Reagan:** None. **A.K. Shetty:** None. **K. Sullivan:** None. **S. Chatterjee:** None.

Poster

P265. Therapeutic Strategy: Preclinical: Small Molecule

Location: Virtual Only

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Program #/Poster #: P265.03

Topic: C.03. Parkinson's Disease

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Ron and Pratima Gatehouse Fund
Anne M. and Phillip H. Glatfelter, III Family

Title: Oral squalamine treatment protects against parkinsonism

Authors: *T. SUBRAMANIAN[1], K. M. LE[2], K. SAUTER[4], K. VENKITESWARAN[3]; [1]Penn State Hershey Med. Ctr. & Col. Med., Hershey, PA; [2]Neurol., [3]Neurol. and NBS, Penn State Col. of Med., Hershey, PA; [4]Neurol., Penn State Univ. Col. of Med., Hershey, PA

Abstract: The aim of the present study was to determine if the novel prokinetic squalamine lactate (SL) would prevent parkinsonism in an environmentally relevant rat model of Parkinson's disease (PD). Exposure to paraquat, has been linked to development of PD in humans. In our previous studies, we demonstrated that oral paraquat plus lectin (P+L) could trigger the formation of misfolded / aggregated α-synuclein (Ser^{129}-P-α-syn) in the gut, which then can travel via the vagus nerve to reach the substantia nigra pars compacta (SNpc) causing degeneration of these neurons accompanied by symmetric parkinsonism that is L-dopa responsive. Bilateral subdiaphragmatic vagotomy prevents parkinsonism in this model and the degenerating dopaminergic neurons demonstrate aggregates of Ser^{129}-P-α-syn that result in Lewy Body-like structures (Anselmi L, et. al., *NPJ Parkinsons Dis.*. 2018;4(1):30). SL is thought to bind to the surface membrane of neurons in the gut and displace cell membrane bound α-syn and thus mitigate misfolding. We tested the hypothesis that oral administration of SL will prevent or mitigate the development of parkinsonism in rats treated with P+L. SL was administered via the water supply to single-housed adult female Sprague Dawley rats (n=7, 300-500 g) immediately following 7 day oral gavage of P+L. A control group (n=6) was not given SL after P+L. The vibrissae evoked forelimb placement test and the stepping task test were performed before and each week after P+L treatments. After 30 days, animals were perfused with 4% PFA and the brains processed for histology. P+L treated animals without SL, exhibited significantly lower vibrissae evoked forelimb placement test scores (*$p<.05$) than animals treated with SL. Preliminary histology showed bilateral TH loss in SNpc of P+L alone treated animals and

protection of TH loss in SL treated animals. Further analysis including assessments of Ser^{129}-P-α-syn in these animals are ongoing. Our data provides first evidence for effectiveness of oral treatments with SL in mitigating the effects of oral gavage P+L induced parkinsonism. Additional studies including male rats and testing of the effects of oral SL administration after symptoms develop 2 weeks after P+L treatment ongoing. These studies suggest that SL may be a very promising new therapeutic agent for the treatment and disease prevention of PD.

Disclosures: T. Subramanian: B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Research Grant from Enterin, Research Grants from Eli Lily, Intec, Pharma2B, NIH and PA Tobacco Settlement funds. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Squalamine is supplied by Enterin. D. Fees for Non-CME Services Received Directly from Commercial Interest or their Agents (e.g., speakers' bureaus); Honoraria from Adamas, Teva, Kyowa Krin, Supernus, Neurocrine, Acorda and Acadia. F. Consulting Fees (e.g., advisory boards); Consulting fees from Enterin, Adamas, Teva, Kyowa Krin, Supernus, Neurocrine, Acorda and Acadia. **K.M. Le:** None. **K. Sauter:** None. **K. Venkiteswaran:** None.

Poster

P265. Therapeutic Strategy: Preclinical: Small Molecule

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P265.04

Topic: C.03. Parkinson's Disease

Title: Evaluation of Anti-Parkinson's activity of Rajat bhasma on MPTP induced Parkinson's disease in C57BL/6 mice.

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Dept. of Pharmacol. & Therapeut., Seth GS Med. Col. & KEM Hosp., Mumbai, India

Abstract: Background and rationale: Parkinson's disease still has no cure or treatment which can halt the progression of disease by preventing neuronal death. It has symptom descriptions in Ayurveda and modern medicine. The study was designed to validate the claims in Ayurveda regarding the efficacy of Ayurvedic drugs in neurodegenerative disorders. It was decided to conduct an efficacy study of a herbomineral drugs (*Rajat bhasma*) on Parkinson's disease induced chemically in C57BL/6 mice. **Methods:** 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine **(**MPTP), 20 mg/kg, twice a day (12 hours in between) for 4 days, i.p. was used as an inducing agent in C57BL/6 mice. The herbomineral drug (*Rajat bhasma*) was used as study drug after carrying out toxicity study in our lab. The mice were fed with the drug for 15 days and MPTP was administered between 7th- 11th day. On the 15th day, the mice were subjected to neurobehavioural tests. Biochemical estimations of Malondialdehyde and reduced Glutathione

was carried out. Neurotransmitter analysis of Dopamine and it's metabolite (HVA and DOPAC) were carried out along with Immunohistochemistry of 5-hydroxy tyrosine. **Results:** The neurobehavioral assays showed an improvement as compared to the disease group. There was a distinct improvement in the inflammatory markers. Neurotransmitter analysis showed a significant improvement in the Dopamine level. Immunohistochemistry showed an distinct reduction in the loss of dopaminergic neurons. **Conclusion:** The drug (*Rajat bhasma*) was found to be effective in reducing the oxidative stress and also slow down the loss of dopaminergic neurons in the striatum.

Disclosures: A. Shankar: None.

Poster

P265. Therapeutic Strategy: Preclinical: Small Molecule

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P265.05

Topic: C.03. Parkinson's Disease

Support: NIH-R56NS109608
ABRC ADHS18-198846

Title: Analysis of the role of opioid receptors in the anti-dyskinetic effects of sub-anesthetic ketamine

Authors: *M. J. BARTLETT[1], C. J. STOPERA[2], A. ESQUEDA[1], S. J. SHERMAN[1], T. FALK[1];
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Abstract: We have previously demonstrated that treatment with sub-anesthetic ketamine (10-hrs; 5x20 mg/kg; *i.p.*; 2-hrs apart) can reduce levodopa (L-DOPA)-induced dyskinesia (LID) in a rat model of 6-hydroxydopamine (6-OHDA) and is acutely anti-parkinsonian. Ketamine is a multifunctional ligand with similar binding affinity to both *N*-methyl-D-aspartate (NMDA) and opioid receptors. Through NMDA receptor antagonism, ketamine modulates synaptic function, leading to brain-derived neurotrophic factor release onto striatal medium spiny neurons and neuroplasticity. However, the striatum is rich in both μ- and δ-opioid receptors and it is known that long-term L-DOPA therapy changes the levels of opioid peptides. Recent data also indicates that opioid agonism is required for the anti-depressive activity of ketamine. This leads to the hypothesis that ketamine may also activate opioid receptors in the basal ganglia, contributing to either the anti-dyskinetic or the anti-parkinsonian action of the drug, or possibly both. In this study, we used the pan-opioid receptor antagonist naloxone to investigate if opioid receptor activation is necessary for reducing LID. In this study, unilateral 6-OHDA lesioned male

Sprague-Dawley rats were treated with 6mg/kg of L-DOPA for 21 days to establish a model of LID. Once abnormal involuntary movements (AIMs) were stable over three testing days, the rats (n=7/group) were then treated on Day 0 with either vehicle, ketamine (20mg/kg), or ketamine (20mg/kg) + naloxone (3mg/kg) via the same 10-hour treatment protocol. L-DOPA was administered immediately following the protocol and AIMs were assessed. All animals received a 6mg/kg dose of L-DOPA every 3-4 days to maintain the LID and AIMs were scored once per week. Similar to our published data, ketamine significantly reduced AIMs by 36% immediately following the treatment protocol ($p < 0.05$). However, the addition of naloxone did not prevent ketamine's effects (mean AIMs ± SEM; ketamine = 24.1 ± 1.9 vs. ketamine + naloxone = 22.9 ± 4.4). By Day 7, ketamine treated animals had a non-significant 23% reduction in AIMs scores as compared to both the vehicle and ketamine + naloxone treated groups, and again there was no significant difference between the two treatment groups. This pilot study provides early evidence suggesting that opioid receptor activation may not be necessary for ketamine's anti-dyskinetic effects. Next, we will escalate the doses of both ketamine and naloxone. And in a follow-up study we will investigate in unilateral 6-OHDA-lesioned rats if the anti-parkinsonian activity of ketamine is dependent on opioid receptor activation using the forelimb adjusting steps and rotarod tests.

Disclosures: M.J. Bartlett: None. **C.J. Stopera:** None. **A. Esqueda:** None. **S.J. Sherman:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); SJS and TF have a pending patent application for the use of ketamine as a novel treatment for levodopa-induced dyskinesia associated with Parkinson's disease. **T. Falk:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); SJS and TF have a pending patent application for the use of ketamine as a novel treatment for levodopa-induced dyskinesia associated with Parkinson's disease..

Poster

P265. Therapeutic Strategy: Preclinical: Small Molecule

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P265.06

Topic: C.03. Parkinson's Disease

Title: Impulsive behavior induced by Dopamine D3 receptor agonist treatment in Parkinson's disease is mediated by β-arrestin

Authors: *S. DAS, W. XU, S. KORTAGERE;
Microbiology, Drexel Univ., Philadelphia, PA

Abstract: Impulsive behavior induced by Dopamine D3 receptor agonist treatment in Parkinson's disease is mediated by β-arrestin**Sanjay Das**, Wei Xu and Sandhya

KortagereParkinson’s Disease (PD) is a movement disorder characterized by progressive degeneration and loss of mid-brain dopaminergic neurons. Pramipexole (PRX), a Dopamine D3 receptor (D3R) agonist, has emerged as a potent therapeutic to treat motor deficits, but chronic therapy leads to impulse control disorders (ICDs) in PD patients. ICDs comprise a group of complex behavioral disorders characterized by failure to resist an impulse or temptation to perform an act that is harmful to the individual or to others. While the etiology of ICDs is not well understood, several clinical and animal studies have demonstrated that D3R agonist activity directly correlates with the development ICDs. D3Rs are highly expressed in the mesolimbic pathways and mediate motor and reward processes in the brain. D3R agonists can signal through multiple signaling pathways and predominantly through the activation of G-proteins, β-arrestin, or both. We hypothesize that activation of β-arrestin signaling pathway by D3R agonists such as PRX can induce ICDs in PD. To test this hypothesis, PD rats were trained to criteria in a 5-choice serial reaction time task (5CSRTT) and their baseline levels of impulsive responses (measured as premature responses), correct responses and omissions were recorded. Animals were then treated with vehicle or PRX or our G-protein biased D3R agonist SK609 for 3 weeks (1x/day; i.p) and their responses in the 5CSRTT were measured at the end of week-3. Results from the study demonstrated that PRX significantly increased premature responses compared to vehicle or SK609 treatment. SK609 reduced premature responses and increased the correct responses and reduced omissions when compared to both PRX and vehicle treatment. The increase in premature responses by PRX treatment correlated with an increase in anxiety as measured by time spent in the closed arms of the elevated plus maze. Current studies are aimed at understanding the molecular mechanisms of β-arrestin mediated signaling that promote ICDs. Our study will augment the understanding of biased D3R signaling and its role in modulating ICDs in PD. This study’s outcome will be directly translatable to optimizing therapy for PD without the risk of developing ICDs seen with existing therapies.

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Poster

P265. Therapeutic Strategy: Preclinical: Small Molecule

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P265.07

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: SC INBRE 21-4275

Title: The selective inhibitors for Heat shock protein 90 beta and tumor necrosis factor type 1 reduce the lipopolysaccharide-induced inflammatory response in BV2 microglial cells

Authors: *V. KLIEBE[1], A. G. SMITH[2], C. G. HANNA[2], B. S. J. BLAGG[3], W. LEI[2];
[1]Dept. of Biol., Presbyterian Col., Clinton, SC; [2]Presbyterian Col. Sch. of Pharm., Clinton, SC;
[3]Dept. of Chem. and Biochem., Univ. of Notre Dame, Notre Dame, IN

Abstract: The selective inhibitors for Heat shock protein 90 beta and tumor necrosis factor type 1 (TRAP1) reduce the lipopolysaccharide (LPS)-induced inflammatory response in BV2 microglial cells Valentin M Kliebe[1], Amanda G Smith[2], Christen Hanna[2], Brian SJ Blagg[3], Wei Lei[21] Department of Biology, College of Art and Science, Presbyterian College, Clinton, SC 29325[2] Department of Pharmaceutical and Administrative Sciences, Presbyterian College School of Pharmacy, Clinton, SC 29325[3] Department of Chemistry and Biochemistry, University of Notre Dame, Notre Dame, IN 46556Neuroinflammation contributes to several health conditions related to the central nervous system, such as Alzheimer's Disease and pain. Understanding the regulation of neuroinflammation is essential for discovering approaches to treat inflammation-related diseases. Previous studies demonstrated that the inhibition of Heat shock protein 90 (Hsp90) using 17-N-allylamino-17-demethoxygeldanamycin (17-AAG) could reduce neuroinflammation. However, the contribution of Hsp90 isoforms, including Hsp90α, Hsp90β, glucose-regulated protein 94 (Grp94), and tumor necrosis factor type 1 (TRAP1), to inflammatory signaling remain poorly understood. This study aimed to determine the impact of selective inhibitors for different Hsp90 isoforms on lipopolysaccharide (LPS)-induced inflammatory responses in microglial cells. A murine microglial cell line, BV-2 cells, were pretreated with selective inhibitors for different Hsp90 isoforms for 1 hour, followed by the stimulation of LPS. The production of Nitric Oxide (NO) was measured using Griess Reagents, and qPCR was performed to detect the expression of tumor necrosis factor alpha (TNF-α) and Interleukin-1 beta (IL-1β). The activation of the nuclear factor-κB (NF-κB) and Nrf2 were investigated. We found that treatment with Hsp90β inhibitors strongly inhibited the production of NO, TNF-α, and IL-1β, and the activation of NF-κB, but showed a modest effect on Nrf2 activation. Meanwhile, a TRAP1 inhibitor, T3, also exhibited anti-inflammatory activities via enhancing the Nrf2 activation. However, the inhibitors targeting Hsp90α and Grp94 had very limited or no effect on the LPS-induced inflammatory responses in the microglial cells. These findings demonstrate that Hsp90β and TRAP1 are the isoforms of Hsp90 which play essential roles in the LPS-induced neuroinflammation in microglial cells.

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Poster

P266. Therapeutic Strategies: Cellular Models

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Program #/Poster #: P266.01

Topic: C.03. Parkinson's Disease

Support: NINDS 1R01NS117968
NINDS R21 NS109505
NIA SBIR 1R43AG063675
UL1TR002494
T32GM132039
RF1AG053951

Title: Advancements in cellular time-resolved FRET biosensor for a high-throughput drug pipeline identifying small molecule inhibitors of alpha-synuclein pathology.

Authors: *A. R. BRAUN[1], E. E. LIAO[1], M. HORVATH[3], P. KALRA[2], K. ACOSTA[4], M. C. YOUNG[1], N. NATHAN KOCHEN[1], C. LO[1], R. BROWN[6], M. D. EVANS[6], W. C. K. POMERANTZ[7], E. RHOADES[4,5], K. C. LUK[9], R. L. CORNEA[8,10], D. D. THOMAS[8,10], J. N. SACHS[1];
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Abstract: Preventing or reversing the pathological misfolding and self-association of alpha-synuclein (aSyn) can rescue a broad spectrum of pathological cellular insults that manifest in Parkinson's disease (PD), dementia with Lewy bodies (DLB), and other alpha-synucleinopathies. Over the past several years, the field's understanding of which forms of aSyn assemblies are most toxic has evolved, shifting away from mature fibrils rich in β-sheets towards structurally heterogeneous oligomers. Many questions remain regarding these oligomers, including what their exact macromolecular constituency is in cells (i.e., what co-mingles with aSyn), whether they possess any well-defined structural motifs—various forms, including a tetramer, have been described in the literature—and how variations in their molecular properties can contribute to toxicity. These unknowns complicate drug discovery, where it remains to be seen whether a unique small-molecule binding site or epitope even exists for oligomers. More generally, it is unclear whether direct binding of an inhibitor to aSyn oligomers, or fibrils for that matter, is more efficient than indirectly targeting the cellular machinery and pathways that can prevent or alter their assembly.
In our recently published study, we demonstrated our aSyn live-cell time-resolved FRET biosensors performance in a high-throughput screening (HTS) campaign with the 1280-compound LOPAC library. Our combination of inter-protomeric and intra-protomeric FRET biosensors provide complementary insight into aSyn oligomerization and conformation. We then tested the efficacy of hit compounds in SH-SY5Y cells overexpressing unlabeled wild-type aSyn, and we report that two small molecules, Demeclocycline HCl (DEM) and Ro 90-7501 (RO), completely protected SH-SY5Y cells from aSyn-induced death. The compounds are active in the low nanomolar range (EC_{50} = 65nM and 78nM respectively) and are an order of magnitude more potent than previously described small molecules.
We have continued to refine our HTS drug discovery pipeline to include novel FLT-FRET

biosensors and functional assays to improve both the scalability of the HTS platform and to dive deeper into the potential mechanisms of action of hit compounds.

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Poster

P266. Therapeutic Strategies: Cellular Models

Location: Virtual Only

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Program #/Poster #: P266.02

Topic: C.03. Parkinson's Disease

Support: Research Grant from The Army Research Office (ARO) # W911NF-16-1-0311
Merit Award from the Veterans Administration (VA) Biomedical Laboratory Research and Development 1I01BX003303-01

Title: Nad^+ biosynthetic pathway enzymes moonlight as molecular chaperones to protect against proteotoxic stress

Authors: *M. PINKERTON[1], A. BARRIENTOS[2];
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Abstract: Neurodegenerative proteinopathies involve the misfolding and aggregation of disease-specific proteins in the brain. Searches for neuronal maintenance factors have identified the NAD+ salvage biosynthetic pathway enzyme NMNAT (nicotinamide mononucleotide (NMN) adenylyltransferase) as an effective suppressor of proteotoxicity. Although there has been some controversy regarding whether the neuroprotective effect of NMNAT is mediated by increased NAD+ levels or a non-catalytic chaperone role of NMNAT, it is now accepted that both may contribute. Screens in yeast models of HD and PD in our lab have allowed us to identify not only the NMNAT homologs Nma1/2 but three additional enzymes of the NAD+ salvage pathway that achieve similar protection against extended polyglutamine- and a-synuclein-induced proteotoxic stress: Npt1, Pnc1, and Qns1. Under proteotoxic stress, the four proteins are recruited to promote the clearance of misfolded and oligomerized proteins. We have shown that the suppression mechanism by NAD+ proteins is independent of their catalytic activity. In vitro and in vivo studies have indicated that the four proteins have the ability to act as molecular chaperones. Our data suggest that they perform holdase and foldase chaperone activities to contribute to the reduction of misfolded toxic proteins, while promoting their refolding. The proteotoxicity suppression mechanism requires the presence of the HSP90 chaperones HSP82/HSC82. In the

case of Nma1, structure-function relationship studies have shown that the C-terminus of the protein is essential for its chaperone activity and presumably for interaction with HSP90. Our data illustrates the existence of an evolutionarily conserved strategy of repurposing housekeeping enzymes under stress conditions, such as age-associated neurodegenerative proteotoxicities.

Disclosures: M. Pinkerton: None. **A. Barrientos:** None.

Poster

P266. Therapeutic Strategies: Cellular Models

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Program #/Poster #: P266.03

Topic: C.03. Parkinson's Disease

Support: Center for Regenerative Nanomedicine in the Simpson Querrey Institute for BioNanotechnology

Title: Gdnf mimic supramolecular nanostructure

Authors: *O. CARBALLO-MOLINA, A. EDELBROCK, *Z. ÁLVAREZ, T. PEREZ-ROSELLO, S. CHIN, T. FYRNER, J. SURMEIER, S. STUPP;
Northwestern Univ., Chicago, IL

Abstract: Parkinson´s Disease (PD) is the second most common neurodegenerative disorder. The administration of glial cell line-derived neurotrophic factor (GDNF) has shown to improve the motor dysfunctions in animal models of PD. However, GDNF has not shown the desired results the clinical outcomes, in part due to a short half-life and the uncontrolled diffusion of this trophic factor. To overcome those obstacles, we report here the incorporation of a GDNF-mimetic peptide on the surface of peptide-based nanofibers. Peptide amphiphiles (PAs) are self-assembling molecules that form nanostructures that have shown the ability to retain and control the release of various growth factors. We show that the nanostructure displaying the GDNF mimetic activates the canonical GDNF receptor RET *in vitro*. Using two-dimensional cell cultures, we demonstrate that GDNF mimetic PA enhanced the cell viability of dopaminergic neurons derived from human induced pluripotent stem cells and promotes their maturation by increasing the expression of MAP2, pre and post-synaptic markers, as well as the improvement of morphometric characteristics. In addition, we observed the improvement of the electrophysiological activity. Like native GDNF, this bioactive nanostructure provides a neuroprotective effect against the neurotoxin 6-OHDA. Furthermore, we used three-dimensional cultures and we found that the neurons encapsulated in the GDNF mimetic PA gels showed better maturation, morphometric and neuroprotective effects, compared to control gels. These results indicate that GDNF mimetic PA is a bioactive matrix capable of mimicking the key properties of GDNF. This bioactive nanostructure has the potential to improve cell replacement

therapy in PD by improving the cell viability of grafted neurons, as well as the morphometric and functional maturation.

Disclosures: O. Carballo-Molina: None. **A. Edelbrock:** None. **Z. Álvarez:** None. **T. Perez-Rosello:** None. **S. Chin:** None. **T. Fyrner:** None. **J. Surmeier:** None. **S. Stupp:** None.

Poster

P266. Therapeutic Strategies: Cellular Models

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Program #/Poster #: P266.04

Topic: C.03. Parkinson's Disease

Support: R01-NS117968

Title: Discovery of small molecule inducers of autophagy using a phenotypic mutant A53T alpha synuclein autophagy biosensor

Authors: *E. E. LIAO, A. R. BRAUN, J. N. SACHS;
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Abstract: Dysregulation of the autophagy pathway has been implicated in various neurodegenerative diseases. As progress is made in understanding the pathogenesis of Parkinson's disease (PD), dementia with Lewy bodies (DLB) and other alpha-synucleinopathies, it is clear that basal autophagy may be a key factor in increasing neuron survival and promoting the clearance of toxic protein aggregates. We designed a high throughput autophagy screening platform that is sensitive to mutant A53T alpha synuclein (aSyn) pathology. Our phenotypic biosensor uses a combination of the LC3 probe with internal control designed by Noboru Mizushima and A53T aSyn expression to screen for small molecule inducers of autophagy that can rescue mutant aSyn pathology. Stable HEK cell clones expressing GFP-LC3-RFP(LC3ΔG) were selected under puromycin and confirmed using flow cytometry and PCR. CRC plates were run using Rapamycin, starvation condition and Bafilomycin A1 to determine the optimal range of the mutant aSyn autophagy biosensor. Coefficient of variance (CV) was also determined on control plates to verify a good HTS screen. Basal GFP/RFP levels in A53T aSyn cells were significantly higher than GFP/RFP levels of HEK-LC3 alone and WT aSyn expressing cells, indicating higher levels of autophagy impairment. Further characterization of the phenotypic biosensor was conducted using immunoblots and LC3 turnover assays, which confirmed blockage of autophagy in A53T aSyn biosensor. Using this mutant aSyn autophagy biosensor, a small molecule drug screen was conducted using the Selleck library of 2,863 compounds with Rapamycin and Bafilomycin A1 as positive and negative tool compounds, respectively. Hit compounds that decreased the GFP/RFP ratio (signifying an increase in the autophagy flux) were further evaluated using a LC3 turnover assay to confirm effects on autophagy. Consequent

analyses using TFEB localization and Lysotracker in conjunction with LAMP1 immunofluorescence were used to confirm and validate the hits. We then tested the hits in a differentiated SH-SY5Y model of PD expressing A53T aSyn and evaluated cellular viability and rescue of aSyn pathology. Hits were further vetted using a HEK-LC3 counter screen to determine pathologic aSyn specific effects. Further evaluation of mechanisms of action of the hits will be conducted in future studies.

Disclosures: E.E. Liao: None. **A.R. Braun:** None. **J.N. Sachs:** None.

Poster

P266. Therapeutic Strategies: Cellular Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P266.05

Topic: C.03. Parkinson's Disease

Title: Development of an in vitro α-synuclein aggregation assay in primary cortical cultures for pharmacological characterization, target discovery and validation

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Abstract: In Parkinson's disease (PD), the formation and propagation of α-synuclein (α-syn) insoluble amyloid structures is a process hypothesized to drive the pathology of disease. As part of this process, impaired protein clearance and protein aggregation within neurons occurs. From a drug discovery perspective, preventing protein aggregation or promoting protein clearance provide interesting opportunities as pharmacological intervention points. However, there is currently a lack of biologically relevant in vitro systems modelling these mechanisms. Such models are much needed for e.g. development of small molecules, or for further increasing the understanding of pathological processes involved in neurodegenerative disease.
With this in mind, we initiated the development and validation of a high-capacity in vitro model based on primary embryonic mouse cortical cultures in the 384-well format, where endogenous α-syn aggregation was induced at 6 or 10 days in vitro (DIV) using recombinant human α-syn pre-formed fibrils. Endogenous α-syn aggregation and cell health was then quantified at 14 or 17 DIV using immunocytochemistry and automated high content imaging and analysis.
Following the development of a robust protocol for induction and quantification of α-syn aggregation, the model was validated using commercially available small molecule modulators of α-syn aggregation, as well as using lentiviral shRNAs for knock-down of a small set of genes (SNCA, LRRK2, GBA and TMEM175) known for their involvement in PD.
The outcome is a robust, sensitive and high-capacity dual assay for assessment of α-syn aggregation, where it is possible to detect both negative and positive modulators of α-syn

aggregation even in single point screening format. While most of the commercially available small molecules tested were only weakly potent regulators of α -syn aggregation, the knock-down of genes relevant for PD did result in the expected effect on α-syn aggregation for all genes tested. We can therefore conclude that the developed model indeed is relevant for characterization of modulators of α-syn aggregation and can be used for screening of small molecule regulators as well as for target discovery and validation applications.

Disclosures: S. Lardell: A. Employment/Salary (full or part-time):; Cellectricon AB. **C. Nodin:** A. Employment/Salary (full or part-time):; Cellectricon AB. **A. Back:** A. Employment/Salary (full or part-time):; Cellectricon AB. **P. Karila:** A. Employment/Salary (full or part-time):; Cellectricon AB. **S. Illes:** A. Employment/Salary (full or part-time):; Institute of Neuroscience and Physiology, The Sahlgrenska Acadamy, University of Gothenburg. **J. Pihl:** A. Employment/Salary (full or part-time):; Cellectricon AB.

Poster

P266. Therapeutic Strategies: Cellular Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P266.06

Topic: C.03. Parkinson's Disease

Title: Patient-derived three-dimensional Cortical Neurosphere to model Parkinson's Disease

Authors: *W. RAJA[1], **E. NEVES**[1], C. BURKE[1], X. JIANG[1], P. XU[1], K. RHODES[1,2], V. KHURANA[3], R. SCANNEVIN[1,4], C. CHUNG[1];
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Abstract: There are currently no preventive or disease-modifying therapies for Parkinson's Disease (PD). Prevalence is consequently rising, as is the substantial burden on patients, caregivers, and the healthcare system. Failures in clinical trials necessitate a re-evaluation of existing pre-clinical models in order to adopt models that better recapitulate underlying disease mechanisms and better predict clinical outcomes. In recent years, models utilizing patient-derived induced pluripotent stem cells (iPSC) offer an attractive alternative, as they better recapitulate disease-relevant neuropathology *in vitro* without exogenous overexpression of disease-related pathologic proteins. Here, we utilize iPSC derived from a patient either harboring a point mutation (A53T) in the α-synuclein gene or copy number variation in α-synuclein (triplication of α-synuclein locus) associated with a strong early-onset PD and dementia phenotype. We generated a three-dimensional (3D) cortical neurosphere culture model to better mimic the microenvironment of the brain. We extensively characterized the differentiation process using quantitative PCR, Western blot, and immunostaining. Differentiation and aging of the neurospheres revealed alterations in fatty acid profiles and elevated total and pathogenic

phospho-α-synuclein levels in both A53T and the triplication lines compared to their isogenic control lines. Furthermore, treatment of the neurospheres with a small molecule inhibitor of Stearoyl Co-A Desaturase (SCD) attenuated the protein accumulation and aberrant fatty acid profile phenotypes. Our findings suggest that 3D cortical neurosphere model is a useful tool to interrogate targets for PD and amenable to test small molecule therapeutics.

Disclosures: W. Raja: A. Employment/Salary (full or part-time):; Full time. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); stock, stock option. **E. Neves:** A. Employment/Salary (full or part-time):; Full time. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); stock, stock option. **C. Burke:** A. Employment/Salary (full or part-time):; Full time. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); stock, stock option. **X. Jiang:** A. Employment/Salary (full or part-time):; Full time. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); stock, stock option. **P. Xu:** A. Employment/Salary (full or part-time):; Full time. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); stock, stock option. **K. Rhodes:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); stock, stock option. **V. Khurana:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); stock, stock option. **R. Scannevin:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); stock, stock option. **C. Chung:** A. Employment/Salary (full or part-time):; Full time. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); stock, stock option.

Poster

P266. Therapeutic Strategies: Cellular Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P266.07

Topic: C.03. Parkinson's Disease

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NRF-2020R1F1A1076616
BCRI21039
Jeollanam-do Science and Technology R&D Project

Title: Autophagy regulation pathways in rotenone-induced Parkinson's disease in SH-SY5Y cells treated with neural induced-human adipose-tissue derived stem cells conditioned medium

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Abstract: Rotenone (ROT) is a naturally occurring piscicide that inhibit mitochondrial complex I causing aggregated forms of alpha-synuclein was most widely used to induce Parkinson's disease (PD). Autophagy is the major intracellular degradation system involving lysosomal degradation of damaged organelles and abnormal aggregated proteins which is regulated by mechanistic target of rapamycin (mTOR)-dependent and -independent pathways. The phosphorylation of mTOR multiprotein signaling complexes (mTORC1 and mTORC2) controls several cellular functions. AMP-activated protein kinase (AMPK) directly regulates phosphorylation of ULK1 and function of ATG13. mTOR-independent MAPK/ERK have also been implicated in the regulation of autophagy. The autophagic marker, LC3B, binds autophagic related (ATG) proteins such as ATG3, 5, 7, and 12 to formation of autophagosome. These autophagosomes fused with functional lysosomes result in the formation of autolysosome for further degradation in the cytosol. Impaired autophagy contributes to the pathogenesis of neurodegenerative diseases including PD via accumulation of abnormally aggregated alpha-synuclein. In this study, we examined the neurogenic differentiation of human adipose tissue-derived stem cells (NI-hADSC) conditioned medium (NI-hADSC-CM) against ROT-induced toxicity in SH-SY5Y neuroblastoma cells. ROT toxicity significantly decreased the phospho-/total- (p-/t-) mTORC1 and p-/t-mTORC2 along with p-/t-ULK1 and ATG13 by increased p-/t-AMPK. Moreover, p-/t-ERK and beclin-1 were decreased by ROT but not changed the p-/t-Akt. ROT promoted the lipidation of LC3BI to LC3BII by inducing autophagy representing an adaptive cellular response. ATG3, 5, 7, and 12 were decreased and p62 level was not changed by ROT. Lysosomal LAMP1, LAMP2, and TFEB were decreased during ROT toxicity leading to lysosomal dysfunction. However, NI-hADSC-CM treatment increased the phosphorylation of mTORC1 and mTORC2 and decreased the phosphorylation of AMPK along with increased phosphorylation of ULK1 and ATG13. p-/t-ERK, p-/t-Akt, and level of beclin-1 were increased by NI-hADSC-CM treatment against ROT. In addition, NI-hADSC-CM treatment restored the ROT-induced increase in the level of LC3BII. Together with increased p62 levels, Atgs and lysosomal proteins were all normalized to the control levels by NI-hADSC-CM treatment. Taken together, our results show that NI-hADSC-CM treatment on autophagy regulating signaling pathways could facilitate the beneficial effects on Parkinson's disease.

Disclosures: M. Ramalingam: None. **J. Hwang:** None. **H. Cho:** None. **B.C. Kim:** None. **S. Jang:** None. **H. Jeong:** None.

Poster

P266. Therapeutic Strategies: Cellular Models

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Program #/Poster #: P266.08

Topic: C.03. Parkinson's Disease

Support: NIH Grant NS105826
Michigan Parkinson Foundation

Title: Interaction of Advancing Age with the BDNF Val66Met Polymorphism (rs6265): Impact on Dopamine Neuron Graft Efficacy and Side-Effect Liability in rs6265 Knock-In Rats

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Abstract: We previously investigated the effects of the Val66Met (aka: rs6265) single nucleotide polymorphism (SNP) in the gene for brain-derived neurotrophic factor (*BDNF*) in a rat model of neural grafting for Parkinson's disease (PD), demonstrating that young mature rats (grafted at 8 m.o.) carrying the variant Met allele exhibited paradoxical enhanced graft function but also developed aberrant graft-induced dyskinesias (GID) in contrast to wild-type (WT) rats (PMID: 33188920). Since aging is a primary risk factor for PD and known to reduce graft efficacy, we extended our studies to investigate whether aging interacts with this SNP to further alter cell transplantation outcomes. We hypothesized that aging would (1) dampen enhancement of graft function associated with this genetic variant and (2) exacerbate GID in all grafted subjects. To test this, middle-aged WT (Val/Val) and Met/Met SNP rats (grafted at 15m.o.) were rendered unilaterally parkinsonian, primed with levodopa, then transplanted with WT embryonic day 14 ventral mesencephalic dopamine (DA) neurons. Graft function was assessed over 10 weeks. Reductions in levodopa-induced dyskinesia (LID) severity served as the primary measure of graft function; OFF-time sensorimotor dysfunction served as a secondary measure. In this experiment we demonstrate, in contrast to our hypothesis, that behavioral enhancement associated with rs6265 is maintained with advanced age; however, behavioral improvement per grafted cell was significantly greater in the young cohort compared to the middle-aged cohort at the final post-graft time point regardless of genotype (2-way ANOVA $F(1,24) = 16.07$, $p = 0.0005$; Šídák's multiple comparisons test: Val/Val: $t(24) = 2.545$, $p = 0.0352$; Met/Met: $t(24) = 3.161$, $p = 0.0084$). In keeping with our hypothesis, we demonstrate using amphetamine mediated GID that advanced age is permissive to its induction. Specifically, DA grafted rats of both genotypes with widespread striatal reinnervation developed robust GID compared to sham grafted rats. However, in these middle-aged rats, GID scores were higher in Met/Met compared to Val/Val rats (2-way ANOVA $F(1,27) = 5.510$, $p = 0.0265$; Šídák's test: $t(27) = 2.990$, $p = 0.0118$). Data related to graft-host synaptic connectivity, which is increased with aging in Met68Met but not Val/Val rats, its association with LID, and the co-localization of vesicular

glutamate in grafted DA neurons and its significant association with GID will be presented and discussed. Understanding the role of aging and the common rs6265 SNP on DA terminal replacement therapies will aid in development of safe and optimized approaches for remodeling the parkinsonian striatum.

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Poster

P266. Therapeutic Strategies: Cellular Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P266.09

Topic: C.03. Parkinson's Disease

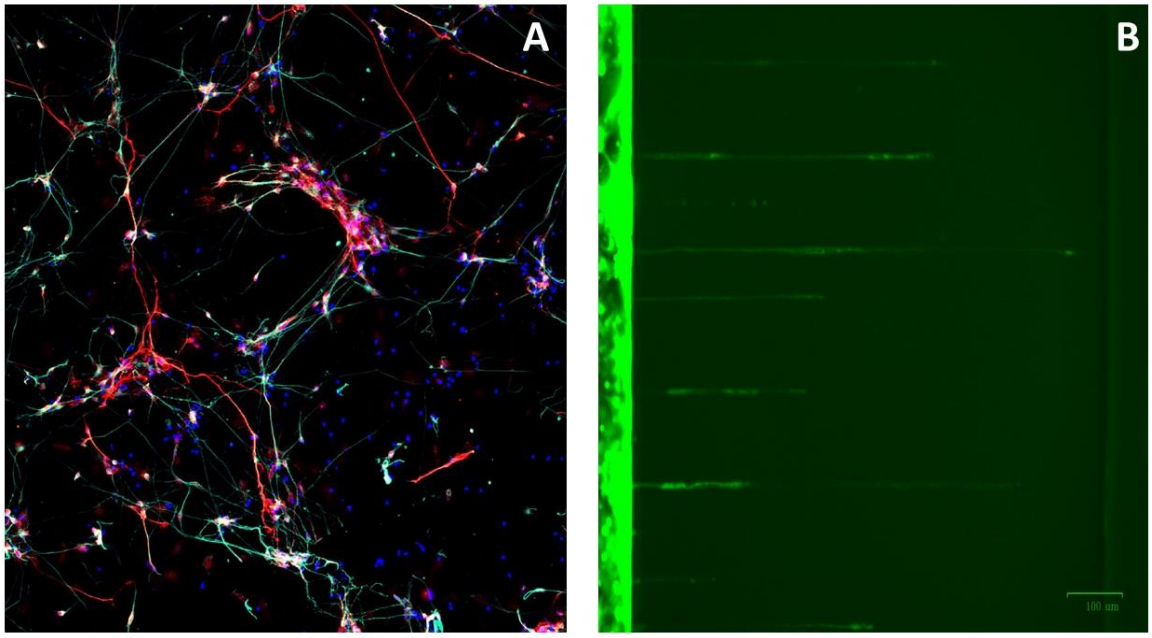
Support: UC MEXUS-CONACYT CN-19-163

Title: Effect of sonic hedgehog on the axonal guidance of human dopaminergic neurons

Authors: *B. GARCIA-LARA[1,2], A. LÓPEZ-ORNELAS[4], E. RAMÓN-GALLEGOS[1], M. URIBE-ROBLES[5], A. A. MARTÍNEZ MORALES[5], E. ORTIZ-ISLAS[3], I. VELASCO[6], E. ESTUDILLO[2];
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Abstract: Effect of sonic hedgehog on the axonal guidance of human dopaminergic neurons Parkinson's disease (PD) is a neurodegenerative disorder characterized by the premature death ofdopaminergic neurons (nDA), which causes motor and non motor symptoms. Currently, the maintreatments are palliative. Cell replacement therapy consists on restoring loss of nDA and isconsidered a promising therapeutic strategy, however, it is necessary for transplanted neurons toextend their axons to their appropriate target of innervation. Chemoattractant molecules act asguidance cues capable of guiding axonal growth for the establishment of accurate connectionswithin the brain. Interestingly, Sonic Hedgehog (Shh) is a molecule capable of attracting nDA axonsduring rodent embryonic development, however, this effect has not been studied in human nDA. Inthis study, we evaluate the effect of Shh on the axonal guidance of nDA derived from humanembryonic stem cells (hESC). We used modified hESC that

constitutively express the GFP protein(H9-GFP), which allows the direct visualization of nDA axons derived from H9-GFP cells. At day 28 ofdifferentiation, we used molecular markers (tyrosine hydroxylase (TH), FOXA2, β Tubulin) tocharacterize nDA by immunofluorescence. Once the presence of nDA markers was confirmed, weseeded the nDA cells in axonal growth chambers, where they were exposed to a Shh gradient. Themicrochannels occupied by axons were counted daily to evaluate the axonal growth. Although wefound the presence of nDA markers, neurons exposed to Shh did not show a defined attractive orrepulsive growth pattern. Therefore, further experiments will be performed to determine the effectof Shh on the axonal growth of nDA.**Figure 1. Experimental models for axonal guidance assays.** A) Inmunofluorescence for the nDAmarker TH (red) and the neuronal marker β Tubulin (cyan) after 28 days of differentiation. B) Axonsexpressing the GFP protein and entering the microchannels of the axonal growth chambers.



Disclosures: B. Garcia-Lara: None. **A. López-Ornelas:** None. **E. Ramón-Gallegos:** None. **M. Uribe-Robles:** None. **A.A. Martínez Morales:** None. **E. Ortiz-Islas:** None. **I. Velasco:** None. **E. Estudillo:** None.

Poster

P266. Therapeutic Strategies: Cellular Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P266.10

Topic: C.03. Parkinson’s Disease

Support: Swedish Research Council
Wallenberg Foundation

Title: Three-dimensional direct reprogramming approach to model human glia conversion

Authors: *J. GIACOMONI, A. BRUZELIUS, M. HABEKOST, P. STORM, A. FIORENZANO, D. OTTOSSON, M. PARMAR;
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Abstract: Parkinson's disease (PD), one of the most common neurodegenerative disorders, is primarily characterized by progressive loss of dopamine (DA) neurons in the ventral midbrain. The relatively focal degeneration in PD makes it a good candidate for cell replacement therapies, and efforts are on their way to use stem cell derived-DA neurons in clinical trials. An emerging alternative approach to cell transplantation is *in vivo* reprogramming, where resident glia is converted into neurons directly within the brain parenchyma. Proof-of concept that *in vivo* conversion can be a viable future option has been provided in rodent studies. However, the inaccessibility of human brain tissue and the lack of an *in vitro* culture system that recapitulates the complexity of the human brain represent major limitations in the development and pre-clinical validation of such approach.We have established a renewable and reproducible stem cell-based system of human glial progenitor cells for direct neural conversion and identified optimal combinations of fate determinants for the generation of functional DA neurons *in vitro*. We developed a 3D culture model from previous 2D protocol allowing cells to interact more efficiently by recapitulating the complexity of a 3D space and evaluated it as a pre-clinical model of human glia conversion. Our approach increases conversion efficiency and accelerates the reprogramming process to generate mature neuronal phenotypes with electrophysiological function within 3 weeks. Furthermore, reprogramming in 3D allows long term cultures, and is applicable for a variety of neuronal conversion protocols. We are currently investigating region-specific molecular identity of glial cells and induced neurons using single-nucleus RNA sequencing. With this we hope to explore neuron lineage diversity after direct cell conversion and to understand the reprogramming competence of different subtypes of glial progenitors.

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Poster

P271. Human Studies: Genetics and Diagnostics PD II

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P271.01

Topic: C.03. Parkinson's Disease

Support: Graham and Carolyn Holloway Foundation

JES Edwards Foundation

Title: Bio-cognitive markers suggest that young veterans with mTBI show premature cognitive decline and may be at-risk for a prodromal onset of Parkinson's disease: A proof-of-concept, matched-control study

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Abstract: Epidemiological reports suggest 56% of veterans with mild traumatic brain injury (mTBI) will eventually develop Parkinson's disease (PD) within ~5 - 7-years post-injury. Cognitive decline is considered a pre-motor prodromal characteristic of PD. However, specific cognitive domains contributing most to an mTBI-related PD onset is unknown. Identifying which cognitive domains will decline first and examining their relationship to neural biomarkers may yield a novel bio-cognitive profile to augment existing hallmark characteristics of prodromal PD that will inform therapeutic strategies. Here, we report the results of a proof-of-concept, match-controlled, cross-sectional study examining age- and IQ-matched veterans with and without mTBI, non-veteran healthy controls, and IQ-matched non-demented early-stage PD. Dependent variables were scores from well-known standardized neuropsychological tests assessing cognitive flexibility, attention, memory, visuospatial ability, and verbal fluency. Serum-based astrocytic biomarkers of central nervous system (CNS) functioning were analyzed in relation to cognitive outcomes. Out of 200 volunteers, 114 provided evaluable data. Groups significantly differed on cognitive tests using multivariate analyses [F (21,299) = 3.09, p<0.0001]. Benjamini and Hochberg False Discovery Rate post-hoc tests showed veterans with mTBI performed significantly worse than matched-control groups on four out of eight cognitive tests (range: p =.009 to .049), and most often performed similarly to decades older subjects with early-stage PD (range: p =.749 to .140). A Spearman correlation showed that out of three astrocytic biomarkers analyzed, glial fibrillary acidic protein (GFAP) significantly and positively correlated with tests of cognitive flexibility and attention (Trail-making A, rs(101) = .29, p = .004; Trail-making B, rs(101) = .28, p = .005). The significant relationship we found between GFAP and Trail-making tests A and B provides preliminary data supporting a bio-cognitive marker of subtle, premature cognitive aging in veterans with mTBI that potentially identifies them as at-risk for continual cognitive decline, the onset of PD, or some other neurodegenerative disease.

Disclosures: V.A. Nejtek: None. **R. James:** None. **M.F. Salvatore:** None. **H. Alphonso:** None. **I. Soto:** None. **G. Boehm:** None.

Poster

P271. Human Studies: Genetics and Diagnostics PD II

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Topic: C.03. Parkinson's Disease

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National Yang Ming Chiao Tung University and the Ministry of Education (Aim for the Top University Plan), Taipei,Taiwan

Title: Identifying structural and functional brain features in PD with and without dementia

Authors: *C.-J. HUANG[1], A.-C. YANG[1,2,3];
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Abstract: Parkinson's disease (PD) is a common neurodegenerative disorder, mainly affecting the motor nervous system. About 35% of patients are combined with dementia in the later stages of the illness, making their brains shrink faster than those without dementia, but no specific structural and functional abnormalities have been found in the brain area that is related to comorbidity of dementia. We hypothesized that brain structural and functional imaging would differ in specific brain regions between PD without dementia (PDnD) and PD with dementia (PDD). Voxel-based morphometry was used to analyze T1 magnetic resonance images. The study cohort included 113 healthy controls and 381 patients with PD, and the latter were further subtyped into 340 cases of PDnD, and 41 cases of PDD. The General Linear Model was used to examine whether there were differences in gray matter volume between groups and after one year of follow-up while controlling for age, sex, education years and Montreal Cognitive Assessment scores, and the voxel cluster was evaluated with family wise error for the correction of multiple comparisons. No study has been conducted to investigate the relationship between PDnD and PDD by combining structural and functional brain images. By identifying structural and functional differences, we expect to identify possible brain regions that are related to progression of dementia at an early stage of the PD and provide early intervention. We found that at baseline, the volume of the caudate nucleus was significantly bigger in PD and PDnD compared to healthy controls, while the volume of cerebellar region was significantly smaller than healthy controls ($P_{FWE} < 0.001$). When comparing PDD to healthy controls, it was found that the volume of insula was significantly smaller than healthy controls ($P_{FWE} < 0.001$). After one year of disease, PD and PDnD showed a significant increase in volume in the cerebellar region compared to baseline volume ($P_{FWE} = 0.001$); the occipital regions showed a significant decrease in volume ($P_{FWE} = 0.015$). While PDD showed no significant increase in volume after one year of disease, but a significant decrease in volume in the insula ($P_{FWE} < 0.001$). Based on structural imaging findings, it was found that the superior temporal gyrus volume was significantly smaller in PDD than in PDnD. Previous studies have shown that localized brain atrophy can also cause dementia. Frontotemporal dementia is a type of dementia caused by

severe temporal lobe atrophy, with symptoms mostly related to verbal expression. Based on structural imaging and previous studies, significant temporal lobe atrophy may be considered a sign of future dementia in PD.

Disclosures: C. Huang: None. **A. Yang:** None.

Poster

P271. Human Studies: Genetics and Diagnostics PD II

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P271.03

Topic: C.03. Parkinson's Disease

Support: The Michael J. Fox Foundation for Parkinson's Research (Grant ID: 13280)

Title: First test of dopamine buffering capacity imaging as a disease severity biomarker in Parkinson disease

Authors: *K. J. BLACK[1], J. M. KOLLER[2], J. S. PERLMUTTER[3];
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Abstract: BACKGROUND: We tested whether a novel pharmacological fMRI approach, dopamine buffering capacity imaging, could quantify disease severity in Parkinson disease (PD). This approach measures how quickly the effect of a dose of levodopa (LD) appears and wears off, using not a clinical effect (which can change e.g. with motivation and fatigue), but LD's objective effect on rCBF (regional cerebral blood flow) in the midbrain and pons. This large rCBF response has been demonstrated repeatedly (e.g. 62-74% increase in DOI: 10.7717/peerj.1381/table-2), but previous studies all averaged over participants and averaged over before- *vs.* after-drug time points. The present study relies on a mathematical model that is well grounded in available information (DOI 10.3389/fneur.2020.00370), but requires measuring for the first time rCBF responses to levodopa over numerous brief time intervals and in individual participants.
METHODS: Details appear in the pre-registered study protocol (DOI 10.17605/OSF.IO/G7DFY). After 200 mg carbidopa by mouth, pCASL fMRI measured rCBF repeatedly for over an hour in 16 people with Parkinson disease, before and during an i.v. infusion of levodopa bioequivalent to about 80 mg by mouth. The effect site rate constant k_e was calculated for a pre-specified midbrain-pons region with a large LD-induced increase in rCBF.
RESULTS: On average, the midbrain region showed a plausible time course of response to LD, with mean rCBF beginning to increase near the end of the i.v. LD loading dose and persisting elevated throughout the 15-40 min. time period during which an increase was expected. However, some participants' time-rCBF curves showed little response, including the participant

who appeared to have the best scan session (minimal head movement, no sleep). The hypothesized correlations of midbrain k_e with disease duration and severity (UPDRS III score and tapping speed in an overnight off state) showed the expected sign but none was significant. DISCUSSION: This novel pharmacological fMRI method to quantify PD disease progression did not perform as expected in this sample using these methodological choices. Specifically, the midbrain region did not give in every subject the expected, pharmacologically sensible time course of increased activity after LD administration. We will discuss possible explanations and potential methodological improvements.

Disclosures: K.J. Black: E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); QuanDyn.com. **J.M. Koller:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); QuanDyn.com. **J.S. Perlmutter:** None.

Poster

P271. Human Studies: Genetics and Diagnostics PD II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P271.04

Topic: C.03. Parkinson's Disease

Support: Bikaintek (Basque Government)
PUE (UPV/EHU)
PTQ (Ministry of Science and Innovation)

Title: Detection of reactive antibodies against mouse cerebral antigens in serum of Parkinson's disease patients

Authors: J. EGIGUREN[1], E. PAREDES-RODRIGUEZ[2], C. DOMÍNGUEZ-FERNÁNDEZ[2,1], R. FERNÁNDEZ[2], E. ASTIGARRAGA[2], C. MIGUELEZ[1], ***G. BARREDA-GÓMEZ**[2]; [1]Pharmacol., Univ. of the Basque Country (UPV/EHU), Leioa, Spain; [2]Res. and Develop., IMG Pharma Biotech, Derio, Spain

Abstract: Detection of reactive antibodies against mouse cerebral antigens in serum of Parkinson's disease patients
June Egiguren, Elena Paredes, Celtia Domínguez, Roberto Fernández, Egoitz Astigarraga, Cristina Miguelez, Gabriel Barreda-Gómez
Parkinson's disease (PD) has become the second most common neurodegenerative disorder, affecting 1% of the individuals older than 60 years. Despite large research efforts, its diagnosis remains a challenge and no ideal biomarkers have been identified to be used in the early phases of the disease. Although the etiology and pathogenesis of PD remains unclear, several evidences

support the autoimmune basis of the disease, highlighting the existence of reactive antibodies against dopaminergic neurons of the *substantia nigra pars compacta*. The complex immunohistochemistry process required to monitor reactive antibodies in tissue sections has limit their potential as PD biomarkers. However, newer techniques as cell membrane microarrays (CMMA) overcome the main limitations regarding reproducibility, handling and quantitation. Thus, the aim of this study was to set up a serological reactive antibody assay using as sensor cell membranes isolated from multiple brain areas of mice and anti-human IgG secondary antibody as fluorescently probe. For this purpose, CMMA were developed using cell membranes isolated from a collection (N = 5) of mouse brain areas (cerebral cortex, frontal cortex, striatum, hippocampus, hypothalamus, thalamus and striatum). A titration was performed incubating the CMMA with increased concentrations of sera from PD patients with autoimmune disease (N = 4) or without (N = 4), together with controls (N = 4) and the reactive anti-neuronal antibodies were quantified in each of the brain areas. The data were calibrated, normalized and standardize before analyzing the panel of selected biomarkers by machine learning algorithms. 63% of the PD patients present a significant change in the reactive antibody profile against the brain regions included in the CMMA. The prevalence in those PD patients suffering also from an autoimmune disease was higher, although further investigation must be performed with a higher cohort of patients, in order to validate the potency of these easily accessible biomarkers. Moreover, the multiplexing capability of the microarray technology provides a novel approach for regular monitoring of patients and, thus, for checking the efficacy of therapies and disease progression, avoiding surgical procedures.
Keywords: Parkinson's disease, microarray, autoantibodies, serum

Disclosures: J. Egiguren: None. **E. Paredes-Rodriguez:** None. **C. Domínguez-Fernández:** None. **R. Fernández:** None. **E. Astigarraga:** None. **C. Miguelez:** None. **G. Barreda-Gómez:** None.

Poster

P271. Human Studies: Genetics and Diagnostics PD II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P271.05

Topic: C.03. Parkinson's Disease

Title: Feasibility of a system for human visual rating of signals from accelerometers on the extremities of people with Parkinson's disease

Authors: *J. R. BRASIC[1], L. ZIEGELMAN[3], K. A. MILLS[2], M. E. HERNANDEZ[4]; [1]Radiology, [2]Neurol., Johns Hopkins Univ., Baltimore, MD; [3]Neurosci., Univ. of Illinois At Urbana-Champaign, Urbana, IL; [4]Kinesiology and Community Hlth., UIUC, Urbana, IL

Abstract: Objective: To assess methodology for trained experts to rate the signals generated by the extremities of people with Parkinson's disease (PD).
Methods: Ten people with PD underwent a low-cost quantitative structured measurement of movements in the extremities with four accelerometers generating a trace of the three location dimensions. Segments of the signals and the fast Fourier and amor continuous wavelet transforms reflecting the structured repetitive movements were expressed as six panels representing the x, y, and z axes for the finger and the wrist (Figure) or the toe and the ankle for each extremity. Raters were instructed as follows:

Interruptions or freezing

1a 1 to 2 interruptions

2a 3 to 5 interruptions

3a 5 or more interruptions or a freeze, a sustained absence of repetitions

4a Worse

Slowing

1b Minimal

2b Mild

3b Moderate

4b Worse

Amplitude reductions

1c End of sequence

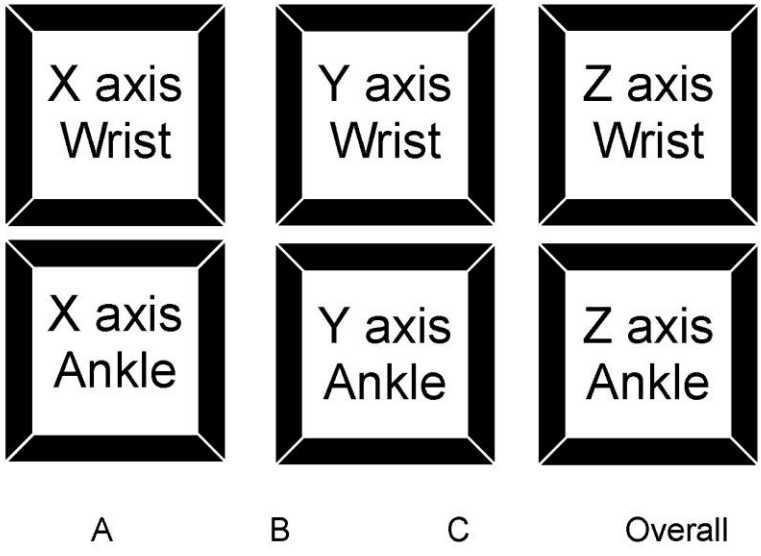
2c Middle of sequence

3c Beginning of sequence

4c Worse

Results: Twenty quizzes representing the output for the (A) signals and fast Fourier transforms and (B) amor continuous wavelet transforms of the left and right assessments of upper and lower extremities were completed and tabulated for 34 trained raters (Ziegelman, *et al.,* 2020).
Discussion:
A methodology for scoring of output of accelerometers on the extremities by an international team of trained raters has generated a valuable dataset (Zieglelman, *et al.,* 2020) for analysis and interpretation. Since this is an inexpensive procedure to quantitatively measure motions in PD and other movement disorders, this will be a valuable resource to colleagues, particularly in underdeveloped regions with limited budgets. The dataset will serve as a template for other investigations to develop novel techniques to facilitate the diagnosis, monitoring, and treatment

of PD, other movement disorders, and other nervous and mental conditions. The procedure will provide the basis to obtain objective quantitative measurements of participants in clinical trials of new agents.
Ziegelman L, et al. 2020. https://doi.org/10.17632/4dp4v7968z.8



Disclosures: J.R. Brasic: None. **L. Ziegelman:** None. **K.A. Mills:** None. **M.E. Hernandez:** None.

Poster

P271. Human Studies: Genetics and Diagnostics PD II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P271.06

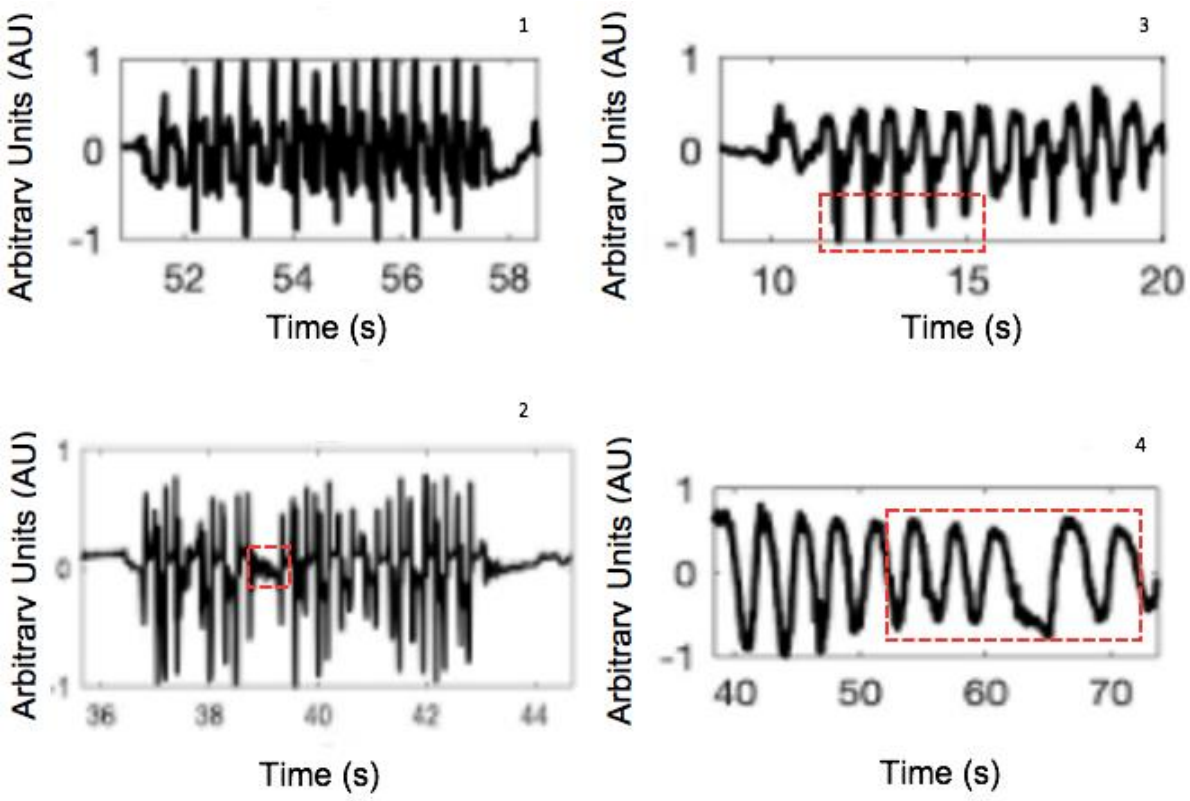
Topic: C.03. Parkinson's Disease

Title: Use of accelerometer signals to improve classification of structured motor assessments of Parkinson's Disease

Authors: *H. HAKIM[1], T. KOSURI[2], L. ZHAO[3], L. ZIEGELMAN[4], K. MILLS[5], A. PANTELYAT[5], J. BANG[5], M. E. HERNANDEZ[6], J. R. BRASIC[7];
[1]Johns Hopkins Univ. Undergraduate Neurosci. Program, Baltimore, MD; [2]Johns Hopkins Univ., Baltimore, MD; [3]Univ. of Illinois, Urbana, IL; [4]Neurosci., Univ. of Illinois At Urbana-Champaign, Urbana, IL; [5]Johns Hopkins Med., Baltimore, MD; [6]Kinesiology and Community Hlth., UIUC, Urbana, IL; [7]The Russell H. Morgan Dept. of Radiology and Radiological Sci., Johns Hopkins Sch. of Med., Baltimore, MD

Abstract: Motivation/Problem Statement: Routine clinical and research assessments of persons with Parkinson's Disease (PD) by visual observation introduces inaccuracies reflecting inherent human limitations, motivating the need to obtain precise measurements with instrumentation. We sought to develop a protocol to utilize accelerometer signals to rate the severity of movements to obtain optimal classifications of people with PD.
Methods: We expressed accelerometry signals of five repetitive movements for ten participants with PD as panels (Figure) for rating by visual inspection. Trained raters were instructed to check a box for an overall rating of
0 normal
1 minimal
2 mild
3 moderate
4 worse
for individual abnormalities
a interruptions
b slowing
c decreased amplitudes
A higher score indicates greater impairments. The representations of the accelerometry signals of a single extremity without identification of laterality were presented as panels to be scored by trained raters. Each panel contained six images corresponding to the x, y, and z axes of the positions of the accelerometers on the finger and wrist for the upper extremity or the toe and ankle for the lower extremity.
Results: Images were constructed to represent signals corresponding to scores of 0, 1, 2, 3, or 4 (Figure). A protocol was developed to rate signals by interruptions, slowing, and amplitude decrements.
(1) Once started, regular rate and rhythm of movement is observed - this waveform indicates no abnormalities in fluency, speed, or amplitude of movement - trace merits a score of 0.

(2) Single interruption (depicted in red outline) merits a rating of 1a.
(3) Amplitude decrement midway through the trace merits a rating of 2c.
(4) Frequency of trace declines toward end of sequence, warranting a rating of 3b.
Discussion: A methodology for scoring of signals of accelerometers on the extremities by an international team of trained raters has generated a valuable dataset (Ziegelman, *et al.*, 2020) for analysis and interpretation. References: Ziegelman L, *et al.*, Mendeley Data. V8. 2020. https://doi.org/10.17632/4dp4v7968z.8



Disclosures: H. Hakim: None. **T. Kosuri:** None. **L. Zhao:** None. **L. Ziegelman:** None. **K. Mills:** None. **A. Pantelyat:** None. **J. Bang:** None. **M.E. Hernandez:** None. **J.R. Brasic:** None.

Poster

P271. Human Studies: Genetics and Diagnostics PD II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

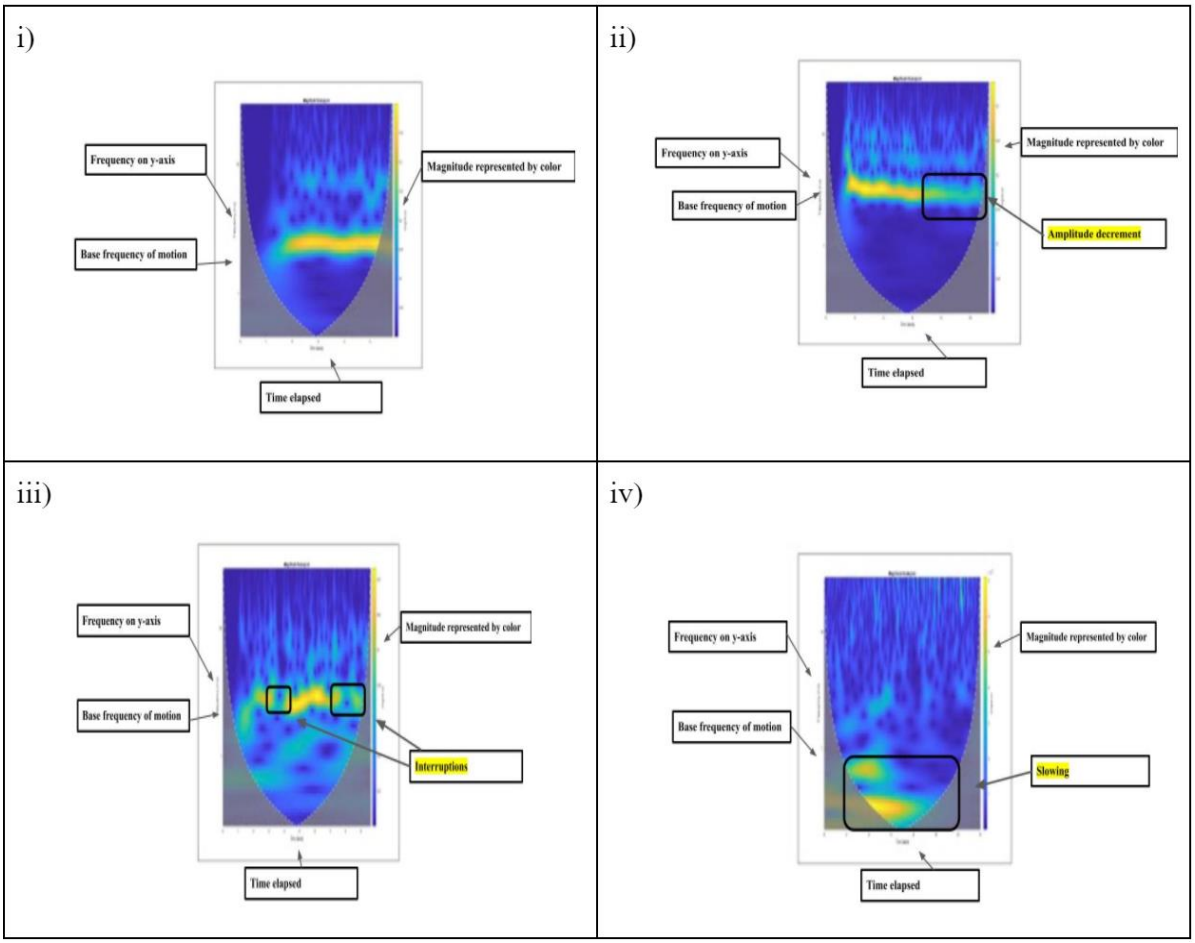
Program #/Poster #: P271.07

Topic: C.03. Parkinson's Disease

Title: Continuous wavelet transforms to improve the accuracy of motor assessments of Parkinson's disease

Authors: *T. KOSURI[1], H. HAKIM[3], L. ZIEGELMAN[4], K. A. MILLS[2], A. PANTELYAT[5], J. BANG[3], M. E. HERNANDEZ[6], J. R. BRASIC[7];
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Abstract: Motivation/Problem Statement: Routine clinical and research assessments of persons with potential Parkinson's Disease (PD) involving the use of visual observation of structured examinations introduces inaccuracies reflecting inherent human limitations, motivating the need to obtain precise measurements with instrumentation. We sought to develop a protocol to utilize amor continuous wavelet transforms (CWTs) to rate the severity of movements to obtain optimal classifications of people with PD.
Methods: We expressed CWTs of five repetitive movements for ten participants with PD as panels (Figure) for rating by visual inspection. Trained raters were instructed to check a box for an overall rating of
0 normal
1 minimal
2 mild
3 moderate
4 worse
For individual abnormalities
a interruptions
b slowing
c decreased amplitudes
The representations of the CWTs of a single extremity without identification of laterality were presented as panels to be scored by trained raters. Each panel contained six images corresponding to the x, y, and z axes of the positions of the accelerometers on the finger and wrist or the toe and ankle.
Results: Raters classified transforms by scoring three particular abnormalities in the CWTs. Interruptions were identified on the CWTs as breaks in continuous horizontal segments of line; slowing was demonstrated by a negative sloping line; amplitude decrements were evident while examining the warmth of color (yellow representing the greatest amplitude). Therefore, lines of diminishing warmth from left to right were deemed to show decreasing amplitude.
i) 0 (no amplitude decrements, slowing, or interruptions seen)
ii) 1 (amplitude decrement seen by decrease in warmth of color)
iii) 2 (3-5 interruptions seen by halts in CWT)
iv) 3 (moderate slowing seen by negative sloping line)
Discussion: A methodology for scoring CWTs of accelerometers on the extremities by trained raters has generated a valuable dataset (Zieglelman, *et al.*, 2020) for analysis and interpretation.
References: Ziegelman L, *et al*., Mendeley Data. V8. 2020.
https://doi.org/10.17632/4dp4v7968z.8



Disclosures: T. Kosuri: None. **H. Hakim:** None. **L. Ziegelman:** None. **K.A. Mills:** None. **A. Pantelyat:** None. **J. Bang:** None. **M.E. Hernandez:** None. **J.R. Brasic:** None.

Poster

P271. Human Studies: Genetics and Diagnostics PD II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P271.08

Topic: C.03. Parkinson's Disease

Title: Characterization of motor impairment rating error of transformed IMU data

Authors: *L. ZIEGELMAN[1], T. HARRIGAN[2], J. R. BRASIC[3], M. E. HERNANDEZ[4]; [1]Univ. of Illinois At Urbana-Champaign, Urbana, IL; [2]REDD, Johns Hopkins Univ. Applied Physics Laborator, Laurel, MD; [3]The Russell H. Morgan Dept. of Radiology and Radiological Sci., Johns Hopkins Sch. of Med., Baltimore, MD; [4]Kinesiology and Community Hlth., UIUC, Urbana, IL

Abstract: This study aimed to characterize the relationship between level of movement impairment and rater accuracy of the movement impairment during a clinical task. These terms are operationalized using the Unified Parkinson's Disease Rating Scale (UPDRS) motor subscale to characterize level of impairment and the absolute value of the percent error of the rater's rating of transforms of inertial measurement unit (IMU) data when compared to a clinical gold standard to represent rater accuracy. The rationale for characterizing this relationship is that it allows insight into ways in which transformations of motion data could be compared to a traditional method of diagnosis of movement impairment by a clinician. Additionally, characterizing typical rating error allows for additional necessary information in the creation of machine learning classifiers of UPDRS motor task ratings, which is a meaningful step in the process of creating telemedicine tools for movement disorder specialists. This is done by creating a parent regression model, where the performance of a linear model, cubic model, and exponential decay model are compared, as well as testing the selected exponential decay model for a task effect. The clinical ratings were scored by 35 raters certified to administer the UPDRS on a series of 24 participants diagnosed with Parkinson's Disease (PD) with transformed motion data. The exponential model, which was fit as $Y=-0.00291e^{1.13124}+0.44694$ is shown to have the best fit with all three estimates statistically significant at \alpha=0.05 and finds that UPDRS scores between 0 and 2 are harder to accurately rate as compared to a 3 or 4. This means that mild symptoms of motor impairment are more difficult to separate from no impairment as compared to severe symptoms. This parent exponential decay model is tested for a task effect, which is found to be present. All together, these models indicate that transformed data is not equally easy to evaluate, where the movement impairment and task type impact a rater's ability to rate motion accurately. This is a necessary first step in the evaluation of the feasibility of using IMU data ratings to build telemedicine tools.

Disclosures: L. Ziegelman: None. **T. Harrigan:** None. **J.R. Brasic:** None. **M.E. Hernandez:** None.

Poster

P311. Neuroprotective Mechanisms: Preclinical: Other Therapeutics III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P311.01

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Title: Protective effects of NAMPT and MAPK inhibitors on Wallerian degeneration of mammalian axons: mechanisms and synergies

Authors: *A. ALEXANDRIS[1], J. RYU[1], R. HARLAN[2], L. RAJBHANDARI[3], S. AJA[2], D. GRAHAM[2], A. VENKATESAN[3], V. KOLIATSOS[1,3,4];
[1]Pathology, Johns Hopkins Univ., Baltimore, MD; [2]The Mol. Determinants Ctr. and Core, Johns

Hopkins Univ., Saint Petersburg, FL; [3]Neurol., [4]Psychiatry and Behavioral Sci., Johns Hopkins Univ., Baltimore, MD

Abstract: Wallerian degeneration (WD) is a highly conserved axon self-destruction program that has been implicated in traumatic axonal injury and other neurological disorders. Several key players have been identified as drivers of WD that are centered on the regulation of the NAD+ metabolome. These include the degradation of nicotinamide mononucleotide adenylyltransferase (NMNAT2), the built up of its substrate NMN, and the activation of the NAD+ degrading Sterile α and TIR motif containing 1 (SARM1) protein, heralding axon fragmentation hours or days after injury. Yet, the time course of these events, their regulation, and their therapeutic targetability are still unclear. Here we utilized an in vitro mouse dorsal root ganglion axotomy model in order to explore pharmacological strategies that modulate NMN and NAD+ metabolism and might protect against WD, namely the inhibition of the NMN-synthesizing enzyme NAMPT and the NMNAT2-degrading MAPK pathway, as well as augmentation of the nicotinic acid riboside salvage pathway. Transected axons were assessed by a complement of techniques including degeneration index analysis, and targeted metabolic and western blot profiling of injury-related signals. We found that the decision point of axon fragmentation occurs within a time window corresponding to 2-4 hours post-injury and is related to reciprocal changes in NMN and NAD+ levels driven in turn by the MAPK-dependent degradation of NMNAT2. NAMPT inhibition was sufficient to partially suppress SARM1 activity, protect axonal NAD+ levels and delay WD in a time-dependent manner whereas its supplementation with nicotinic acid riboside led to further suppression of SARM1 activity and complete repression of axon fragmentation. Metabolite analyses revealed complex and dynamic effects, which however indicated the ratio of NMN/NAD+ as the main driver of SARM1 activity. The existence of a time window of commitment and the successful pharmacological modulation of NMN/NAD+ metabolism, may have translational significance for a wide spectrum of axonopathies.

Disclosures: A. Alexandris: None. **J. Ryu:** None. **R. Harlan:** None. **L. Rajbhandari:** None. **S. Aja:** None. **D. Graham:** None. **A. Venkatesan:** None. **V. Koliatsos:** None.

Poster

P311. Neuroprotective Mechanisms: Preclinical: Other Therapeutics III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P311.02

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Training Grant 5T32NS096050-24

Title: The potential of astrocyte-derived extracellular vesicles to modulate microglial activation

Authors: *S. L. REED[1], L. SHAPIRO[1], J. C. WONG[1], R. MA[2], G. HU[2], A. ESCAYG[1]; [1]Dept. of Human Genet., Emory Univ., Atlanta, GA; [2]Dept. of Pharmacol. & Exptl. Neurosci., Univ. of Nebraska Med. Ctr., Omaha, NE

Abstract: Extracellular vesicles (EVs) are small vesicles released by cells into the extracellular space. EVs play a key role in intercellular communication and are believed to be responsible for mediating at least some of the regenerative benefits of stem-cell based therapies. In fact, the administration of EVs has been shown to elicit neuroprotective and anti-inflammatory effects and reduce neuropathological, cognitive, and behavioral phenotypes in some disease models. Most studies on the therapeutic potential of EVs have utilized stem-cell derived EVs, but there is evidence that EVs from some terminally differentiated cell types, such as astrocyte-derived EVs (astro-EVs), may also have beneficial effects. The contents of astro-EVs include molecules with anti-inflammatory and neuroprotective properties, and astro-EVs have been shown to be efficacious in animal models of stroke and traumatic brain injury. However, the ability of astro-EVs to reduce features common to many neurological disorders, such as gliosis, is not well characterized. Microglia are the resident immune cells of the nervous system. While microglial activation can initiate reparative processes, it is also associated with pathological increases in neuroinflammation and neuronal death. In animal models of neurological injury, such as stroke or status epilepticus (SE), reducing microglial activation can improve recovery and reduce long-term deficits. In this study, we investigated the ability of astro-EVs to modulate the response of microglia in a mouse model of neurological injury. Mouse astrocytic EVs were isolated from conditioned media using ultracentrifugation. Isolated astro-EVs or vehicle were administered to mice following SE. Mice were sacrificed 4-5 days following SE and hippocampi were collected for gene expression analysis. SE was associated with robust increases in the expression of inflammatory genes including pro-inflammatory cytokines and microglial markers. Treatment with astro-EVs significantly attenuated increases in the microglial and macrophage markers CD11b and CD68 in the hippocampus suggesting that astro-EVs can modulate the microglial response to SE. Research is ongoing to determine whether astro-EVs can provide additional therapeutic benefits in this model.

Disclosures: S.L. Reed: None. **L. Shapiro:** None. **J.C. Wong:** None. **R. Ma:** None. **G. Hu:** None. **A. Escayg:** None.

Poster

P311. Neuroprotective Mechanisms: Preclinical: Other Therapeutics III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P311.03

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: This work was supported by the Adelman Multiple Sclerosis Fund

Title: Effects of genetically engineered therapeutic biopolymers on S100B protein mediated demyelination in an organotypic cerebellar slice culture model of multiple sclerosis.

Authors: *P. VIG[1], M. LOPEZ[2], L. KENNEDY[2], J. RYU[2], D. RAUCHER[2], M. WILLIS[2]; [1]Univ. Mississippi Med. Ctr., Jackson, MS; [2]Univ. of Mississippi Med. Ctr., Jackson, MS

Abstract: The demyelinating lesions in multiple sclerosis (MS) are often associated with an increased release of calcium binding protein S100B from reactive astrocytes. The excessive amounts of this pro-inflammatory protein impairs oligodendrogenesis, resulting in reduced myelination. It has been demonstrated that administration of S100B protein can induce inflammatory demyelinating disease in animals. Therefore, this study was initiated to assess if blocking S100B with our genetically engineered therapeutic peptide (TP) could suppress demyelination and neurodegeneration in an experimentally induced demyelination ex vivo model. Our TPs were developed with three key elements: An elastin-like polypeptides (ELP) carrier, S100B inhibitory peptide TRTK as TP and Synb1, to penetrate extracellular matrix. The cerebellar slice cultures were prepared from seven to eleven-day-old enhanced green fluorescent protein (GFP) expressing transgenic mice, transgene under the control of Purkinje cell specific-2 promotor. After one week in culture, slices were treated with various concentrations of demyelinating agent lysolecithin (LL) overnight to optimize its neurotoxicity profile as well as demyelination potential. The slices were treated with various concentrations of control Synb1-ELP peptide or Synb1-ELP-TRTK in the presence or absence of optimized concentration of LL overnight followed by incubations with either control peptide or TP in LL-free medium for additional 72 to 96 hours and processed for immunohistochemical and image analysis. We observed reduced dendritic arborization and numerous dendritic swellings of the Purkinje cells (PCs) in LL treated slices. In addition, axonal myelination was markedly reduced in a concentration dependent manner. The control Synb1-ELP peptide did not protect PCs from the effects of LL; however, Synb1-ELP-TRTK treatment showed remarkable suppression. Even the white matter regions were endogenous GFP fluorescence was low showed enhanced levels of immunofluorescence for myelin basic protein immunostaining. These preliminary data demonstrate that our TP could be checked as a candidate to disrupt the S100B-RAGE axis and may further be tested for potential in treating MS lesions.

Disclosures: P. Vig: None. **M. Lopez:** None. **L. Kennedy:** None. **J. Ryu:** None. **D. Raucher:** None. **M. Willis:** None.

Poster

P311. Neuroprotective Mechanisms: Preclinical: Other Therapeutics III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P311.04

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: Adelson Medical Research Foundation
MTEC (LB: J.L. Goldberg, Stanford Univ., PI
NIH IDDRC imaging core (HD018655)
Viral Vector Core (P30EY012196)

Title: Identification of a putative receptor for the neutrophil-derived growth factor oncomodulin and its role in CNS regeneration

Authors: *L. XIE[1], S. L. PETERSON[2], Y. YIN[3], L. I. BENOWITZ[4];
[1]Boston Children's Hospital, Harvard Med. Sch., Boston, MA; [2]Boston Children's Hosp., Boston, MA; [3]Harvard Med. Sch., Boston, MA; [4]Labs Neurosci Res. Neurosurg, Boston Children's Hosp/Harvard Med. Sc, Boston, MA

Abstract: Identification of a putative receptor for the neutrophil-derived growth factor oncomodulin and its role in CNS regenerationLili Xie[1,2], Sheri Peterson[1,2], Yuqin Yin[1,2], Larry Benowitz[1,21]Department of Neurosurgery, Boston Children's Hospital and Harvard Medical School, Boston, MA 02115; [2]F.M. Kirby Neurobiology Center, Boston Children's Hospital, Boston, MA 02115;
AbstractAlthough the optic nerve, like other pathways in the mature mammalian CNS, can not regenerate when injured. This failure can be partially reversed by inducing sterile inflammation. Previous work from our lab showed this phenomenon is mediated in large part by oncomodulin (Ocm), a 12 kDa Ca++-binding protein, secreted by myeloid cells. Ocm also induces outgrowth from dorsal ganglion neurons (DRGs) and may play a role in the conditioning lesion effect, whereby injuring to periphery nerve enhances the ability of DRGs to extend their central axons in the spinal cord. We now report the identification of a potential Ocm receptor (Ocm-R). We identified Ocm-R using biotinylation by antibody recognition (BAR) followed by mass spectrum. Co-immunoprecipitation studies reveal the direct interaction between Ocm and Ocm-R. Knock-down of Ocm-R using CRISPR-Cas9 leads to loss of Ocm binding to retinal ganglion cells (RGCs) and a substantial loss of inflammation-induced regeneration after optic nerve injury without affecting regeneration induced by other means (*pten* delection in RGCs). Conversely, ectopic expression of Ocm-R in cells with low baseline expression results in high Ocm binding. Ongoing studies are investigating the role of Ocm-R in the condition lesion effect and downstream mechanisms by which Ocm binds to Ocm-R contributes to optic nerve regeneration.
Support: Adelson Medical Research Foundation (LB), NIH IDDRC imaging core (HD018655) and Viral Vector Core (P30EY012196) of Boston Children's Hospital, the Neurobiology Department and Neurobiology Imaging Facility of Harvard Medical School for consultation and instrument use for some of this work (NINDS P30 Core Center grant #NS072030), MTEC (LB: J.L. Goldberg, Stanford Univ., PI)
Competing interests The authors declare that they have no competing interests.

Disclosures: L. Xie: None. **S.L. Peterson:** None. **Y. Yin:** None. **L.I. Benowitz:** None.

Poster

P311. Neuroprotective Mechanisms: Preclinical: Other Therapeutics III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P311.05

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: JP20K09820
JP20K18404
JP21K09688
JP19K09943
JP20K07751
JP19KK0229
JP21H02819

Title: AAV-mediated delivery of constitutively active TrkB promotes CNS axon regeneration and neuroprotection

Authors: *A. KIMURA, E. NISHIJIMA, Y. KITAMURA, S. HONDA, X. GUO, C. HARADA, K. NAMEKATA, T. HARADA;
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Abstract: Severed axons in the CNS do not usually regenerate. Therefore, reversing the damage to CNS axons (for example, spinal cord injury) is currently a major clinical challenge. Recently, studies indicated that AAV delivery of certain genes to neurons could bring out neurons' intrinsic ability to regenerate, providing hope that functional restoration of injured CNS may be possible in the near future. The aim of this study was to develop a gene therapy vector that is effective in promoting CNS axon regeneration and neuroprotection. The roles of brain-derived neurotrophic factor (BDNF)- tropomyosin receptor kinase B (TrkB) signaling in neurite outgrowth and neuroprotection are known; however, whether or not this signaling pathway also plays a role in regeneration of injured CNS axons is unknown. Therefore, we focused on TrkB and generated constitutively active TrkB. For this, we extracted the intracellular domain of TrkB (iTrkB) and attached a farnesyl group (F-iTrkB) for promoting membrane association. Transfection of F-iTrkB to Cos7 cells demonstrated increased levels of pERK and pAkt, which are downstream targets of TrkB signaling. In order to determine the effects of F-iTrkB on axon regeneration and neuroprotection *in vivo*, we used the mouse eye and optic nerve as experimental models. We used AAV serotype 2 (AAV) to deliver F-iTrkB into retinal ganglion cells (RGCs) and examined its effects in an optic nerve crush (ONC) injury model. Intravitreal injection of AAV-F-iTrkB reduced RGC death at 4 weeks after ONC compared with AAV-control, demonstrating its neuroprotective effects on RGCs. Furthermore, we traced RGC axons with intravitreal injection of cholera toxin subunit B and we found that AAV-F-iTrkB treatment promoted robust RGC axon regeneration at 2 and 4 weeks after ONC. We also used an optic tract transection model, in which RGC axons are cut near their projection site, the superior colliculus. We found that RGC death was not observed in this model, indicating that this model allows examination of the effects on axon regeneration without the reduced cell number caused by the

axonal damage. Intravitreal injection of AAV-F-iTrkB promoted robust RGC axon regeneration beyond the transection site in the brain. Excitingly, optokinetic responses were partially recovered in some mice, indicating that AAV-F-iTrkB treatment promoted partial recovery of visual behavior. Taken together, our findings demonstrate that AAV-F-iTrkB promotes CNS axon regeneration and neuroprotection, suggesting that F-iTrkB may be useful for treatment of axonal injuries in the CNS and neurodegenerative diseases.

Disclosures: A. Kimura: None. **E. Nishijima:** None. **Y. Kitamura:** None. **S. Honda:** None. **X. Guo:** None. **C. Harada:** None. **K. Namekata:** None. **T. Harada:** None.

Poster

P311. Neuroprotective Mechanisms: Preclinical: Other Therapeutics III

Location: Virtual Only

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Program #/Poster #: P311.06

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

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RPB unrestricted Departmental grant
P30 EY001792 Core Grant for Vision Research and Eversight to Victor Guaiquil

Title: Role of Netrin-4 in neuronal growth and cornea wound healing

Authors: *C. XIAO, D. LARA, Q. ZHOU, G. DIMAILIG, M. ROSENBLATT, V. H. GUAIQUIL;
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Abstract: The cornea is the highest innervated tissue in the human body, receiving axonal projections from the trigeminal ganglia (TG). The cornea acts as a protective barrier against the environment, and its transparency is essential for maintaining normal visual acuity. Injury of the corneal epithelium, its more anterior layer, is clinically relevant, as loss of neuronal innervation may result in the disruption of several modalities of sensation and alterations in vision. Several growth factors, lipids, and microRNAs have been described to play a role in cornea wound healing. Axon guidance proteins can also induce nerve regeneration in the adult cornea. Here, we study the role of Netrin-4 in neuroepithelium regeneration using in vitro and in vivo models. Netrins are a family of secreted proteins that serve as guidance signals during development. Netrin-4 acts through its receptors *deleted in colorectal cancer* (DCC), *uncoordinated family members* (UNC5H1-4), and *neogenin*. Netrin-4 has been shown to prevent inflammation in diabetic retinopathy and promote retinal angiogenesis, but less is known of its role in the cornea. This study aims to determine the expression and localization of Netrin-4 and its receptors, the direct functional effects of Netrin-4 on neuron and epithelium regeneration, and its potential preclinical effects in mice.

We found endogenous expression of Netrin-4, DCC, UNC5H1, and neogenin in the corneal epithelium (mouse and human) and trigeminal ganglia through immunofluorescence staining. After corneal epithelium injury in mice (n=3), the expression of Netrin-4 and its receptors quickly recovered and initially localized to the basal layers of the epithelium. The level of expression increased as the epithelium healed. Our observations were quantitatively corroborated by qRT-PCR on injured mice corneas (n=6). Additionally, we observed neuronal growth induction in isolated TG neurons exposed to recombinant Netrin-4. Our dose-response data shows that 50 ng/ml induced the most potent neuronal growth. We are currently evaluating the direct effects of Netrin-4 on human corneal limbal epithelial cells using scratch assays. Moreover, we initiated studies to determine if the observed neuronal growth induced by Netrin-4 can be seen in vivo. For this, we are using a corneal epithelium debridement model which damages the subbasal nerve plexus in mice that then receive vehicle or recombinant Netrin-4 treatment. Our preliminary data demonstrate that Netrin-4 could play a significant role in cornea wound healing and may be of importance for PNS regeneration.

Disclosures: C. Xiao: None. **D. Lara:** None. **Q. Zhou:** None. **G. Dimailig:** None. **M. Rosenblatt:** None. **V.H. Guaiquil:** None.

Poster

P311. Neuroprotective Mechanisms: Preclinical: Other Therapeutics III

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Program #/Poster #: P311.07

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: R44EY031642
R01EY025717
R44EY025905

Title: Optical Gene Delivery for Neuroprotection

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Abstract: Millions of people in the US suffer from vision loss and blindness due to degeneration of retinal cells (e.g. Glaucoma, diabetic retinopathy: DR, Age-related macular degeneration: AMD). The currently available pharmacological treatments based on anti-VEGF (for exudative AMD), steroid therapies (e.g. Dexamethasone for anti-angiogenesis; and triamcinolone for DR), as well as ongoing clinical trials on complement factors (for dry-AMD) and neurotrophic factors (e.g., Ciliary neurotrophic factor for AMD) for inhibiting neurodegenerations, have significant

limitations with respect to the requirement of multiple injections and related adverse-effects including systemic reactions to medications, and eye infections. We present an optical coherence tomography (OCT)-guided near-infrared laser-based non-viral delivery method for spatially localized delivery of therapeutic genes with two distinct approaches. 1) Ultrafast pulsed laser delivery: optoporation and 2) Low power continuous wave laser in combination to functionalized gold nanorods (fGNRs): Nano-enhanced optical delivery. We used the optical delivery method to deliver engineered the pigment epithelium-derived factor (PEDF) gene (a serpin superfamily neurotrophic factor) to targeted retinal cells for efficient and enhanced long-lasting expression. Our results demonstrated in-vivo OCT-guided laser-based delivery of PEDF genes leads to robust expression in the RGC layer in mice. The efficacy of the neuroprotection was assessed by ERG and VEP, which showed that PEDF expression preserves the electrophysiology in the treated eye as compared to the control eye. Also, the number of RGCs in the acute mice model was found to be high in the optically delivered PEDF treated eyes. We also evaluated the anti-angiogenic effect of PEDF in the choroidal neovascularization mouse model and found that PEDF expression in the targeted retinal region (delivered by OCT guided laser) lead to a significant reduction of CNV as compared to non-treated groups. In addition to PEDF gene delivery and neuroprotection, we also evaluated the successful delivery of other genes such as ceMCO or large ABCA4 genes using Optical delivery platform for the treatment of eye indications such as Retinitis Pigmentosa, Dry-AMD, Stargardt disease. Large animals (NHPs, Pigs) safety, toxicity, and efficacy study associated with optical gene delivery are being conducted. The development of a safe, effective, long-lasting, single-dose, delivery and therapy for the treatment of retinal diseases would be transformative in clinical ophthalmology and would improve the lives of millions of people worldwide.

Disclosures: S. Batabyal: A. Employment/Salary (full or part-time):; Nanoscope Technologies LLC. **S. Kim:** A. Employment/Salary (full or part-time):; Nanoscope Technologies LLC. **A. Dibas:** A. Employment/Salary (full or part-time):; Nanoscope Technologies LLC. **B. Mahato:** A. Employment/Salary (full or part-time):; Nanoscope Technologies LLC. **D. Narcisse:** A. Employment/Salary (full or part-time):; Nanoscope Technologies LLC. **H. Al-Saad:** A. Employment/Salary (full or part-time):; Nanoscope Technologies LLC. **M. Carlson:** A. Employment/Salary (full or part-time):; Nanoscope Technologies LLC. **S. Mohanty:** A. Employment/Salary (full or part-time):; Nanoscope Technologies LLC. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Nanoscope Technologies LLC.

Poster

P311. Neuroprotective Mechanisms: Preclinical: Other Therapeutics III

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Program #/Poster #: P311.08

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant R01NS104117
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Brazilian CAPES (88881.311939/2018-01)

Title: Neuroprotection by PAF-receptor antagonist plus a lipid mediator against experimental focal cerebral ischemia

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Abstract: Neuroinflammation and oxidative stress play a key role in the pathophysiology of ischemic stroke. We tested the hypothesis that blocking pro-inflammatory platelet-activating factor receptor (PAF-R) plus administering a selected docosanoid after middle cerebral artery occlusion (MCAo) would lead to neurological recovery. Two different types of small bioactive molecules were investigated: LAU-0901 (LAU), PAF-R antagonist that blocks pro-inflammatory signaling, and aspirin-triggered neuroprotectin D1 (AT-NPD1), which activates cell-survival pathways, and their combination that exerts potent anti-inflammatory activity in the brain. This study investigated the dose-response and therapeutic window in a model of focal cerebral ischemia. In each series described below, male Sprague-Dawley rats were subjected to 2h of MCAo. The behavior was evaluated on days 1, 2, 3, and 7, and an *ex vivo* MRI of the brains was conducted using 11.7 T MRI on day 7. In series 1 (Dose-response), rats were treated with LAU (45 and 60 mg/kg, IP, at 3h), AT-NPD1 (111, 222, and 333 µg/kg, IV, at 3.15 h after onset of stroke), LAU+AT-NPD1 (LAU at 3h and AT-NPD1 at 3.15 h) or vehicle (n=5-6 per group). In series 2 (Therapeutic window), vehicle, LAU (60 mg/kg, IP), AT-NPD1 (222 µg/kg, IV), LAU+AT-NPD1 were administered at 3, 4, 5, and 6 h after onset of MCAo. Physiological variables showed no significant differences among groups. Treatments with LAU-0901 (45 and 60 mg/kg) alone improved behavior on days1-7 by 34-36%; AT-NPD1 (222 and 333 µg/kg) by 23-29% compared to vehicle groups. AT-NPD1 in a low dose (111 µg/kg) was not different from the vehicle group. The neuroprotective effect was enhanced using the LAU-0901+AT-NPD1, which resulted in improved behavior up to 32-38% on days 1-7. Total lesion volumes, which were computed using T2WI, were significantly reduced with LAU-0901 (45 and 60 mg/kg) by 73-90% and AT-NPD1 (222 and 333 µg/kg) alone by 83-85% compared to the vehicle group. Ischemic core, penumbra, and total lesion volumes were dramatically reduced with LAU-0901+AT-NPD1 by 94, 92, and 93% compared to the vehicle. Treatment with LAU and AT-NPD1 alone, when administered at 3h, improved behavior on day 7 by 35 and 28%. In contrast, LAU-0901+AT-NPD1 improved behavior by 46% when administered at 3h, by 52% at 4h, 34% at 5h, and 19% at 6h. In conclusion, these data suggest that LAU-0901 and AT-NPD1 alone in moderate doses provide high-grade neuroprotection in the MCAo model. Combination therapy with LAU-0901 and AT-NPD1 is more effective than the single therapy, affording synergistic neuroprotection with improved neurological recovery when administered up to 6 h after focal cerebral ischemia in rats.

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Poster

P311. Neuroprotective Mechanisms: Preclinical: Other Therapeutics III

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Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH-R01NS091238
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Title: Evaluation of a systemically delivered PACAP glycopeptide as a neuroprotective agent in 2 rodent models of Parkinson's Disease

Authors: K. BERNARD[1], A. LUJAN[2], H. MORRISON[3], M. J. BARTLETT[4], C. LIU[5], G. MOLNAR[6], C. APOSTOL[5], L. SZABÒ[5], S. SHERMAN[4], L. MADHAVAN[4], J. STREICHER[6], M. HEIEN[5], R. POLT[5], T. FALK[7];
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Abstract: Parkinson’s Disease (PD) is the second most common neurodegenerative disease, yet lacks any disease modifying therapeutics to stop or reverse the ongoing pathology occurring in the brain. Pituitary adenylate cyclase-activating peptide (PACAP) is an endogenous neuropeptide hormone that has been extensively studied as a potential protective therapeutic in a variety of neurodegenerative settings due to its potential for initiating cellular processes that promote survival and inhibit potentially harmful processes such as inflammation. This multi-faceted response to PACAP is due to its interactions with PAC1, as well as VPAC1 and VPAC2 receptors found in both the CNS and periphery. However, despite PACAP’s trophic and anti-inflammatory properties, it has not realized its translational potential due to poor stability and limited blood-brain barrier (BBB) permeability. We have designed and synthesized a glycosylated PACAP analog (2ls98lac) that is able to stimulate cAMP production *in vitro* using individual CHO cell lines expressing PACAP receptors. Glycosylation of a peptide plays two roles in increasing the ability of a peptide to reach the CNS; increasing peptide stability and therefore half-life, and enhancing BBB penetration**.** To test the protective ability of the 2ls98lac, we conducted a study using a progressive unilateral 6-hydroxydopamine (6-OHDA) rat PD model (n=16/group). In the treatment group rats were injected (*i.p.*) with 15 mg/kg of 2ls98lac 6-hrs prior, 48-hrs and 72-hrs post 6-OHDA injection. Analysis of a subset of animals at 4 weeks post-6-OHDA lesion PACAP treated animals exhibited reduced ipsilateral turning behavior in

response to the dopamine agonist amphetamine, consistent with reductions in overall lesion size. Unbiased stereology of dopaminergic neurons in a subset of animals (n=8) revealed an increased number on the lesioned side of 2ls98lac treated animals (10159 in treated, 8020 in vehicle, p=0.0277). Microglial analysis revealed a full restoration of the activated microglia phenotype, that was reduced in the vehicle group, characterized by process length, number of branches per cell and the % area/frame ($p<0.01$ each). Additional motor behavior scoring and stereological analysis is ongoing.We have also begun a pilot in the Thy1-aSyn transgenic mouse model of PD. These mice display early progressive motor deficits and early cognitive impairments. We treated mice every other day for 1 month starting at 2 months of age with 2ls98lac (1 mg/kg, s.c.) or vehicle, and performed motor and cognitive assessments. Brains were harvested for immunohistological analysis of alpha synuclein spread. Data analyses are currently ongoing.

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Poster

P311. Neuroprotective Mechanisms: Preclinical: Other Therapeutics III

Location: Virtual Only

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Program #/Poster #: P311.10

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: 1R21NS118223-01A1

Title: Cns protection by repetitive conditioning: proteomic analyses of induced retinal phenotypes

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Abstract: CNS Protection by Repetitive Conditioning: Proteomic Analyses of Induced Retinal Phenotypes

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Previous preclinical research from our lab has demonstrated that repetitive hypoxic conditioning (RHC) induces long-lasting neuroprotection in the retina (Zhu Y et al., IOVS 2007; Zhu Y et al., Mol Med 2012) and brain (Stowe et al., Ann Neurol 2011). Herein, we performed quantitative proteomics to elucidate the underlying molecular phenotypes. At 12 wks of age, male and female Swiss-Webster ND4 outbred mice were exposed to 2 wks of RHC; a matched normoxia cohort served as controls. Three days after RHC, retinae were harvested and processed for MS3 analysis on a Fusion Orbitrap Mass Spectrometer using TMTpro labels for quantification, and 1.5 fold-change and <1% FDR thresholds. Subsequent bioinformatic analysis was performed on these differentially expressed proteins using Ingenuity Pathway Analysis (IPA) software. MS3 identified a total of 7,305 proteins, of which 6,253 were quantified. 160 proteins exhibited significant abundance differences in RHC-treated mice, with 31 upregulated and 129 downregulated relative to normoxic controls. 10 of these proteins were differentially expressed in both sexes. The retinal proteome of females was significantly more responsive to RHC than that of males. IPA revealed broad changes in cellular pathways and molecular functions involved in inflammatory responses, including handling of reactive oxygen species. Diverse metabolic functions were also up- and down-regulated, including phase 1 metabolism, protein metabolism, and lipid transport. These epigenetically-mediated changes in the retinal proteome, at both the single protein and pathway/network level, reveal the potential mechanistic underpinnings of neuroprotection, and in turn serve as potential therapeutic targets. Moreover, the unique sex-dependent changes we observed likely contribute to the sexually dimorphic degree of resilience to ischemic injury in RHC-treated mice (Harman et al., IOVS 2020).

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Poster

P321. Neuroinflammation: Oligodendrocytes, Pericytes, and Other Cells

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Program #/Poster #: P321.01

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

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Title: Dolutegravir inhibition of matrix metalloproteinases affects mouse neurodevelopment

Authors: *A. N. BADE[1], J. M. MCMILLAN[1], Y. LIU[2], B. J. EDAGWA[1], H. E. GENDELMAN[1];
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Abstract: Dolutegravir (DTG) is a first-line antiretroviral drug (ARV) used in combination therapy for the treatment of human immunodeficiency virus type-1 (HIV-1) infection. The drug is effective, safe, and well tolerated. Nonetheless, concerns have recently emerged for its usage in pregnant women or those of child-bearing age. Notably, DTG-based ARV regimens have been linked to birth defects seen as a consequence of periconceptional usages. To this end, uncovering an underlying mechanism for DTG-associated adverse fetal development outcomes has gained clinical and basic research interest. We now report that DTG inhibits matrix metalloproteinases (MMPs) activities that could affect fetal neurodevelopment. DTG is a broad-spectrum MMP inhibitor and binds to Zn++ at the enzyme's catalytic domain. Studies performed in pregnant mice show that DTG readily reaches the fetal central nervous system during gestation and inhibits MMP activity. Postnatal screenings of brain health in mice pups identified neuroinflammation and neuronal impairment. These abnormalities persist as a consequence of *in utero* DTG exposure. We conclude that DTG inhibition of MMPs activities during gestation has the potential to affect prenatal and postnatal neurodevelopment.

Disclosures: A.N. Bade: None. **J.M. McMillan:** None. **Y. Liu:** None. **B.J. Edagwa:** Other; Co-founder of Exavir Therapeutics, Inc. **H.E. Gendelman:** Other; Co-founder of Exavir Therapeutics, Inc..

Poster

P321. Neuroinflammation: Oligodendrocytes, Pericytes, and Other Cells

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P321.02

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant MH098742
NIH Grant MH118121
NIH Grant AI007632

Title: Bictegravir-mediated lysosomal de-acidification inhibits oligodendrocyte maturation and autophagic flux

Authors: *L. FESTA[1], A. E. CLYDE[1], C. C. LONG[1], L. ROTH[2], J. B. GRINSPAN[2], K. L. JORDAN-SCIUTTO[1];
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Abstract: Antiretroviral therapy (ART) has led to a reduction in the most severe forms of HIV-associated neurocognitive disorders (HAND); however, cognitive impairment continues to persist in approximately 30-50% of HIV+ patients. As the severity of HAND has shifted with the advent of ART, so has the underlying pathology, though certain deficits continue in the post-ART era. In particular, white matter (WM) alterations persist despite peripheral viral suppression by ART, suggesting that antiretrovirals (ARVs) may directly contribute to WM structural and functional deficits. Work in our laboratory has revealed that ARVs from numerous classes, including integrase strand transfer inhibitors (INSTIs), inhibit oligodendrocyte (OL) differentiation through distinct cellular stress pathways, including oxidative stress and the integrated stress response. We sought to determine whether bictegravir (BIC), a member of the INSTI class, prevents oligodendrocyte maturation. Using our well-established cell culture system of OPC purification and differentiation, we demonstrate that BIC inhibits oligodendrocyte maturation and reduces the expression of mature myelin proteins. In order to determine the potential mechanism underlying this phenomenon, we focused on lysosomal de-acidification since our previous work has revealed that ARVs from other classes can disrupt lysosomal pH, leading to deleterious effects on OPC maturation. BIC de-acidified lysosomes in both OPCs and mature OLs, as well as significantly impaired lysosomal degradative capacity; however, lysosomal membrane integrity was unaffected. De-acidification of lysosomes led to an inhibition of autophagic flux, a process known to be essential for proper OL function. Co-administration of the TRPML1 agonist, ML-SA1, which extrudes Ca^{2+} out of the lysosomal lumen and re-acidifies lysosomes, blocked the ability of BIC to inhibit OL differentiation. Ongoing studies are examining the potential impact of BIC on remyelination following a demyelinating insult. Taken together, our data reveal the critical role of proper lysosome acidification in modulating OL differentiation and maturation. Furthermore, this work has implications for other disorders characterized by WM injury, including multiple sclerosis, where OPC differentiation is impaired and remyelination is insufficient. Thus, our work suggests therapeutic targeting of lysosomal acidification or Ca^{2+} efflux may promote oligodendrocyte differentiation and myelin repair.

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Poster

P321. Neuroinflammation: Oligodendrocytes, Pericytes, and Other Cells

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Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant MH098742
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Title: Effects of Antiretroviral Bictegravir on Differentiating and Mature Oligodendrocytes

Authors: *A. E. CLYDE[1], L. FESTA[2], L. ROTH[3], K. L. JORDAN-SCIUTTO[2], J. B. GRINSPAN[4];
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Abstract: HIV-associated neurocognitive disorder (HAND) develops in approximately 50% of HIV (+) people and causes cognitive, motor, and behavioral impairments. Treatment with combined antiretroviral therapy (cART) led to a decrease in the most severe forms of HAND, but manifestations still occur in 30-50% of the HIV (+) population on cART. Despite successful viral suppression in the post-cART era, neuropathologic changes persist including white matter abnormalities and myelin loss. Previous studies from our lab have shown that some antiretroviral (ARV) drugs inhibit the differentiation of oligodendrocytes (OL), the myelin-producing cells in the brain, and could be contributing to the cognitive and white matter deficits seen in HAND. Bictegravir (BIC) is an integrase strand inhibitor (INSTI) front-line ARV drug that was approved in 2018 along with emtricitabine and tenofovir alafenamide in a drug cocktail called Biktarvy for treatment of HIV-1 infection. We hypothesized that bictegravir would decrease oligodendrocyte differentiation and increase cell death in differentiating oligodendrocytes but not in mature oligodendrocytes. To determine the effect of BIC on OL differentiation, oligodendrocyte progenitor cells (OPCs) were placed in differentiation medium (DM) and treated with three different concentrations of BIC for 72 hours. Cells were then immunostained with GalC and propidium iodide to examine immature OLs and cell death, respectively. Cells treated with BIC showed significantly lower numbers of GalC+ cells and lower cell viability compared to vehicle. To determine the effect of BIC on mature OLs, the cells were incubated in DM for 72 hours then treated with the highest dose of BIC for an additional 72 hours. There was no significant decrease in GalC+ cells or viability for BIC treated cells. Interestingly, the mature OLs also showed a smaller decrease in cell viability compared with OPCs when treated with thapsigargin (THAP), an inhibitor of the calcium ATPase on the endoplasmic reticulum, which serves as a positive control for cell death. We found that the mature OLs had significantly higher numbers of GalC+ cells and cell viability when treated with BIC or THAP than the simultaneously treated differentiating OPCs. These data suggest that mature OLs are more resistant to BIC induced changes in viability or differentiation as well as the cell death effects of THAP. Future studies will explore the potential mechanisms underlying the differential effects of BIC on stages of differentiation of the oligodendrocyte lineage.

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Poster

P321. Neuroinflammation: Oligodendrocytes, Pericytes, and Other Cells

Location: Virtual Only

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Program #/Poster #: P321.04

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

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Title: Pre-exposure prophylaxis (PrEP) inhibits oligodendrocyte differentiation

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Abstract: Adolescents comprise one-fourth of individuals diagnosed with HIV in the United States each year. Even with effective peripheral viral suppression by antiretroviral therapy (ART), this population is at risk for significant life-long behavioral, cognitive, and motor impairments as approximately 30-50% of HIV+ individuals develop HIV-associated neurocognitive disorders (HAND). White matter abnormalities are a hallmark of HAND and transcriptome analysis have found a significant decrease in the expression of genes associated with oligodendrocytes (OL), cells that produce myelin, in the white matter of ART treated, HIV+ individuals. Data from our lab has demonstrated that even in the absence of HIV, select antiretroviral drugs of the integrase inhibitor (INSTI) and protease inhibitor (PI) classes inhibit OL differentiation *in vitro*. PrEP, a combination of emtricitabine (FTC) and tenofovir disoproxil fumarate (TDF) two antiretrovirals of the NRTI class, is an effective method to prevent the transmission of HIV in adolescents at substantial risk for acquiring HIV. However, as myelination continues through adolescence and into adulthood, it is important to understand the effect of PrEP on developing white matter, which has not been investigated. Using our well-established cell culture system for purification and differentiation of primary rat OLs, we demonstrated that treatment with FTC and TDF, alone or in combination, inhibited OL differentiation. OL were treated at the time of differentiation with concentrations of FTC or TDF equivalent to that of the plasma C_{max} of patients on PrEP. Treatment with FTC (7uM), TDF (0.6uM), or in combination reduced the number of GalC and PLP positive cells after 3 days of differentiation. Known mechanisms by which ARV drugs affect myelination include ER stress

and lysosome stress, but we focused on lysosome deacidification as FTC and TDF are slightly basic drugs. Treatment with FTC and TDF, alone or in combination, significantly decreased the number of acidic lysosomes and increased the pH of lysosomes in a dose dependent manner. Lysosomes are critical in OLs as they must process and transport large quantities of lipids and proteins necessary for the generation of the myelin membrane and low intralysosomal pH is essential for the function of the lysosomal proteases. These findings suggest that FTC and TDF impair lysosome function and inhibit OL differentiation. Finally, reacidifying lysosomes restores OL differentiation in the context of FTC and TDF treatment. Our findings suggest that PrEP inhibits OL differentiation and the lysosome may be a therapeutic target to mitigate white matter damage in individuals on PrEP.

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Poster

P321. Neuroinflammation: Oligodendrocytes, Pericytes, and Other Cells

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P321.05

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Title: Occludin provides HIV-1 protection in human brain pericytes by regulating the expression of interferon stimulated genes

Authors: *S. TORICES[1], S. RAMASWAMY[1], T. MORENO[2], L. BERTRAND[1], R. CABRERA[1], A. KLUTTZ[1], O. NARANJO[1], M. TOBOREK[1];
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Abstract: HIV-1 associated blood brain barrier (BBB) alteration and neurocognitive disorders are typical clinical manifestations in HIV-1 infected patients. The functional and structural changes of the BBB closely relate with variations in expression of the tight junction protein occludin (ocln). Several studies have shown the capacity of ocln to control HIV-1 infection of human pericytes, ones of the main regulatory cells of the BBB integrity. Our study identified changes in gene expression-related to ocln modifications in human brain pericytes. After ocln

silencing, high-throughput RNA sequencing analyzed possible changes in a total of 14415 genes. We found that after downregulate ocln, there is an alteration in several genes of the INF-stimulated signaling pathway when compared with non-treated cells. This study focused on the genes that compose the ISG15 antiviral mechanism pathway given the capacity of these genes to inhibit infection of different viruses. Using qpcr analysis, we verified that ocln can modulate the expression levels of DDX58, USP18, ISG15, IFIT1, HERC5, MX1 and MX2 genes. Mechanistically, we show that ocln may regulate INF-stimulated genes though JAK/STAT signaling. We show that overexpressing ocln can significantly up-regulate INF-5α, INF-β, IRF3, IRF7 and IRF9 genes. Furthermore, we demonstrated that ocln induces the expression of STAT-1 at RNA and protein levels as well as increase the levels of phosphorylated STAT-1. Additionally, our results show for the first time that DDX58, ISG15, IFIT1, HERC5, and MX2 but not USP18 or MX1 genes regulate HIV-1 infection rate in human brain pericytes. These results potentiate the understanding of the molecular mechanisms for HIV-1 infection in the human brain.

Disclosures: S. Torices: None. **S. Ramaswamy:** None. **T. Moreno:** None. **L. Bertrand:** None. **R. Cabrera:** None. **A. Kluttz:** None. **O. Naranjo:** None. **M. Toborek:** None.

Poster

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Title: Brain Pericytes and the Formation of HIV-1 reservoirs in the CNS

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Abstract: Successful antiretroviral therapy (ART) has significantly decreased the mortality and morbidity of individuals infected with HIV. Still, low levels of HIV replication persist in viral reservoirs and lead to immune activation and chronic inflammation. The CNS was thought to be

protected from HIV infection. However, experiments on microglia and astrocytes indicated that these cells are all capable of active and latent viral infection. Even on ART, HIV-infected individuals are at a higher risk for non-AIDS related co-morbidities, including cardiovascular and neurological disease. We have pioneered research on HIV-1 infection in brain pericytes and indicated that these cells possess the receptor profile enabling latent and active HIV-1 infection. BBB pericytes have been shown to regulate brain paracellular and transendothelial fluid transport at the BBB, maintain homeostasis of the CNS microenvironment, and maintain BBB integrity. **Due to the position of BBB pericytes in the interplay between HIV and the CNS, we hypothesize that BBB pericytes are a key cell type for CNS viral reservoirs of HIV.** To test this hypothesis, we use a novel HIV reporter, named HIV_{GKO}, that allows for purification of latently infected cells in absence of reactivation. Fluorescence-activated cell sorting was performed to isolate active, latent, and uninfected populations of BBB pericytes. Transcriptome analysis on isolated cell populations indicates key differences in angiogenesis, glycolysis, and several inflammatory, and cell cycle pathways. Combined with in-vivo modeling of the BBB our data will help to understand in the mechanisms that might be triggering pericytes to become latent reservoirs and provide key targets in developing novel protective therapeutics for those suffering from HIV.

Disclosures: O. Naranjo: None. **S. Torices:** None. **M. Toborek:** None.

Poster

P321. Neuroinflammation: Oligodendrocytes, Pericytes, and Other Cells

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Program #/Poster #: P321.07

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: CAPES
CNPq
FAPERJ

Title: Mesenchymal Stromal Cells Protect the Blood-Brain Barrier, Reduce Astrogliosis, and Prevent Cognitive and Behavioral Impairments in Experimental Sepsis

Authors: *M. C. BARBOSA-SILVA[1], A. Y. O. SILVA[1], É. A. AMORIM[1], M. N. LIMA[1], M. G. GRANJA[1], R. CAMPOS[2], C. A. MORAES[1], P. M. FAGUNDES[1], H. A. OLIVEIRA[1], P. R. M. ROCCO[2], H. C. CASTRO-FARIA-NETO[1], T. MARON-GUTIERREZ[1];
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Abstract: Sepsis is a systemic inflammatory host-response against a pathogen; it is the largest cause of admission to intensive care units (ICUs) in the United States. Sepsis-associated

encephalopathies (SAEs) are neurological complications that occur during or after sepsis events. SAE causes long-term neurological consequences affecting memory, cognition and mood. Currently, there is no prevention or treatment for neurological damage resulting from SAEs. Thus, investigating new therapies is necessary. Both clinical and experimental studies have been show the effects of cell therapy in neurodegenerative diseases. Indeed, the immunomodulatory capacity of mesenchymal stromal cells (MSCs) is well established. The aim of the present study was to evaluate the effects of MSC therapy on blood-brain barrier (BBB) maintenance, astrocyte activation, neuroinflammation and cognitive/ behavioral damage in an experimental model of sepsis. In order to investigate that, we used male adult Swiss mice and cecal ligation and puncture (CLP) surgery for sepsis induction and sham-operated animals were used as control. Six hours after surgery, mice were treated with MSC intravenously (1×10^5 cells in 0.05 mL of saline/mouse) or saline. At day 1 the clinical score and plasma levels of Interleukin (IL) -1β, IL-6, and MCP-1 (assessed by ELISA) were increased in CLP animals. Cell therapy did not affect clinical score or survival rate, but led to a reduction in systemic levels of IL-1β, IL-6, and MCP-1 and also led a reduction in cortical and hippocampal cytokine levels. MSC therapy led a BBB protection (evaluated by Evans Blue Dye Assay) and a reduction in astrogliosis (evaluated by immunofluorescence) at 72h. In cultured astrocytes stimulated with lipopolysaccharide (LPS) conditioned media from MSCs reduced astrogliosis at 24h, suggesting a paracrine mechanism of action. These results might be related with an improvement in spatial memory (Morris Water Maze test), aversive memory (fear-conditioned test) and anxiety-like behavior (Elevated plus maze test). Thus, we can conclude that a single dose- therapy with MSC led to an improvement in cognitive damage, behavioral impairments, protection of the BBB, reduction in local levels of cytokines and astrogliosis and these effects might be associated with a decrease in systemic levels of inflammatory mediators.

Disclosures: M.C. Barbosa-Silva: None. **A.Y.O. Silva:** None. **É.A. Amorim:** None. **M.N. Lima:** None. **M.G. Granja:** None. **R. Campos:** None. **C.A. Moraes:** None. **P.M. Fagundes:** None. **H.A. Oliveira:** None. **P.R.M. Rocco:** None. **H.C. Castro-Faria-Neto:** None. **T. Maron-Gutierrez:** None.

Poster

P321. Neuroinflammation: Oligodendrocytes, Pericytes, and Other Cells

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Program #/Poster #: P321.08

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Title: Cerebral malaria-related factors heme and Histidine Rich Protein 2 (HRP2)-induced brain organoids damage is Toll Like Receptors 1 and 2-dependent

Authors: *A. HARBUZARIU, J. C. CESPEDES, A. NTI, K. HARP, A. DRISS, J. K. STILES; Morehouse Sch. of Med., Atlanta, GA

Abstract: Rationale: Despite available treatment, *Plasmodium falciparum (Pf)* infection remains a significant global health burden. Over 400,000 deaths mainly due to cerebral malaria (HCM) complications in children under age 5 are reported annually. Previous studies with a 3D *in vitro* model (human brain cortical organoids) indicated that heme and HRP2 released from lysed *Pf*-infected erythrocytes cause brain inflammation (e.g., CXCL10) and loss of neuronal viability and development. Comparative analyses of Human Cerebral Malaria (HCM; *Pf*) and experimental cerebral malaria (ECM; *Plasmodium berghei* ANKA in C57BL/6) revealed that both models activate brain microvascular endothelial cells, cause blood brain barrier (BBB) leakage and perivascular neuronal injury in the late stages. Circulating Neuregulin-1 (NRG1; cytoprotective) declined in HCM, whereas tissue expression of NRG1 and its receptor ErbB4 in damaged brain areas were elevated. Adjunctively administering NRG1 with artesunate (ART) in ECM mice eliminated parasite burden and reduced brain damage and ECM mortality. The molecular pathways mediating heme- and HRP2-induced brain damage are unknown. Transcriptome analysis revealed key roles for toll like receptors 1 and 2 (TLR 1& 2). Hypothesis: heme and HRP2-induced brain inflammation associated with cerebral malaria are TLR (1, 2) dependent. We functionally assessed heme- and HRP2-induced inflammation as well as the neuroprotective effects of NRG1 in brain organoids using IHC, WB and gene microarrays. The involvement of TLR 1 and 2 signaling was also assessed in the brain of Plasmodium berghei ANKA (PbA) infected mice and in post-mortem HCM brain samples by IHC. Results indicate that exposure of human brain organoids to physiologically relevant concentrations of HRP2 (4.2 μg/ml; HCM) and heme (30 μM; HCM) increased TLR 1 and 2 expression in microglia, neurons and astrocytes. Furthermore, heme and HRP2 effects were attenuated by exogenously augmenting NRG1. We conclude that heme and HRP2-induced brain organoids damage is TLR1 and TLR2 dependent and is attenuated by NRG1.

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Poster

P321. Neuroinflammation: Oligodendrocytes, Pericytes, and Other Cells

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Program #/Poster #: P321.09

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

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Title: Dendrimer-conjugated N-acetyl cysteine prevents neurobehavioral defects in a pediatric rabbit model of tuberculous meningitis

Authors: *J. KIM[1], C. ERICE[1], L. PIETERSE[1], A. SHARMA[2], S. JAIN[3], R. KANNAN[2], S. KANNAN[1], E. TUCKER[1];
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Abstract: Introduction Tuberculous meningitis (TBM) is the most aggressive form of extrapulmonary TB, particularly among children. Despite standard-of-care treatment with antimicrobials and host-directed therapy (HDT; i.e. corticosteroids), mortality and morbidity are unacceptably high and new therapies are needed urgently. N-acetyl cysteine (NAC) is a promising HDT with antioxidative and anti-inflammatory properties, albeit with poor bioavailability. However, when conjugated to dendrimer (D-NAC) nanoparticles for targeted delivery to activated microglia, it improves behavior deficits and decreases microglia activation in models of neuroinflammation (e.g. cerebral palsy, traumatic brain injury). Our previously established pediatric rabbit model of TBM demonstrated both neurobehavioral defects and increased activated microglia associated with pathophysiology. Here we test the ability of D-NAC to rescue neurobehavioral defects associated with TBM and to determine if behavioral data correlates with decreased neuroinflammation. **Methods** Young New Zealand White rabbits (post-natal day 5) were injected with live *Mycobacterium tuberculosis* H37Rv (infected) or PBS (uninfected control). After 3 weeks, infected rabbits received weekly intravenous D-NAC or PBS for 2 weeks. Neurobehavior testing was done by automated video tracking of open field tests recorded for 2 mins on day 0, 3, 7, 10, and 14 of treatment. A comprehensive score was calculated based on 15 components of motor behaviors from an automated tracking software (Noldus EthoVision XT^{TM}) and manual scoring (by 2 blinded reviewers) of 11 salient neurobehavioral deficits (e.g. limb paresis, head tilt, balance). All scores were normalized to the corresponding day's baseline score established by the control mean score. Neuroinflammation was assessed by ionized calcium binding adaptor molecule 1 (Iba-1) immunostaining of paraffin-embedded brain sections which was semi-quantitively analyzed with the HALO Image Analysis Platform. **Results** Infected rabbits treated with PBS (n=9) showed significantly worse neurobehavioral outcomes at 14 days compared to control (n=8; P=0.0013). However, a decline in behavior was not observed in D-NAC treated rabbits that demonstrated no behavioral deficits compared to control (n=5; P=0.442). Iba-1 staining has been optimized and quantification is underway (n=3 per condition). **Conclusions** D-NAC treatment prevents worsening neurobehavioral outcomes and possibly neuroinflammation in a pediatric rabbit model of TBM. These data suggest D-NAC is a promising HDT candidate and additional studies in combination with antimicrobial regimens would be beneficial.

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Poster

P321. Neuroinflammation: Oligodendrocytes, Pericytes, and Other Cells

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Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: ASCEND: RL5GM118972
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R29 NS31857-01A1

Title: Evidence of Reactive Oxygen Species in the HIV-1 Transgenic Rat Model of Cardio Pathology

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[1]Biol., Morgan State Univ., BALTIMORE, MD; [2]Biol. and Imaging Core Director, Morgan State Univ., Baltimore, MD; [3]Animal Model Div., Inst. of Human Virology, BALTIMORE, MD

Abstract: Because of improvements in antiviral medicines, HIV/AIDS has become a chronic disease. The survival rate has been improved markedly. However, HIV is not eliminated from the patient but exists in a restricted population of cells. These reservoir cells continue to produce very low levels of virus and viral proteins (GP-120, Tat, Nef). It has been hypothesized that this low level of HIV or the HIV proteins contribute to the development of noninfectious HIV comorbidities (CM). The longer the patients survive, the greater the probability of development of the CMs. There are a number of organs that are affected. Of particular interest are the heart and brain. There are several theories on how cell death and dysfunction can occur. One is the production of Reactive Oxygen Species (ROS). We have identified the presence of nitrotyrosine in the HIV-1 Transgenic Rat heart (Microscopy and Microanalysis, V26, S2, 2020 pp. 1342-1344) suggesting that free radial damage is occurring. This model produces measurable levels of GP-120. It therefore models chronic viral protein production in treated HIV infection. One mechanism by which ROS can be produced is if the enzyme NOX 2 is activated and up regulated. One way this might happen is if GP-120 binds to CXCR4 positive cells in the heart. Immunocytochemical staining of the HIV-1 rat heart for CXCR4 reveals a positive dendritic cell type. Immunocytochemical staining in the HIV-1 rat heart for NOX2 reveal more positive cells than controls. Also, of a dendritic type. Increased numbers of NOX2 positive cells suggest an inflammatory process. Labels to cell type will reveal if the cells are macrophages (infiltrating) or dendritic cells (resident cells). The production of ROS by these cells may affect the myocytes. Cardiac muscle also presents with an addition mechanism which uses ROS. This is the XROS

system. In this system the microtubules act as a mechanotransducer to activate NOX2. But GP-120 has been reported to bind to tubulin. If this is the case the generation of ROS by the ROS-X system may be compromised. Alteration of XROS signaling could contribute to myopathy in addition to extracellular exposure by macrophages or dendritic cells. Future studies will examine this possibility.

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Poster

P325. Cellular Mechanisms of Stroke

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Program #/Poster #: P325.01

Topic: C.08. Ischemia

Support: NIH Grant R01NS120322
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Title: Agent-based modeling of neuronal mitochondrial dynamics and quality control in ischemia/reperfusion injury

Authors: *G. M. FOGO, T. H. SANDERSON;
Univ. of Michigan, Ann Arbor, MI

Abstract: Cerebral ischemia continues to be a leading cause of death and disability. In this injury paradigm, both ischemia and reperfusion induce extensive damage to the brain. A prominent component of ischemia/reperfusion (I/R) injury is mitochondrial dysfunction. While chiefly known as essential generators of cellular energy, mitochondria also act as dynamic agents fusing to form sprawling healthy networks and undergoing fission to separate unhealthy segments. Mitochondrial homeostasis is tightly regulated by the fission and fusion forces of mitochondrial dynamics, as well as mitophagy. Cerebral I/R injury leads to disruptive events of mitochondrial dynamics and mitophagy that exacerbate and drive neurological damage. To better understand the patterns of mitochondrial dysfunction in I/R injury, we aimed to construct a predictive computational model of mitochondrial dynamics and quality control. Utilizing the open-source agent-based modeling platform NetLogo, we have designed a simulation model of single cell mitochondrial dynamics in basal and ischemic conditions. The model simulates several intrinsic factors of individual mitochondria and their influences on dynamic behavior, including: morphology, size, age, connectivity, and local membrane potential. The probabilistic equations that drive the model's behaviors were generated from live cell imaging data of mouse primary cortical neurons. Primary neurons were cultured to DIV14 and exposed to oxygen-

glucose deprivation and reoxygenation (OGD/R) in our live cell imaging setup. In order to correlate intrinsic factors with dynamic events of fusion and fission, we developed an automated machine learning-based methodology for detecting individual fission and fusion events over time during imaging. To extract targeted intrinsic parameters from the mitochondria of these cells, we utilized the genetic reporters MitoTimer and MitoQC, as well as TMRM staining. Our agent-based model closely simulates the dynamic behavior of neuronal mitochondria and is currently in further development to mimic the patterns of mitochondrial dysfunction seen in cerebral I/R injury. Future work will be focused on implementing known proteins of mitochondrial dynamics into the model (i.e., Drp1, Opa1), as well as other intracellular variables including calcium and reactive oxygen species (ROS) signaling. We expect further optimization and utilization of our model will inform the development of potential therapeutic interventions and predict the optimal intervention time to promote cell survival in cerebral I/R injury.

Disclosures: G.M. Fogo: None. **T.H. Sanderson:** None.

Poster

P325. Cellular Mechanisms of Stroke

Location: Virtual Only

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Program #/Poster #: P325.02

Topic: C.08. Ischemia

Title: Cd38 activation of the trpm2 ion channel contributes to post-stroke hippocampal dysfunction

Authors: *A. BURCH[1], D. MITCHELL[1], J. ORFILA[2], A. L. DINGMAN[1], N. QUILLINAN[1], P. S. HERSON[2];
[1]Univ. of Colorado Anschutz Med. Campus, Aurora, CO; [2]Ohio State Univ. Col. of Med., Columbus, OH

Abstract: Emerging clinical data reveals post-stroke cognitive impairment (PSCI) as one of the leading causes of disability in the country. Recently, our lab has identified the TRPM2 ion channel as a potential molecular target for the reversal of cognitive symptoms following global cerebral ischemia, without conferring neuroprotection. These results suggest a potential neurorestorative function of the TRPM2 channel independent of its well-established role in neuronal toxicity and death. Whether TRPM2 channel modulation is of benefit in the context of focal ischemia and PSCI remains an open question. To address this, we perform a 60-minute transient middle cerebral artery occlusion (MCAO) in adult (8-12wk) male mice as a preclinical model for stroke and assess synaptic plasticity in combination with neurobehavior with a focus on the hippocampus, a region known to contribute to PSCI but is not directly supplied by the MCA. To measure hippocampal synaptic plasticity, we employ field electrophysiology and assess long-term potentiation (LTP) in the CA1-Schaffer collateral pathway at delayed (30 day)

timepoints following MCAO. We find that LTP is reduced following MCAO (n=3, 15.0±4.0%) compared to sham (n=6, 47.2±9.2%; $p<0.05$). However, i.v. treatment with tat-M2NX (20mg) on day 29, a potent and specific inhibitor of TRPM2, fully restores LTP on day 30 (n=6, 65.3±8.1%). Consistent with the electrophysiology data, we also observe reduced levels of contextual fear conditioning (a measure of hippocampal-dependent short-term memory), 30 days following MCAO (n=3, 23.6±3.3%), that are reversed following i.v. treatment with 20mg tat-M2NX on day 29 (n=5, 73.4±5.3%). We next investigate potential mechanisms of stroke-induced TRPM2 activation with a focus on CD38, an ectoenzyme and potential source of the TRPM2 ligand, ADP-ribose (ADPr). Using immunohistochemical (IHC) staining, we show CD38 is localized to activated astrocytes in the hippocampus following MCAO. We also find impaired hippocampal LTP on MCAO acute brain slices is restored to sham levels following bath application of apigenin (vehicle n=7, 23.9±7% vs. 10μM apigenin n=7, 65.5±2.5%; $p<0.05$), and 78c (vehicle n=6, 4.0±8.5% vs. 100nM 78c n=6, 78.3±15.8%; $p<0.05$), phenocopying TRPM2 inhibition. Taken together, these data identify the TRPM2 channel as a contributor to stroke-induced hippocampal dysfunction, suggesting a role for TRPM2 beyond neuronal death. Furthermore, we also provide data demonstrating the astrocyte-localized ectoenzyme, CD38, as an ADPr source and potential upstream regulator of TRPM2, uncovering a novel neuroglial pathway underlying PSCI.

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Poster

P325. Cellular Mechanisms of Stroke

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Program #/Poster #: P325.03

Topic: C.08. Ischemia

Support: Corporación Universitaria Remington 2017' (Project 4000000115-17)
Cellular and Molecular Neurobiology Area of Group of Neuroscience, University of Antioquia.

Title: High fructose diet-induced obesity worsens post-ischemic brain injury

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Abstract: Cerebral ischemia is caused by a reduction of the blood flow in a specific area in the brain, triggering cellular cascades in the tissue that result in neuronal death. This process leads to

a neurological decline in patients who suffer cerebral ischemia. The extent of the injury after cerebral ischemia could be related to the comorbidity condition. Therefore, the main aim of this study was to analyze the effect of obesity induced by a high fructose diet (HFD) on the brain after cerebral ischemia in rats. We induced the obesity model in Wistar rats with 20% fructose in water for 11 weeks (n=12). After this time, we performed the cerebral ischemia surgery (2VO) (n=3-4), we also carried out the neurological test at 6-, 24- and 48-hours post-ischemia and we analyzed the histological markers of neurodegeneration (n=3-4). Finally, we treated bend.3 endothelial cell line with 20% of plasm from obese rats and analyzed endothelial damage (n=3-4, 1 replica). Data were analyzed using Shapiro-wilk normality test, T-test and two-way ANOVA. The HFD induced an obese phenotype without insulin resistance. The obese rats exhibited worse neurological performance at 6 hours post-ischemia. Due to the ischemia surgery was global, we observed a different infarct zone in each animal analyzed (caudate-putamen, motor cortex, amygdala, and hippocampus). However, independent of the infarct zone we found the same pattern of neurodegeneration markers related to the damage in the infarct zone; such as higher neuronal loss area and astroglial and microglial hyperreactivity at 48 hours post-ischemia. We also found that the hippocampus was a commonly affected area, where we found loss of neurons, an increase in interleukin 1 â (IL-1B) in the blood vessels of the Dentate gyrus, a remarkable disruption of MAP-2+ dendrites, a loss of BDNF staining, and the presence of PHF-tau. Although, we observed a tendency to increase the IL-1B expression. In the same way, the plasm from obese rats induced more endothelial damage characterized by increased condensed nuclei, and, cPLA2 and phalloidin fluorescence intensity. In conclusion, an HFD induces an obese phenotype, which produces an increase in neuronal loss, inflammation, and plasticity impairment in the hippocampus after cerebral ischemia. Additionally, the endothelial alteration is a possible cellular mechanism of damage in obesity.

Disclosures: P. Pérez Corredor: None. **G. Cardona Gómez:** None. **N. Balcázar Morales:** None. **J. Gutierrez Vargas:** None.

Poster

P325. Cellular Mechanisms of Stroke

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Topic: C.08. Ischemia

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NIH Grant NS088555

Title: Pilot study of calcium imaging in the contralesional hippocampus of aged and young synapsin-Cre/GCaMP6S male mice after stroke

Authors: *T. A. UJAS[1], T. L. ANDERSON[1], M. XIA[1], A. GORNET[1], V. O. TORRES[4], S. MESSMER[1], J. TURCHAN-CHOLEWO[1], M. K. MALONE[1], P. I. ORTINSKI[2], A. M. STOWE[3];
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Abstract: Background: Stroke injury following a transient middle cerebral artery occlusion (tMCAo) induces a rapid neuroinflammatory response in the injured brain that lasts for weeks. The present study focuses on investigating the effects of neuroinflammation on spatiotemporal synaptic hippocampal activity long-term post-stroke. We sought to establish a protocol using calcium imaging in the contralesional hippocampus to test long-term modulation of adaptive immune responses after stroke. Methods: C57BL/6 synapsin-Cre/GCaMP6S$^{+/-}$ aged mice (12-15 months; n=10) were depleted of B cells (100µg mCD20 or isotype (Iso) control; Genentech) 1 day prior to 60-min tMCAo, with weekly injections to deplete B cell turnover. The second cohort (young, 1.5-3 months; n=10) were split into uninjured and 60-min tMCAo groups. Brains were sectioned into 300µm slices, oxygenated, and kept at 37ºC in aCSF. Wide-field calcium imaging was conducted in 100µM of picrotoxin using a stimulating electrode within the Schaefer collaterals of the stratum radiatum, 5-10µm from the CA1 pyramidal cell layer. Cells were stimulated at increased intensities (0.1-0.5 mA) for 10ms intervals at 100Hz. For each cell, a fluorescence trace was generated by averaging all pixels within ROI outline in each recording frame. The associated background signal was removed from each cell's fluorescence trace. Calcium transients were identified using a wavelet ridgewalking algorithm. Statistical analyses were performed in GraphPad Prism. Results: In the aged cohort, imaging of CA1 pyramidal cells did not occur in the absence of picrotoxin, and few cells were found in the Iso-treated mice (n=2 mice with 13 cells detected) when compared to mCD20-treated mice (n=3 mice with ~100 cells). The amplitude and duration of the fluorescent signal were both significantly affected by stimulus intensity ($p<0.001$) regardless of B cell depletion. The Iso-treated group exhibited a trend for lower amplitude ($F(1,104)=3.2$; $p=0.08$). Young synapsin-Cre/GCaMP6S$^{+/-}$ exhibited low fluorescence expression, with 2/5 uninjured mice having no expressed cells despite a confirmed genotype by PCR. The young cohort is currently being analyzed by the ridgewalking algorithm to determine age-effects after tMCAo. Conclusions: While calcium imaging is useful in determining synaptic activity after stroke, particularly as it may relate to the development of dementia, it has proven challenging using C57BL/6 synapsin-Cre/GCaMP6S mice. This may be secondary to heterozygosity for GCAMP6S, animal age, and injury status. Further optimization is needed to determine the usefulness of this transgenic line in pre-clinical stroke studies.

Disclosures: T.A. Ujas: None. **T.L. Anderson:** None. **M. Xia:** None. **A. Gornet:** None. **V.O. Torres:** None. **S. Messmer:** None. **J. Turchan-Cholewo:** None. **M.K. Malone:** None. **P.I. Ortinski:** None. **A.M. Stowe:** None.

Poster

P325. Cellular Mechanisms of Stroke

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P325.05

Topic: C.09.Stroke

Support: KHIDI Grant HI19C0481
KHIDI Grant HC19C0028

Title: Protective effects of preconditioning with carbon dioxide inhalation against acute focal cerebral ischemia/reperfusion-induced brain injury in a rat middle cerebral artery occlusion model.

Authors: C. YOON[1], H. LEE[2], I. KWON[2], Y. PARK[1,2,3], Y. SHIM[1,2,3], Y. KIM[1,2], J. HEO[1,2,3], *H. NAM[1,2];
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Abstract: Stroke is a major cause of mortality and permanent neurological deficits all over the world. Despite its high incidence and death rate, clinical therapeutic options are still limited. The potential therapeutic target is to keep the ischemic penumbra alive and to prevent destruction of cerebral tissue. Ischemic preconditioning is a promising strategy to save the ischemic penumbra and reduce tissue damage. Carbon dioxide (CO_2) is the most potent vasodilator of cerebral vessels in the earth and CO_2 inhalation dramatically increases cerebral blood flow. However, it is not clear whether preconditioning treatment with CO_2 can effectively reduce cerebral tissue damage and functional deficits. Hence, we evaluated the effects of preconditioning with CO_2 inhalation on focal ischemia-induced brain injury and neurological deficits in a rat ischemic stroke model. Adult male Wistar rats (270-320g, n=8) were inhaled with 20% CO_2 mixed gas (applied in 20% CO_2, 20% oxygen, and 60% nitrogen mixture) with 3 cycles of inhalation for 5 min followed by 10 min recovery (room air) before transient middle cerebral artery occlusion (tMCAO) for 90 min. To evaluate the neurological deficits, behavioral tests were blindly performed with the modified Neurological Severity Scores (mNSS), Longa and Garcia score at 24 h after tMCAO. In order to assess motor coordination and balance after stroke, we also performed the rotarod test and recorded the latency of rats to fall from the accelerating rotarod. We found that the mNSS was 0 in the sham and CO_2 pretreatment sham condition (n=5 per group). The CO_2 inhaled preconditioning tMCAO rats exhibited significantly lower mNSS compared to tMCAO rats (6±2.0 vs. 11±1.3, n=8, $p<0.0001$). Rats in both sham group and CO_2 pretreatment sham group had the latencies in 300 sec. The latency was dramatically increased in CO_2 preconditioning tMCAO rats compared to tMCAO rats (99.5±53.5 sec vs. 27.5±8.7 sec, $p=0.0006$). Furthermore, preconditioning tMCAO rats with CO_2 showed significantly small infarction size compared to tMCAO rats (17.4±4.1% vs. 30.8±3.7%, $p<0.0001$). We demonstrated that CO_2 pretreatment plays an important role in reducing ischemic brain damage. In the sham animals, CO_2 inhalation did not show harmful effects. Taken together, these findings suggest that preconditioning with CO_2 inhalation protects against ischemic-induced brain injury

and improves neurological outcomes without complication after acute ischemic stroke. Therefore, inhaled CO_2 preconditioning treatment may provide a novel therapeutic approach for ischemic stroke.

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Poster

P325. Cellular Mechanisms of Stroke

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P325.06

Topic: C.09.Stroke

Title: Poststroke lipid droplet accumulation in residing microglia and its influence on inflammation and phagocytosis

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Abstract: Inflammation is a hallmark of ischemic brain injury, for which resident microglia play a pivotal role. Recent evidence from non-stroke disease conditions suggests microglia to be also involved in abnormal lipid metabolism and lipid droplet (LD) formation. Such processes are highly regulated by lipid-associated proteins like Plin2, a member of the perilipin family. Whereas the role of Plin2 under stroke settings remains elusive, Plin2 has recently been found to promote LD production in Alzheimer's Disease. Using preclinical stroke models in both cell culture and C57BL6 mice, the present work aimed to elucidate the role of LD formation under stroke condtions in general and Plin2 in particular. Indeed, LD formation was significantly increased in microglia exposed to in vitro hypoxia conditions, as was the case in stroke mice. The extent of LD formation correlated with the extent of cell death and brain injury. Likewise, Western blotting against Plin2 revealed increased protein abundance in the aforementioned cells and brain tissue. Using ELISA and RT-PCR techniques, cytokine secretion patterns from microglia displayed increased levels of TNFalpha, IL6, and IL1beta under stroke conditions. The latter was associated with high formation rates of reactive oxygen species. Further analysis of microglia resulted in enhanced M1 phenotypes that displayed high expression rates of the ABCA1 transporter, thus blocking cholesterol efflux into the extracellular matrix. The plethora of observations made herein finally resulted in a stimulation of the proinflammatory NfKb signaling pathway, further increasing cell injury. In conclusion, interfering with lipid metabolism disorders in microglia exposed to stroke rather than to directly exert neuroprotection, may provide a new perspective for the treatment of cerebral ischemia.

Disclosures: W. Wei: None. **D. Fitzner:** None. **M. Bähr:** None. **T. Doeppner:** None.

Poster

P325. Cellular Mechanisms of Stroke

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P325.07

Topic: C.08. Ischemia

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Killam fellowship
Michael Smith Foundation for Health Research

Title: Fluorescence lifetime imaging reveals KCC2-dependent chloride microdomains in dendritic blebbing

Authors: *N. L. WEILINGER[1], L. E. WICKI-STORDEUR[1], C. J. GROTEN[1], J. M. LEDUE[2], K. T. KAHLE[3], B. A. MACVICAR[1];
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Abstract: Intracellular chloride ion ($[Cl^-]_i$) homeostasis is critical for neurotransmission and volume regulation. We recently showed that calcium (Ca^{2+})-independent cytotoxic swelling in neuronal somata requires intracellular Cl^- loading through the Cl^- channel SLC26A11. However, our understanding of Cl^--dependent swelling in dendrites (termed “blebbing”), a hallmark of stroke and traumatic brain injury, is poorly understood. Here, we investigated the spatiotemporal $[Cl^-]_i$ dynamics in dendritic subcompartments to elucidate mechanism of synaptic swelling that engenders synaptic lysis. We combined whole-cell patch clamp electrophysiology with simultaneous fluorescence lifetime imaging (FLIM) of the Cl^- dye MQAE to quantitatively map dendritic Cl^- levels in N-methyl-D-aspartate (NMDA) excitotoxicity. We validated FLIM-based $[Cl^-]_i$ readouts from rat and mouse neurons, and found basal $[Cl^-]_i$ measures closely matched expected values from the literature and were corroborated by Rubi-GABA uncaging to measured E_{GABA}. Low baseline $[Cl^-]_i$ in dendrites required Cl^- efflux via the K^+-Cl^- cotransporter KCC2 (*SLC12A5*). To assess the possibility of pathological Cl^- microdomains in dendrites, we applied NMDA which generated spatially heterogeneous and persistent subdomains of high $[Cl^-]_i$ that formed dendritic blebs. Unlike somatic swelling, dendritic $[Cl^-]_i$ dysregulation required local Ca^{2+} influx and Cl^- loading caused by reversed KCC2 transport. Together, our data show that neuronal swelling is mechanistically distinct between somatic and dendritic subcompartments. These data also establish a novel role for KCC2 to generate subcellular $[Cl^-]_i$ microdomains in dendritic blebbing, with relevance for multiple neurological disorders.

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Poster

P325. Cellular Mechanisms of Stroke

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P325.08

Topic: C.08. Ischemia

Support: NIH grant AA026756

Title: Prenatal alcohol exposure elevates stroke severity and increases gut permeability in adult rats

Authors: *S. BAKE[1], S. PANDEY[3], D. HURST[4], M. PINSON[2], R. C. MIRANDA[5], F. SOHRABJI[6];
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Abstract: Adverse prenatal experience is linked to poor health outcomes in postnatal life. There is very little information on the developmental origin of adult-onset diseases, particularly prenatal alcohol exposure (PAE) and stroke outcomes. The present study tested the hypothesis that PAE will lead to greater stroke severity and will disrupt the integrity of the gut barrier in adult offspring. Pregnant Sprague Dawley rats were exposed to 10% ethanol vapor for 1 h from GD 8-18 using the rodent alcohol inhalation system. Control animals were exposed to air. The 5 month old adult offspring were subjected to endothelin-1 induced unilateral middle cerebral artery occlusion (MCAo) and outcomes were evaluated after 2 days. Stroke outcomes were assessed from mortality rates, behavioral assays, infarct volume, and gut permeability markers. Interestingly, the PAE males had significantly higher mortality within 48 h of stroke compared to PAE females ($p<0.03$). Behavioral analysis indicates that PAE animals exhibited greater neurological deficits as determined by their higher composite score compared to controls ($p<0.02$). Brain damage assessed by infarct volume was not significantly different from control animals. However, serum LPS was increased in PAE-males ($p<0.03$), and there was also an elevation in serum intestinal fatty acid binding protein (iFABP, $p<0.03$) in PAE-females compared to their control counterparts, suggesting that PAE increased intestinal barrier permeability after stroke. The present findings indicate that PAE contributes to long-lasting adverse effects on adult health as evidenced by greater mortality and poor functional recovery during the acute phase of stroke.

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Poster

P325. Cellular Mechanisms of Stroke

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P325.09

Topic: B.10. Epilepsy

Support: NIH Grant NS18400
NSF 1901344

Title: Novel approach to analyzing steady-state intracellular pH and the recovery from NH_4^+-induced acidosis in rat hippocampal neurons and astrocytes during acute hypoxia

Authors: *V. A. RUFFIN[1], W. F. BORON[2];
[1]Natural Sci., Virginia Union Univ., Richmond, VA; [2]Physiol. & Biophysics, Case Western Reserve Univ., Cleveland, OH

Abstract: Regulation of intracellular pH (pHi) in neurons and neuroglial is necessary for optimal brain function, especially in the hippocampus (HC), an area involved in learning and memory. In the absence of CO2/HCO3−, the major family of proteins regulating acid/base balance and pHi are the Na-H exchangers (NHEs). In the present study, we used the pH-sensitive dye BCECF to examine the regulation of steady-state pHi and pHi recovery from NH4+ -induced intracellular acid loads during acute hypoxia in HC neurons and astrocytes, co-cultured from embryonic (E18-20) Sprague Dawley rats, and studied in CO2/HCO3− -free HEPES-buffered ("HEPES") solutions. After at least 14-days in a CO2 incubator, cells were removed, loaded with BCECF, and placed in a recording chamber with flowing HEPES at 37°C. We measured pHi at the beginning of each experiment (checkpoint A), and again after 5 min of stabilization (checkpoint C). The "initial pHi"— the value at checkpoint C, (pHi)C—was 7.423 (N=42) for neurons (astrocytes: 7.184, N=46). We used a twin, unpaired NH4+-pulse protocol (1st pulse in HEPES/21% O2, 2nd pulse in HEPES/1% O2) to examine the acute transition to hypoxia. After each NH4+ removal, pHi recovers (increases) following an ~exponential time course. During pHi recovery from the 1st acid load in HEPES/21% O2, the quasi-single exponential rate constant (kqSExp) averaged 0.0048 s−1. The new average neuronal steady-state pHi, now at checkpoint D, (pHi)D, was 7.293 (astrocytes: 7.154 /0.0096 s−1). After switching to HEPES containing 1% O2, neuron pHi shifts upward to 7.415 (astrocytes: 7.340) at checkpoint E, (pHi)E. We then impose the 2nd NH4+ pulse. During the pHi recovery, kqSExp was 0.0060 s−1;

the new steady-state pHi in 1% O2, now at checkpoint F, (pHi)F, was 7.382 (astrocytes: 7.347/0.0072 s−1). We hypothesize that (pHi)C exceeds (pHi)D because, without HCO3− -dependent acid loaders, cells—after they initially alkalinize as they come out of the CO2 incubator—cannot fully acidify . For each recovery from an acid load, we use a novel analysis in which we plot pHi recovery rate (dpHi/dt) vs pHi. We bin kqSExp values according to (pHi)D, using pHi intervals of 0.2. We find that 1) kqSExp is greater (faster) for astrocytes than neurons; 2) kqSExp tends to be low for cells in the lowest pHi bins, highest in moderate-pHi bins (more dramatically for astrocytes), and low again at the highest-pHi bins; and 3) acute transition from a HEPES solution containing 21% O2 to a solution containing 1% O2 alkaline raises pHi in both neurons and astrocytes. These new analytical approaches may ultimately provide mechanistic insight into cell-to-cell heterogeneity of pHi regulation in the nervous system.

Disclosures: V.A. Ruffin: None. **W.F. Boron:** None.

Poster

P325. Cellular Mechanisms of Stroke

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P325.10

Topic: B.11. Glial Mechanisms

Support: LaCroute Fellowship
NIH Grant R01NS110690

Title: The role of mGluR5 in neurovascular coupling during development and after stroke

Authors: *T. STACKHOUSE[1], Z. LI[4], H. MCCONNELL[1], W. ZHANG[2], A. MISHRA[3,1]; [1]Dept. of Neurology, Jungers Ctr. for Neurosciences Res., [2]Dept. of Anesthesiol. and Peri-operative medicine, [3]Knight Cardiovasc. Inst., Oregon Hlth. & Sci. Univ., Portland, OR; [4]Gen. hospital of Ningxia Med. Univ., Ningxia, China

Abstract: Cerebral blood flow (CBF) is regulated to match local neural activity with high temporal and regional specificity via a process termed neurovascular coupling. In adult humans and rodents, increases in neural activity reliably induces capillary dilations, resulting in an increase in local CBF. However, neurovascular coupling is much more variable in young humans and rodents, suggesting that the underlying mechanisms are different in adults and the developing brain. We studied this developmental change in neurovascular coupling in acute cortical slices using electrical stimulation to evoke neuronal activity. In P6-10 rat slices, stimulation evoked a dilation in 78% and a constriction in 22% of capillaries. The proportion of dilating capillaries increased to 100% by P21. The magnitude of stimulation-evoked capillary dilation also increased with age, from 3.3±2.4% in P6-10 slices to 11.9±3.2% in P11-15 ($p<0.14$) and 15.4±2.0% in >P21 ($p<0.003$). A loss or attenuation of neurovascular coupling is also

reported in many neurological conditions including stroke. Using the middle cerebral artery occlusion (MCAO) model of stroke, we found that activity-dependent capillary response is reduced by ~75%, to 3.3 ± 1.0% dilation, in otherwise healthy ipsilesional cortical regions outside the infarct compared to contralateral hemisphere or naïve brains. Neurovascular coupling at capillaries is mediated by astrocytes in a Ca^{2+}-dependent manner; however the source of this Ca^{2+} is debated, with some findings supporting a role for metabotropic glutamate receptor 5 (mGluR5) while others suggest a role for purinergic P2X1 channels. We investigated whether mGluR5, which is expressed in immature and reactive astrocytes but not healthy adult astrocytes, is responsible for the suppression of neurovascular coupling during early development and after stroke. Inhibiting mGluR5 rescued stimulation-evoked capillary dilation in the stroke hemisphere of MCAO rats, and a similar effect was observed in young (<P21) slices. Importantly, mGluR5 inhibition did not alter neurovascular coupling in healthy adult slices. Our data offer a resolution to the debate surrounding the role of mGluR5 in neurovascular coupling and suggest that an mGluR5-dependent mechanism is responsible for the reduced neurovascular coupling observed in development and after stroke, but not in healthy adulthood. This concept aligns with previously published patterns of mGluR5 expression in astrocytes. Future studies will focus on the specific role of astrocytic mGluR5 in neurovascular regulation and the downstream Ca^{2+}-dependent pathways involved.

Disclosures: T. Stackhouse: None. **Z. Li:** None. **H. McConnell:** None. **W. Zhang:** None. **A. Mishra:** None.

Poster

P335. Stroke Imaging

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Program #/Poster #: P335.01

Topic: C.09.Stroke

Support: ORF-RE 04-047

Title: Measures of directional and general visuospatial impairments following stroke using a centre-out reaching task

Authors: *K. PARK[1], S. P. DUKELOW[2], S. SCOTT[1];
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Abstract: Visuospatial neglect is a disorder characterized by an impairment of spatial awareness, most commonly to the contralesional side of space in individuals with stroke. Reaching provides a useful framework to quantify one’s ability to generate motor actions in different spatial directions. Previous studies suggest that individuals with visuospatial neglect

can exhibit delayed reaction times when initiating reaches towards their contralesional side of space. However, no studies have explored the breadth of neglect beyond reaching left or right. In this study, we use an 8 target centre-out visual-guided reaching task on the Kinarm Exoskeleton lab to quantify the ability of individuals with stroke to reach in different spatial directions using their affected and unaffected arms. We assessed 162 left-affected and 103 right-affected individuals following sub-acute stroke and 204 healthy control participants. We developed metrics to quantify reaction time across spatial directions: Asymmetry Score to quantify how reaction time varied across spatial directions, Asymmetry Direction to determine the direction with the slowest reaction time, and a General Score to quantify the fastest response across all directions. Impairments were defined as scores greater than 95th percentile of healthy controls. For the left-affected individuals with stroke, we found 34% were impaired for Asymmetry Score, and 32% were impaired for General Scores for the right unaffected arms. The direction with the slowest reaction time tended to be to the left and slightly away from the body. Interestingly, the direction with the slowest reaction time for the affected left arm tended to be to the left and slightly towards the body, suggesting a general rotation in reaction time impairments between left and right arms. Right affected individuals showed a similar pattern of impairments although the overall number of individuals impaired were slightly lower (left-affected arm: 24% were impaired for Asymmetry Score and 12% were impaired for General Scores). We also found that our metrics correlated with a commonly used measure of visuospatial neglect, the Behavioural Inattention Test (BIT). Most individuals that failed the BIT also were impaired for reaction time measures, whereas some individuals that passed the BIT (>129) were still identified as impaired for reaction time measures.

Disclosures: K. Park: None. **S.P. Dukelow:** None. **S. Scott:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Kinarm Cofounder.

Poster

P335. Stroke Imaging

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Program #/Poster #: P335.02

Topic: C.09.Stroke

Support: NRF-2020R1A2C3010304
NRF-2020R1C1C1011688

Title: Sharing of Large-scale Brain Network Between Cognitive Function and Motor Recovery in Stroke Patients

Authors: *J. LEE[1], H. KIM[2], J. KIM[2], W. CHANG[1], Y.-H. KIM[1];
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Abstract: Cognitive and motor functions are known to be linked with each other. The primary motor cortex is activated during motor imagery and movement observation in the absence of any movement, while the corticospinal excitability is altered at an initial intention to motor action. In stroke patients, preserved cognitive function is known to play a role in motor recovery, but there is not enough evidence. In this study, we investigated the relationship between cognitive function and motor recovery in terms of sharing a large-scale brain network. One hundred eight subacute ischemic stroke patients participated. All patients underwent resting-state functional MRI (rs-fMRI) and cognitive/motor function assessments at two weeks after stroke onset using the Fugl-Meyer assessment (FMA) and Korean version mini-mental state examination (K-MMSE). The FMA score was obtained again at three months after stroke onset to assess motor recovery. To construct a large-scale brain network using the rs-fMRI, cortical surface parcellation was performed using Gordon atlas whose parcellation includes 333 regions of interest in twelve resting-state networks. This process included brain extraction, segmentation, generation of white matter and pial surface, inflation of the surfaces to a sphere, and surface-based spherical registration. Within-networks consisting of connections within each resting-state network and between-networks consisting of connections between twelve resting-state networks were extracted in the brain network. These networks related to the initial K-MMSE score (cognitive network) and FMA upper extremity (FMA-UE) recovery score (motor recovery network) was investigated and compared. Strength of within-networks, such as the somato-motor, auditory, cingulo-opercular, salience, cingulo-parietal, and dorsal attention networks, showed a significant correlation with the FMA-UE recovery score. Strength of between-networks, such as somato-motor and fronto-parietal networks, auditory and cingulo-opercular networks, auditory and cingulo-parietal networks, cingulo-opercular and default networks, and dorsal attention and default networks, also demonstrated positive correlation with FMA-UE motor recovery. Interestingly, most of these motor recovery networks were shared by the cognitive network at an early stage of stroke. Particularly, all of the significant within-networks in the motor recovery network were shared by the cognitive network. These results demonstrated that large-scale brain networks shared between cognitive function and motor recovery give some clues of interaction between two functions in the recovery phase of stroke.

Disclosures: J. Lee: None. **H. Kim:** None. **J. Kim:** None. **W. Chang:** None. **Y. Kim:** None.

Poster

P335. Stroke Imaging

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Program #/Poster #: P335.03

Topic: C.09.Stroke

Support: TACC Computing Grant

Title: Automatic segmentation of stroke lesions on MR images for voxel-based lesion-symptom mapping

Authors: *K. VERMA[1], S. KUMAR[2];
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Abstract: Over the past century, clinical studies on patients with lesions have provided key insights into the relationship of brain areas to behavior. As a result of this approach, we have gained a better understanding of several functional areas in the brain. However, lesions are often focal and typically follow vascular territories, and involve concomitant brain regions. Due to this, these early studies could not precisely link specific brain regions to behavior. To overcome these limitations, voxel-based lesion-symptom mapping (VLSM) is used to evaluate the role of lesions in cognitive deficits. VLSM is an imaging analysis technique that establishes a relationship between precise lesion location and clinical deficits. It is a major challenge for VLSM to avoid false positives and maintain statistical power (missing true effects). This technique benefits from the directed analysis method by using pre-defined binarized lesions maps. To automate lesion segmentation on MR images, we trained 3D deep convolutional segmentation models with residual learning (3D U-Net). Based on the open-source dataset ATLAS (Anatomical Tracings of Lesions After Stroke), we developed our algorithm using T1-weighted MRIs and manually segmented lesions. A subset of the data (n=229) was standardized, intensity-normalized, and defaced before being used for testing and training the model. The volume of the scans is 197 x 233 x 189 mm^3, and the canonical voxel size is 1 mm^3. Lesion size in the dataset ranges from 10 mm^3 to 2.8×10^5 mm^3. Data was augmented with scaling, brightness adjustments, and elastic deformations to enhance generalization. We used a spatial overlap index - Dice similarity coefficient (DSC) to quantify the segmentation performance. The average DSC across the cross-validated 5-folds was 0.74 (0.61- 0.85; 95% bootstrapped CI). On the ATLAS dataset, our model demonstrated the effectiveness of automated volumetric segmentation of stroke lesions. This approach will be used together with VLSM in future studies to study how precise lesion location relates to clinical deficits caused by stroke lesions on MR images

Disclosures: K. Verma: None. **S. Kumar:** None.

Poster

P335. Stroke Imaging

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Program #/Poster #: P335.04

Topic: C.09.Stroke

Support: IK2RX002837

Title: Corticospinal tract diffusivity is increased within the lesion volume in chronic stroke

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Abstract: White matter anisotropy and diffusivity of the corticospinal tract (CST) exhibit broad change at multiple stages after stroke, but adaptation focal to the portion of the CST falling within the lesion volume (CST_{LV}) is less well understood. Here, we compared CST_{LV} to the entire CST segment originating in primary motor cortex and extending to brainstem ($CST_{M1\text{-}BS}$) inclusive of CST_{LV} on voxel-based (fractional, FA) and population-based (quantified, QA) metrics of anisotropy, and metrics of diffusivity (mean, MD; radial, RD). Stroke survivors (n=11, 64.6±6.1 years, 4 female) with longstanding hand impairment (107.6±73.4 months) underwent MRI using a Siemens Prisma 3T scanner (64-channel coil). T1 and T2 images were acquired for anatomical reference and lesion segmentation using FslTools. A Diffusion Spectrum Imaging scheme acquired a total of 259 diffusion directions (maximum b-value=4000 s/mm^2) via a single-shot, twice-refocused echo planar imaging sequence (~12 min scan time, 2 mm isotropic voxel, repetition/echo time 2490/99.2 ms, multiband factor 3, phase encoding in A>P and P>A, vertex to brainstem coverage). Data were processed using *dsi_studio* software. The CST and lesion were segmented by two raters, and the mean value of each anisotropy and diffusivity metric was computed from CST_{LV} and $CST_{M1\text{-}BS}$. Two-way analysis of variance was used to test for effects of side (contralateral, ipsilateral to the paretic hand) and segment (CST_{LV}, $CST_{M1\text{-}BS}$). FA ($F_{1,10}$=15.9, p=.003, η^2=.61) and QA ($F_{1,10}$=8.8, p=.014, η^2=.47) were reduced in both CST segments on the contralateral side, but there was no difference between segments (p=.43 and .38, respectively) nor a side by segment interaction (p=.06 and .62, respectively). Diffusivity metrics also showed asymmetries with higher MD ($F_{1,10}$=38.8, p=<.001, η^2=.80) and RD ($F_{1,10}$=25.9, p=<.001, η^2=.72) in both segments on the contralateral side. Side by segment interactions were also detected (MD: $F_{1,10}$=32.1, p<.001, η^2=.76; RD: $F_{1,10}$=7.8, p=.019, η^2=.44), indicating an increase restricted to the CST_{LV}. Our preliminary findings demonstrate asymmetries in CST anisotropy and diffusivity at the chronic stage of stroke. However, the portion of the tract falling within the lesion volume exhibits changes in diffusivity, but not anisotropy, relative to the entire CST contralateral to the paretic hand. A complex evolution of pathological processes at the chronic stage might account for less restricted movement of water molecules within the lesion despite maintaining microstructure that is homogenous with the entire CST.

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Poster

P335. Stroke Imaging

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P335.05

Topic: C.09.Stroke

Support: NIH R00HD091375

Title: Corticomuscular Coherence and Corticospinal Tract Injury Association During Early Stroke Recovery

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Abstract: Introduction Stroke heterogeneity has propelled the development of neuroimaging biomarkers to enhance stroke outcome prediction. Structural neuroimaging biomarkers, such as corticospinal tract (CST) injury, are well-established in the literature. Whereas, biomarkers from functional neuroimaging require additional development. Corticomuscular coherence (CMC) is a functional measure representing synchrony between brain and muscle signals recorded during simultaneous electroencephalography (EEG) and electromyography (EMG) collection. Given the role of CST mediating communication between brain and muscle, this study determined associations between CMC and CST injury in individuals early post-stroke.**Methods** Participants with hemiparetic stroke hospitalized in an inpatient rehabilitation facility (IRF) completed a behavioral battery (Upper Extremity Fugl-Meyer, UEFM; Action Research Arm Test, ARAT), a diffusion MRI, and CMC testing using a 256-lead EEG system at around IRF admission. During CMC testing, muscle activity from bilateral biceps brachii, flexor and extensor digitorum (FD, ED), and first dorsal interossei (FDI) was recorded while participants performed a submaximal isometric grip task with visual feedback provided. CMC measurements involving leads overlying ipsilesional primary motor cortex (iM1) with the above muscles were computed across alpha-mu (11-14 Hz), low beta (13-19Hz), and high beta (20-30Hz) frequency bands. CST injury was determined by computing fractional anisotropy values at ipsi- and contralesional cerebral peduncles and calculating a laterality index.**Results** Eight individuals (3 females, 67±7.2 years, 11.7±4 days post-stroke) with moderate motor deficit (UEFM=47.9±19.8; ARAT=37.1±18.2) resulting from ischemic or hemorrhagic stroke completed all study procedures. Decreased CST injury (higher laterality index values) correlated with greater CMC between leads overlying iM1 and EMG leads over affected biceps in alpha-mu ($r=0.80$, $p=0.05$) and high beta ($r=0.80$, $p=0.006$) frequency bands and also between leads overlying iM1 and EMG leads over affected FDI in the high beta ($r=0.68$, $p=0.03$) frequency band. **Conclusion** Preliminary findings suggest relevant structural and functional connectivity associations, supporting the notion that the CST facilitates communication between central and peripheral nervous systems as captured by CMC. Validation of these findings in a larger sample and determination of longitudinal associations between CMC and CST injury are important next steps.

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Poster

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Title: Preliminary findings regarding a link between atypical corticospinal tract development, as determined with diffusion weighted MRI, and hand impairments in early-onset pediatric hemiplegia

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Abstract: Introduction: Early-onset pediatric hemiplegia (PH) is caused by a unilateral brain injury occurring before or during birth. PH severely limits the functional ability of one side of the body, causing reduced strength in the paretic hand (weakness) and involuntary movements in the paretic hand during use of the non-paretic hand (mirror movements). PH often involves atypical corticospinal tract (CST) development due to a combination of damage to the lesioned hemisphere and lack of pruning of atypical ipsilateral projections from the contralesional hemisphere, which we investigated using diffusion tensor MRI (DTI) and tractography analyses of the CST.
Methods: DTI data were collected and processed with steps of brain extraction, denoising, motion and eddy current correction, and tensor fitting to generate maps of diffusivity metrics. Anatomical landmarks were used to guide probabilistic tractography of the CST for analysis in both the lesioned and non-lesioned hemispheres. To quantify grasp weakness and mirroring severity, participants completed a bilateral assessment of grip strength using handheld force measurement devices and custom MATLAB data acquisition software. Spearman correlation analyses were performed between diffusivity metrics and hand impairment measures with age and sex as covariates.
Results: Preliminary results include five participants with early-onset PH (2M, 16±7.8 years) and six controls (2M, 12±3.5 years). For the lesioned hemisphere CST, there were significant positive correlations between grasp weakness severity and mean diffusivity (MD) (ρ=0.66, p=0.038) and between grasp weakness severity and axial diffusivity (AD) (ρ=0.68, p=0.030). For the non-lesioned hemisphere CST, there was a significant positive correlation between mirroring

severity and radial diffusivity (RD) (ρ=0.70, p=0.023). No significant correlations with fractional anisotropy (FA) were found in these preliminary analyses.
Conclusions: For the paretic hand weakness impairment, the increased diffusivity (MD and AD) in the lesioned hemisphere CST may be sensitive to decreased white matter tract density or myelination. For the mirror movement impairment, the increased diffusivity (RD) in the contra-lesioned hemisphere CST may indicate that unpruned, disordered cellular structure is sensitive to the inability for independent hand function. While these relationships between CST diffusivity properties and hand function in PH are preliminary, they provide the first steps to better understand underlying neural mechanisms for motor impairments in early onset PH.

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Poster

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Topic: C.09.Stroke

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Title: Skull-meninges connections: a new getaway to diagnose and treat neurological diseases

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Abstract: Neurological diseases affect about one in six people in the developed countries and can have profound and even debilitating impact. They have become a major socio-economic burden and costing trillion Euros of treatment per year. Given the complexity of the nervous system, understanding of the disease would require rather holistic biological approach and analysis of tissues as whole. Using whole-body vDISCO clearing technique (Cai⋯Ertürk, Nature Neuroscience, 2018), we can reliably image and quantify cellular even subcellular structures in intact mice. Imaging intact mouse heads, we recently discovered short vascular channels connecting the skull's bone marrow to the meningeal surface of the brain. These channels, named skull-meninges connection (SMC), were filled with CSF tracer, unraveling the

accessibility of the skull's bone marrow, which is filled with effective immune cells. A cerebral ischemic stroke in mice, mobilized LysM+ immune cells from skull into SMC and then into meninges. Subsequently, these immune cells invaded the brain parenchyma. These findings suggest that the skull is more than a physical barrier and is a dynamic site that responds to brain injury. The close proximity and accessibility of skull bone marrow's immune cell reservoir to the brain surface suggest that the skull might be involved in diverse brain functions in addition to making it an attractive target site for either diagnostics or therapeutic approaches.

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Poster

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Program #/Poster #: P335.08

Topic: C.09.Stroke

Title: Probabilistic anatomically-constrained tractography enhances correlation between structurofunctional resting-state networks and motor function in chronic stroke

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Abstract: Post-stroke, crucial anatomical and functional brain networks involved in creating and refining movements may be destroyed or altered. Surviving anatomical connections and functional connectivity (FC) are vital for functional motor and cognitive recovery. Combining diffusion weighted magnetic resonance imaging and functional Magnetic Resonant Imaging (fMRI), presence and strength of brain networks can be extracted. Structural connectivity provides insight into structural integrity of white matter (WM) connections between brain regions. FC alludes to temporal correlations of fluctuations in blood-oxygen-level-dependent (BOLD) signals between voxels or brain regions. Structurofunctional connectivity (SFC) combines the techniques to provide insight of the effects of anatomical reconfigurations and loss of brain network hubs on functional recovery. Ten stroke participants with age matched controls underwent T1-weighted (T1), resting-state fMRI, and high angular resolution diffusion imaging scans. Via Mrtrix3, performing spherical deconvolution on fiber-orientation distributions generated a probabilistic, anatomically-constrained tractography (PACT). Allowing complex modeling of WM tracts—enabling a more accurate SFC from subcortical structures compared to

previous deterministic methods. PACT SFC assigned each voxel a scaled BOLD time series from the sum of its structural connections weighted by the mean fiber length of all connections projected to connected voxels. The voxel's original time series was correlated with the connected time series to derive the structurofunctional coefficient. Voxel-wise application provided a metric of probabilistic intrinsic SFC (piSFC). Also, Probabilistic network SFC (pnSFC) was calculated to identify networks of interest from resting-state FC. piSFC and pnSFC maps were compared between groups via voxel-based Student's T-test with multiple comparisons correction. Linear regression was performed between SFC metrics and clinical measures of lower extremity motor function. Significant decreases ($p<0.01$) in piSFC were found in the stroke group within key functional hubs of sensory and motor areas. pnSFC showed strong connectedness amongst prefrontal regions and cerebellum in controls, and decreases in the stroke group, supporting previous findings indicating a decrease in their FC. pnSFC also showed higher connectedness in the contralesional hemisphere of lower-function stroke participants.The results illustrated changes in piSFC and pnSFC post-stroke correlated with motor function, highlighting importance of anatomical effects on functional recovery within brain networks.

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Poster

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Title: Lesion location and lesion-associated networks predict post-stroke outcomes

Authors: *M. BOWREN, Jr.[1], J. E. BRUSS[1], K. MANZEL[1], D. EDWARDS[2], C. LIU[3], M. CORBETTA[4], D. TRANEL[1], A. D. BOES[1];
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Abstract: Being able to make accurate empirical predictions about chronic post-stroke cognitive and motor outcomes could lead to improvements in the healthcare of patients with stroke.

Lesion-behavior mapping (LBM), and structural (sLNM) and functional (fLNM) lesion-network mapping are statistical techniques that link complex functions (e.g., cognitive and motor abilities) to brain regions and networks. Few studies have evaluated the potential of such techniques for predicting stroke outcomes. Here, using data from two large cohorts of individuals with focal brain lesions, we evaluated whether LBM, sLNM, and fLNM can be used to predict cognitive and motor outcomes in a third cohort of patients with stroke, and whether sLNM and fLNM add predictive value beyond LBM. We used data from the Iowa Neurological Patient Registry and the NICHE clinical trial to calculate LBMs for expressive language (n = 437), receptive language (n = 210), verbal memory (n = 477), and motor function (n = 103). We used the local maxima from the LBMs as seed regions-of-interest for sLNM and fLNM analyses based on structural and functional connectivity data from the Human Connectome Project. Using latent growth curve modeling, we evaluated the amount of variance in outcomes and recovery rates predicted by each map in a separate cohort of stroke patients who also completed tests of expressive language (n = 135), receptive language (n = 135), verbal memory (n = 103), and motor function (n = 133). We compared the predictive utility of each map using hierarchical modeling and model fit indices. We found that LBM, sLNM, and fLNM predicted a statistically significant amount of variance in cognitive and motor outcomes. fLNM and sLNM predicted variance in outcomes and/or recovery rates beyond LBM. Though the best predictor varied by domain, the combination of LBM, sLNM, and fLNM consistently performed the best. Our results support the conclusion that LBM, sLNM, and fLNM can be used to predict post-stroke outcomes, and that lesion location, and structural and functional brain networks, capture distinct information. Our work provides some of the largest LBM and LNM analyses of cognitive abilities to date, and represents a step toward the clinical translation of tools from cognitive neuroscience.

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Poster

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Title: Multimodal in vivo imaging reveals differences in post-stroke recovery in Tlr2-deficient mice

Authors: *S. GAJOVIC, P. JOSIć, S. SRAKOčIć, R. ISTER, A. GLASNOVIć, S. ŠKOKIć, M. DOBRIVOJEVIć RADMILOVIć;
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Abstract: Inflammatory response after ischemic brain injury is mediated by Toll-like receptors (TLR) located on the microglia. Tlr2-deficient mice represent a model of the modified neuroinflammation, which is hypothesized to result in the modified post-stroke recovery. The aim of the current study was to characterize the dynamic of post-stroke events by performing a longitudinal *in vivo* multimodal magnetic resonance imaging and bioluminescence imaging of the ischemic injury in Tlr2-deficient mice. The consequences of the transient middle cerebral artery occlusion (MCAO) were compared between Tlr2-deficient and C57Bl/6 (WT) mice. The animals were imaged at multiple time points by a 7T BioSpec 70/20 USR MRI system and with optical imager Perkin Elmer IVIS Spectrum. Gap43-luc/gfp bioluminescence was imaged to measure neurorepair and caged luciferase VivoGlo (Promega) was imaged to reveal caspase 3 and 7 activity and measure apoptosis. The functional consequences were assessed by behavioral testing and neurological scoring. At day 28 after the ischemia the animals were sacrificed, and the brains were processed for histological analysis. The volume of the ischemic lesion measured by MRI did not differ between Tlr2 and WT animals. However, the functional outcomes were different, and the Tlr2-deficient animals showed higher neurological deficit than the WT mice. Using cumulative linear mixed-modeling (CLMM) as a method of ordinal regression, taking neurological score as a dependent variable, we have managed to explain 59% of variance by including relative stroke volume in the model. Both the stroke size and the experimental group have shown to significantly determine neurological score, with statistically significant differences on both variables. In conclusion, longitudinal multimodal imaging combined with the functional outcome assessment revealed clear differences between Tlr2-deficient and WT animals.

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Poster

P372. Spinal Cord Injury: Training, Rehabilitation, and Repair IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P372.01

Topic: C.11. Spinal Cord Injury and Plasticity

Support: DVA RR&D grant RX002316
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Rebecca F. Hammond Endowed Chair

Title: Recovery of Yield Adaptation In Early Stance Mediated by Downslope Training post-SCI

Authors: *L. MONTGOMERY[1,5,2], R. L. VAN SANDT[1,2,6], A. N. TRELL[5,3], S. F. MCMURTRY[7], A. M. DE BOEF[9], T. NICHOLS[8], D. R. HOWLAND[1,2,3,5,4];
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Abstract: Locomotion requires controlled knee and ankle flexion in early stance as the hindlimb yields with weight acceptance. Yield is controlled by eccentric activity in knee and ankle extensors, ensuring appropriate limb compliance for efficient and stable gait. This eccentric activity is regulated, in part, by Golgi tendon organs (GTOs) and extensive force feedback circuitry. We have shown that after a T9 or 10 lateral hemisection (lHx) in the adult cat, inhibitory force feedback (iFFb) is disrupted, causing exaggerated and biased inhibition of ankle extensors (Niazi et al. 2020). After lHx, animals also have greater ankle flexion at yield (Montgomery et al. 2019) and clinically, changes in yield are observed after spinal cord injury (SCI). If iFFb disruptions lead to deficits in yield then tasks that enhance iFFb, such as downslope walking (Nichols et al. 2014), may facilitate improvements in weight support. We incorporated downslope walking into a rehabilitation program to identify if this task could improve yield after lHx. Changes in iFFb after lHx and training are being investigated by collaborators at Georgia Tech. A companion poster (McMurtry et al. 2021) details changes in iFFb following this training paradigm.

Animals were trained to voluntarily cross an overground (OG) runway and walk bipedally on a treadmill (TM) set to various declines from 0 (level) to -26° (steep). Baseline data was collected for each animal on all tasks. Two weeks following lHx, animals were randomly assigned to one of three groups - non-trained (NT), eccentric trained (ETr), and level trained (LTr). ETr and LTr animals received training 5x a week for 12 weeks. ETr animals trained on TM and OG on various declines, LTr animals trained only on level TM and OG.

Spinally intact animals adapted to steep declines on OG by increasing knee and ankle yield and increasing ankle excursion on -26°. They however, do not show a significant change in yield in response to declines on TM. After lHx, animals displayed exaggerated yield and lost the ability to adapt to declines. Following training, ETr and LTr animals also exhibited greater yield after lHx but in contrast to the NT group, ETr animals retained their ability to adapt to declines.

This study shows that downslope walking allows the nervous system to effectively adapt yield responses to challenging environments. As deficits in yield continue despite current rehabilitative strategies, incorporating downslope walking into rehabilitation programs presents an effective modality to facilitate the recovery of yield.

This presentation does not represent the views of the DVA, NIH, US gov.

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Poster

P372. Spinal Cord Injury: Training, Rehabilitation, and Repair IV

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Program #/Poster #: P372.02

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH R01 NS097781
Rebecca F. Hammond Endowment for Spinal Cord Injury Research

Title: Evidence for reticular and vestibular regulation of strength and distribution of short latency force feedback

Authors: *A. DE BOEF[1], S. MCMURTRY[1], L. R. MONTGOMERY[2,4], D. R. HOWLAND[2,4], A. TRILL[3], R. VAN SANDT[5], R. NICHOLS[1];
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Abstract: Coordinated movement results from feedforward motor signals and sensory feedback. One important source of sensory information is the population of Golgi tendon organs (GTO). GTO's distribute inhibitory force information to muscles that cross different joints and axes of rotation. This system, along with length feedback from muscle spindles, is thought to regulate the mechanical properties of the entire limb. While the strength and distribution of force related inhibition has been observed to vary, the source of its regulation has not been identified. Changes in the organization of these networks and the concomitant behavioral affects have been assessed following selective lesions to the spinal cord to interrupt different supraspinal tracts as part of a collaborative study based out of Georgia Institute of Technology and the University of Louisville.We investigated the intermuscular distribution of force related inhibition in decerebrate animals by recording muscular forces in response to differing stretch combinations. Previous work has shown three main patterns of intermuscular force feedback (Lyle and Nichols(2018) J Neurophysiol 119:668). Following T10 lateral spinal hemisection, in which both dorsal and ventral spinal quadrants on one side are interrupted, only a single pattern of feedback is observed (Niazi et al., 2020, J Neurosci Res, 98: 1646) in which the inhibition onto the ankle extensor muscles is amplified (convergent pattern). This pattern is observed bilaterally.Animals with dorsal quadrant lesions show disruption of the iFFB system primarily on the ipsilesional side, while ventral quadrant lesions lead to bilateral disruption of the iFFB system as is observed following lateral hemisection. The combined results suggest that not only are ventral neural pathways (vestibulospinal and reticulospinal) potential sources for regulation of the distribution

of force feedback. The disruption of interneuronal processing due to the grey matter component of the lesion as well as the medullary aspect of the reticulospinal tract may lead to the convergent pattern of the force feedback network in the case of the dorsal quadrant lesions.

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Poster

P372. Spinal Cord Injury: Training, Rehabilitation, and Repair IV

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Topic: C.11. Spinal Cord Injury and Plasticity

Support: Spt DVA RR&D RX002316
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Rebecca F. Hammond Endowed Chair

Title: Downslope training induces plasticity in the force feedback system following incomplete spinal cord injury

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Abstract: Sensory feedback is imperative for smoothly coordinated movements. Spinal cord injuries (SCI) disrupt motor commands descending from the brain, as well as cause malfunctions in spinal circuitry. One component of sensory feedback, which arises from Golgi Tendon Organs (GTOs), encodes changes in force within the muscles. This feedback creates a network that is widely distributed across the feline hindlimb to regulate whole limb stiffness through inhibitory actions on motoneurons. We evaluate the structure of this network by recording forces from individual muscles in response to combinations of stretches delivered to muscle pairs in decerebrate animals. Spinally intact decerebrate felines may show 1 of 3 patterns of feedback. Following lateral hemisection at the low thoracic level, we have shown loss of the flexibility between patterns and expression of a single, strongly convergent pattern of inhibition focused on the ankle musculature (Niazi et al, 2020; Kajtaz 2019). The loss of flexible patterns and subsequent targeted rehabilitation are being investigated in a cross-collaboration at Georgia Tech as part of a larger study at the University of Louisville looking at the effects of SCI and training on gait, as well as associated structural changes in the spinal cord. A companion poster (Montgomery et al, 2021) addresses concomitant changes under various gait conditions for the

same groups of animals. Based on our previous findings that downslope walking modulates force feedback, we had provided preliminary evidence that training animals with lateral hemisection on downslope walking re-establishes the multiple patterns of force feedback observed in controls (McMurtry et al, 2019). Current work further strengthens this conclusion. Further, animals with lateral hemisections who had undergone training on level ground walking did not show the beneficial effects observed with downslope training. Using our expanded database, we now show that thoracic lateral hemisections produce bilateral effects on the FFB system, which are more pronounced ipsilesionally. Finally, the impact of downslope training initiated at two weeks post-injury is apparent at both 7 and 13 weeks post-SCI, indicating a stable response to this rehabilitation strategy.
This presentation does not represent the views of the DVA, NIH, US gov.

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Poster

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P372.04

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NINDS R01 NS097880
Craig H. Neilsen Foundation #457508

Title: Nociception impedes grasping recovery in the spinal cord injured rat.

Authors: *J. WALKER, T. KIM, S. GISZTER, M. R. DETLOFF;
Neurobio. & Anat., Drexel. Univ., Philadelphia, PA

Abstract: Significant deficits in motor control and sensory function diminish an individual's quality of life following spinal cord injury (SCI). A top priority of the injured population is regaining upper limb function. It has been shown that descending motor commands and primary afferent input, such as cutaneous feedback and proprioception, drive plasticity and recovery of function after SCI. Our lab and others have shown that primary nociceptors sprout and facilitate the maladaptive plasticity seen in chronic pain development, but their role in motor control is critically overlooked. The purpose of this experiment was to better understand the role of nociceptive input on recovery of motor function after SCI. Mechanosensitive, non-peptidergic nociceptors were ablated in Sprague-Dawley rats via intraganglionic injections of rIB4 - conjugated saporin or unconjugated (vehicle) saporin into the C7-8 dorsal root ganglia (DRGs). During the same surgery, rats received an ipsilateral C5 hemicontusion and the implantation of a braided multi-electrode probe for recording in the ipsilateral C8 gray matter. The von Frey test

for allodynia and immunohistochemistry, in which cervical DRG and spinal cord sections were stained with antibodies against CGRP and isolectin-B4, were performed to confirm nociceptor ablation. The following battery of food motivated behavioral tests were performed to assess the components of the reach and grasp: single pellet retrieval, isometric pull, paw preference, Montoya staircase, and the cereal manipulation task (IBB). Results indicate that ablation of nociceptors following SCI improved the rats' ability to grasp chocolate food pellets when compared to vehicle controls. Intraspinal recordings of local field potentials in the dorsal horn, intermediate gray, and ventral horn were obtained to measure changes in intraspinal activity in awake behaving rats. Multivariate statistical analyses of the behavioral, anatomical, and electrophysiological outcomes are underway. Increasing our understanding of the role nociceptors play in the spinal plasticity related to motor control following SCI will help guide future research and development of rehabilitative techniques.

Disclosures: J. Walker: None. **T. Kim:** None. **S. Giszter:** None. **M.R. Detloff:** None.

Poster

P372. Spinal Cord Injury: Training, Rehabilitation, and Repair IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P372.05

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH Common Fund DP2 NS106663
NYS DoH C33610GG

Title: Rehabilitation dependent remodeling of intracortical networks during recovery from spinal cord injury

Authors: *H. MOHAMMED, E. R. HOLLIS, II;
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Abstract: Motor cortex in rodents undergoes reorganization after spinal cord injury. Spared motor representations expand into the deafferented motor representations depending on the level of spinal lesion. Following C5 dorsal column lesions in mice, the forelimb muscle representations expands into the deafferented hindlimb motor representation. Rehabilitation regains the balance of forelimb flexor and extensor muscle representations previously demonstrated by optogenetic motor mapping. In this study, we focused on the intracortical circuit mechanisms mediating recovery of motor function with rehabilitation. Intracortical hindlimb to forelimb axons show bouton remodeling in CFA in mice receiving rehabilitation in a skilled reach task, demonstrated by in vivo 2 photon imaging. Optogenetically silencing the forelimb circuit and intracortical hindlimb to forelimb projections during recovery reduced the

success rate of skilled reach task in mice. Our results suggests a role for remodeling of intracortical networks in recovery from spinal cord injury with rehabilitation.

Disclosures: H. Mohammed: None. **E.R. Hollis:** None.

Poster

P372. Spinal Cord Injury: Training, Rehabilitation, and Repair IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P372.06

Topic: C.11. Spinal Cord Injury and Plasticity

Support: U01-EB029374
R01-NS104291
Lisa Dean Moseley Foundation
F32-NS119348

Title: Engineering human neural precursors to repair the injured cervical spinal cord

Authors: *L. V. ZHOLUDEVA[1], M. A. LANE[2], T. C. MCDEVITT[1];
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Abstract: Emerging technologies, such as cell-based repair strategies, offer new promise for some of the most devastating medical conditions that currently lack treatments. However, to harness the full therapeutic potential of stem cells, it will be necessary to understand how to direct their differentiation to appropriate cell phenotypes and ensure that their phenotype and function persist after transplantation into a pathologic environment. Using pre-clinical spinal cord injury (SCI) as a testbed, we tested the therapeutic potential of transplanted human induced pluripotent stem cell (hiPSC)-derived spinal interneurons (SpINs). Neuroanatomical tracing and immunohistochemistry were used to assess transplant integration and connectivity with injured host tissue. Optogenetic activation of hiPSC-SpINs was used to assess development of synaptic connectivity to injured host circuits with time (1-, 2- and 3-months post-transplantation). Bilateral terminal diaphragm electromyography was used to assess functional contribution of transplanted human SpINs to the recovery of phrenic function with time (1-, 2- and 3-months post-transplantation). These studies demonstrated that transplanted human SpINs survived and integrated with injured cervical spinal cord circuits, displayed anatomical (e.g., pre- and post-synaptic proteins) and functional (intratransplant and intraspinal cord recordings) evidence of connectivity. Ongoing work is focused on silencing transplanted hiPSC-SpINs to further elucidate mechanisms by which transplanted cells may contribute to motor recovery post-SCI.

Disclosures: L.V. Zholudeva: None. **M.A. Lane:** None. **T.C. McDevitt:** None.

Poster

P372. Spinal Cord Injury: Training, Rehabilitation, and Repair IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P372.07

Topic: C.11. Spinal Cord Injury and Plasticity

Support: JSPS KAKENHI 19K23773
JSPS KAKENHI 21K07290

Title: The patterns of axon rewiring of multiple corticospinal tract pathways after stroke in different cortical areas

Authors: *T. SATO, Y. NAKAMURA, M. UENO;
Brain Res. Institute, Niigata Univ., Niigata, Japan

Abstract: Brain injuries such as stroke and trauma often lead to deficits in sensorimotor functions. Previous studies have demonstrated that reorganization of neural network in the spared brain region is one of the key processes to recover functions. The corticospinal tract (CST) that controls voluntary movements is one of the essential neural pathways for functional recovery. We and others previously showed that the CST axons have different trajectories to the spinal cord from multiple cerebral origins such as the motor cortex and the somatosensory cortex, and each has distinct functions during the movements in rodents. However, the patterns of reorganization of those multiple CST pathways are not comprehensively understood across different types of lesion in size and location. In this study, we systematically examined the rewiring patterns of CST pathways of multiple cerebral origins in a mouse stroke model. We first established four different stroke models that vary in size and location in the sensorimotor cortex, and then injected with an anterograde tracer into the ipsi- or contralesional area of the sensorimotor cortex to investigate the projection patterns of remnant CST fibers. We find that the spared contralesional motor and sensory CST axons cross the midline and sprout into the denervated side of cervical cord after stroke with a large cortical region. In contrast, the contralesional CST fibers do not sprout in a small stroke, whereas the ipsilesional axons from the spared motor area grow in the denervated side. Furthermore, we demonstrate that motor and sensory CST axons do not compensate the projections each other when either one is injured. In conclusion, the present study reveals that brain injuries spontaneously induce characteristic patterns of CST rewiring depending on the types of CST, lesion size and location, showing the neural principles in which the rewiring is predominantly induced in functionally homologous and ipsilesional pathways. Our data indicate the importance to target different neural substrates to restore functions among the types of injury.

Disclosures: T. Sato: None. **Y. Nakamura:** None. **M. Ueno:** None.

Poster

P372. Spinal Cord Injury: Training, Rehabilitation, and Repair IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P372.08

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH Grant R01 NS097781
Rebecca F. Hammond Endowed Chair

Title: Overground gait, but not hindlimb stepping on a treadmill, recovers in animals with clasp knife responses following dorsal hemisection

Authors: *R. L. VAN SANDT[1], A. DE BOEF[2], L. R. MONTGOMERY[3], A. N. TRELL[4], S. MCMURTRY[2], T. R. NICHOLS[2], D. R. HOWLAND[3];
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Abstract: A major obstacle in neurorehabilitation is providing training for patients with inadequate lower extremity strength. Understanding control of muscle activation during gait is imperative for rehabilitation program design. This collaborative study used subjects with thoracic dorsal spinal hemisection (dHx) to assess post-injury changes in quadrupedal and bipedal locomotion as well as muscle stretch and force, which play major roles in regulating limb stiffness. Purpose bred animals (n=5) were conditioned to perform gait tasks during level surface walking on an overground (OG) runway and motorized treadmill (TM). Data were captured pre- and post-dHx with a Vicon Motion Analyses system. Independent voluntarily gait was assessed 2- and 6-weeks (wks) post-dHx. Terminal experiments were conducted at 7 wks to investigate intermuscular distribution of force related inhibition. Animals underwent decerebration, were fixed in a standing position, and muscular forces were recorded in response to combinations of stretches of different muscles acting on the knee and ankle. Histological analyses confirmed lesion level and extent. By 2 wks post-dHx, all subjects recovered OG locomotion. Performances were characterized by full weight supported steps with consistent delays in transition from stance to swing relative to pre-dHx data. This delay was the result of an increase in the stance phase duration along with delayed ankle extension and increased late stance hip extension. In contrast, only 1 of 5 subjects could perform bipedal walking on the TM at 2 wks. This did not improve by 6-wks post-dHx. On the TM, 4 of 5 subjects assumed a posture in which both hindlimbs were moderately abducted, extending symmetrically behind the pelvis. These extended limbs full weight supported but did not step at either walking speed (0.5 and 0.8 meters/sec) tested. In these same subjects, muscles stretched during evaluation of force feedback inhibition exhibited clasp-

knife inhibition. Hindlimb sensory input alone was insufficient to promote bipedal stepping in subjects with dHx. Clasp-knife inhibition has a profound inhibitory effect on the muscles in the hindlimb, which likely causes sensory feedback to fail at initiating locomotion on the TM. However, the addition of descending modulation is sufficient to produce voluntary OG walking. Understanding why there are striking differences in muscle activation and limb stiffness between more voluntary (overground) versus more spinally-mediated (TM) walking and standing will be beneficial for the improvement of rehabilitation programs for patients with incomplete spinal cord injury. The abstract does not represent NIH, VHA or NIH views.

Disclosures: R.L. Van Sandt: None.

Poster

P380. Pain: Nociceptors: Cell and Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P380.01

Topic: D.03. Somatosensation – Pain

Support: R01NS109936

Title: The mitochondrial uncoupling agent dinitrophenol decreases nociceptor excitability and inflammatory hyperalgesia

Authors: *D. C. MOLLIVER[1], R. GEGUCHADZE[1], B. M. DAVIS[2], C. STRAUB[3];
[1]Biomed. Sci., Univ. of New England, Biddeford, ME; [2]Dept. of Neurobio., Univ. of Pittsburgh, Pittsburgh, PA; [3]Biomed. Sci., Univ. of New England, Col. of Osteo., Biddeford, ME

Abstract: 2,4-dinitrophenol (DNP) is an uncoupler of the mitochondrial proton gradient used in the 1920s as an effective diet drug, with a high incidence of morbidity and mortality. Recently there has been interest in potential neuroprotective effects of DNP at low doses. Given evidence that mitochondrial dynamics impact nociceptor responsiveness, we tested the impact of DNP on nociceptor response properties and behavioral thresholds to noxious stimuli. To examine actions of DNP on acute hyperalgesia, the prostaglandin PGE2 was injected into the hindpaw of mice injected 30 min prior with 5mg/kg DNP or vehicle, i.p. PGE2 induced heat hyperalgesia (Hargreaves assay) that resolved within 3 hours. DNP reduced heat hyperalgesia in male but not female mice, with no change in baseline thresholds. Next, hindpaw injection of 1% carrageenan (CAG) was used as a model of persistent inflammatory hyperalgesia. DNP or vehicle was injected daily (i.p., 5mg/kg) in males, and mice received a hindpaw injection of 1% carrageenan on day 3. Withdrawal thresholds for heat and mechanical (von Frey) stimuli were tested daily. In vehicle-treated mice, CAG caused heat and mechanical hyperalgesia that resolved within 7 days. In contrast, DNP administration reduced both heat and mechanical hypersensitivity. Despite the impact of DNP on hyperalgesia, the dose was not sufficient to reduce body weight. To determine

whether the anti-hyperalgesic effects of DNP could be mediated by a direct action on sensory neurons, DNP was applied to dissociated dorsal root ganglion (DRG) neurons. l00 micromolar DNP caused a dramatic increase in mitochondrial respiration and reduction in mitochondrial ATP production, as measured by oxygen consumption rate (OCR) in the Seahorse mitochondrial stress test, and completely occluded the effect of a maximal dose of the uncoupler FCCP, indicating collapse of the mitochondrial membrane potential (MMP). In contrast, 20 micromolar DNP increased OCR, but cells remained responsive to FCCP, indicating partial depolarization of MMP. Subsequently, patch clamp recordings were performed in isolated neurons using 20 micromolar DNP. DNP (10 min) resulted in hyperpolarization and increased membrane permeability, together reducing excitability and increasing action potential threshold. This effect was observed in a subset of neurons and was not activity dependent. Together, the results indicate that DNP acutely decreases sensory neuron excitability in vitro and exerts anti-hyperalgesic effects in models of acute (PGE2) and persistent (CAG) inflammatory pain. Experiments are underway to identify molecular mechanisms that mediate the effects of DNP on nociceptor excitability.

Disclosures: D.C. Molliver: None. **R. Geguchadze:** None. **B.M. Davis:** None. **C. Straub:** None.

Poster

P380. Pain: Nociceptors: Cell and Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P380.02

Topic: D.03. Somatosensation – Pain

Title: Single neuron profiling using GeoMx Digital Spatial Profiler platform for transcriptomic analysis of human Dorsal Root Ganglia

Authors: J. BROUGHER[1], L. PAN[2], S. SHIERS[3], D. TAVARES FERREIRA[3], S.-L. J. PHELAN[2], Y. LIANG[2], S. WARREN[2], V. KUMAR[1], ***C. PAIGE**[1], T. J. PRICE[3]; [1]Doloromics, Inc., Foster City, CA; [2]Nanostring, Seattle, WA; [3]Sch. of Behavioral and Brain Sci., Univ. of Texas At Dallas, Richardson, TX

Abstract: Novel drug targets are desperately needed for the treatment of chronic pain, and RNA sequencing provides unparalleled insight into transcriptomic changes underlying the development of chronic pain in patient populations. However, bulk RNA sequencing lacks cell-type specific information, and most methods of single cell sequencing for human DRG rely on cell dissociation and cell sorting, which may distort transcriptomes. Here, we used the Whole Transcriptome Atlas (WTA) on GeoMx® Digital Spatial Profiler (GeoMx DSP) platform to quantify RNA from single human Dorsal Root Ganglia (DRG) neurons. The GeoMx DSP platform uses fluorescence-based region of interest (ROI) imaging to select specific cell-types

and populations for downstream molecular profiling. Fresh frozen preparations of 6 human DRG were sectioned to 8µm, processed with standard antigen retrieval protocols, and incubated with the WTA probes and an anti-peripherin antibody to visualize the neuronal cell bodies. Fifty-nine single cell ROIs that were positive for peripherin immunostaining were selected for transcriptional profiling, and serially exposed to UV light, which releases an indexing oligo from the WTA probes. Oligos were quantitated on an Illumina Novaseq and data was analyzed on GeoMx DSP Control Center. Following initial QC, 42 single cell ROIs were selected for downstream analysis, and the remaining ROIs were excluded due to low RNA content based on the data of over 18,000 genes' expression, We were able to identify neurons from 12 neuronal populations in the human dorsal root ganglia (DRG) previously identified through Visium spatial sequencing. Additionally, differentially expressed neuronal genes were conserved when compared to previously published work. Following this initial quality control analysis, data was analyzed using a previously published interactome algorithm where neuronal and fibroblast ligand-receptor interactions were identified in patients with fibromyalgia. We have validated the use of GeoMx DSP as a platform suitable for sequencing RNA from both single cells and clusters of cells in the human DRG.

Disclosures: **J. Brougher:** A. Employment/Salary (full or part-time):; Doloromics, Inc.. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Nanostring. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Doloromics, Inc. **L. Pan:** A. Employment/Salary (full or part-time):; Nanostring. **S. Shiers:** None. **D. Tavares Ferreira:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Doloromics, Inc. **S.J. Phelan:** A. Employment/Salary (full or part-time):; Nanostring. **Y. Liang:** A. Employment/Salary (full or part-time):; Nanostring. **S. Warren:** A. Employment/Salary (full or part-time):; Nanostring. **V. Kumar:** A. Employment/Salary (full or part-time):; Doloromics, Inc. **C. Paige:** A. Employment/Salary (full or part-time):; Doloromics, Inc.. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Nanostring. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Doloromics, Inc. **T.J. Price:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Doloromics, Inc..

Poster

P380. Pain: Nociceptors: Cell and Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P380.03

Topic: D.03. Somatosensation – Pain

Support: Donation from Dr. John M. Davis

Title: Metabolites of Linoleic Acid, A Common Fatty Acid in the Western Diet, as Pain Mediators

Authors: *J. WHEELER[1], A. DOMENICHEILLO[2], G. KEYES[2], K. MAIDEN[2], J. DAVIS[3], C. RAMSDEN[2], S. MISHRA[1];
[1]North Carolina State Univ., Raleigh, NC; [2]Lipid Peroxidation Unit, National Institute on Aging, MD; [3]Psychiatry, Univ. of Illinois at Chicago, Chicago, IL

Abstract: Omega-6 (ω-6) metabolites are well established mediators of pain and inflammation. Pain mediators derived from n-6 arachidonic acid are well recognized; however, linoleic acid is the most abundant n-6 fatty acid in modern industrialized diets. Possibly due to this dietary prevalence, several oxidized linoleic acid metabolites (OxLAMs) have been mechanistically linked to pathological conditions ranging from cardiovascular disease to chronic pain. Rodents fed a diet enriched in linoleic acid to mimic the modern western diet had an increased concentration of OxLAMs in tissues associated with idiopathic pain syndromes. In clinical trials, decreasing the amount of dietary ω-6 fatty acids resulted in a decrease in the number of headache days per month. Further, several OxLAMs were found to have decreased concentrations in headache patients with this decreased ω-6 fatty acid diet. Here, we characterize the functional role of one of these mediators, 13-hydroxy-9,10-epoxyoctadecenoate, and its metabolite, 13, 9, 10-trihydroxyoctadecenoate. Using calcium imaging, we first determined that these two compounds are capable of activating sensory neurons. Next, we determined, using the subcutaneous cheek and intraplantar injection, that these OxLAMs induce pain behaviors and thermal hypersensitivity. Additionally, we used these in vitro and in vivo assays to determine the potentially active pharmacophore of these OxLAMs. Taken together, our results suggest a role in pain and hypersensitivy for these OxLAMs.

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Poster

P380. Pain: Nociceptors: Cell and Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P380.04

Topic: D.03. Somatosensation – Pain

Title: Single nucleus transcriptomic profiling of hiPSC populations

Authors: *J. BROUGHER[1], V. TRUONG[2], P. WALSH[3], A. HOVLAND[3], C. PAIGE[1], T. J. PRICE[4];

[1]Doloromics, Foster City, CA; [2]Anatomic Inc., Brooklyn Park, MN; [3]Anatomic Inc., Minneapolis, MN; [4]Sch. of Behavioral and Brain Sci., UTD, Richardson, TX

Abstract: Therapeutic development for the treatment of chronic pain requires access to functional screening platforms to ensure target validity. The pain therapeutics field has long relied on animal models, such as rodents, to test upcoming therapeutics; however, recent transcriptomic publications reveal key differences in gene expression between humans and rodents. Further complicating the situation is the difficulty of collecting living human sensory tissue. Here, we address this problem by profiling a scalable RealDRG™ human induced pluripotent stem cell (hiPSC)-derived nociceptor population that can meet the demands of high-throughput screening. Our approach utilized a single nucleus SPLiT-Seq approach to functionally cluster and characterize hiPSC-derived nociceptor populations. Our findings reveal the presence of multiple subpopulations that differentiate from the same hiPSC culture. Within these subpopulations, we have highlighted key genes that are also found within set human nociceptor populations. Together, this data shows that hiPSC-derived nociceptor populations are a transcriptomically similar screening platform to nociceptive populations found within the human dorsal root ganglion.

Disclosures: J. Brougher: A. Employment/Salary (full or part-time):; Full, Doloromics, Inc.. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Anatomics, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Doloromics, Inc. **V. Truong:** A. Employment/Salary (full or part-time):; Anatomi Corp. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Anatomi Corp. **P. Walsh:** A. Employment/Salary (full or part-time):; Anatomi Corp. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Anatomi Corp. **A. Hovland:** None. **C. Paige:** A. Employment/Salary (full or part-time):; Doloromics, Inc.. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Anatomics, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Doloromics, Inc. **T.J. Price:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Doloromics, Inc..

Poster

P380. Pain: Nociceptors: Cell and Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P380.05

Topic: D.03. Somatosensation – Pain

Support: Swedish Research Council
Uppsala University
The Brain Foundation

Title: Transcriptional analysis to study molecular diversity of sensory modality-specific neurons

Authors: *H. M. WEMAN, M. M. CEDER, J. E. T. JAKOBSSON, M. C. LAGERSTRÖM; Neurosci., Uppsala Univ., Uppsala, Sweden

Abstract: Pain and itch are unpleasant but unquestionably useful sensations, having evolved to induce withdrawal behavior to allow injuries to heal and to scratch off potentially harmful parasites. Unfortunately, both these sensations can develop into chronic conditions, causing the previous protective roles to become maladaptive. Presently, studies have used behavioral and histological approaches targeting specific neuronal populations and signaling pathways in attempts to find targets for novel treatment in different pain and itch models. The present study aims to detect modality-specific sensory neurons in the spinal cord, and will achieve this by identifying neurons activated by certain sensory stimuli using single-cell based and spatial transcriptional profiling. Firstly, mice are microinjected with a virus expressing a reporter gene under the control of the immediate early gene *fos* promotor. This allows detection and isolation of activated neurons (fos-EYFP) in multiple sensory-modality pathways for subsequent single cell mRNA sequencing (scRNA-seq). In the second approach, *in situ* sequencing (ISS), where the expression of multiple genes can be detected spatially in an intact tissue section, is used to study the molecular identity of *fos* expressing neurons. This study will examine which lumbar spinal cord dorsal neurons that are activated by noxious heat and further, how nerve growth factor b (NGFβ)-induced hyperalgesia affects the activity in terms of neuronal recruitment and gene expression. Histological analysis using the viral approach showed that thermally-induced fos-EYFP neurons could be detected ipsilaterally in the medial dorsal laminae of the L5/L6 segments following hind paw stimulation (91.2 ± 2.6 % of fos-EYFP located laminae I-IV and 7 ± 2.1 % in laminae V-X, n animals: 2 females & 1 male, 8 weeks old, n sections: 12) a pattern not visualized in the contralateral dorsal horn ($p < 0.0001$, two-tailed unpaired t-test). The activated cells were overlapping with NeuN (96.8 ± 0.3 %, n animals: 1 female & 1 male, 8 weeks old, n sections: 8) and both inhibitory (37.0 ± 9.3 % overlap with Pax2, n animals: 2 females, 8 weeks old, n sections: 8) and excitatory neurons were identified. The subsequent exploratory analyses will focus on assigning the activated neurons to previously described spinal cord cell types (Häring et al.) and to map these cell types to spinal laminas using ISS. Identifying modality-specific neurons in the spinal cord is a critical and natural next step as scRNA-seq experiments have identified a vast diversity of spinal interneurons.

Disclosures: H.M. Weman: None. **M.M. Ceder:** None. **J.E.T. Jakobsson:** None. **M.C. Lagerström:** None.

Poster

P380. Pain: Nociceptors: Cell and Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P380.06

Topic: D.03. Somatosensation – Pain

Support: BPD NIH GM137863-01
MARC NIH GM008395
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CSUCOAST-LACROL-CSUN-AY2021

Title: A molecular approach to elasmobranch nociception

Authors: *R. LACAP, M. TESORO, M. VAN DER PLAS, M. DE BELLARD;
Biol. Dept., California State University, Northridge, Northridge, CA

Abstract: To perceive pain, specialized sensory neurons, specifically nociceptor neurons, must be present to sense noxious stimuli. This process known as nociception is used by animals to avoid injury. In vertebrates, nociceptor neurons projecting from sensory ganglia are either C-fiber or A-δ fiber neurons. However, sharks have been proposed to lack full-scale nociception because of their low numbers of C-fiber neurons and their disregard for serious bodily injury. Sharks belong to a group called elasmobranchs, along with skates and rays. There exists a gap of knowledge regarding elasmobranchs nociception. In the present study, we aim to determine the existence of true nociceptor neurons in elasmobranchs. We conducted RNA sequencing with the trigeminal and dorsal sensory ganglia, followed by RT-PCR and histology. Our results show that elasmobranchs express a larger set of nociceptor genes in their trigeminal ganglia in comparison to their dorsal root ganglia (DRG). Interestingly, we found the presence of the two key transcription factors for nociceptor differentiation (Runx1 and PRDM12) in significant levels in the trigeminal ganglia, while almost no detectable levels in the DRGs.

Disclosures: R. Lacap: None. **M. Tesoro:** None. **M. van der Plas:** None. **M. de Bellard:** None.

Poster

P380. Pain: Nociceptors: Cell and Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P380.07

Topic: D.03. Somatosensation – Pain

Support: Duke University Anesthesiology Research Fund

Title: Macrophage-produced IL-23 Enhances Light and Capsaicin-induced Spontaneous Pain in Female Mice

Authors: ***J. JI**[1], Q. HE[2], X. LUO[3], A. MCGINNIS[2], A. G. NACKLEY[4], R.-R. JI[5];
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Abstract: The incidence of chronic pain is especially high in women. However, pain signaling in females remains poorly understood. IL-23 is a pro-inflammatory cytokine, but little is known about its role in pain regulation. Here, we examine the role of interleukin-23 (IL-23) in sex dimorphism in pain, using both behavioral and optogenetic methods. We employed transgenic mice expressing channelrhodopsin-2 (ChR2) in TRPV1-positive nociceptive neurons. Blue light was used to stimulate the hindpaw of the transgenic mice at varying intensities. IL-23 was injected into the hindpaw of mice of both sexes, and the mechanical pain threshold was measured using the von-Frey up-down test. Additionally, we assessed capsaicin-induced spontaneous pain following IL-23 pre-treatment. We also examined the phosphorylation of p38 MAP kinase (p-p38) in dorsal root ganglion (DRG) neurons. We found that blue light stimulation in the transgenic mice induced intensity-dependent spontaneous pain. At the highest intensity, females show more intense spontaneous pain than males. Intraplantar injection of IL-23 induced mechanical allodynia in females only. Furthermore, IL-23 potentiates light-induced spontaneous pain as well as capsaicin-induced spontaneous pain in females but not males. Of note, TRPV1 mRNA expression in a subset of small TRPV1-positive cells (100-200 μm2) was greater in females than males. We observed that IL-23 is expressed by macrophages in mouse DRGs. IL-23 also induced significantly greater p38 phosphorylation in DRG neurons of female mice than male mice. Our findings suggest that IL-23 potentiates spontaneous pain predominantly in females. Given the fact IL-23 is produced by macrophages and can activate p38 in nociceptors, our study may present a new form of cytokine-induced macrophage-nociceptor interaction in a sex-dependent manner.

Disclosures: J. Ji: None. **Q. He:** None. **X. Luo:** None. **A. McGinnis:** None. **A.G. Nackley:** None. **R. Ji:** None.

Poster

P380. Pain: Nociceptors: Cell and Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P380.08

Topic: D.03. Somatosensation – Pain

Support: NIH Grant R01NS107569

Title: Deficient AMPK activity in dorsal root ganglion leads to hyper-excitability in nociceptive sensory neurons and hyperalgesia in mice with systemic lupus erythematosus.

Authors: *V. VIATCHENKO-KARPINSKI, L. KONG, H.-R. WENG;
Mercer Univ., Macon, GA

Abstract: Systemic lupus erythematosus (SLE) is an autoimmune disease in which the autoantibodies attacks its own tissues, causing widespread inflammation and tissue damage in the affected organs. Patients with SLE often suffer from chronic pain. We previously reported that overactivation of spinal macrophage colony-stimulating factor-1 receptor causes chronic pain via suppressing glial glutamate transporter in female MRL lupus-prone (MRL/lpr) mice, a SLE mouse model. In this study, we characterized the development of chronic pain and its peripheral mechanisms in a male MRL/lpr mice. We found that male MRL/lpr mice developed thermal hyperalgesia at the age of 10 weeks, which is similar to the time course found in female MRL/lpr mice. Using patch-clamp recording techniques, we compared the membrane properties of nociceptive sensory neurons (membrane capacitance = 23.67 ± 1.82 pF) in L4-5 dorsal root ganglions obtained from MRL/lpr mice with chronic pain and normal controls. We found that in comparison with normal controls, nociceptive sensory neurons from mice with chronic pain had significantly lower rheobase and action potential thresholds, and more positive resting membrane potential (RMP), which indicates an increased excitability in these nociceptive neurons. Our western blot data show that dorsal root ganglions obtained from MRL/lpr mice with chronic pain had significantly lower levels of phosphorylated AMP-activated protein kinase (AMPK). This was concurrently accompanied with increased levels of TNFα and ERK/MAPK activities. More importantly, our patch clamp experiments found that pharmacological suppression of AMPK activity in normal nociceptive neurons by perfusing compound C (10 μM) into the recording bath recapitulates the membrane properties found in mice with chronic pain. Furthermore, subcutaneous injection of compound C (25 μM, 25 μl) in control mice (n=6) induced significantly heat hyperalgesia while activation of AMPK using AICAR (1 mM, 25 μl) significantly attenuated heat hyperalgesia in mice (n=5) with chronic pain. In conclusion, our study reveals that deficiency of AMPK activity in DRG neurons plays a critical role in the increased excitability of nociceptive sensory neurons and chronic pain in male SLE mice.

Disclosures: V. Viatchenko-Karpinski: None. **L. Kong:** None. **H. Weng:** None.

Poster

P380. Pain: Nociceptors: Cell and Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P380.09

Topic: D.04. Somatosensation – Touch

Support: Grant-in-Aid for Scientific Research(A) 19H00633

Title: Signal transduction between keratinocytes and dorsal root ganglion neurons via extracellular ATP

Authors: *K. FUJITA, M. TANAKA, Y. SHINDO, K. HOTTA, K. OKA;
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Abstract: The skin is exposed to various external and internal stimulation: heat, several chemicals, oxidative stress, mechanical stimuli, and noxious stimuli including pain. Keratinocytes, which are the main cell type in the epidermis, interact with peripheral sensory neurons and modulate the signal transduction. It has been reported that adenosine triphosphate (ATP) released from keratinocytes contributes to the mechanical sensations (Koizumi *et al*., 2004), however, the detailed mechanisms of ATP-mediated signal transduction is still unknown. To directly visualize and analyze the spatio-temporal dynamics of the ATP and Ca^{2+}, we performed the live imaging of extracellular ATP and intracellular Ca^{2+}. We utilized iATPSnFR (Lobas *et al*., 2019), a genetically encoded ATP sensor localized on the outside of the cell membrane, and Fura Red, fluorescent Ca^{2+} indicator. Mechanical stimulus was applied to the keratinocyte by touching with a fine tip of a glass pipette and the release of ATP from the stimulated keratinocyte was confirmed. In adjacent keratinocytes, we observed transient increase in ATP level on its cell surface and subsequent increase in intracellular Ca^{2+} level. To our knowledge, this is the first report to directly visualize ATP during mechanical sensations. Next, to reveal the signal transduction from keratinocytes to peripheral sensory neurons in vitro, we co-cultured human keratinocytes and rat dorsal root ganglion (DRG) neurons. To visualize the signal transduction from keratinocytes to DRG neurons upon mechanical stimuli, both keratinocytes and DRG neurons were stained with a Ca^{2+} indicator, fluo-4, and mechanical stimulus was applied to a keratinocyte, then the cell types and their contacts were confirmed with immunostaining by anti-keratin14 and anti-βIII-tubulin. The touched keratinocyte showed Ca^{2+} increase followed by large Ca^{2+} increase in DRG neurons contacting with the stimulated keratinocyte and small Ca^{2+} increase in neighboring keratinocytes. To examine the difference between DRG neurons and keratinocytes, the sensitivity of keratinocytes and DRG neurons to ATP was estimated. The half-maximal effective concentration (EC_{50}) was estimated to be 7.3×10^{-7} M for keratinocytes and 4.3×10^{-8} M for DRG neurons, respectively. Our results suggest that the difference of Ca^{2+} response upon mechanical stimuli between keratinocytes and DRG neurons attributes to high sensitivity of DRG neurons to ATP compared to keratinocytes.

Disclosures: K. Fujita: None. **M. Tanaka:** None. **Y. Shindo:** None. **K. Hotta:** None. **K. Oka:** None.

Poster

P428. Taste II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P428.01

Topic: D.05. Olfaction and Taste

Support: NIH Grant 5R90DA033463-10

Title: A Model for Overshadowing in Conditioned Taste Aversion

Authors: *A. LEPSKY[1], P. MILLER[2];
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Abstract: Conditioned taste aversion is a psychological phenomenon in which a subject develops a strong negative association with a certain taste because they felt sick or nauseous afterwards. It has been suggested that the formation of these associations in the brain is competitive, since a more recent taste stimulus can reduce the degree of aversion to a prior taste. We investigated whether a model based on competing units, representing distinct populations of neurons, could describe conditioned taste aversion and its key features. Our model was composed of two self-excitatory and cross-inhibitory units in a winner-takes-all circuit and an input stimulus applied equally to both units. We then added a series of intermittent stimuli to represent taste recall and a decay function to represent the decay in synaptic plasticity between the presentation of a taste and the arrival of nausea. Incorporating the decay in synaptic plasticity over time led to a bias in the formation of aversions towards the most recent taste stimulus without fully blocking the aversion to the oldest taste. Overall, the model is able to predict qualitative features of conditioned taste aversion. Further additions to the model will help reproduce experimental results by tuning the time at which each taste was presented to the degree of aversion.

Disclosures: A. Lepsky: None. **P. Miller:** None.

Poster

P428. Taste II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P428.02

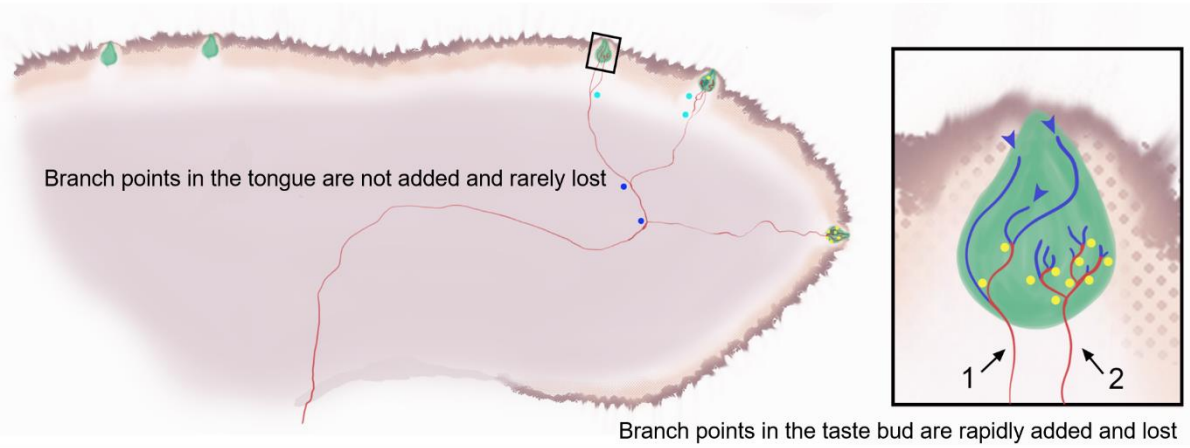
Topic: D.05. Olfaction and Taste

Support: DC018878
DC019050

Title: Structural remodeling of peripheral taste neurons

Authors: *Z. WHIDDON, D. ALSTON, *J. MARSHALL, A. MCGEE, R. KRIMM;
Univ. of Louisville, Louisville, KY

Abstract: Taste neurons receive information from taste-transducing cells that undergo continual turnover. Given that taste neurons must connect with new cells, we asked if they undergo structural remodeling over time using intravital imaging. Because the half-life of taste-transducing cells is 10 days, we speculated that half of the arbors (portion of the peripheral taste axon that innervates the taste bud) would remodel within 10 days in order to form connections with new cells. We sampled arbor structure every 12 hours for 10 days. Since synapses occur on distal (terminal) branches of the arbor, we quantified the change in the number of terminal branches over time. We found that all arbors altered their complexity by adding or pruning terminal branches within the first 5 days, and 19 of 31 arbors changed terminal branch number in the first 24 hours. On average 1 terminal branch was gained or lost every 16.0 hours. Given this surprising speed, we imaged arbors at 4 hours for 12 hours. We found that 42.4% of arbors gained or lost a terminal branch more than once in a 12-hour period. and that a terminal branch was gained or lost once every 6.8 hours. This finding indicates terminal branches remodel much faster than would be predicted by taste bud cell turnover. To investigate if terminal arbor size is altered over time, we measured the volume the arbor occupies within the taste bud (convex hull). Minimum hull volume predicts maximum hull volume (R^2=0.937), indicating that arbor size does not change over the full dynamic range in 10 days. Lastly, we examined 26 taste buds every 10 days for 100 days and found that no taste buds added new arbors and only ~5% of arbors were lost from fungiform papillae. Together these data demonstrate that during adulthood arbor structure within the taste bud is plastic, with terminal branches undergoing rapid, continuous remodeling, and that taste neurons can prune whole arbors from taste buds, but this type of refinement is not common.



Disclosures: Z. Whiddon: None. **D. Alston:** None. **J. Marshall:** None. **A. McGee:** None. **R. Krimm:** None.

Poster

P428. Taste II

Location: Virtual Only

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Program #/Poster #: P428.03

Topic: D.05. Olfaction and Taste

Support: Productos Medix 3247
Fundación Miguel Alemán
Catedra Marcos Moshisky

Title: Obesity alters palatability neurons and decreases lick-spike coherent activity in the rat Lateral Hypothalamus

Authors: *O. X. GUERRERO GUTIERREZ, M. A. VILLAVICENCIO, E. FONSECA, C. I. PEREZ, R. GUTIERREZ;
Pharmacol. Dept., CINVESTAV, CDMX, Mexico

Abstract: The prevalence of overweight and obesity has scaled in the last decades. Despite the consensus that the Lateral Hypothalamic Area (LHA) is important for regulating sleep-wake cycles, feeding, and motivated behaviors, how neural responses in the LHA are altered by a diet-induced obesity (DIO) model remains poorly understood. Mounting evidence indicates that multiple cell types in the LHA are affected by consuming HFD. An equally untouched question is how diet-induced obesity alters LHA neuronal activity in response to sucrose's palatability. To answer this question, we used Wistar rats fed (after weaning) with either a high-fat diet (HFD, 45% fat) to induce DIO or a standard diet (Chow, 8.4% fat) for 12 weeks. We characterized LHA extracellular single-unit activity while DIO and lean rats performed a Brief Access Taste Test (BATT). During training, DIO rats initially exhibited a larger inter-trial-interval (ITI) with a lower trial rate than the lean group, suggesting that initially sucrose motivates less to DIO rats than lean rats. However, after training, the difference disappears, and during recordings, both groups achieved the same level of motivation performing BATT. During recordings, we still found differences in the licking microstructure. Specifically, DIO exhibited a shorter inter-lick interval (ILI), which resulted in a faster fragmentation of licking; that is, DIO rats exhibited a lower lick rate, bout size, and less trial completeness than lean rats, with no changes in the trial rate or total calories consumed. As a neural correlate of the shorter ILIs, a spike-lick coherence analysis revealed that the DIO rats had lower coherence values than the lean rats while maintaining the same proportion of lick-coherent neurons. Therefore, LHA spiking synchronization with rhythmic licking is directly affected in a diet-induced obesity model. Moreover, LHA neuronal population analyses unveiled that in DIO rats, sucrose evoked a different population trajectory dynamic than lean rats, suggesting that LHA neurons processed sucrose concentrations in a fundamentally different manner. We performed a palatability analysis to identify the palatability-related neurons and found that DIO decreased the fraction of negatively correlated palatability neurons but increased those that positively correlated with palatability. These palatability-related neurons were used in a sucrose concentration decoding analysis showing that DIO rats have an earlier decoding onset than lean rats. In sum, we show that DIO alters the licking microstructure, lick-spike coherent activity, and the activity and proportion of the palatability-related neurons in the rat LHA.

Disclosures: O.X. Guerrero Gutierrez: None. **M.A. Villavicencio:** None. **E. Fonseca:** None. **C.I. Perez:** None. **R. Gutierrez:** None.

Poster

P428. Taste II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P428.04

Topic: D.05. Olfaction and Taste

Support: NIDCD RO1-DC018273

Title: Processing of multimodal chemosensory stimuli by the mediodorsal thalamus

Authors: *K. FREDERICKSEN, C. SAMUELSEN;
Univ. of Louisville, Louisville, KY

Abstract: The mediodorsal thalamus (MD) is a higher-order thalamic region important for stimulus-outcome associations and decision-making. Previously, we found that immediate early gene expression in the mediodorsal thalamus differed with experience for an orally consumed odor, taste, or odor-taste mixture. However, how neurons in the mediodorsal thalamus represent odors, tastes, and odor-taste mixtures, as well as its role in consummatory choice for experienced chemosensory stimuli remains unclear. To determine how neurons in the mediodorsal thalamus represent chemosensory signals, rats were given repeated experience with a palatable odor-taste mixture (isoamyl acetate-sucrose), and an unpalatable mixture (benzaldehyde-citric acid). Then, single-unit activity in the mediodorsal thalamus was recorded in behaving rats during the intraoral delivery of individual odors, individual tastes, and odor-taste mixtures. Our preliminary experiments revealed a subpopulation of chemo-responsive neurons in the mediodorsal thalamus (62.9%, 85/135) that are broadly tuned, respond with excitation and suppression, and represent mixtures differently than their odor and taste components. A population decoding analysis revealed that this subpopulation accurately represents the identity of odors, tastes, and odor-taste mixtures, with a quarter of them representing taste palatability (25.8%, 22/85). To determine the role of the mediodorsal thalamus in consummatory choice based on prior chemosensory experience, we used a two-bottle brief-access task to measure preferences for previously experienced odors, tastes, and odor-taste mixtures during the pharmacological inactivation of the mediodorsal thalamus. Pharmacological inactivation (NBQX) of mediodorsal thalamus significantly reduced consumption during both the odor and taste sessions and eliminated the initial odor preference for the saccharin paired odor. Taken together, these preliminary results begin to discern the role of mediodorsal thalamus in processing chemosensory signals to guide consummatory choice.

Disclosures: K. Fredericksen: None. **C. Samuelsen:** None.

Poster

P428. Taste II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P428.05

Topic: D.05. Olfaction and Taste

Support: PAPIIT IN219720

Title: Neural code of taste processing ethanol in the Orbitofrontal Cortex in mice

Authors: *E. PERRUSQUIA[1,3], K. ZEPEDA REYES[2], R. ANDRADE GONZÁLEZ[1,3], I. PEREZ MARTINEZ[1];
[1]UNAM, Tlalnepantla, Mexico; [2]UNAM, Cuautitlan Izcalli, Mexico; [3]Sip, IPN, Mexico City, Mexico

Abstract: In most people, consuming ethanol for the first time results in an aversive or unpleasant taste experience that requires peripheral and central neuroadaptation processes to increase consumption in subsequent consumption opportunities. This is how constant intake induces neuronal alterations that result in the development of alcohol use disorder (AUD). This is associated with electrophysiological changes in the reward circuit, specifically in prefrontal regions such as the Orbitofrontal Cortex (OFC); which participates in anticipation of both aversive and appetitive stimuli for decisión-making; and is precisely the adaptation to the appetitive and aversive taste qualities of ethanol, which induces the compulsion to ingest it. Before the results presented in this work, it was unknown if the taste code was processed in OFC when ethanol was presented for the first time at the orofacial level, in such a way that it can be determined what the changes that underlie the development of AUD by changes in the gustatory response area. To answer this question, we recorded neuronal OFC activity in C57BL/6 mice under sedation to monitor neuronal modulation without motor elements of ingestión, and we performed extracellular neuronal recordings with a 16-electrode microarray. Different concentrations of ethanol (.6, 1, 2.1, 3.2, 4.3, and 8.6 M) were administered to the togue extensión of the mice and water, quinine, and sucrose as control substances for neutral, bitter, and sweet flavors, respectively. 73 OFC neurons were registered, of which their electrophysiological characteristics were determined for functional classification. Fort the first time, we demonstrate that neuronal modulations or changes in firing rate (Wilcoxon signed rank test, $p<0.001$) encode the neural taste code previously proposed by Katz; a) Early or somatosensory stage, b) middle or identification stage, and c) late or palatability stage. We then observe that OFC neurons form different neuronal assemblies for each type of taste stimulation and low and high ethanol concentrations. Finally, we classify neuronal activity based on changes in the z-score from baseline activity; and we show that the taste coding of ethanol is represented in the activity of neuronal groups that are activated similarly. It makes sense to conclude a robust

sensory integration in OFC of orofacial stimulation with ethanol. Alterations in this processing are fundamental for the initiation, maintenance, and development of addiction to ethanol.

Disclosures: E. Perrusquia: None. **K. Zepeda Reyes:** None. **R. Andrade González:** None. **I. Perez Martinez:** None.

Poster

P428. Taste II

Location: Virtual Only

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Program #/Poster #: P428.06

Topic: D.05. Olfaction and Taste

Title: Histological and Functional Characterization of Gastrin-Releasing Peptide-Expressing Neurons in the Gustatory Cortex of Mice

Authors: *L. A. CZARNECKI, O. K. SWANSON, S. SWAMINATHAN, J. F. KOGAN, A. MAFFEI, A. FONTANINI;
Neurobio. and Behavior, SUNY Stony Brook, Stony Brook, NY

Abstract: Gastrin-releasing peptide (GRP) is a peptide expressed both peripherally and centrally. In the periphery, GRP is involved in enteric activities related to digestion and metabolism. GRP has been explored centrally for its role in itch, circadian rhythm, memory, and ingestion. Interestingly, GRP is highly expressed in the gustatory cortex (GC) although its role is unknown. Here, we investigated the anatomical and electrophysiological properties of GC neurons expressing GRP, as well as their taste-evoked responses. First, we show that GRP-expressing neurons constitute an approximate 6% of the total neural population in GC with densest expression in superficial layers, particularly in the dysgranular subdivision. We recorded from GRP-expressing neurons in acute slice preparations of GC and found that all recorded neurons had intrinsic properties consistent with pyramidal cells. To assess taste-evoked responses, we performed two-photon calcium imaging in awake GRP-tdTomato mice. These mice also expressed GCaMP7f under the synapsin promoter which allowed us to visualize calcium-based fluorescent transients in all neurons and to identify GRP-expressing cells. GRP-expressing neurons in GC display characteristic taste-evoked and lick-evoked activity. To assess the connectivity of these neurons, we used anterograde tracing approaches and found the most prominent projections to frontal regions, to contralateral GC, and to the basolateral amygdala (BLA). Since the BLA is involved in taste valence and taste learning, we sought to further investigate the influence of GRP in the BLA. Consistent with previous reports, GRP bath application increased spontaneous inhibitory activity in the BLA. To assess the synaptic properties of GRP+ neurons projecting to BLA, we expressed ChR2 in GRP+ neurons in GC. After 2-3 weeks, we prepared acute slices of BLA and performed whole-cell recordings of excitatory neurons. Optogenetic stimulation revealed that GRP-expressing GC cells make

excitatory synaptic connections with neurons in BLA. Future work will further elucidate the contribution of neurotransmitter versus neuropeptide contributions to synaptic responses as well as evaluate the contribution of the GRP-BLA projection in taste-guided ingestive decisions.

Disclosures: L.A. Czarnecki: None. **O.K. Swanson:** None. **S. Swaminathan:** None. **J.F. Kogan:** None. **A. Maffei:** None. **A. Fontanini:** None.

Poster

P428. Taste II

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P428.07

Topic: D.05. Olfaction and Taste

Support: NIDCD R01-DC018273

Title: Functionally diverse populations of chemoresponsive neurons in the gustatory cortex identified by photostimulation of fibers from the posterior piriform cortex.

Authors: *S. STOCKE, C. SAMUELSEN;
Univ. of Louisville, Louisville, KY

Abstract: The perception of flavor is a multisensory experience requiring smell and taste. Sampling an odor-taste mixture associates an odor with a taste's quality and hedonic value, resulting in behavioral preferences for odors paired with palatable tastes and avoidance of odors paired with unpalatable tastes. The gustatory cortex (GC) receives convergent gustatory and olfactory signals and is considered a principle site for processing multimodal chemosensory signals. Recent electrophysiological and behavioral studies show that cortico-cortical interactions between the gustatory cortex and posterior piriform cortex (pPC) influence functional and behavioral responses to odors and tastes. However, how neurons in the gustatory cortex represent odor-taste mixtures, when multiple odors are paired with the same taste, as well as the functional connectivity between the posterior piriform cortex and chemoresponsive neurons in the gustatory cortex remains unclear. To test this, we injected the posterior piriform cortex in rats with a virus to drive the expression of channelrhodopsin-2 (ChR2) and implanted a drivable multielectrode optrode into the gustatory cortex. Following surgery, rats were given repeated experience with four odor-taste mixtures: two palatable (isoamyl acetate-sucrose and methyl valerate-sucrose) and two unpalatable (benzaldehyde-citric acid and ethyl butyrate-citric acid). Next, we recorded single-unit activity in the gustatory cortex of alert rats during the intraoral delivery of the individual odors, the individual tastes, and the odor-taste mixtures. Immediately following each chemosensory delivery session, we photo-activated ChR2-expressing posterior piriform fibers in the gustatory cortex to identify the population of neurons likely modulated by input from the posterior piriform cortex. We found that ~40% of chemoresponsive neurons in the gustatory

cortex are modulated by photostimulation of fibers from the posterior piriform cortex. Both the light-responsive and non-responsive populations respond to multiple different odors, tastes, and odor-taste mixtures. Interestingly, a significantly greater proportion of the laser-responsive neurons selectively represent odors and odor-taste mixtures as compared to the non-responsive population. Our preliminary findings support the hypothesis that the gustatory cortex is a key component of the network processing of multimodal chemosensory signals and suggest that cortico-cortical connections with the posterior piriform cortex may underlie the functional diversity of chemoresponsive neurons in the gustatory cortex.

Disclosures: S. Stocke: None. **C. Samuelsen:** None.

Poster

P428. Taste II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P428.08

Topic: D.05. Olfaction and Taste

Support: ISF 2895/20

Title: The dynamics of neuronal activity and network connectivity changes that underlie the formation and consolidation of a gustatory associative memory

Authors: E. ARIELI[1], O. RAHAMIM[1], A. ITZKOVITCH[1], ***A. MORAN**[1,2];
[1]Neurobio., Tel Aviv Univ., Tel Aviv, Israel; [2]Sagol Sch. of Neurosci., Tel Aviv, Israel

Abstract: Acquiring new memories is a multi-stage process. Many studies have demonstrated that initially acquired memories are labile, and only stabilized by later consolidation processes. These multiple phases of memory formation are known to involve modification of both cellular excitability and synaptic connectivity, which in turn change neuronal activity at both the single neuron and ensemble levels. However, the specific mapping between the known phases of memory and the observed changes in neuronal activity, as well as the reorganization of the neuronal network remain unknown. We address this unknown in the context of conditioned taste aversion (CTA) learning in which a novel taste is paired with malaise to induce taste avoidance. Following CTA induction, we continuously tracked the activity of gustatory cortex (GC) neurons of rats for 24 hours during which CTA is acquired and consolidated. We found that the progression of neuronal activity changes in the GC depend on the neuronal organizational level. The population response changed continuously; these changes, however, were only reflected in the population mean amplitude during the acquisition and consolidation phases, and in the known quickening of the ensemble state dynamics after the time of consolidation. Preliminary analyses of the activity of hundreds of GC neurons recorded simultaneously using Neuropixels probes showed that network connectivity changes were also time-locked to the memory

acquisition and consolidation phases. Together our results demonstrate how complex dynamics in different representational level of cortical activity underlie the formation and stabilization of memory within the cortex.

Disclosures: E. Arieli: None. **O. Rahamim:** None. **A. Itzkovitch:** None. **A. Moran:** None.

Poster

P442. Auditory Processing: Neural Coding, Experiment, and Theory

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P442.01

Topic: D.06. Auditory & Vestibular Systems

Support: NIDCD R01DC015138
UConn OVPR

Title: Categorical perception and auditory midbrain representations of chimeric sound textures

Authors: M. SADEGHI[1], X. ZHAI[2], *D. PEDRICK[3], H. L. READ[1], I. STEVENSON[1], **M. A. ESCABI**[1];
[2]Electrical & Computer Engin., [1]Univ. of Connecticut, Storrs, CT; [3]Univ. of Connecticut, Marlborough, CT

Abstract: Categorical perception, the ability to group sounds or images into discrete perceptual categories, enables humans and other animals to rapidly and robustly recognize and respond to stimuli. Categorical representations have been observed for speech and other vocalized sounds along relatively simple time and frequency dimensions. However, the high-order acoustic features and neural computations underlying categorical representations for more general sound categories are not well understood.Here we used a texture synthesis (McDermott & Simoncelli 2011) to synthesize ‘chimeric’ auditory textures. Synthetic chimeric textures are generated by morphing summary statistics of runningwater andcracklingfire sounds. The chimeric sounds were first used in human perception tasks were participants were either required toidentify a chimeric texture as fire or water or alternately required todiscriminate two chimeric textures. For both tasks, the chimeric textures statistics were varied by adjusting the included statistics during the synthesis and the morph ratio (MR) from 0 (water extreme) to 1 (fire extreme). Parallel studies were also carriedout to study the neural representation of chimeric textures in the inferior colliculus (IC) of unanesthetized rabbits. Neural activity was obtained using multi-channel silicon probes inserted along the principal frequency axis during passive listening of the chimeric textures. We demonstrate that shifting high-order summary statistics by changing the morph ratio of fire and water produces a robust shift in human listener’s perception. Participants readily identified water and fire sounds in a categorical-like fashion with increasing morph ratio where the sound correlation structure was the critical statistical cue responsible for the categorical

effect. The categorical-like shift is accompanied by lower discrimination accuracy and larger just noticeable difference limen (JND) for within category as compared to across category discrimination. We then demonstrate how the response statistics of neural ensembles in the IC can be used to decode the sound category. Neural decoders were used to assess how population response metrics can categorize or discriminate the chimeric textures. Shifting the morph ratio produces a similar categorical-like shift in neural decoding performance and similar larger JNDs and lower accuracy trends as for human participants. The findings suggest that high-order statistical sound cues can drive categorical-like perception for textures and that the neural response statistics in IC can contribute towards such phenomenon.

Disclosures: M. Sadeghi: None. **X. Zhai:** None. **D. Pedrick:** None. **H.L. Read:** None. **I. Stevenson:** None. **M.A. Escabi:** None.

Poster

P442. Auditory Processing: Neural Coding, Experiment, and Theory

Location: Virtual Only

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Program #/Poster #: P442.02

Topic: D.06. Auditory & Vestibular Systems

Support: NIDCD R01DC015138
UConn OVPR Foundation

Title: Modulation content of interfering natural background sounds affects neural population synchrony and response power during vocalization processing

Authors: D. PEDRICK[1], X. ZHAI[2], I. STEVENSON[1], ***M. ESCABI**[1];
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Abstract: Selectively listening to a target sound within a noisy environment is a challenge to the auditory system. Studies using simple sounds such as a tone in noise have attempted to understand how sound segregation is accomplished during normal listening. However, in everyday settings natural background sounds confront listeners with far more complex listening conditions. During sound masking, low-order sound structure such as overlapping power spectra can inhibit the strength of neural responses. However, the contribution of higher-order statistics in the modulation content of natural backgrounds to masking at the neural level is less understood. Here we manipulated the spectrum and modulation content of five natural backgrounds (babble, machine noise, water, fire, birds) and white noise to determine their contribution to energetic and modulation masking. To dissociate masking attributed to the spectrum versus modulation content, we generated phase randomized (PR) backgrounds which have the power spectrum of the original sounds but a whitened modulation spectrum. Furthermore, to compare masking effects attributed to the modulation content of different

background sounds, we used spectrum equalized (SE) backgrounds that have a power spectrum identical to pink noise, but retain the original sound modulation content. Electrophysiological recordings were carried out in the inferior colliculus of awake Dutch Belt rabbits. Neural population responses to speech and animal vocalizations were measured in the presence of the natural and perturbed backgrounds.We demonstrate that vocalizations in the presence of PR backgrounds reduce the neural population signal strength and synchrony of the encoded foreground vocalizations, when compared against the unaltered backgrounds. Despite this, and paradoxically, the neural output signal-to-noise ratio of the encoded foreground is higher for the PR conditions. This suggest that PR backgrounds sparse the neural activity, yet they produce a higher fidelity neural representation of the foreground vocalization. In contrast, SE backgrounds have little effect on neural responses when compared to unaltered backgrounds. Masking differences observed across the original background sounds remained for the SE condition suggesting that the background modulation content dominated the observed masking patterns for individual backgrounds.Collectively, these results suggest that modulation statistics in natural background sounds can have varied interfering effects and that the unique modulation content of each of the interfering background sounds can strongly influence the encoding of masked vocalizations.

Disclosures: D. Pedrick: None. **X. Zhai:** None. **I. Stevenson:** None. **M. Escabi:** None.

Poster

P442. Auditory Processing: Neural Coding, Experiment, and Theory

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P442.03

Topic: D.06. Auditory & Vestibular Systems

Support: NIDCD R01DC015138

Title: The role of rate-based and correlation-based neural activity towards the recognition and discrimination of natural sound textures

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Abstract: Time-averaged summary statistics in sound textures, such as stream and a noisy restaurant, contribute to sound perception and are known to modulate neural activity in auditory midbrain. Yet, the role of different neural codes (rate, timing, and correlation based) towards the neural representation of textures remains largely unknown. Here we recorded neural activity in the inferior colliculus (IC) of unanesthetized rabbits using a linear multi-channel silicon probe

during passive listening to natural sound textures and synthetic variants with perturbed statistics. An analog representation of multi-unit activity (aMUA) and spike-sorted single-unit data were evaluated from the neural recordings and the results were compared to human psychoacoustic tests using the same sounds.We demonstrated that summary statistics strongly influence coordinated firing across the IC neural ensembles and that neural ensemble correlations converged to those measured the original textures as statistics were added to the synthetic variants. By comparison, a rate-based representation of the neural population activity was largely unaffected by adding high-order statistics to the synthetics variants. Single-unit response properties also varied with changing sound summary statistics. Shuffled autocorrelograms exhibited a sharper and stronger peak upon adding higher-order statistics, indicating enhanced temporal coding, higher temporal precision and higher reliability. By comparison, firing rates were relatively constant and unaffected by the summary statistics. Furthermore, correlated firing between single-units showed diverse structures across sounds and increased upon adding statistics to the synthetic stimuli. Next, single-trial neural decoders were used to determine whether the neural correlations and spectrum in IC could allow sound textures to be identified and discriminated. The neural correlation-based classifier performance improved with increasing sound duration and added sound statistics, with an evidence accumulation time scale of ~1 sec, resembling trends observed in human psychoacoustics (Zhai et al 2020). By comparison, neural spectrum-based codes provided a substantially faster neural signal which contributed much more towards texture discrimination, analogous to human participants.The findings suggest that neural response statistics in auditory midbrain are modulated in unique ways by statistical regularities in sounds and that rate-based and correlation-based neural codes may have dissociable roles and time scales for the recognition and discrimination of natural texture sounds. (supported by NIDCD R01DC015138)

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Poster

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Topic: D.06. Auditory & Vestibular Systems

Support: ERC - DEEPEN
FRM - PostDoc France
Fondation pour l'audition

Title: Transformation of population representations of sounds throughout the auditory system

Authors: *S. BAGUR[1], J. BOURG[2], A. KEMPF[3], T. TARPIN[1], E. GOSSELIN[1], S. A. CEBALLO[1], K. BERGAOUI[1], Y. GUO[1], A. MULLER[1], J. BOURIEN[4], J. PUEL[4], B. BATHELLIER[1];
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Abstract: Auditory perception relies on robust spectrotemporal pattern separation. Yet the step-by-step computational transformations through which this occurs in the auditory system are still elusive, partly due to the lack of systematic comparison of sound representations at different stages. To address this, we compared population representations based on extensive two-photon calcium imaging and electrophysiology recordings in the auditory cortex, thalamus, inferior colliculus in mice and on a detailed biophysical model of the cochlea. Using a noise-corrected correlation metric, we measured the similarity of activity patterns generated by diverse canonical spectrotemporal motifs (pure tones and chords, amplitude and frequency modulations, broad band noise). This measure evidenced a gradual decorrelation of population responses across sounds, accompanied by a convergence of the information carried by temporal and rate codes in cortex. Rate code representations that were most similar at cochlear level, for example for time symmetric sounds were decorrelated mainly between thalamus and cortex, while many other sound pairs underwent significant decorrelation earlier. Strikingly, representations of different intensities of the same sounds were also decorrelated. While this transformation was related to response sparsenning in the cortex and strong non-linearities, the sound representations remained continuous, preserving local similarity relationships at all stages. Gradual decorrelation of time-independent representations could be reproduced by a deep network trained to detect in parallel basic perceptual attributes of sounds, including estimates of frequency and intensity ranges as well as temporal modulation types. In contrast networks trained on tasks ignoring some perceptual attributes (e.g. intensity) did not decorrelate the related acoustic information. The auditory thalamus was the site of recorrelation of population representations and very dense neural coding, without loss of accuracy in sound encoding. Modifying the architecture of CNN networks to reproduce the anatomical bottleneck of the thalamus resulted in a recorrelation of representations, similar to the data. The contrasting representations of cortex and thalamus, both of which connect to behavioural output regions, suggest complementary roles in the broadcast of auditory information. Together these results establish that the mouse auditory system transforms sound representations to make information about diverse perceptual attributes generically accessible in a rate-based population code.

Disclosures: S. Bagur: None. **J. Bourg:** None. **A. Kempf:** None. **T. Tarpin:** None. **E. Gosselin:** None. **S.A. Ceballo:** None. **K. Bergaoui:** None. **Y. Guo:** None. **A. Muller:** None. **J. Bourien:** None. **J. Puel:** None. **B. Bathellier:** None.

Poster

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Program #/Poster #: P442.05

Topic: D.06. Auditory & Vestibular Systems

Support: KIBM IRG
Eugene Cota-Robles

Title: Relating Mesoscale Local Field Potential and Macroscale Electroencephalography (EEG) Signals in the Rat Primary Auditory Cortex

Authors: *T. JOHNSON, A. GALLAGHER, S. COULSON, L. M. RANGEL;
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Abstract: Electroencephalography (EEG) signals incorporate summated currents from a range of different sources, making it challenging to precisely identify their local circuit underpinnings. In this study, we acquired skull EEG from the rat primary auditory cortex (A1) while simultaneously recording contralateral local field potential (LFP) activity using silicon probe electrodes that spanned cortical layers. Repetitive auditory stimuli presented at 20, 30, 40, 50, and 80Hz were used to assess the local circuit underpinnings of the Auditory Steady State Response (ASSR) in rats. These stimuli effectively pace networks involved in auditory processing, creating predictable large scale currents at the frequency of the presented stimulus that are reliably detected in electrophysiological measurements in a range of species. In this study, the ASSR manifested prominently in all cortical layers and in the EEG signal of the rat, enabling an assessment of the degree to which features in the LFP (e.g. the magnitude of currents in each layer and the degree of synchrony across layers) related to the EEG signal. Our findings suggest that a resonant response occurs when networks are paced by 40Hz gamma frequency range stimuli, with both current magnitudes and the degree of synchrony across layers contributing to this larger response. By relating signals across these different scales, we provide mechanistic insight into the unique local circuit dynamics underlying distinct EEG signals and auditory processing states in the rat.

Disclosures: T. Johnson: None. **A. Gallagher:** None. **S. Coulson:** None. **L.M. Rangel:** None.

Poster

P442. Auditory Processing: Neural Coding, Experiment, and Theory

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Program #/Poster #: P442.06

Topic: D.06. Auditory & Vestibular Systems

Support: Fondation pour l’Audition

European Research Council (ERC CoG DEEPEN)

Title: Encoding of complex natural sounds in mouse auditory cortex

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Abstract: The encoding of simple and complex sounds in the large neural ensembles of the mouse auditory cortex (AC) remains a puzzle. Our preliminary data indicate that large clusters of AC neurons respond specifically to complex or natural sounds and not to the simpler sounds (e.g. pure tones) that share the same frequencies. To investigate further how complex sounds are encoded in the AC and which of their features are specifically extracted at this level of auditory pathway, we have exposed animals to the extended set of sounds that not only consists a variety of simple sounds (i.e.: pure tones, intensity ramps, chirps, sine amplitude modulated sounds, chords, and different versions of white noise), but also decomposition of several complex sounds into simpler and yet specific blocks highlighting their features. Mice were also presented with an additional set of natural sounds required for cross-validation of obtained results. Data have been collected throughout all the layers of AC in 14 adult (4-5 months old) awake male mice using large scale multi-plane two-photon calcium imaging that enabled us to record simultaneously neuronal activity in 3 or 4 plans distributed across 200um of vertical extent of AC. Acquired data have been used to test whether our models can predict cortical responses to complex natural sounds.
Our methodology coupled with computational modelling is aimed to establish a better understanding of the contributions of auditory cortex representations to natural sounds perception.

Disclosures: J. Schwenkgrub: None. **S. Bagur:** None. **E. Gosselin:** None. **S. Jamali:** None. **A. Renard:** None. **B. Bathellier:** None.

Poster

P442. Auditory Processing: Neural Coding, Experiment, and Theory

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Title: Distinct synaptic plasticity mechanisms determine the diversity of cortical responses during behavior

Authors: M. INSANALLY[1], B. F. ALBANNA[2], **J. M. TOTH**[1], B. DEPASQUALE[3], S. FADAEI[4], T. GUPTA[4], K. RAJAN[5], R. C. FROEMKE[4,6,7];
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Abstract: Heterogeneous neuronal responses are found in many cortical areas including visual, auditory, parietal, and frontal cortex. The extent of this heterogeneity is vast. A fraction of cells exhibit trial-averaged responses with obvious task-related features such as pure tone frequency tuning in auditory cortex (classically responsive). However, a substantial number of cells do not appear to fire in a task-related manner and are often neglected from analysis (non-classically responsive). Our recent study used a novel spike-timing-based analysis to show that both classically and non-classically responsive cortical neurons contain significant task information suggesting that non-classically responsive cells may play an underappreciated role in perception and behavior (Insanally et al. 2019). Where do these diverse response profiles come from and what is their contribution to behavior? What are the synaptic inputs that might drive such diverse responses? To address these questions, we trained a novel spiking recurrent neural network model that incorporates spike-timing-dependent plasticity mechanisms and FORCE learning to perform the same task as behaving animals. By leveraging excitatory and inhibitory plasticity, this model reproduces neurons with response profiles consistent with published in vivo data including classically and non-classically responsive neurons which both encode task information in their spike times as seen in vivo. Using simulated perturbation experiments to assess the functional contribution of these subpopulations, we found that classically responsive cells contribute more to behavior via output connections while non-classically responsive cells contribute via recurrent connections by increasing the dimensionality of activity during the choice period. These results provide strong evidence for a double-dissociative function of classically and non-classically responsive neurons. Interestingly, excitatory plasticity played a primary role in establishing the diversity of response profiles observed; however inhibitory plasticity had a strong modulatory effect when excitatory plasticity was active. Local patterns of connectivity predicted single-unit responsiveness and placed constraints on the synaptic patterns required for non-classically responsive activity to emerge making predictions that we compare to in vivo whole-cell recordings taken from the auditory cortex of behaving animals. Our approach recapitulates the heterogeneous neural responses found in behaving animals and provides a powerful lens for exploring large-scale neuronal dynamics and the plasticity rules that shape them.

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Poster

P442. Auditory Processing: Neural Coding, Experiment, and Theory

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Topic: D.06. Auditory & Vestibular Systems

Support: NIH R01DC018055

Title: Reconstruction of natural auditory stimuli from neuronal population activity

Authors: *B. THEILMAN[1], T. S. MCPHERSON[1], M. TURVEY[2], S. RUDRARAJU[2], T. Q. GENTNER[2,1,3,4];
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Abstract: The collective activity of neural populations in higher-order sensory brain regions is thought to support the representation of the complex natural stimuli that guide behavior. Encoding properties of single neurons and populations within these higher-order regions of the sensory hierarchy are difficult to characterize, however, because they can respond selectively to multiple, complex stimulus features. In principle, the ability to reconstruct natural stimuli from spiking activity across a neuronal population can provide valuable insight into the nature of natural stimulus representation at the neural population level. Here, we present a method to achieve spectrographic reconstructions of natural birdsong from spiking activity in neural populations in the Caudal Medial Nidopallium (NCM), an auditory region in songbirds analogous secondary auditory cortex. Using the maximum noise entropy method to model linear and non-linear components of NCM neurons' spiking responses, we show that high-fidelity reconstructions can be achieved with out a priori knowledge of the stimulus autocorrelation structure, marking an advance over previous auditory reconstruction efforts. This suggests the presence of an underlying representational "feature space" of lower dimension than the natural birdsongs. To investigate this, we trained a novel autoencoder to yield a nonlinear compression of natural birdsong into a low-dimensional latent-space. We show that these latent spaces capture relevant sensory dimensions for this secondary auditory population enabling high-quality spectrographic stimulus reconstructions. Finally, we relate the geometry of these reconstructions to the combinatorial structure of the spiking activity. Collectively, these results suggest that the information required for reconstruction of any conspecific vocal communication signal in songbirds is intrinsic to spatiotemporal firing patterns in the NCM population.

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Poster

P442. Auditory Processing: Neural Coding, Experiment, and Theory

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Title: Predictive processing in auditory cortex without consciousness

Authors: *Y. XIONG[1], J. A. DONOGHUE[2], M. K. MAHNKE[3], M. LUNDQVIST[5], E. N. BROWN[4], E. K. MILLER[6], A. BASTOS[1];
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Abstract: There is mounting evidence that the brain inhibits processing of predictable stimuli and responds more to unpredicted “oddballs”. Neuronal spiking as well as gamma-band frequency (40-100 Hz) power in the local field potential (LFP) are reduced to predictable stimuli, while alpha/beta frequency band (8-30 Hz) LFP power is enhanced to predicted stimuli. In sensory cortex, these modulations occur in a stimulus-specific manner. It has been suggested that these mechanisms for prediction are a feature of awake (conscious) processing. If this is the case, these prediction mechanisms should be reduced or eliminated during unconscious states. To test this, we recorded spiking and LFP activity from auditory (area STG) cortex of two macaque monkeys in both awake and unconscious states. We induced unconsciousness using the anesthetic propofol. Monkeys passively listened to pure tone auditory sequences that were either predictable or unpredictable. In the awake state, we reproduced previous findings that unpredictable stimuli evoke more gamma and less alpha/beta than predictable stimuli. In the unconscious state, baseline levels of both gamma and alpha/beta were strongly reduced relative to the awake state. However, when unpredictable stimuli occurred they enhanced LFP power in

both the gamma and alpha/beta bands relative to predictable stimuli. This power enhancement by unpredictable stimuli was even larger in the unconscious compared to the conscious state. Both awake and unconscious states featured sensory responses that were reduced by predictable stimuli in a stimulus-specific manner. Our findings challenge the hypothesis that inhibition of sensory responses by prediction is a key feature of conscious processing.

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Poster

P442. Auditory Processing: Neural Coding, Experiment, and Theory

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Topic: D.06. Auditory & Vestibular Systems

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Title: Pv neurons enhance cortical coding in the cocktail party problem

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Abstract: The human brain is a powerful computational device, capable of feats yet to be matched by machines. An impressive example is its ability to solve the cocktail party problem. For example, at a crowded cocktail party, we can hear what a friend is saying among other speakers and background noise. The integrative multisensory nature of such complex scene analysis suggests that the circuits of the cerebral cortex hold the key to solving this problem. How cortical circuits achieve this solution remains poorly understood. Complex auditory scenes typically contain multiple sound sources at different spatial locations. Here, we report auditory cortical neurons in mouse A1 with spatial response properties that are well suited for segregating competing sounds from different locations. We discovered that these neurons are sensitive to the spatial configuration of “target” and “masker” sound sources from different locations. Specifically, we quantified neural discrimination performance over a spatial grid of all possible combinations of target and masker locations and found that cortical neurons display “hotspots” of high performance at particular grid positions. At most hotspots, both rate modulations and spike timing-based coding contribute towards discrimination performance. Individual sites show diverse spatial grids, which enable the population representation of distinct spatial configurations. Finally, we found that optogenetic silencing of parvalbumin-positive (PV)

interneurons resulted in an expected increase in average firing rate that coincided with a decrease in discrimination performance at hotspots. This decrease in performance is accompanied by a reduction in the lifetime sparseness and reproducibility of responses across trials. We used several spike distance metrics to dissect the neural code and found that the decrease in performance at most hotspots is driven by a decrease in spike timing reproducibility and rapid temporal modulations of firing rate, independent of changes in overall spike count. Our findings indicate that PV neurons improve cortical representation of auditory stimuli in the cocktail party problem by promoting a sparse, temporally modulated, and reproducible code.

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Poster

P444. Auditory Processing: Arousal State

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Topic: D.06. Auditory & Vestibular Systems

Title: Probing semantic violations via the N400 event related potential component: provision of a publicly available auditory stimulus set and twenty high-density electrophysiological datasets.

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Abstract: When listening to speech, context is used to predict sentence outcome by limiting subsequent word options. How well a word matches a predicted word is quantified by a measure known as ‘cloze probability’. Neurophysiologically, cloze probability is represented at the level of the brain and can be measured using electroencephalography (EEG). The N400 event-related potential (ERP) derived from EEG data is an established outcome measure for semantic processing and integration which is highly sensitive to semantic congruency. The purpose of this study was to create the first publicly-available stimulus set that evokes the N400 ERP through auditory presentation, in order to aid in the comparability and reproducibility of future research examining semantic comprehension. This stimulus set utilizes the semantic anomaly paradigm, provides the cloze probability for all stimuli, and was developed to be used across a wide set of age ranges. This stimulus set was validated with 20 neurotypical adults and was found to elicit the N400 as well as two additional semantically-related ERPs: the late positivity component (LPC) and the recognition potential (RP). This standardized and validated stimulus set can be used to investigate several populations, and future semantic studies that use this stimulus set may benefit from the increased comparability and reproducibility within this field of research.

Disclosures: K. Toffolo: None. **E. Freedman:** None. **J. Foxe:** None.

Poster

P444. Auditory Processing: Arousal State

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Topic: D.06. Auditory & Vestibular Systems

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Title: A toolkit of modeling approaches for interpreting behavior and its neural correlates in quasi-continuous decision-making paradigms

Authors: M. MCGINLEY[1,2,3], S. CHEN[3], J. DE GEE[1,2];
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Abstract: Decision-making and its neural correlates are typically studied in laboratory tasks with rigid trial-based structures. A major appeal of these designs is existent powerful modeling frameworks like signal detection theory (SDT; Green and Swets, 1966) and drift diffusion modeling (DDM; Ratcliff and McKoon, 2008). However, in the real world, stimuli of interest arrive at unpredictable and sometimes long intervals. It may be that the neural circuit implementation of decision-making, including factors like attention and arousal, may differ substantially between rigid trial-based and natural decisions. Naturalistic tasks and efficient and interpretable modeling frameworks are needed. To fill this gap, we have developed a novel quasi-continuous sensory detection paradigm and a toolkit of modeling approaches based on SDT, DDM, and survival analysis.

In the first approach, which we call real-time signal detection theory (rt-SDT), SDT is applied in a sliding time-window, and then take the limit as the window duration goes to zero. We show, analytically, that the resultant equations for sensitivity (d') and bias (c) are equivalent to those derived by applying SDT at the per-stimulus level while ignoring the alternating stimulus sequencing and varying stimulus durations, and can be interpreted in the usual fashion after accounting for an estimated empirical temporal prior of the animal's decisions, using tools from survival analysis.

In a second approach, we modify the conventional DDM by using a single bound (instead of two) and adding leak to the diffusion process, mirroring a recent approach in human studies (Ossmy et al., 2013). With extensive simulations, we show that rt-SDT and this leaky DDM give consistent results; rt-SDT provides simple interpretations that work on small data sets, while the DDM requires more extensive data but provides a richer and behaviorally mechanistic account of the decision process.

As a final approach, we apply GLM models of choice as a binary variable (DeCarlo, 1998) or of

the hazard function for response using survival analysis. We find that these GLM approaches are intermediate between rt-SDT and leaky DDM in terms of constrainability, can be the most opaque to interpretation, but are powerful at incorporating physiological and behavioral covariates. In ongoing work, we are refining the GLM modeling to improve interpretability, thus establishing a firm foundation for our modeling toolkit.
Green and Swets (1966). Signal detection theory and psychophysics.
Ratcliff and McKoon (2008). Neural computation, 20, 873-922.
Ossmy, Moran, ⋯ Donner (2013). Current Biology, 23(11), 981-986.
DeCarlo (1998). Psychological methods, 3(2), 186.

Disclosures: M. McGinley: None. **S. Chen:** None. **J. de Gee:** None.

Poster

P444. Auditory Processing: Arousal State

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Program #/Poster #: P444.03

Topic: D.06. Auditory & Vestibular Systems

Title: Behavioral and computational approaches to mouse foraging behavior in physical and virtual patchy environments

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Abstract: Despite the large body of work in key areas of decision making, the neural circuits governing behavior of animals in natural environments has been relatively understudied. In particular, a rich literature in behavioral ecology has highlighted patch-based foraging, in which animals harvest from geographically clumped areas of finite resources, as a central task for many animal species, and thus one for which the brain presumably has evolved. Moreover, a plethora of studies have linked native animal behavior to mathematical models of patch-based foraging tasks, including the marginal value theorem, which states that the animal should leave the current patch and incur the search cost of traveling to a replenished patch when the incremental reward in the current patch falls below the average reward rate across the environment. The similarity to natural behavior and existence of proven behavioral models imply that foraging represents a robust framework for studying natural decision-making.
Here, we extrapolated these ideas to a probabilistic foraging task in both physical and virtual acoustic environments that use custom systems based in Python and LabVIEW, respectively. Reward ports were located at the endpoints of the physical maze or at evenly-spaced intervals in virtual space. When an animal arrived at a reward location, an auditory stimulus (tone cloud) was continuously played to indicate the “patch” is active. Potential rewards accumulated at intervals

which followed a modified non-homogeneous Poisson process with an exponentially decaying Poisson rate, which we term a “Poisson drip process,” and were given in 2 uL droplets in response to licking. A pure tone played when reward was available, and its frequency indicated the number of available reward droplets. The animal could leave the current reward zone at any point, allowing for a continuous background leaving decision, and move to the next active location. While moving between reward locations, a distinct auditory stimulus (pink noise) was played until the animal arrived in the next “patch”, at which point tone cloud played to indicate a new, replenished patch was active.
To analyze this task, we first developed theoretical deterministic and probabilistic models of foraging behavior. Our analysis underscores the importance of parameter selection for foraging environments and highlights potential strategies animals may employ. Fitting our models to preliminary data suggests that the animal's behavior was consistent with the marginal value theorem for both travel time and reward decay rate manipulations. Further analysis will compare learning and performance between the physical and virtual tasks.

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Poster

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Program #/Poster #: P444.04

Topic: D.06. Auditory & Vestibular Systems

Support: Japan Society for the Promotion of Science KAKENHI 18K18066

Title: Temporal Correlation between medial olivocochlear reflex and electroencephalographic activity during sleep

Authors: *S. OTSUKA, Y. SUZUKI, S. NAKAGAWA;
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Abstract: The medial olivocochlear reflex (MOCR) is a feedback response that controls the gain of the outer hair cells. It has been reported that the MOCR becomes weaker in a state of sleep. Given this, it is possible that MOCR strength fluctuates depending on awareness. However, no studies have evaluated awareness level quantitatively and examined the relationship with MOCR. In this study, we evaluated stages from awakening to sleep using electroencephalogram (EEG) and electrooculogram (EOG) and investigated the relationship with MOCR strength. The amplitude of β (13-30 Hz), α (8-13 Hz), θ (4-8 Hz) and δ wave (0.5-4 Hz), which reflects each stage from wakefulness to deep sleep, were monitored during the two-hours measurement session. We measured the contralateral MOCR in terms of the suppression of click-evoked otoacoustic emissions (CEOAEs) induced by noise presented to the contralateral ear. Click trains

were played into the right ear canal to generate CEOAEs from within the cochlea. Each click had a duration of 100 μs. Clicks were presented at a 55-dB peak-equivalent sound pressure level (SPL) and at a rate of 50 times per second. The MOCR was elicited by a noise presented to the left ear, referred to as the MOCR elicitor, during the CEOAE recording. The noise was band-pass filtered between 100 and 10,000 Hz, with a duration of 0.5 s, including a 10-ms raised-cosine ramp. The noise was presented at 60-dB SPL and at intervals of 5 s. The CEOAE waveforms measured from one minute before and one minute after each time point were extracted, and then the waveforms measured in the interval with and without contralateral noise were averaged, respectively.
The amount of OAE suppression (MOCR strength) had a statistically significant positive correlation with the time variation of α waves (appearing mainly from awakening to sleep onset), but not with other EEG components and with the time variation of slow eye movement (appears in the sleep onset period), which was calculated from EOG. This result indicates that MOCR strength becomes weaker at a relatively early stage of sleep onset, which has a large change in awareness level, regardless of the depth of sleep. Further, no correlation was found between the auditory brainstem response and the α waves. This result indicates that activities from the inner ear to brainstem are not affected by awareness level, but the activity of the efferent pathway is affected by awareness level.

Disclosures: S. Otsuka: None. **Y. Suzuki:** None. **S. Nakagawa:** None.

Poster

P444. Auditory Processing: Arousal State

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P444.05

Topic: D.06. Auditory & Vestibular Systems

Support: CONICYT-FONDECYT Regular Grant N° 283 1160258
CONICYT/ANID-PFCHA National Grant for Doctoral Studies N° 2018-21181786

Title: Pupil size and prefrontal Theta/Beta power reflect phasic adaptation of arousal during subconscious processing of auditory novelty

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Abstract: The ability to extract statistical regularities from sensory input and detect sudden changes in incoming sensory patterns mediates both autonomic and goal-directed responses. The

pupil response, considered a proxy of noradrenergic modulation of arousal, is sensitive to the detection of sensory novelty. However, how conscious perception modulates the brain's response to novel sensory stimuli and the subsequent adaptation of arousal remains unclear. In this study, we presented subjects (n = 24, mean age = 25.5, range = 13) sequences of sinusoidal tones and asked them to decide whether the last stimulus was the same or different from the previous standard ones. Target stimuli could be another standard tone or a deviant tone that could either be above or below each participant's threshold for conscious auditory discrimination. We collected behavioral (accuracy and reaction times), pupillometric (pupil size) and neurophysiological (Electroencephalography) data. We found faster reaction times for targets that were consciously detected and correctly reported as different (n = 21, $p < 0.05$, Bonferroni-corrected, two-tailed). While the pupil responded to all targets, increased pupil size was associated to the processing of targets that were not consciously detected as novel and therefore incorrectly reported as standards (300-400ms, n=21, $p < 0.05$, one-tailed). A Mismatch Negativity Response was elicited by both types of targets, but only targets that were consciously perceived as novel were associated to a P3 response (n = 20, $p < 0.05$, Bonferroni-corrected, one-tailed). Time-frequency decomposition analyses from source-reconstructed signals revealed increased power in the Beta (15-30hz) and Theta (4-8Hz) frequency range (n = 20, $p < 0.05$, cluster-corrected, one-tailed) within temporal and prefrontal regions for targets that were not consciously perceived as novel. In turn, increased theta power was observed within cingulate and insular regions for targets that were consciously perceived as novel (n = 20, $p < 0.05$, cluster-corrected, one-tailed). These findings suggest that phasic adaptation of arousal can operate independently of conscious perception. We discuss these results in the context of the adaptive-gain framework, where increased phasic arousal following undetected novel targets presumably reflects top-down modulation of neuronal gain functions to optimize performance and satisfy the demands imposed by the task at hand.

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Poster

P444. Auditory Processing: Arousal State

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Topic: D.06. Auditory & Vestibular Systems

Support: NIH Grant R01-DC04290
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Title: Arousal state dependence of short-and long-term auditory novelty response interactions: An intracranial electrophysiology study

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Abstract: According to the predictive coding hypothesis, sensory processing occurs through bidirectional information flow. The local/global deviant paradigm engages auditory predictive coding over short ('local deviance'; LD) and long ('global deviance'; GD) time scales. Intracranial electroencephalography (iEEG) allows information flow within the human auditory cortical hierarchy to be examined with high spatiotemporal resolution. LD and GD effects differ in their sites of origin, timing, and sensitivity to general anesthesia. Recent findings indicate that LD and GD effects are not independent; their interactions reflect overlapping processing networks that subserve novelty detection. This study examined local-by-global interactions using iEEG in 7 neurosurgical patients (3 female) who performed a GD target detection task before and during induction of general anesthesia with propofol. Recordings were made from auditory cortex, surrounding auditory-related areas, and prefrontal cortex. High gamma (70-150 Hz) activity was used to measure the interaction between responses to short- and long-term auditory novelty, defined as (LDGD-LSGD)-(LDGS-LSGS) where LS and GS denote local and global standard conditions respectively. In the awake state, subjects detected LSGD targets less accurately than LDGD targets. There were two types of neural interactions. Positive interaction corresponded to a greater response to LDGD than LSGD above and beyond LD effect. Negative interaction corresponded to a greater response to LSGD. Positive interaction was more frequent and robust than negative and was largely confined to temporal cortex. Negative interaction typically occurred in prefrontal and temporo-parietal auditory-related cortex and had longer onset latencies. Sedation and subsequent loss of responsiveness led to a progressive reduction in high gamma interactions. General anesthesia had a greater effect on negative interaction. These results show GD to be more behaviorally salient when paired with LD feedforward error signals, accompanied by a positive neural interaction throughout auditory cortex. The behaviorally more challenging LSGD target condition is associated with a negative neural interaction in prefrontal cortex. Temporo-parietal auditory-related cortex exhibits both types of interaction. Positive interaction is partially preserved during sedation and thus may serve as a biomarker of conscious perception in the evaluation of depth of anesthesia and disorders of consciousness.

Disclosures: K.V. Nourski: None. **M. Steinschneider:** None. **A.E. Rhone:** None. **R.N. Mueller:** None. **H. Kawasaki:** None. **M.I. Banks:** None.

Poster

P444. Auditory Processing: Arousal State

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Topic: D.06. Auditory & Vestibular Systems

Support: NIH GM109086
NIH DC004290

Title: Graph theoretic measures indexing arousal state transitions during sleep and anesthesia in human subjects

Authors: *D. CAMPBELL[1], B. M. KRAUSE[3], D. G. BERGER[2], C. K. KOVACH[4], K. V. NOURSKI[5], M. I. BANKS[6];
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Abstract: Introduction: Consciousness is associated with a balance between integration and differentiation in cortical networks, but measurements of changes in these properties during sleep and anesthesia using graph-theoretic analyses have been inconsistent. Increases in network segregation [measured as network modularity or local efficiency (E_L)] have been reported during NREM sleep, while decreases have been reported under anesthesia. Decreased integration (measured as global efficiency, E_G) is reported under anesthesia, but whether this also occurs for sleep is unclear. We investigated these issues using diffusion map embedding (DME) applied to resting-state functional connectivity derived from intracranial electroencephalography (iEEG) in neurosurgical patients. DME provides a geometric framework for understanding changes in network structure. Segregation is reflected in the clustering of nodes in embedding space, quantified by the effective dimensionality (D_E) of the normalized connectivity matrix. Integration is reflected in the volume of the data cloud in embedding space.
Methods: iEEG recordings from adult neurosurgical patients to identify seizure foci were obtained during propofol anesthesia (N=15) and natural sleep (N=13). Three arousal states were compared during anesthesia experiments: pre-drug wake (WA), sedated/responsive (S) and unresponsive (U), and five in sleep: wake (WS), N1, N2, N3, and REM. These 8 arousal states were segregated into conscious {WA, S, WS, N1, REM} and reduced consciousness {U, N2, N3} states. Data were segmented (60 sec) and channel-by-channel adjacency matrices were computed as pairwise gamma (30-70Hz) orthogonalized power envelope correlations. Connectivity matrices were thresholded (top 33%) and normalized, and graph-theoretic metrics [D_E, E_G, E_L (or equivalently clustering coefficient, C)] computed and compared across arousal state.
Results: During U, N2, and N3 compared to conscious states, E_G was slightly elevated, C and E_L were substantially higher, and D_E was reduced. D_E and C best tracked transitions in awareness between conscious and reduced consciousness states. The embedding volumes were lower during the reduced consciousness states, reflecting the decreased network integration associated with unconsciousness.
Conclusions: Consistent changes in graph-theoretic measures of network organization during sleep and anesthesia suggest overlapping mechanisms of arousal state transitions. The geometric framework of DME facilitates comparisons of large cortical networks across arousal states.

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Poster

P444. Auditory Processing: Arousal State

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Program #/Poster #: P444.08

Topic: D.06. Auditory & Vestibular Systems

Support: NIH GM109086
NIH DC004290

Title: Changes in the functional geometry of human cortical networks across arousal states

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Abstract: Introduction: Leading theories of consciousness predict that loss of consciousness (LOC) under sleep and anesthesia is precipitated by reorganization of brain networks critical for consciousness. Diffusion map embedding (DME) maps data into a space in which distance represents similarity in connectivity to the rest of the sampled network, and has been used previously to demonstrate the hierarchical structure of cortical networks and to track changes in that structure in response to treatments, disorders, and aging. We investigated changes in cortical networks across arousal state using DME applied to functional connectivity derived from resting state intracranial electroencephalography (iEEG) recordings in neurosurgical patients.
Methods: Resting state intracranial recordings were obtained during induction of propofol anesthesia or during sleep from adult neurosurgical patients implanted with electrodes (propofol: n = 15 total; sleep: n = 13 total) in temporal, parietal, and frontal cortex to identify epileptic foci. Three arousal states were compared during anesthesia [pre-drug wake (WA), sedated/responsive (S) and unresponsive (U)] and five in sleep [wake (WS), N1, N2, N3, and REM]. Channel-by-channel adjacency matrices were computed as pairwise gamma (30-70Hz) orthogonalized power envelope correlations. Adjacency matrices were thresholded (top 33%) and normalized to yield the transition probability matrix **P**, which was analyzed using DME. The eigenvalue spectrum of **P** ($|\lambda_i|$) was used to calculate effective dimensionality (estimated as the quadratic entropy: $(\Sigma \lambda_i)^2/(\Sigma \lambda_i^2)$), a measure of network redundancy, and pairwise distances in the embedding space were calculated and compared across states.
Results: Embeddings in states involving reduced consciousness (U, N2, N3) had lower effective dimensionality than corresponding states (S/WA and N1/WS). REM sleep had similar effective

dimensionality to WS. Most node pairs showed reduced distances in the U, N2, and N3 states compared to S/WA or N1/WS, while there was little difference between WA and S, WS and N1, or WS and R. No individual brain region was responsible for these changes, suggesting a global rather than centralized functional reorganization with LOC. More variation in effective dimensionality was observed in U, N2, and N3.
Conclusions: The effective dimensionality of diffusion map embeddings robustly indexes changes in consciousness, and these changes occur across both sensory and higher-order brain regions. Variability of effective dimensionality may reflect fluctuations in arousal in states of reduced consciousness.

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Poster

P529. Reaching Control: Motor Learning: Human Psychophysics II

Location: Virtual Only

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Program #/Poster #: P529.01

Topic: E.04. Voluntary Movements

Support: NSF Grant 1823889

Title: Quantitative measures of task similarity to assess transfer in motor learning

Authors: L. NELSON[1], L. A. MROTEK[1], M. CASADIO[2], R. A. SCHEIDT[1], *R. RANGANATHAN[3];
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Abstract: It has long been recognized that one of the fundamental aspects that influences transfer of motor skills is the similarity between the two skills. However, task similarity has been difficult to define quantitatively in most motor tasks, especially when they involve coordination of multiple degrees of freedom. The goal of this study was to examine the effect of task similarity on transfer of motor learning. We use a novel experimental approach that allows a clear quantitative measure of similarity between coordination patterns, based on the dimensional overlap of coordination patterns. Participants (N = 36) wore a data glove, and the 19 signals from the glove were linearly mapped onto the 2-D position of a cursor on a display screen. The participants were asked to move their fingers to guide the cursor to specific targets as quickly and accurately as possible. The coordination required to perform this task was critically dependent on the 'map' between the glove signals and the cursor motion. To examine transfer, we used a design where participants initially practiced different versions of the map (i.e., the 'prior' task) depending on group assignment. However, halfway through the experiment, all

participants switched to using a common map (i.e., the 'criterion' task). Participants were split into three groups based on the amount of overlap between the prior and criterion tasks - (i) 0% overlap, (ii) 50% overlap and (iii) 100% overlap. Participants performed a total of 480 trials (240 in the prior task, 240 in the criterion task), and a final block of 60 trials on a set of different targets to examine generalization. Results show that the amount of transfer to the criterion task was influenced by task similarity - i.e., the 0% overlap group showed the highest movement times in the criterion task when compared to the 50% or 100% overlap groups. Moreover, this difference between the 0% and 100% groups was seen until the end of the criterion task and in the generalization block, indicating that the interference from the prior task was prolonged. These results not only highlight the importance of task similarity, but also provide a way to systematically manipulate task similarity to fully understand its role in motor transfer.

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Poster

P529. Reaching Control: Motor Learning: Human Psychophysics II

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Program #/Poster #: P529.02

Topic: E.04. Voluntary Movements

Support: NSF 1823889

Title: Bowing a re-strung violin: a real-world test of motor re-learning

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Abstract: • Motor re-learning is central to performing arts, athletics and movement rehabilitation after neuromotor injury. Motor re-learning involves reorganizing movement coordination patterns to find alternate solutions in the presence of a previous solution. We partnered with the Chicago Suzuki Institute to explore how task similarity influences motor re-learning in the context of violin bowing, a skill that requires many hours of practice to learn. We used violins with either a standard string arrangement (CONTROL: G-D-A-E) or with 1 of 2 alternate arrangements (INVERTED: E-A-D-G; SHUFFLED: A-G-E-D) to explore re-learning of a familiar motor task. Whereas INVERTED and SHUFFLED violins are both novel, only the

INVERTED and CONTROL violins share identical neighbor relationships between the strings, making them more similar. The SHUFFLED violins shares no neighbor relationships with either the CONTROL or INVERTED violins.
• 51 participants with prior experience playing the violin repeatedly played a 13-note G-scale arpeggio at 140 beats/minute. The first 10 arpeggio trials were performed with a CONTROL violin ("baseline"). The next 30 "test" trials were performed with either an INVERTED (N=19 participants), SHUFFLED (N=21), or CONTROL violin (N=11). The last 10 "washout" trials were performed with the CONTROL violin. We recorded audio at 10000 samples/s and analyzed 3 performance measures (number of notes played per trial; accuracy and variability of note timing) to test the hypothesis that the rate and extent of motor re-learning depend systematically on the degree of similarity between the prior (CONTROL) and novel (INVERTED or SHUFFLED) tasks.
• The INVERTED and SHUFFLED groups demonstrated motor re-learning for each of the 3 dependent variables. Average performances in the first 5 trials of the test block were more highly degraded in SHUFFLED vs. INVERTED, which were in turn worse than CONTROL. By the end of the test block, SHUFFLED still attempted more notes and played them more unevenly than either INVERTED or CONTROL groups, which did not differ from each other. We conclude that the rate and extent of motor re-learning does indeed depend on the degree of similarity between novel and prior tasks. Interestingly, the INVERTED group increased the mean number of notes across the first 5 test arpeggios whereas performance for the remaining test arpeggios declined back toward baseline values, suggesting that participants likely used a pattern of exploration and exploitation to solve the re-learning problem posed by the manipulated violins.

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Poster

P529. Reaching Control: Motor Learning: Human Psychophysics II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P529.03

Topic: E.04. Voluntary Movements

Title: Lack of beneficial effects of moderate-intensity aerobic exercise on visuomotor adaptation and savings

Authors: *R. JAMES, J. WANG;
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Abstract: Acute exercise performed prior to training is thought to have a positive impact on motor skill acquisition and memory retention. In young adults, exercise is linked with biological and behavioral changes due to increased cortical excitability via long-term potentiation (LTP)-like mechanisms. It has also been shown that a single bout of moderate-intensity aerobic exercise can enhance visuomotor adaptation, though not interlimb transfer (Neva et al. 2019). However, the effects of a single bout of moderate-intensity aerobic exercise on aftereffects and savings (faster when individuals are re-exposed to a previously experienced condition) following visuomotor adaptation are unknown. In this study, thus, 16 neurologically intact right-handed young adults completed two conditions (25 minutes of rest and cycling counterbalanced across subjects) on two separate days with a minimum washout period of 14 days between the two conditions (rest vs. exercise). The exercise or rest condition was followed by an adaptation session with the right hand, in which the visual display was rotated 30° counterclockwise about the start circle; then it was followed immediately by washout and readaptation sessions with the same arm. Results indicated similar patterns of adaptation, aftereffects, and savings between the two conditions. These findings suggest that a single bout of aerobic exercise (cycling) before visuomotor adaptation may not be sufficient to enhance visuomotor adaptation or savings in young adults. Further work is needed to determine the effects of aerobic exercise on visuomotor adaptation.

Disclosures: R. James: None. **J. Wang:** None.

Poster

P529. Reaching Control: Motor Learning: Human Psychophysics II

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Program #/Poster #: P529.04

Topic: E.04. Voluntary Movements

Support: NIH Grant HD055931

Title: Bayesian statistical modeling of the relationships between error size, proprioception, and the magnitude of implicit adaptation

Authors: *H. E. KIM[1], J. S. TSAY[2];
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Abstract: Bayesian statistics provides a unified framework for combining prior knowledge with data in order to compute joint probability distributions of different world states (i.e., conjectures). Unlike frequentist statistics, Bayesian methods can readily quantify support for null hypotheses and be combined with graphical models to aid causal inference. Here, we leverage these two advantages of Bayesian statistics to reanalyze, and reinterpret, some of our recent published studies on implicit motor adaptation. We recently reported that implicit motor

adaptation in reaching converged to a common asymptote in response to a wide range of error sizes (1.75° - 15°) (Kim et al 2018). Instead of a dose-dependent response, group-averaged motor aftereffects in response to these errors fell within a surprisingly narrow range of 23°-27°, with no reliable differences detected by a frequentist ANOVA. While originally interpreted as evidence of convergence in asymptotic adaptation, upon re-analyzing these data using a Bayesian linear model with weakly informative priors, only 39-49% of the credible differences in asymptotic adaptation fell within a region of practical equivalence (ROPE) of ±3°. That is, while our Bayesian re-analysis revealed only small expected differences in the asymptotic levels of adaptation (1.2° - 3.4°), due to high uncertainty around these estimates there was slightly more evidence for divergence than convergence in asymptotic adaptation, contrary to our original interpretation. We also recently reported a novel link between measures of proprioception (variability and recalibration) and implicit adaptation (Tsay et al 2021). When re-analyzing these data using directed acyclic graphs and Bayesian regression models, we found strong relationships between both proprioception variables with the asymptotic level of adaptation, with over 99% of credible slope estimates having non-zero values. The results of our analyses bolster our original claim and imply a causal relationship between proprioception and the extent of implicit adaptation. Taken together, our application of Bayesian statistics has sharpened our inferences regarding previously published results and demonstrated the merits of this approach in assessing the credibility of different hypotheses. The clear alignment between research question and inference that Bayesian methods afford motivate moving beyond frequentist null hypothesis testing and p-values.

Disclosures: H.E. Kim: None. **J.S. Tsay:** None.

Poster

P529. Reaching Control: Motor Learning: Human Psychophysics II

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Program #/Poster #: P529.05

Topic: E.04. Voluntary Movements

Support: 418589

Title: The counterintuitive effects of overlearning on subsequent learning (A - A)

Authors: *E. DE LA FONTAINE[1], R. HAMEL[2], J.-F. LEPAGE[3], P.-M. BERNIER[1];
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[3]Pediatrics, Sherbrooke Univ., Sherbrooke, QC, Canada

Abstract: Although overlearning (i.e. extending practice at performance asymptote) has been known to enhance retention (Postman, 1962), most studies have examined its effect on retention (Krueger, 1929; Rohrer et al., 2005) or subsequent learning of a different task (A->B; Shibata et

al., 2017). The latter argues that practice at performance asymptote increases neural inhibitory activity, stabilizing and protecting the newly acquired memory but more importantly, making the system refractory to subsequent learning (i.e. detrimental effect of prior learning on future learning; A->B). Interestingly, recent work from our laboratory (Hamel et al., 2021) reported that, if constrained into a close temporal window, learning twice the same task (A->A) also leads to anterograde interference. This leads to a re-evaluation of the classic finding that exposure to a perturbation necessarily leads to faster re-adaptation the second time (i.e. savings; Krakauer, 2009). The objective of this work was to elucidate if increasing the duration of practice would increase savings and lead to faster re-adaptation, or on the contrary would increase the degree of anterograde interference. In a fully within-subject and counterbalanced design, four conditions were carried out where participants (n = 24) adapted to a gradually introduced 20° visual deviation while the length or type of the performance asymptote were manipulated. The performance asymptote at 20° was either Short (40 trials), Moderate (160 trials), Long (320 trials) or kept on changing around 20° for 160 trials (Jagged). Following a 2 min break, participants adapted to the same visual deviation and practiced at performance asymptote for 40 trials. Immediate retention was assessed through reach aftereffects in the absence of corrective visual feedback. Results revealed that increasing the duration of asymptote enhanced immediate retention. Interestingly, however, increasing the duration of asymptote from Short to Moderate also increased anterograde interference. There was no difference between Moderate and Long, suggesting the presence of a ceiling effect. One interpretation is that the increased anterograde interference is prompted by the accumulation of adenosine due to the energy consuming metaplasticity of sensorimotor adaptation.

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Poster

P529. Reaching Control: Motor Learning: Human Psychophysics II

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Program #/Poster #: P529.06

Topic: E.04. Voluntary Movements

Support: JSPS-JP16H06566

Title: Static and dynamic gaze states are separately linked with motor learning of reaching movements

Authors: *N. ABEKAWA, H. GOMI;
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Abstract: Learning novel motor actions depends on forming and updating internal models. It has been a critical issue in neuroscience how the brain represents internal models with regard to their

association with spatial frames. We recently used a context-dependent learning paradigm to examine the involvement of gaze-related spatial coding in the learning of reaching movements (Abekawa et al. 2019, SfN Abstract). We focused on two types of eye-hand coordination: reach to a target while looking at the target (Foveal-reach: FOV) and while looking elsewhere (Peripheral-reach: PER). The results revealed that two opposing motor skills (e.g. different visuomotor rotations) can be learned simultaneously, which usually interfered with each other, when each skill was associated with FOV and PER. This suggests that there is a link between static representation about eye-hand coordination and internal models of reaching. Meanwhile, reaching movements are usually accompanied with eye movements in our natural behaviors. Here we investigated whether such dynamic chain of eye-hand movements is engaged in the learning of reaching. In the experiment, participants (n=12) performed reaching movements under three different eye-hand conditions presented randomly across trials. In the first two static conditions, as in our previous study, participants reached to the target while looking at a fixation point (FOV and PER). In the last dynamic condition, the trial started in the same way as in PER, but the fixation jumped to the target location before reaching was initiated. Participants had to make a saccade as quickly as possible, and then perform FOV (Sac-Reach). We applied visuomotor rotation with its direction varied across trials, but was uniquely associated with the static or dynamic condition. For half of the participants, CW rotation was applied in FOV and PER, and CCW rotation was in Sac-Reach (For the other half, CCW was in FOV and PER, and CW was in Sac-Reach). The results showed that in all three conditions, the reaching error decreased throughout the training session (1200 trials), with clear aftereffects in the post-learning session where the rotation removed. The control experiment, in which the fixation jump was given without making saccades, showed no progress in learning, especially for the Sac-Reach condition. These results indicate that saccade before reaching can work as the context for motor learning. Taken together, static and dynamic gaze states that are chained with reaching behavior would activate distinct neural states in the sensorimotor pathway, which can be inherently linked with forming and retrieving internal models of reaching.

Disclosures: N. Abekawa: None. **H. Gomi:** None.

Poster

P529. Reaching Control: Motor Learning: Human Psychophysics II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P529.07

Topic: E.04. Voluntary Movements

Support: NS116883

Title: Contextual interference in implicit sensorimotor adaptation

Authors: *C. IRVING, R. IVRY, J. S. TSAY;
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Abstract: A widely observed phenomenon in skill acquisition is the contextual interference effect: motor skills are learned slower but show better retention when learning takes place under a random practice schedule compared to a blocked practice schedule. A common account of contextual interference is that random practice engages greater attentiveness and more deliberate top-down control, leading to more elaborate and accessible memories. Whether the principles of contextual interference are relevant for implicit sensorimotor adaptation remains unknown.
To explore this question, we used a method that isolates implicit motor adaptation (clamped feedback task), instructing participants to reach to one of three target locations, each paired with a different sized visual clamp, feedback in which the radial position was aligned with the hand but angular position that was rotated by a constant 30º, 45º, or 60º from the target. Participants were divided into two groups: Random Training in which reaches to the three targets were interleaved in a single block of 270 trials, or Blocked Training where each target was tested in a serial manner across three blocks of 90 trials each. Retention was assessed in a no feedback washout block in which, for both groups, the three targets were interleaved. Consistent with previous studies, participants exhibited an implicit and gradual drift in hand angle in the opposite direction of the visual error. The magnitude of this effect was approximately 20 degrees and near asymptotic value by around 100 trials for the Blocked group and 180 trials for the Random group (per target location). A pronounced contextual interference effect was found: Random Training led to slower changes in hand angle during the training block yet a larger aftereffect in the washout block.
Randomizing the targets in the washout phase introduces a context change for the Blocked group. To assess the relevance of this factor, we conducted a second experiment in which the targets were blocked in the washout block. The magnitude of the aftereffect decreased for the second and third locations probed, presumably due to passive decay. Nonetheless, the retention advantage persisted for the Random training group. In a final experiment, aftereffects were probed after each training block in a Blocked group and compared to the aftereffect probed at the end of a single training block for a Random group. Again, random training led to stronger aftereffects.
These results demonstrate that the principles of contextual interference are not only applicable to the development of motor skills demanding explicit control, but also extend to the way we implicitly recalibrate our movements.

Disclosures: C. Irving: None. **R. Ivry:** None. **J.S. Tsay:** None.

Poster

P529. Reaching Control: Motor Learning: Human Psychophysics II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P529.08

Topic: E.04. Voluntary Movements

Support: FRIA (F.R.S.-FNRS, Communauté française de Belgique)

Title: Online control influences visual and proprioceptive contributions to motor adaptation

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Abstract: To date, little is known about how the brain integrates multisensory signals in dynamic contexts, such as during a movement. Here, we investigated the contribution of visual and proprioceptive error feedback as well as their potential interaction on online control and motor adaptation in two frequently used reaching paradigms. We conducted two experiments (N=14 and N=19) on healthy volunteers aged 20 to 34. Participants performed visually guided reaches with a robotic manipulandum while their arm was hidden from view. We randomly applied clockwise or counterclockwise velocity-dependent force-fields in combination with visual cursor shifts. These perturbation trials were interleaved with baseline trials to measure single-trial adaptation by extracting the amplitude of the after effect in the baseline trials. Importantly, to investigate the influence of online control on adaptation we additionally manipulated the instruction to participants asking them to either correct or not correct their movement when encountering a perturbation. We recorded movement kinematics as well as EMG signals from shoulder and elbow muscles. For our analyses we defined proprioceptive error and adaptation as the maximum limb displacement on perturbation and subsequent baseline trials, respectively. The size of the visual error was quantified as the gain applied to shift the visual cursor relative to the true hand position. We observed a scaling of muscle responses with the size of the visual error resulting in a decrease of proprioceptive error with increasing visual error. Using a linear mixed effect analysis, we further found that adaptation was negatively related to the size of the proprioceptive error but showed no consistent relationship with visual errors across the two experiments. Visual and proprioceptive errors exhibited no interaction in their contribution to adaptation. Importantly, in the second experiment we observed an influence of the task instruction on the relationship between sensory errors and adaptation. Our results demonstrate that the interplay between different sensory errors shapes feedback responses during a movement which in turn influences how errors are mapped into updates for the next trial. Thus, explaining the integration of multisensory signals for motor adaptation requires close attention to how task instructions influence participants' engagement with different sources of sensory feedback. Ultimately, understanding if and how sensory integration rules differ between online control and adaptation will be an important step towards extending models of multisensory integration to dynamic contexts.

Disclosures: A. Hoffmann: None. **F. Crevecoeur:** None.

Poster

P529. Reaching Control: Motor Learning: Human Psychophysics II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P529.09

Topic: E.04. Voluntary Movements

Support: NSERC PDF 546022 2020
BrainsCAN

Title: The influence of action observation on fast feedback control

Authors: N. MANGOS[1], *C. FORGAARD[1], A. PRUSZYNSKI[2], P. L. GRIBBLE[1];
[1]Brain and Mind Inst., [2]Physiol. and Pharmacol., Western Univ., London, ON, Canada

Abstract: Many sensorimotor areas of the brain are engaged while watching others move. This "action observation network" underlies an important mechanism by which humans often learn motor skills: through the observation of a tutor (Mattar & Gribble 2005). Studies to date have focused on how movement observation influences voluntary (feedforward) movement control. Here we tested whether movement observation also influences feedback-based control. When external forces (mechanical perturbations) are abruptly applied to the arm, short- (SLR; 20-50 ms) and long-latency (LLR; 50-100 ms) feedback responses are elicited in the stretched muscles. While the SLR is generated purely by spinal circuitry, the LLR is produced by a transcortical pathway involving primary sensorimotor cortex (Pruszynski et al. 2011). Due to the cortical contributions, the LLR displays many similarities with feedforward control (Forgaard et al. 2021). The purpose of this study was to test whether action observation can also influence the LLR. Sixteen participants performed reciprocal elbow flexion/extension (30 deg) movements in a horizontal plane with strict timing constraints (0.83–1 Hz) using a KINARM exoskeleton robot. Participants then observed a video of an expert performing the same task. During observation participants maintained their right hand in a home target against small background loads that pre-activated elbow flexor (biceps and brachioradialis) or extensor (triceps) muscles. At two different time points in the expert's movement cycle (peak elbow flexor and peak elbow extensor activity), unpredictable torque perturbations were applied at the elbow joint to evoke feedback responses in participants' elbow muscles. Participants were instructed to not intervene in response to the perturbations and to remain focused on the video. The mechanical perturbations successfully evoked an SLR and an LLR in elbow muscles; however, compared to a control condition in which participants observed the expert sitting still with no movement of their arm, neither the SLR nor the LLR was facilitated by observing the expert moving. We did observe a 12% decrease in the brachioradialis LLR at the time point in the video when the expert's antagonist muscle (triceps) was most active, compared to the static control condition. These preliminary findings thus suggest that action observation has a negligible or a weak inhibitory influence on feedback responses.

Disclosures: N. Mangos: None. **C. Forgaard:** None. **A. Pruszynski:** None. **P.L. Gribble:** None.

Poster

P529. Reaching Control: Motor Learning: Human Psychophysics II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P529.10

Topic: H.10. Human Learning and Cognition

Support: T32 NS047987-04

Title: Transfer of implicit perceptual-motor sequence knowledge across spatially-unique cue colors and shapes

Authors: *P. SHU, R. CHEN, Y. HAN, C. MCROBERT, P. REBER;
Northwestern Univ., Evanston, IL

Abstract: Implicit learning, which is considered fundamental to skill acquisition, has been demonstrated to be largely inflexible. When knowledge acquired in one context cannot be expressed when perceptual conditions have been altered, we infer that these perceptual elements have become part of the learned, inflexible representation.Here we report studies measuring transfer across changes in cue color and shape within a perceptual-motor sequence learning paradigm, the Serial Interception Sequence Learning (SISL) task. In the SISL task, moving cues scroll down the screen to signal a precisely-timed motor response corresponding to the spatial location of the cue. Participants learned a covertly-embedded 12-item sequence during training before completing a test of sequence knowledge under the same condition and transfer conditions. Transfer conditions only changed perceptual aspects of the moving cue irrelevant to the motor response and the motor sequence was constant across all test conditions. In Experiment 1 (n=30), participants completed four blocks of training with one cue shape (circle or square) for all four locations and then completed tests with the trained cue shape, untrained cue shape, and four novel, unique shapes. Robust transfer to the novel shape conditions was observed , indicating that cue shape was not seemingly part of the sequence representation .In Experiment 2 , participants practiced the repeating sequence with either four different cue colors (n=30) or shapes (n=25), each assigned to a specific spatial location. Transfer of sequence knowledge was tested with four completely novel spatially-unique colors/shapes or with the same trained colors/shapes re-assigned to different locations. Transfer was particularly impaired when the trained colors/shapes were re-assigned, $F(2,106)=10.82$, $p<.001$, indicating significantly less expressed sequence knowledge when the specific association between perceptual elements and spatial locations was disrupted at test. The results of the second experiment indicate that when color or shape are uniquely associated with spatial location, this perceptual information becomes part of the sequence representation and interferers with transfer when the cue location is re-associated with a novel feature during test. In contrast, when the color/shape is consistent across locations during training, sequence knowledge can fully transfer to novel perceptual conditions.

This implies that there is no simple rule for what context features become part of the learned representation but rather that implicit learning is sensitive to whether perceptual information is predictive of cued responses.

Disclosures: P. Shu: None. **R. Chen:** None. **Y. Han:** None. **C. McRobert:** None. **P. Reber:** None.

Poster

P529. Reaching Control: Motor Learning: Human Psychophysics II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P529.11

Topic: H.10. Human Learning and Cognition

Support: T32 NS047987-04

Title: Examining the time course of forgetting for implicitly learned auditory perceptual-motor sequence knowledge

Authors: *Y. C. HAN, P. J. REBER;
Northwestern Univ., Evanston, IL

Abstract: Implicit statistical knowledge, which is thought to underlie skill learning, has been demonstrated to be resistant to forgetting over time and may be forgotten more slowly. Compared to the well-studied explicit forgetting curve, which demonstrates a steep exponential decline over time, the rate at which implicit memory decays is hypothesized to follow a similar, though shallower, decay curvature. Implicit knowledge retention has primarily been studied in visually-based implicit learning paradigms. Here, we report a study providing a quantitative measurement of the forgetting curve for an auditory-based implicit learning task, which may provide insight into memory decay associated with domains such as statistical aspects of language. To measure the rate at which auditory implicit knowledge decays, we utilized the auditory Serial Interception Sequence Learning (SISL) task, which produces a robust, quantitative, implicit learning signal that allows for measuring performance across a range of time points after acquisition. Auditory cues signaled precisely-timed motor responses based on cue pitch, with keys D, F, J, and K mapped to distinct starting frequencies. Motor responses were timed to the third onset of a three-cue sequence. During training, participants were not informed that there was a covertly embedded repeating sequence of tones (80% of trials). Knowledge of this sequence was quantitatively assessed during a test (33% sequence; 67% foils) by the increased task accuracy for cues within the repeating sequence over foils (SSPA). After completing a training and test phase, participants were randomly assigned to complete a second test after delay periods of 11 hours (n=20), 24 hours (n=17), or one week (n=21). Group-level retention of sequence-specific knowledge was measured as the percentage of SSPA retained

from first to second test for participants who demonstrated robust learning during the first session. A linear regression indicated log-transformed hours significantly predicted retention amount ($F(1,57)=20.6$, $p<.001$, $R2=.25$) across delay intervals (11h, M=73.5%, SE=17.1%; 24h, M=45.9%, SE=11.6%; 1 week, M=63.9%, SE=11.3%). The regression-estimated log-linear implicit forgetting rate follows a fairly shallower curve (slope = -9.6%), suggesting that implicit knowledge decays at slower rate and would persist potentially for years. The forgetting rate for implicit learning estimated here was considerably shallower than classic studies of explicit forgetting (e.g., Ebbinghaus, 1885, log-linear slope = -12.4%), consistent with our hypothesis that implicitly acquired knowledge decays much more slowly than explicit memory.

Disclosures: Y.C. Han: None. **P.J. Reber:** None.

Poster

P529. Reaching Control: Motor Learning: Human Psychophysics II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P529.12

Topic: D.06. Auditory & Vestibular Systems

Support: The Australian Research Council - DP180103081

Title: Audiomotor adaptation

Authors: *B. MILLER-MILLS[1], K. MCANALLY[2], L. LEOW[3], B. F. KEANE[3], P. GROVE[4], T. J. CARROLL[3];
[1]Sch. of Human Movement and Nutr. Sci., Univ. of Queensland, Brisbane, Australia; [3]Sch. of Human Movement and Nutr. Sci., [4]Sch. of Psychology, [2]The Univ. of Queensland, Brisbane, Australia

Abstract: Sensorimotor adaptation has been extensively studied in the visual domain, but the degree to which altered sensorimotor mappings occur in modalities other than vision remains less well understood. Here we manipulated the modality of stimulus presentation to compare motor updates in response to perturbations of visual and auditory feedback location. We compared a) the extent of learning from perturbed sensory feedback for the visual and auditory sensory modalities and b) the magnitude of reach direction aftereffect when the perturbation was removed. Errors related to violations of expected sensory outcomes (sensory prediction errors), as well as errors related to the successful completion of a clearly defined goal (task errors), have been shown to drive motor adaptation. Thus, in order to isolate the contribution of sensory input during motor adaptation, we held sensory prediction errors and task errors constant via a task-irrelevant clamp of sensory feedback. In this paradigm, visual or auditory feedback of the reach endpoint was systematically displaced, but participants were informed of the perturbation and instructed to ignore it and reach directly toward the target. These experiments involved target

and reach angle feedback presented by loudspeakers (Experiment 1), headphones (incorporating head-related transfer functions) (Experiment 2) and a visual display (Experiment 3). Although we appropriately matched auditory location cues between the loudspeaker and headphone conditions, participants of the loudspeaker condition showed substantially greater reach accuracy than participants of the headphone condition during a familiarization block where reaches were made to seven targets separated by 20 degrees. The headphone condition also showed greater adaptation with respect to baseline than the loudspeaker condition (Exp 1 M = 4.4° ± 9, Exp 2 M = 16° ± 21.6). In Experiment 3, the visual stimuli were presented under conditions similar to those of the auditory experiments; participants saw only a transient visual cursor following a given reach displayed in either the target location or the perturbed location. Consistent with our predictions and previous literature, a significant change in reach direction was observed (Exp 3 M = 3.1° ± 4.8), with an aftereffect that persisted when participants were instructed that the perturbation had been removed. This study provides new evidence that motor adaptation occurs in response to perturbed feedback of reach endpoint across sensory modalities. It remains an open question if these visual and auditory motor adaptations are dependent upon underlying processes that are shared or independent.

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Poster

P543. Neurophysiology: Non-Invasive Techniques I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P543.01

Topic: E.05. Brain-Machine Interface

Support: Kahn, Dan D. and Betty, Foundation
Israel Ministry of Science and Technology

Title: Error related potentials in response to erroneous movements generated by Brain Computer Interfaces for hand control

Authors: I. DEMCHENKO, M. BENYAMINI, *M. ZACKSENHOUSE;
Tech. IsraelInstTech, Haifa, Israel

Abstract: Background: In recent years, brain computer interfaces (BCIs) studies and applications, which provide direct communication between the brain and the computer, have grown rapidly. A widely used technique for measuring brain activity is electroencephalogram (EEG). EEG is a relatively inexpensive, easy to use, and provides a high-temporal resolution. BCI for closing\opening the hand are very important for comfortable and intuitive brain-controlled hand prosthesis and may help 3 million upper limb amputees worldwide. Recently it

has been shown that Error related potentials (ErrP), which are evoked as a reaction to erroneous hand movements significantly improve the final decision of a movement.
Methods: Here we investigate ErrPs in response to different erroneous BCI decisions of closing\opening hand.
Experiment design: Participants controlled two virtual hands that appeared on a computer screen by moving their hands (hand control). The posture of the hand was determined by bend-sensitive sensors embedded in the gloves wore by participants. A ball was places in front of each virtual hand and participants were asked to close/open their hands if the color of the ball changed from red to green or from green to red, respectively. Participants were asked to maintain the hand posture if the color did not change. Data collected during hand controlled was used to train a convolutional neural network (CNN) BCI-classifier, which was used to control with virtual hands in the second part of the experiment. The experiment was designed to investigate the response to three types of errors: 1. Virtual hands-maintained posture even though participant changed the posture of one hand. 2. Virtual hand changed posture even though participant-maintained hands' posture. 3. The wrong virtual hand changed posture.
Results: The accuracy of the CNN-based BCI classifier for 3 different options (change the posture of the right or left hand, or maintain posture) was between 50 to 60 % for all participants. The three types of errors evoked ErrPs with different amplitudes, with largest amplitude in response to the second error-type and smallest for the first error-type.
Conclusions: ErrPs can be detected while using EEG-based BCI to control hand closing/ opening. ErrPs evoked in response to different error-types have different latency and amplitudes.

Disclosures: I. Demchenko: None. **M. Benyamini:** None. **M. Zacksenhouse:** None.

Poster

P543. Neurophysiology: Non-Invasive Techniques I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P543.02

Topic: E.05. Brain-Machine Interface

Support: NSF/SBE-BSF #1632259
DHHS/NIDILRR 90REGE0005-01
NINDS R041NS053603

Title: A nonlinear body machine interface for controlling assistive robotic arms

Authors: F. RIZZOGLIO[1,3], M. GIORDANO[2], ***F. A. MUSSA-IVALDI**[3], M. CASADIO[4]; [2]DIBRIS, [1]Universita` di Genova, Genova, Italy; [3]Northwestern University/Shirley Ryan Ability Lab., Chicago, IL; [4]Univ. of Genova, Genova, Italy

Abstract: One of the consequences of spinal cord injury is the loss of the ability to coordinate body motions and perform functional movements. Common activities, such as grooming and object manipulation, could be facilitated through the use of assistive robotic manipulators. However, designing interfaces for controlling such devices can be challenging when dealing with the control of many degrees of freedom (DoFs). Here, we propose an approach based on the framework of body machine interfaces (BoMIs). BoMIs exploit the fact that, even after a severe injury, most individuals retain the ability to perform some movement, especially with their head and shoulders. These residual movements, can be used to control external devices. BoMIs typically rely on Principal Component Analysis (PCA) to extract the lower dimensional control space from information in voluntary movement signals, such as as body kinematics and muscle activity. Despite its widespread use, PCA might not be suited for controlling devices with a large number of DoFs, as the variance explained by successive components drops precipitously after the first. As a consequence most signal information is captured by the first two to three PCs.Here, we propose the use of non-linear autoencoder (AE) networks to overcome these difficulties. Not only non-linear AEs are able to estimate the dimensionality of their input more parsimoniously than PCA, but their structure can also be customized to obtain a variance that is more uniformly distributed across its latent dimensions. The hypothesis of this study is that the customizability offered by AEs makes them an ideal candidate for operating devices with more than two DoFs, such as an assistive robotic arm.To test this hypothesis, we developed a BoMI based on non-linear AEs that mapped kinematic signals recorded from the participants' upper arms into joint angles of a 4D virtual robotic manipulator. First, we performed a validation procedure that aimed at selecting an appropriate AE structure that would allow to distribute the variance uniformly across the dimensions of the control space. Then, we assessed the users' proficiency practicing a 3D reaching task by operating the robot with the validated AE over the course of training across two non-consecutive days.All participants managed to acquire a high level of skill when operating the 4D robot as a consequence of training with the interface. Moreover, they retained the performance they had at the end of the first day while converging towards a more consistent solution over the course of training. We consider these findings as supporting a future implementation of our interface as an assistive tool for people with motor impairments.

Disclosures: F. Rizzoglio: None. **M. Giordano:** None. **F.A. Mussa-Ivaldi:** None. **M. Casadio:** None.

Poster

P543. Neurophysiology: Non-Invasive Techniques I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P543.03

Topic: E.05. Brain-Machine Interface

Title: An adaptive sequence-based stimulus selection in ERP-based brain-computer interface by thompson sampling in a multi-armed bandit problem

Authors: *T. MA[1], J. E. HUGGINS[2], J. KANG[1];
[1]Biostatistics, [2]Physical Med. & Rehabilitation, Biomed. Engin., Univ. of Michigan, Ann Arbor, MI

Abstract: Background: A P300 speller is a non-invasive communication tool for people with severe neuromuscular diseases. It displays a series of stimuli on the computer screen and searches for the elicited P300 event-related potential (ERP) response. The checkerboard paradigm (CBP) is a common stimulus presentation standard [1].
Although a few past studies have proposed data-driven methods for stimulus selection, they suffer from either the intractable decision rule, large computation complexity, or error propagation in real-time use. In addition, no methods have been applied to CBP directly.
Thompson sampling is a heuristic for sequential decision making that addresses the exploration-exploitation dilemma in the multi-armed bandit (MAB) problem. It chooses the optimal action by maximizing the expected reward function with respect to the posterior distribution of the parameters. This Bayesian-like procedure provides a natural framework for data-driven stimulus selection.
Method: We develop a sequence-based algorithm for adaptive stimulus selection by Thompson sampling in the MAB problem with multiple selections.
First, we use the CBP to specify a candidate set of stimulus groups of size 26, where the keyboard contains 84 keys. During each sequence, the algorithm selects a random subset of stimulus groups with fixed size by the posterior probability, aiming to identify all target stimuli and to improve the spelling speed by reducing non-target stimuli. We compute “clean” stimulus-specific rewards from raw classifier scores via the Bayes rule.
We perform simulation studies to evaluate our method, considering the ideal and the practical scenarios. We generate raw classifier scores from two normal distributions with different means but the same standard deviation and record the number of stimuli needed according to two separate criteria: 1) the probability of selecting all target stimuli exceeds 0.9 and, 2) the probability of the target character exceeds 0.9. We compare the results to the conventional offline design. For the practical scenario, we adapt our method to address the observational delay and the refractory effect in real-time BCI use.
Results: For all scenarios, the subset sizes were 3, 8, 13, and 26. In the ideal scenario, we needed 28, 35, 36, and 39 stimuli, respectively, to meet criterion one and 33, 43, 47 and 54 stimuli, respectively, to meet criterion two. In the practical scenario, we needed 33, 43, 141 and 143 stimuli, respectively, to meet criterion one and 36, 46, 148 and 157 stimuli, respectively, to meet criterion two.
References:[1] Townsend, et al. A novel P300-based BCI stimulus presentation paradigm, *Clin Neurophysio*, 121(7), 1109-1120, 2010.

Disclosures: T. Ma: None. **J.E. Huggins:** None. **J. Kang:** None.

Poster

P543. Neurophysiology: Non-Invasive Techniques I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P543.04

Topic: E.05. Brain-Machine Interface

Support: BrainQ Technologies Ltd.

Title: Improvement in motor function with combination therapy of Brain-Computer Interface based Electromagnetic Field Treatment with upper extremity physical therapy in people with chronic cervical spinal cord injury.

Authors: *N. DATTA, D. C. CILIEN, D. CILIEN, K. L. GANT, D. W. DIETRICH;
The Miami Project To Cure Paralysis, Univ. of Miami, Miami, FL

Abstract: In recent years modern, noninvasive brain stimulation (NIBS) techniques have been extensively explored in the arena of motor recovery and neuronal plasticity. However, as a subset of NIBS, electromagnetic field exposure (EMF), and especially extremely low-frequency electromagnetic field (ELF-EMF) exposure is a relatively unprobed field though studies suggest there are beneficial effects of EMF exposure in-vitro, animal models of CNS injuries, and even humans. There are hypotheses purporting regenerative effects of EMF on the spinal cord through modulation of neuroinflammation, stimulation of nerve regeneration, and mesenchymal stem cell proliferation. The aim of this study is to determine the safety and efficacy of a novel brain-computer interface (BCI) based electromagnetic field (EMF) treatment (BrainQ Technologies Ltd., Jerusalem-Israel) in improving function and self-care in persons with chronic SCI. 3 subjects with Chronic (1-year post-injury) Incomplete cervical spinal cord injuries (SCI) received 1 hour treatment sessions of ELF-EMF with BrainQ (thrice a week for 14 weeks) combined with upper extremity physical therapy (PT), after undergoing 4 weeks (12 sessions) of PT alone. Assessments including Graded Redefined Assessment of Strength, Sensibility and Prehension (GRASSP), Modified Ashworth Scale for Spasticity (MAS) and Spinal Cord Independence Measure (SCIM III selfcare) were administered at baseline (week 0), 2nd baseline (after 4 weeks of PT only), and at 4 weeks intervals till end of treatment (week 18 of study). At week 18 of study, analysis showed definite improvement in SCIM III selfcare score in all subjects (3 ± 1.69) with combination therapy as compared to no improvement with PT alone, especially in self-care activities like feeding, grooming and mobility. The change in GRASSP strength sub scores was variable for each subject over the course of the study. There was a gain in quantitative prehension sub scores in all 3 subjects both with PT alone (3± 4.58) and with combination therapy (2.3 ± 1.53) on the dominant side. No change in qualitative prehension or sensibility sub scores were noted either with PT alone or with combination therapy. There was a reduction in spasticity of the wrist in 2 subjects who presented with spasticity at baseline, both at the end of PT alone and after combination therapy. BCI-based EMF treatment in combination with PT is both safe and shows promise regarding improvement in fine motor skills as evidenced

by improvement in SCIM III scores. The ability to perform selfcare with little or no help will significantly improve the quality of life of persons with SCI and warrants further research into ELF-EMF based treatments.

Disclosures: N. Datta: None. **D.C. Cilien:** A. Employment/Salary (full or part-time):; The University of Miami. **D. Cilien:** A. Employment/Salary (full or part-time):; The University of Miami. **K.L. Gant:** None. **D.W. Dietrich:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; BrainQ Technologies Ltd..

Poster

P543. Neurophysiology: Non-Invasive Techniques I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P543.05

Topic: E.05. Brain-Machine Interface

Title: Toward High Gamma (70+ Hz) from Scalp Electroencephalography for Brain-Computer-Interface

Authors: *G. MILSAP, C. SCHOLL, W. COON, M. OGG, E. MCDOWELL, J. WATHEN, P. PERANICH, J. PITTMAN;
Johns Hopkins Univ. Applied Physics Lab., Columbia, MD

Abstract: Brain-computer-interfaces (BCI) that use scalp electroencephalography (EEG) typically operate on signals with <40 Hz bandwidth. This may be due as much to tradition as it is to a number of real limitations, inherent in the physics of EEG signal acquisition, that make higher frequency content less salient. High frequency activation (>70 Hz broadband activity, a.k.a. “high gamma”) was discovered around the turn of the century within electrocorticographic (ECoG) recordings and continues to be one of the most useful neural features in these datasets. While high gamma activity can be easily observed within streaming ECoG data, in some cases without the need for any trial averaging, the signals are very difficult to record at the scalp due to the low signal-to-noise ratio (SNR) of high gamma in traditional EEG recording setups. In a 2008 article, Ball et. al. performed experiments designed to elicit well-characterized and spatiotemporally constrained high gamma activation across many trials recorded in a shielded environment, and demonstrated that high gamma activity is present at the scalp, albeit more spatially distributed and at significantly reduced SNR. The research performed here is an effort to understand the sources of noise that diminish the presence of high gamma at the scalp, and additionally to explore modifications to traditional scalp EEG to optimize detection of high gamma activity at the scalp with less integration time (fewer trials). The ability to distinguish high gamma activity with short integration times may facilitate next-generation BCI technology

based on scalp EEG, which remains one of the least expensive and most extensible non-invasive neural recording modalities. In this research, we characterized the amplitude of task-based high gamma modulation at the scalp, the noise floor present in the recordings, the various sources of noise present in shielded and unshielded environments that contaminate the high gamma frequency range, and iterated our recording technique and post-processing to achieve a modest gain in high gamma SNR at the scalp. We describe our methods and custom recording apparatus, as well as suggest some of the fundamental limitations and alternative approaches to recording high gamma from electrodes on the scalp.

Disclosures: G. Milsap: None. **C. Scholl:** None. **W. Coon:** None. **M. Ogg:** None. **E. McDowell:** None. **J. Wathen:** None. **P. Peranich:** None. **J. Pittman:** None.

Poster

P543. Neurophysiology: Non-Invasive Techniques I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P543.06

Topic: E.05. Brain-Machine Interface

Support: The Republic of Korea's MSIT Grant 2021-0-01537)
IITP Grant 2021R1I1A3060828

Title: A hybrid approach for ERP and ERDS in motor imagery BCI

Authors: *D. GWON[1], M. AHN[2];
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[2]Computer Sci. and Electrical Engin., Handong Global Univ., Pohang, Korea, Republic of

Abstract: A motor imagery (MI) is an imagination of movement and known as producing power change at certain frequencies (e.g., alpha, beta and gamma) which is called event related (de)synchronization (ERD/S) [1]. Since different body limbs involve with different spatial patterns in the brain, this feature is used in building a Brain-Computer Interface (BCI) that decodes user's intention. Indeed, most algorithms in MI-BCI are designed to focus this feature. However, only a few studies pay attention to the lower frequency which may be related to event-related potential (ERP). In this study, we investigate the existence of ERP in motor imagery data and propose a hybrid approach to use both ERP and ERD/S features in classification of motor imagery types (left and right). ERD/S features from common spatial pattern (CSP) [2] and ERP features from Stepwise regression were combined and fed into a linear discriminant analysis (LDA). The average classification accuracy was obtained by 10x10 fold cross validation technique. For this study, we used an open electroencephalogram (EEG) dataset (64 channels, sampling rate 1000 Hz) recorded from 54 subjects [3]. Each subject conducted two motor imagery sessions. As a result, we found the presence of ERP by motor imagery and further the

different spatial pattern of ERP according to imagery types. ERP is likely to be stronger and faster at C3 channel during left MI. This pattern was also observed at C4 during right MI. Statistical test revealed that this spatial difference is significant (Student's T test, Bonferroni correction $p < 0.05$). In the classification analysis, the proposed hybrid approach shows 79.43/80.13% (session 1 and 2 respectively), which were statistically higher (Student's T test $p < 0.05$) than the conventional methods (CSP+LDA: 73.84/75.49% and ERP+SWLDA: 67.08/68.23%). In conclusion, ERP is also important feature to understand motor imagery process and this feature should be included in building an advanced MI BCI. [1]G. Pfurtscheller and F. H. Lopes da Silva, "Event-related EEG/MEG synchronization and desynchronization: basic principles," Clinical Neurophysiology, vol. 110, no. 11, pp. 1842-1857, Nov. 1999, doi: 10.1016/S1388-2457(99)00141-8.[2]S. Lemm, B. Blankertz, G. Curio, and K.-R. Muller, "Spatio-spectral filters for improving the classification of single trial EEG," IEEE Transactions on Biomedical Engineering, vol. 52, no. 9, pp. 1541-1548, Sep. 2005, doi: 10.1109/TBME.2005.851521.[3]M.-H. Lee et al., "EEG dataset and OpenBMI toolbox for three BCI paradigms: an investigation into BCI illiteracy," GigaScience, vol. 8, no. giz002, May 2019, doi: 10.1093/gigascience/giz002.

Disclosures: D. Gwon: None. **M. Ahn:** None.

Poster

P543. Neurophysiology: Non-Invasive Techniques I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P543.07

Topic: E.05. Brain-Machine Interface

Support: NIH DC009834

Title: Target-related alpha attenuation in a brain-computer interface (BCI) rapid serial visual presentation (RSVP) calibration task

Authors: *D. KLEE[1], T. MEMMOTT[2], B. S. OKEN[3];
[1]Neurol., [2]Neurol. & Inst. on Develop. and Disability, Oregon Hlth. & Sci. Univ., Portland, OR; [3]Neurol. & Behavioral Neurosci., Oregon Hlth. and Sci. Univ., Portland, OR

Abstract: Brain-computer interfaces (BCIs) are systems designed to leverage neurophysiological signals to assist users with various tasks, including communication and movement. It is well-established that amplitude of EEG alpha-band activity is sensitive to a variety of cognitive and sensory processes. The objective of this exploratory study was to identify attention-related changes in posterior alpha amplitude following the presentation of target and non-target letter stimuli during a visual attention task as part of an extant BCI for communication (BciPy). EEG data were collected from healthy adult participants as they

completed multiple instances of a standardized rapid serial visual presentation (RSVP) calibration paradigm in BciPy. Recordings during the calibration sequence consisted of 100 randomized sequences of one target stimulus and nine non-target stimuli. Time-frequency analysis using continuous wavelet transformations showed a reduction in alpha amplitude approximately 400 ms following the onset of target stimuli. A paired-samples t-test confirmed a significant decrease in alpha amplitude following targets compared to non-targets ($p < .001$). Expected individual differences were observed in peak alpha frequency and average alpha amplitude. These results suggest that posterior alpha oscillations are good candidate signals for use in attention-based BCIs.

Disclosures: D. Klee: None. **T. Memmott:** None. **B.S. Oken:** None.

Poster

P543. Neurophysiology: Non-Invasive Techniques I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P543.08

Topic: E.05. Brain-Machine Interface

Support: NINDS/NIH R01 NS106094

Title: Skilled independent control of individual motor units enabled by a non-invasive neuromuscular-machine interface

Authors: *P. BOTROS, E. FORMENTO, J. CARMENA;
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Abstract: Brain-machine interfaces (BMIs) have the potential to augment human functions and restore independence in people with disabilities, yet a compromise between non-invasiveness and performance limits their relevance. Here, we hypothesized that individual motor units of the same muscle can be independently controlled to an extent enabling high-performance BMI applications. Through a non-invasive neuromuscular-machine interface (NMI) providing real-time neurofeedback of motor unit activity, 8 participants learned to skillfully and independently control three biceps brachii motor units to complete a two-dimensional center-out task, with marked improvements in control over 6 days of training. Concomitantly, dimensionality of the motor unit population increased significantly relative to naturalistic behaviors, largely violating recruitment orders displayed during stereotyped, isometric muscle contractions. Finally, participants demonstrated the potential of such skilled motor unit control to power general applications by navigating a virtual keyboard in a spelling task. These results highlight a largely unexplored level of flexibility of the sensorimotor system and show that an NMI leveraging skilled independent motor unit control has great potential to enable a new class of devices that can both provide translational benefit and augment motor functions.

Disclosures: P. Botros: None. **E. Formento:** None. **J. Carmena:** None.

Poster

P543. Neurophysiology: Non-Invasive Techniques I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P543.09

Topic: E.05. Brain-Machine Interface

Support: Houston Methodist Foundation
Cullen Foundation

Title: Neural correlates of kinesthetic motor imagery during brain-machine interface training to control a walking exoskeleton

Authors: *K. NATHAN[1,2], A. KILICARSLAN[1], R. GROSSMAN[2], J. L. CONTRERAS-VIDAL[1,2];
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Abstract: Kinesthetic imagery is a subset of motor imagery that involves the feeling and perception of effort exerted during imagined movement, consequently invoking more motor-specific areas of the brain compared to purely visual imagery. It has been shown to be elicited even in patients with paralysis, thus making it a hypothetical control signal for intuitively controlling brain-machine interfaces (BMIs) for restoration of movement through cognitively-controlled robotic prostheses and orthoses. But we do not fully understand how the typical neural signatures of kinesthetic imagery are affected by long-term BMI training or how their representation may relate to BMI performance. Here we present results from a longitudinal BMI training paradigm with 7 able-bodied subjects to control the Rex lower-limb robotic exoskeleton. The paradigm consisted of nine sessions in which users developed their ability to use kinesthetic motor imagery to initiate the walking and stopping of the Rex's gait as a Go-NoGo task. The BMI consisted of active EEG with dynamic eyeblink and motion artifact removal processed through a Localized Fisher Discriminant Analysis dimensionality reduction and a Gaussian Mixture Model classifier on time-lagged delta band amplitudes. Subjects underwent a final session with simultaneous EEG-fMRI recording while watching video playback of themselves walking in the Rex performing the same motor imagery, allowing for fMRI-constrained source localization of the EEG. While BMI decoding for controlling the gait was variable among subjects, EEG and fMRI analysis revealed neural signatures for the kinesthetic imagery during the course of the training. The fMRI scans showed contrasts in activation between the Walk and Stop conditions localized in sensorimotor (SMA) and visual processing (IOG) areas associated with imagery. Offline EEG analysis identified the expected readiness potential negativity in response to the walk cue prior to movement onset overlying central motor channels, and a separate biphasic ERP response in posterior occipital channels. Localized EEG dipole sources

were found to be clustered within motor, visual processing, and prefrontal areas. Together, these results can be used to retune BMI decoders relying on kinesthetic imagery to better capture these features.

Disclosures: K. Nathan: None. **A. Kilicarslan:** None. **R. Grossman:** None. **J.L. Contreras-Vidal:** None.

Poster

P543. Neurophysiology: Non-Invasive Techniques I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P543.10

Topic: E.05. Brain-Machine Interface

Title: Transcutaneous stimulation of the cervical spinal cord promotes recovery of upper limb muscle strength in tetraplegia

Authors: *S. CHANDRASEKARAN[1], R. RAMDEO[2], A. STEIN[3], D. GRIFFIN[3], S. J. HARKEMA[4], C. BOUTON[5];
[1]Feinstein Inst. for Med. Res., Manhasset, NY; [2]Feinstein Inst. for Med. Res., New York, NY; [3]Northwell Hlth., New York, NY; [4]Dept Neurol Surgery, Univ. Louisville, Frazier Rehab Inst, KSCIRC, Louisville, KY; [5]Ctr. for Bioelectronic Med., The Feinstein Inst. For Med. Res., Manhasset, NY

Abstract: Spinal cord injury (SCI) affects approximately 18,000 individuals every year in the United States, with over half of the cases resulting in upper limb impairment. Most persons with tetraplegia rank regaining hand function as being of utmost importance. However, functional recovery after SCI tends to plateau beyond 12-18 months post-injury. Therefore, bringing about long-lasting recovery of upper limb function is a priority in SCI clinical research. It has been shown that epidural lumbar spinal cord stimulation can help restore lower limb movement in humans. Recently, this has also been shown for the upper limb with epidural spinal cord stimulation at the cervical level in primates. However, persistent stimulation of the spinal cord is typically required to evoke movements of the paralyzed limb. Thus, long-term restoration of volitional control of movement is yet to be achieved. Recently, transcutaneous spinal cord stimulation (tSCS) was used to promote long-term recovery of function of the upper-limb in people with motor-incomplete spinal cord injury. With the aim of promoting such recovery of function, we targeted transcutaneous stimulation to the cervical spinal cord in two participants with C4/C5 motor complete spinal cord injuries. We combined physical activity of muscles with spinal cord stimulation. As a proof of concept, we assessed the progress of strength of a muscle for which the participants retained some residual level of volitional control of movement, namely the triceps. In this study, we delivered constant stimulation to the cervical spinal cord for approximately 1-2 hours per session, one session per week while the participant performed

moderate physical activity targeted to the muscle. In two patients with tetraplegia, we show up to 200% increase in the force generated by upper arm muscles within 3 months of starting stimulation. Interestingly, the participants maintained this gain in strength even when stimulation sessions of stopped for up to 6 weeks. We also observed up to a 2-point increase in the sensibility component of the GRASSP test in most areas of the hand. In future experiments, we intend to pair tSCS with user intent conveyed by gesture control for recovery of function of the hand.

Disclosures: S. Chandrasekaran: None. **R. Ramdeo:** None. **A. Stein:** None. **D. Griffin:** None. **S.J. Harkema:** None. **C. Bouton:** None.

Poster

P543. Neurophysiology: Non-Invasive Techniques I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P543.11

Topic: E.09. Motor Neurons and Muscle

Support: NIH Grant 5R01DC018691-02
Howard Hughes Medical Institute
Keck Foundation

Title: Connectivity and Neuroanatomy of the Orofacial Motor Cortex and the Laryngeal Motor Cortex for Vocal Modulation in Mice

Authors: *R. AGRAVAT[1], C. D. M. VARGAS[1], E. JARVIS[1,2];
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Abstract: Vocal communication is a complex behavior that can be innate or learned. In humans, learned vocalizations and speech are the basis of our spoken language, and is a rare behavior that has only been described in five groups of mammals (cetaceans, pinnipeds, elephants, bats, humans) and three groups of birds (songbirds, parrots, hummingbirds). All other species have been shown or assumed to be vocal non-learners. Songbirds have been the standard model of vocal learning, but they lack the experimental tractability found in other species. Recent work in mice, which are tractable and more closely related to humans, shows evidence of a rudimentary vocal plasticity system, including deafness induced vocal deterioration and a sparse direct projection from the motor cortex to the brain stem motor neurons controlling the larynx. This direct projection is thought to be fundamental to the ability to learn vocalizations and finely modulate vocal output. This direct projecting cortical region in humans has been termed the Laryngeal Motor Cortex (LMC). Here we found that the stimulation of the mouse rudimentary LMC and a more anterior Orofacial Motor Cortex (OFC) causes laryngeal muscle contractions with latencies indicative of both direct and indirect projections. We used retrograde and

anterograde viral tracers in the LMC and OFC regions and in vocal muscles. We found labeled neurons from the anterograde tracing experiments in areas such as the posterior portion of the primary motor cortex, dorsal and medial striatum, primary somatosensory cortex, ventral nucleus of the thalamus, hypothalamus, and pyramidal nucleus. Labeled neurons from the retrograde tracing experiments were identified in areas such as the secondary motor cortex, hypothalamus, periaqueductal gray, reticular nucleus, and facial nucleus. Results from these tracing experiments indicate that the two regions are distinct, parallel pathways that influence laryngeal and jaw muscles. The two regions may both act to coordinate vocal behavior through distinct but overlapping circuits.

Disclosures: R. Agravat: None. **C.D.M. Vargas:** None. **E. Jarvis:** None.

Poster

P569. Motor Neurons: Activity, Sensory, and Central Control: Exercise, Injury, and Disease

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P569.01

Topic: E.09. Motor Neurons and Muscle

Support: MIH Grant NS105982

Title: Enhancement of axon regeneration after peripheral nerve injury using the small molecule trkB ligand prodrug, R13.

Authors: *A. W. ENGLISH[1], D. CARRASCO[1], R. ISAACSON[1], D. HOFFMAN[1], X. LIU[2], K. YE[3];
[1]Cell Biol., [2]Pathol & Lab. Med., Emory Univ., Atlanta, GA; [3]Pathol & Lab. Med., Emory Univ. Sch. of Med., Atlanta, GA

Abstract: Axon regeneration after peripheral nerve injury is slow and inefficient, leading to generally poor functional recovery. Activity-dependent experimental therapies that increase expression of brain derived neurotrophic factor (BDNF) and its trkB receptor enhance regeneration, suggesting that treatments with BDNF might also be effective. However, recombinant human BDNF, as well as 7,8 dihydroxyflavone (7,8 DHF), a small molecule BDNF mimetic, may have limited treatment applications because of their modest oral bioavailability and a moderate pharmacokinetic profile. R13 is a 7,8 DHF prodrug. Upon oral administration it is converted in the liver to 7,8 DHF. A single oral treatment with R13 (43.6 mg/Kg) to a mouse immediately following sciatic nerve transection and repair produced increased trkB phosphorylation and phosphorylation of its downstream targets, AKT and MAP kinase, at the site of nerve injury. These increases lasted for more than four hours, an improvement in bioavailability over a comparable dose of 7,8 DHF. In thy-1-YFP-H mice treated daily with this

oral dose of R13 for two weeks following transection and repair of the sciatic nerve, regenerating axon profiles were more than three times longer than controls and significantly longer than in mice treated with 7,8 DHF. More than 40% of these axons were longer than the longest regenerating axon found either in controls or 7,8 DHF-treated mice. Direct muscle EMG responses (M waves) were significantly larger in R13 treated mice four weeks after injury than vehicle treated controls. Oral treatments with the pro-drug, R13, could be a potent therapy for stimulating axon regeneration after peripheral nerve injury.

Disclosures: A.W. English: None. **D. Carrasco:** None. **R. Isaacson:** None. **D. Hoffman:** None. **X. Liu:** None. **K. Ye:** None.

Poster

P569. Motor Neurons: Activity, Sensory, and Central Control: Exercise, Injury, and Disease

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P569.02

Topic: E.09. Motor Neurons and Muscle

Support: Greehey Family Foundation Endowment for Alzheimer's Research
Bartell and Mollie Zachry Endowment for Alzheimer's Disease

Title: Explanatory Artificial Intelligence Uncovers Specific Response related White Matter Connectivity Surrounding DBS contacts in Parkinson Disease and Essential Tremor

Authors: *J. TORANZO[1], M. USCAMAYTA AYVAR[1], F. ARNEDO[2], G. POBLETE[3], R. SALAS[3], I. ZWIR[4], G. A. DE ERAUSQUIN[1];
[1]Glenn Biggs institute for Alzheimer's & neurodegenerative diseases, Univ. of Texas Hlth. San Antonio, San Antonio, TX; [2]Univ. of Granada, Granada, Spain; [3]Baylor Col. of Med., Houston, TX; [4]Washington Univ. in St. Louis, St. Louis, MO

Abstract: Deep Brain Stimulation (DBS) is a well established therapy for advanced Parkinson Disease (PD) and disabling Essential Tremor (ET), but clinical improvement after optimization of the surgical lead placement remains less than fully predictable and can only be established posthoc. Diffusor Tensor Imaging (DTI) tractography allows identification of white matter tracts surrounding active contacts using simulated models of the volume of tissue activated by the DBS current, but this methodology has not been broadly applied, and knowledge regarding the specific networks responsible for therapeutic response is very limited. Yet, recent work provides rationale for targeting networks rather than individual anatomic structures. We used explanatory Artificial Intelligence (xAI) to characterize white matter connections and recruited networks surrounding each active contact to identify connections resulting in greatest therapeutic response in a series of 40 patients who underwent DBS surgery for PD (20) or ET (20). Preoperative MRI

images were fused to postoperative high definition computerized tomography (CT) using Standard Parametric Mapping (SPM) and Advance Normalization Tools (ANTs). Following corregistration volumes were normalized using SPM12 segment nonlinear and Hybrid SPM and Brainsfit methods, and Brainshift Correction with Coarse Masks were applied. A fully automated method for electrode trajectory and contact reconstruction in deep brain stimulation was applied (PaCER), followed by localization, and volume of activated tissue (VAT) calculations (SimBio toolbox) for each of the 4 different settings of the stimulation parameters. Structural connectivity was calculated with fibertracking/normalization using the Generalized Q Sampling method, overlapped to parcellation schemes following Schaefer 2018 (1000 parcels. 17 networks). The connectivity network was characterized for each contact using outcome independent clustering (many to many relation analysis) of preoperative data. The fiber track files were analyzed using unsupervised machine learning techniques to identify groups of stimulations sharing particular fibers, specifically, a biclustering method based on Non-negative Matrix Factorization (NMF) identified 5 networks of white matter connectivity based on corresponding overlapping fibers in PD, and 3 in ET. The outcome information associated with each stimulation in the groups was later used to calculate the clinical response of each group. Networks uncovered are generally associated with better clinical responses, consistent with the good clinical outcomes with DBS, which were distinct for ET and PD.

Disclosures: J. Toranzo: None. **M. Uscamayta Ayvar:** None. **F. Arnedo:** None. **G. Poblete:** None. **R. Salas:** None. **I. Zwir:** None. **G.A. de Erausquin:** None.

Poster

P569. Motor Neurons: Activity, Sensory, and Central Control: Exercise, Injury, and Disease

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P569.03

Topic: E.09. Motor Neurons and Muscle

Title: Identity of the hindbrain neurons activated by inflammation

Authors: *M. JALIL[1], T. COVERDELL[2], J. CAMPBELL[3];
[1]Univ. of Virgina, Charlottesville, VA; [2]Pharmacol., [3]Biol., Univ. of Virginia, Charlottesville, VA

Abstract: Inflammation in the body activates neurons of the dorsal vagal complex (DVC) in the hindbrain, some of which form a circuit for suppressing inflammation - the vagal anti-inflammatory reflex. This reflex circuit is being investigated as a treatment for diseases such as irritable bowel disease (IBS), rheumatoid arthritis, and acute kidney reperfusion injury. Yet, in spite of its clinical importance, little is known about the identity of the DVC neurons involved in the circuit. To identify these neurons, we analyzed co-localization of the neuronal-activation

marker, c-Fos (gene, Fos), with different neuronal subtype markers (Phox2b, Chat, Atf3) in DVC neurons in mice 90 minutes after acute treatment with a bacterial endotoxin, lipopolysaccharide (LPS; 5mg/kg, i.p.), or vehicle control. We found out that 70% of the c-Fos immunoreactive neurons were also positive for Phox2b when injected with LPS compared to 39% of the c-Fos immunoreactive neurons in the vehicle control. The total number of Phox2b-immunoreactive neurons in the DVC did not change significantly between treatment conditions. Together, these results suggest that LPS induces cFos immunoreactivity preferentially in the Phox2b+ population of DVC neurons. To further characterize the LPS-activated neurons, we then performed mRNA fluorescence insitu hybridization for Fos, Chat (a marker of cholinergic vagal neurons), and Atf3 in the DVC 30 and 90 minutes after LPS treatment. Strikingly, whereas very few Chat+ DVC neurons were Fos+, essentially all Atf3+ DVC neurons were Fos+. Overall, our results provide deeper insight into the identity of hindbrain neurons which are activated by inflammation and may mediate the anti-inflammatory circuit. Future studies identifying the neuronal population which sense and reflexively suppress inflammation will help in targeting this circuit for treating autoimmune and inflammatory diseases.

Disclosures: M. Jalil: None. **T. Coverdell:** None. **J. Campbell:** None.

Poster

P569. Motor Neurons: Activity, Sensory, and Central Control: Exercise, Injury, and Disease

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P569.04

Topic: E.09. Motor Neurons and Muscle

Title: 松芝靈芝,脈衝電磁場和乾細胞治療肌肉減少症引起的神經元運動系統行為改變的應用.

Authors: *S. CHEN;
Chung Yuan Christine Univ., Taoyuan City, Taiwan

Abstract: <META NAME="author" CONTENT="阿思">**The application of Ganoderma tsugae, pulsed electromagnetic fields, and stem cells to treat the sarcopenia induced behavioral changes of the neuronal motor system.** Szu-Yu Chen[1], Ming-Wei Chao[2], Chia-Yi Tseng[11]Department of Biomedical Engineering, and [2]Department of Bioscience Technology, Chung Yuan Christine University, No. 200, Zhongbei Rd., Zhongli Dist., Taoyuan City 320314, Taiwan
AbstractThe decrease in muscle mass and muscle strength accelerates with age, leading to the emergence of aging-induced sarcopenia. The etiology of aging-induced sarcopenia is due to accumulation of reactive oxidative species and chronic inflammation of muscles or motor

neurons, false communication in the neuromuscular junction, or lesion in the motor cortex, further leading to changes in motor behavior. This study adopted the integrative medical method with *Ganoderma tsuga*e (GT), pulsed electromagnetic field (PEMF), and human umbilical cord blood mesenchymal stem cells to treat the aging-induced sarcopenia. To explore whether this integrative medical method would add to each other's therapeutic effects and improve motor neuronal behavior performance. Sprague Dawley (SD) rats were divided into six groups: (1) control group, (2) stem cell group, (3) PEMF and stem cell group (4) PEMF and GT group (5) GT group and stem cell group (6) stem cell group, GT and PEMF group. Each group induced aging with subcutaneous D-galactose injections (200 mg/kg). Motor behavior analysis, including rotarod test, rearing count, and tight rope, were conducted to observe muscle strength and activity changes to confirm the induction of aging before treatments were applied. The above behavioral tests are repeated each month after treatment to verify the effectiveness of the treatment. Observations are expected to take up to half a year. After two months of treatment, we currently found that the groups with stem cell injection have significant protective effects against aging in behavioral testing, especially in the rotarod test. However, GT and PEMF show no noticeable protection effect. Based on current data, stem cells show a significant effect on preventing changes in the motor behavior induced by aging sarcopenia.
Funding source: Ministry of Science and Technology (108-2221-E-033-018-)Keywords: *Ganoderma tsuga*e (GT), pulsed electromagnetic field (PEMF), mesenchymal stem cells, sarcopenia

Disclosures: S. Chen: None.

Poster

P569. Motor Neurons: Activity, Sensory, and Central Control: Exercise, Injury, and Disease

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P569.05

Topic: E.09. Motor Neurons and Muscle

Support: R15NS099983

Title: Estrogen signaling is required for treadmill exercise mediated effects on synaptic plasticity around axotomized spinal motoneurons

Authors: *J. C. WILHELM, V. L. KENNEDY, Jr., S. T. WILSON, G. M. BADER;
Col. of Charleston, Charleston, SC

Abstract: After peripheral nerve transection, changes in spinal cord circuitry occur including the withdrawal of synaptic inputs from the somata and proximal dendrites of axotomized spinal motoneurons. Moderate daily treadmill exercise after transection injuries has been shown to

mitigate the reduction in synaptic coverage. The mechanisms that underlie the effect of exercise are not well understood. Estrogen receptor (ER) signaling has been shown to have an important role in the effects of exercise on regeneration of injured motoneuron. Therefore, in this study we tested the hypothesis that ER signaling also is a part of the mechanism by which treadmill exercise mediates its effects on synaptic inputs. Gonadally intact male and female C57BL/6 mice underwent a unilateral sciatic nerve transection. Injured motoneurons were retrogradely labeled using a fluorescent dye immediately after transection. Mice then were divided into groups and treated for two weeks with 1) treadmill exercise, 2) ER antagonist Fulvestrant, or 3) treadmill exercise and Fulvestrant. Lumbar spinal cord tissue was harvested fourteen days after surgical injury. Tissue sections containing injured motoneurons were reacted with antibodies to vesicular glutamate transporter 1 (vGLUT1) and glutamate decarboxylase (GAD67). The average synaptic coverage of glutamatergic and GABAergic inputs onto labeled motoneurons was assessed using Image J. We found no significant reduction in coverage in mice treated with treadmill exercise. Blocking ER signaling during treadmill exercise prevented the sustaining effects of the exercise and resulted in an increased loss of inputs compared to mice not receiving the ER antagonist. Based on these results we suggest that ER signaling is an important part of the machinery required for the exercise-mediated effects on synaptic inputs onto axotomized motoneurons after sciatic nerve transection.

Disclosures: J.C. Wilhelm: None. **V.L. Kennedy:** None. **S.T. Wilson:** None. **G.M. Bader:** None.

Poster

P569. Motor Neurons: Activity, Sensory, and Central Control: Exercise, Injury, and Disease

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P569.06

Topic: E.09. Motor Neurons and Muscle

Support: DMDRP W81XWH

Title: Is too much REST a bad thing? Characterizing a master transcriptional regulator in motor neurons after peripheral nerve injury

Authors: *A. K. YASZEMSKI[1,2], N. MADIGAN[1], A. J. WINDEBANK[1], L. NESTI[2];
[1]Dept. of Neurol., Mayo Clin., Rochester, MN; [2]Dept. of Surgery, Uniformed Services Univ., Bethesda, MD

Abstract: Introduction: Peripheral nerve injuries (PNIs) present complex challenges for repair as they include large areas of neuronal damage with trauma to the surrounding muscle and vasculature. Re-innervation of muscle tissue after injury is necessary for functional recovery, but

motor neurons may not regenerate sufficiently, and permanent loss of function can occur. A persistent decline in support elements such as neurotrophic factors (NTFs) and improper reinnervation of muscle targets from axonal misguidance can limit regenerative capacity, both of which have been observed in parallel with injury-induced expression of the Repressor Element-1 Silencing Transcription Factor (REST), a master transcriptional regulator in neurons. How elevation of REST expression affects motor nerves and subsequent recovery after PNI has yet to be investigated. We are developing a novel approach to peripheral nerve regeneration by targeting the motor neuron nucleus to enhance the natural proximal to distal regeneration pattern by modulating REST. This work characterizes injury-induced expression of REST in peripheral motor neurons and its effects on the regenerative process in a rodent model PNI. **We hypothesize that REST expression is elevated in motor neurons after PNI and that its activation is inhibiting important neurotrophic factors and neuronal genes that affect regenerative potential.** This repressive transcriptional block represents a potential therapeutic target to accelerate recovery from peripheral nerve injury by enhancing neuroregenerative programming.

Methods: Male Sprage-Dawely rats, 8-9 weeks old were grouped into facial nerve transection or sham injury groups at 6-hour and 48-hour timepoints (n=12/group). The facial nerve transection injury involved a complete transection at the nerve trunk, sham surgery received a skin incision with the nerve trunk exposed but left intact. REST protein and gene expression levels were assessed via immunohistochemistry and RT-PCR, respectively. Relevant target gene expression was analyzed by in situ hybridization.

Results: Significant upregulation of REST protein expression exists in injured animals compared to sham and naïve groups at both time points. RT-PCR showed no significant changes in mRNA expression at either timepoints. Data analysis is in process for in situ hybridization.

Discussion: This is the first evidence of REST activation after peripheral nerve injury in motor neurons. The direct regulation of various neurotrophic factors and regenerative-associated genes makes REST a potential therapeutic target for improving functional recovery after peripheral nerve injury.

Disclosures: A.K. Yaszemski: None. **N. Madigan:** None. **A.J. Windebank:** None. **L. Nesti:** None.

Poster

P569. Motor Neurons: Activity, Sensory, and Central Control: Exercise, Injury, and Disease

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P569.07

Topic: E.09. Motor Neurons and Muscle

Support: NIH Grant 5R00NS089824

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Simons Foundation 402047
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Title: Motor training improves coordination and anxiety in Mecp2-null mice despite impaired functional connectivity within the motor circuit

Authors: *Y. YUE[1], P. XU[1], Z. LIU[1], X. SUN[1], J. SU[1], H. DU[1], L. CHEN[1], R. ASH[2], S. SMIRNAKIS[3], R. SIMHA[1], L. KUSNER[1], C. ZENG[1], H. LU[1];
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Abstract: Mutations in the X-linked gene Methyl-CpG-binding protein 2 (*MECP2*) causes a severe neurodevelopmental disorder, Rett syndrome (RTT). Children with RTT (typically girls) develop normally until about 6-18 months of age when they undergo a period of regression wherein they lose acquired motor, cognitive, and social milestones and do not make further gains. Male *Mecp2*-null mice model more severe forms of RTT and replicate the course of the disease. Since little is known about how motor learning is encoded by L2/3 and L5 ensembles in the motor cortex, we sought insight into this process by comparing wild-type and *Mecp2*-null mice on a very specific motor learning task: running on a computerized wheel whose speed is controlled by the experimenter. Thus, the skill to be learned is to adapt to increasing or decreasing speeds. While head-fixed mice ran on the wheel, we conducted two-photon calcium imaging to record thousands of neurons in L2/3 and L5a and study changes in event rates and functional connectivity over the course of two weeks of training. We found that the mutant motor cortex, despite their reduced cross-layer connectivity, a ~20% overall lower firing rate, and impairment in maintaining functional connectivity more than a couple of days, retains sufficient plasticity to support motor learning. Interestingly, overall functional connectivity of neuronal pairs in both layers decreased after training but the top 5% of neuronal pairs that were most strongly connected actually increased their functional connectivity with training. More surprisingly, a mere two weeks of training on the speed-changing wheel caused the mutant mice to improve their coordination on different motor assays (ladder-walking and rotarod), reduce anxiety (in the open field and elevated plus maze tests), and survive about 20% longer than untrained null mice. We found a correlation, albeit a weak one, between forepaw coordination improvement and extension of survival in the null mice, but not between any other measures and lifespan; this raises the possibility that learning confers benefits beyond those of mere exercise. Our results support a model in which motor skills are learned by judiciously strengthening the functional connectivity of a subgroup of neurons while decreasing connectivity among the rest of the population to maintain flexibility for learning new skills. These results underscore the benefit of motor training, especially given that motor deficits and anxiety are core symptoms of Rett, which is not diagnosed until well after symptom onset.

Disclosures: Y. Yue: None. **P. Xu:** None. **Z. Liu:** None. **X. Sun:** None. **J. Su:** None. **H. Du:** None. **L. Chen:** None. **R. Ash:** None. **S. Smirnakis:** None. **R. Simha:** None. **L. Kusner:** None. **C. Zeng:** None. **H. Lu:** None.

Poster

P569. Motor Neurons: Activity, Sensory, and Central Control: Exercise, Injury, and Disease

Location: Virtual Only

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Program #/Poster #: P569.08

Topic: E.09. Motor Neurons and Muscle

Title: Retrospective Review of SMN1 Full Gene Sequencing at Mayo Clinic Laboratories Reveals Two Novel Variants and Two Cases of Homozygous Point Mutations

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Mayo Clin., Rochester, MN

Abstract: Spinal muscular atrophy is caused by homozygous deletions of the *SMN1* gene in 95% of affected patients. The remainder of SMA diagnoses is heterozygous with a *SMN1* deletion in one allele and a small more subtle mutation in the other allele. SMA cases resulting from homozygous point mutations are rare. Mayo Clinic Laboratories offers SMA Del/Dup testing and *SMN1* full-gene sequencing. Our SMA Del/Dup assay uses novel droplet digital PCR technology to accurately quantify *SMN1* and *SMN2* copy numbers between 0 to 5 copies, whereas other methods cannot report higher than 3. Our *SMN1* sequencing assay identifies variants in all exons of *SMN1*, but cannot discriminate between exon 1 *SMN1* or *SMN2* variants. From October 2018 to March 2021, our *SMN1* full gene sequencing tested a total of 50 patients - age ranged from 5 days to 35 years old. We identified seven missense variants: c.43C>T (p.Q15* known pathogenic), c.46delG (p.E16Rfs*24 novel predicted pathogenic), c.278A>C (p.K93T novel VUS), c.683T>A (p.L228* known pathogenic), c.549delC (p.K184Sfs*29 predicted pathogenic), c.597dupC (p.M200Hfs*56 novel predicted pathogenic), and c.735dupA (p.P246Tfs*10 known pathogenic). Interestingly, two variants occurred as homozygous mutations. In one case, a 17 year old boy presented with symptoms consistent with motor neuron disease. Molecular analysis reported 2 copies of *SMN1*, 2 copies of *SMN2*. Importantly, microarray analysis revealed consanguinity. Our *SMN1* full gene analysis revealed a homozygous pathogenic mutation in exon 6 (c.683T>A). In the other case, a 6 month old girl presented with symptoms consistent with SMA. Molecular analysis reported 2 copies of *SMN1* and 2 copies of *SMN2*.The patient's parents are known to be consanguineous. Our *SMN1* full gene analysis revealed a homozygous pathogenic mutation in exon 6 (c.735dupA). Two of seven variants are novel and not previously reported in the literature. Lastly, two of seven variants occurred in exon 1. Our method uses long-range-PCR of *SMN1* exons 2-8, followed by bidirectional Sanger sequence analysis for nucleotide variants in *SMN1*. Exon 1, however, is PCR-amplified and bidirectionally Sanger-sequenced alone using primers which amplify exon 1 of both *SMN1* and *SMN2*. As a result, clinical correlation is needed to render diagnosis. Our

retrospective review of 50 *SMN1* full gene analyses revealed seven incidences in which *SMN1* variants were detected. Two variants occurred as rare homozygous mutations.Two variants are novel. Lastly, two variants are in exon 1. Our test analyzes exon 1 separate from exon 2-8 as variants detected in *SMN1* exon 1 cannot be distinguished from variants in *SMN2* exon 1.

Disclosures: S. Bellampalli: None. **S. Cook:** None. **N. Vidal-Folch:** None. **H. Cabral:** None. **R. Rowsey:** None. **L. Hasadsri:** None. **D. Oglesbee:** None.

Poster

P569. Motor Neurons: Activity, Sensory, and Central Control: Exercise, Injury, and Disease

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P569.09

Topic: E.09. Motor Neurons and Muscle

Support: ALS Therapy Alliance
UIC Startup fund
UIC Neurology department Seed Fund
AYZ Family Foundation
AKG global Foundation

Title: Tracking axonal loss and unique microglia in the corticospinal tract in ALS patients

Authors: J. LIU[1], S. MARTIN[1], K. JOHN[1], J. LEI[1], J. M. RAVITS[2], J. LOEB[1], ***F. SONG**[1]; [1]Univ. of Illinois at Chicago, Chicago, IL; [2]Neurosciences, Univ. of California San Diego Dept. of Neurosciences, La Jolla, CA

Abstract: Amyotrophic lateral sclerosis (ALS) is a progressive neurodegenerative disease that selectively involves both upper and lower motor neurons. The mechanism that links upper and lower motor neurons is unknown. However, a common pathological finding in the spinal cord is corticospinal tract (CST) degeneration. We found the myelin loss in the CST in patients with upper motor neuron (UMN)-dominant clinical symptoms at disease onset (1). UMN-dominant ALS is defined as predominantly UMN signs but still with minor lower motor neuron (LMN) signs at disease onset. To determine the anatomical relationships of the connection of patients with UMN–dominant vs LMN-dominant clinical signs at the disease onset, we developed quantitative methods to measure different sizes of axons and glial morphology in well-characterized postmortem spinal cord at three levels (cervical, thoracic, and lumbar) from ALS patients. Patients with UMN–dominant showed myelin loss in the lateral CSTs in the spinal cord at three levels. We found an ‘All or None’ effect in that all levels of the CST were equally effected at the time of death in ALS patients with UMN-dominant clinical symptoms at disease. In contrast, LMN-dominant ALS patients had significantly less CST involvement. LMN loss in

the ventral horn showed in both UMN- and LMN-dominant ALS patients with no clear pattern related to the site of disease onset. Quantification of axon density showed severe loss of axons in both small and large diameter axons in the lateral CSTs in UMN-dominant ALS patients. Interestingly, significantly more unique foamy-shaped microglia were associated with myelin and axon loss in the lateral CST in UMN-dominant ALS patients. This 'All or None' effect of myelin loss in the lateral CST's associated with the increased unique microglia suggests the possibility of aberrant axon-glial interactions contributing to pathogenesis and axonal degeneration in ALS patients with prominent UMN signs at disease onset.
Reference: 1.Song F, Chiang P, Ravits J, Loeb JA. Activation of microglial neuregulin1 signaling in the corticospinal tracts of ALS patients with upper motor neuron signs. Amyotroph Lateral Scler Frontotemporal Degener. 2014, 15:77-83.

Disclosures: J. Liu: None. **S. Martin:** None. **K. John:** None. **J. Lei:** None. **J.M. Ravits:** None. **J. Loeb:** None. **F. Song:** None.

Poster

P569. Motor Neurons: Activity, Sensory, and Central Control: Exercise, Injury, and Disease

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Program #/Poster #: P569.10

Topic: E.09. Motor Neurons and Muscle

Support: NIH grant 1 R15 AG022908-01A2
NSF grant DBI 0552517

Title: Effects of sedentary aging and exercise on structural plasticity of motor neurons and NMJ structures.

Authors: *A. CINTRÓN-COLÓN, J. FANIZZA, J. M. SPITSBERGEN;
Biol. Sci., Western Michigan Univ., Kalamazoo, MI

Abstract: Neuromuscular weakness has been associated with age progression. A possible reason is a disconnection between muscle and neuron, causing components from both the central and peripheral nervous systems to deteriorate. Exercise, however, has beneficial effects for both muscle and nervous tissue, inducing neuroprotection. This study aims to evaluate changes in structural plasticity of motor neurons in the lumbar spinal cord and NMJ structures in the skeletal muscle with sedentary aging and exercise. We hypothesize that motor neuron area size and possibly subgroups decrease with sedentary aging, but in exercised age-matched animals, the area size will increase, and subgroup diversity will remain stable. At the site of the skeletal muscle, we hypothesize that NMJ structures get degraded with sedentary aging, but exercise can maintain those structures even in advanced age. Spinal tissues were taken from sedentary and

exercised Sprague-Dawley rats between 4 weeks and 24 months of age. Sedentary groups consisted of the following: 4-week-old, 14-week-old, 12-month-old, 18-month-old, and 24-month-old rats, while exercised groups consisted of 14-week-old rats that had access to running wheels for ten weeks; and 12-, 18-, and 24-month-old rats that had access to running wheels for 24 weeks. Lumbar spinal tissue and hindlimb skeletal muscle (soleus and plantaris) was used for immunohistochemical analysis. In the spinal tissue, antibodies against choline acetyltransferase (ChAT), neuronal nuclei (NeuN), and the fluorescent dye DAPI were used to identify neurons in the ventral horn of the spinal cord. In skeletal muscle, α-bungarotoxin was used to identify motor end plates and acetylcholine receptors. Results suggest a decrease in stained area size of neurons in the ventral horn in adulthood. Exercised animals, however, had a larger area size when compared to their respective sedentary age-matched group. Histogram analysis suggests that larger neurons tend to be less numerous in adulthood, suggesting a possible loss of motor neuron subgroups. The area size of motor endplates in skeletal muscle from exercised rats was more prominent than their respective sedentary age-matched counterpart. Our results aid in understanding the structural changes that can occur with aging in the neuromuscular system in a mammalian model of aging. Furthermore, this work adds to the pool of knowledge of how exercise can impact at different points in the lifespan of an aging mammalian model and its potential benefits.

Disclosures: A. Cintrón-Colón: None. **J. Fanizza:** None. **J.M. Spitsbergen:** None.

Poster

P569. Motor Neurons: Activity, Sensory, and Central Control: Exercise, Injury, and Disease

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P569.11

Topic: E.09. Motor Neurons and Muscle

Support: Economic Social Science Research Council ES/R0100072/1
Wellcome Trust Institutional Strategic Support1516ssfira4
China Scholarship Council

Title: Tap to the beat: synchronization between auditory entrainment and motor beta bursts during finger tapping

Authors: *Q. CHEN[1], C. MCALLISTER[1], K. SHAPIRO[1], S. HANSLMAYR[2];
[1]Univ. of Birmingham, Birmingham, United Kingdom; [2]Univ. of Glasgow, Glasgow, United Kingdom

Abstract: Humans can synchronize their movements to an externally presented rhythm, but little is known about the underlying neural mechanisms allowing for such rhythmic behaviour. On the

one hand, it has been shown that the auditory cortex can be entrained to a rhythmic signal. On the other hand, transient bursts of beta oscillations in the motor cortex have been shown to play a critical role in predictive rhythmic action. Arguably, the two processes must interact to allow for movements to be synchronized to an auditorily presented beat, but little is known about this interaction. To shed light on this issue, we recruited 15 young healthy participants to perform a finger-tapping task following rhythmic tones at 1 Hz and 2Hz in an EEG study. Transient motor beta bursts were identified according to established detection algorithms (Staresina et al., 2015) at both scalp and source levels. Preliminary results at the scalp level show that motor beta bursts are significantly more closely related to motor output (i.e., taps) than auditory entrainment (i.e., tones). Surprisingly, behavioural results indicate that the process of adaptation to rhythmic auditory stimuli evolves very quickly, in only 3 consequent tones. In a further analysis, we are planning to investigate the relation between auditory entrained EEG activity and motor beta burst at the source level. Motor beta burst timing is predicted to be modulated by the phase of auditory entrained activity and the degree of modulation is expected to correlate with the performance precision. Overall, our study explores the dynamic configuration of the sensorimotor network in the maintenance of rhythmic motion and timing prediction.

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Poster

P569. Motor Neurons: Activity, Sensory, and Central Control: Exercise, Injury, and Disease

Location: Virtual Only

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Program #/Poster #: P569.12

Topic: E.09. Motor Neurons and Muscle

Support: NIH R01 HD084009
NIH R01 HD039343

Title: A novel time-based surface EMG measure for quantifying hypertonia in hemiparetic arm muscles during daily activities after stroke

Authors: *M. SOHN[1], J. DEOL[1], J. P. A. DEWALD[1,2];
[1]Physical Therapy and Human Movement Sci., Northwestern Univ., Chicago, IL; [2]Biomed. Engin., Northwestern Univ., Evanston, IL

Abstract: After stroke, paretic arm muscles are constantly exposed to abnormal neural drive from the injured brain. As such, hypertonia, broadly defined as an increase in muscle tone, is prevalent especially in distal muscles, which impairs daily function or in long-term leads to a deformed resting posture. However, there currently is no quantitative measure that can reliably track how hypertonia is expressed on daily basis. In this study, we propose a novel time-based

surface electromyography (sEMG) measure that can overcome the limitations of the coarse clinical scales often measured in functionally irrelevant context and the magnitude-based sEMG measures that suffer from signal non-stationarity. We postulated that the key to robust quantification of hypertonia is to capture the "true" baseline in sEMG for each measurement session, by which we can define the relative duration of activity over a short time segment continuously tracked in a sliding window fashion. Preliminary results demonstrated that the proposed approach can well characterize the clinically observed differences in hypertonia expressed across different muscles and impairment levels. Specifically, we recorded sEMG from 6 distal wrist/finger and 2 proximal elbow muscles while individuals with stroke (n=3; FMA-UE score: 13-25-38) were nominally 'awake', completely 'relaxed', and executing a 'ambulatory' motor task. We quantified active duration of sEMG within a time segment (5s sliding window), defined as the percent of time signal is above a threshold $\mu+3\sigma$ (μ: minimum segment mean across all recordings; σ: standard deviation in that segment). We found that hypertonia was more prevalent (i.e., greater active duration) in distal than proximal muscles during 'awake' ($t_{254}=5.45$, $p<0.001$) and 'ambulatory' states ($t_{282}=3.54$, $p<0.001$). There was no difference across muscles ($t_{254}=0.99$, $p=0.32$) or participants during the 'relaxed' state, indicating that our measures robustly quantify the "true" baseline (i.e., absent hyperactivity) that is comparable across muscles and participants. Finally, the amount of reduction in active duration from 'awake' to 'relaxed' state increased with impairment level, more so in distal muscles ($t_{254}=-3.51$, $p<0.001$), indicating that individuals with more severe impairment would experience a longer period of continuous involuntary activity, especially in the distal muscles, even during the ordinary awake hours. The proposed measure can be used for continuous and quantitative monitoring of hypertonia while at home, which will allow for the study of practical effect of pharmacological and/or physical interventions that try to combat its presence.

Disclosures: M. Sohn: None. **J. Deol:** None. **J.P.A. Dewald:** None.

Poster

P632. Sleep and Rhythms: Cellular and Molecular Contributors I

Location: Virtual Only

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Program #/Poster #: P632.01

Topic: F.08. Biological Rhythms and Sleep

Title: Diurnal rhythm of dendritic spine density and per1 expression in the infralimbic and prelimbic areas: a potential role for glucocorticoid regulation

Authors: *A. M. MILLER, J. A. SHENG, R. M. DANIELS, R. J. HANDA;
Biomed. Sci., Colorado State Univ., Fort Collins, CO

Abstract: Title: Diurnal rhythm of dendritic spine density and per1 expression in the infralimbic and prelimbic areas: a potential role for glucocorticoid regulation. Circadian disruption caused

by shift work is associated with a myriad of poor health outcomes ranging from cardiovascular disease susceptibility to depression. The ventromedial prefrontal cortex (vmPFC) is a critical regulator of the stress response, autonomic function, and mood. Additionally, vmPFC dendrites are remodeled by adrenal glucocorticoid (GCs), which are critical for stress signaling and the entrainment of circadian rhythms. Due to vmPFC functional overlap with pathologies presented in circadian disruption, we assessed whether the mouse vmPFC displays rhythmicity in cortical dendrite morphology across 24-hour periods. Additionally, we assessed whether core clock genes are regulated by GC signaling in the vmPFC. To measure vmPFC rhythmicity, the present study assessed layer V pyramidal neuron dendritic spine density and core clock gene *Per1* mRNA expression in two key vmPFC subregions, the infralimbic (IL) and prelimbic (PL) areas, across the light:dark cycle. All mice (n=4-6/group) were sacrificed at ZT3 (early light phase), ZT12 (dark phase onset), and ZT16 (middle dark phase). For spine studies, Thy1-eGFP transgenic mice were used to visualize eGFP-labeled pyramidal dendritic arbors in IL and PL. A robust spine rhythm was detected in the IL, showing baseline spine density at ZT3, an increase at ZT12 ($p<.001$ vs ZT3), and peak spine density at ZT16 ($p<.0001$ vs ZT3, $p<.05$ vs ZT12). The PL showed a slight, but significant, upregulation in spine density at ZT16 ($p<.05$), but not at ZT12 or ZT3. In SHAM mice, IL *Per1* mRNA measured by droplet digital (dd)PCR showed peak expression at ZT12 and baseline expression at ZT3 and ZT16. In adrenalectomized (ADX'd) mice, IL *Per1* expression was phase-shifted, displaying peak expression at ZT16 ($p<.05$ vs SHAM ZT16) and baseline expression at ZT3 and ZT12 ($p<.01$ vs SHAM ZT12). SHAM PL *Per1* mRNA showed peak expression at ZT12 and ZT16, and baseline expression at ZT3. ADX'd PL *Per1* expression displayed no rhythmicity, remaining at baseline values at all three timepoints ($p<.0001$ vs SHAM ZT12, $p<.05$ vs SHAM ZT16). These findings demonstrate a daily rhythm of dendritic spine density and core clock gene expression in the vmPFC. Additionally, changes in phase and amplitude of *Per1* expression in ADX conditions implicates a role for diurnal CORT rhythmicity in synchronizing molecular clocks within the vmPFC.

Disclosures: A.M. Miller: None. **J.A. Sheng:** None. **R.M. Daniels:** None. **R.J. Handa:** None.

Poster

P632. Sleep and Rhythms: Cellular and Molecular Contributors I

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Program #/Poster #: P632.02

Topic: F.08. Biological Rhythms and Sleep

Support: NIH Grant R03 AG062883-01
NIH Grant 5P20 GM121310-05

Title: A potential pathway for pain-related disruption of circadian rhythms

Authors: A. E. WARFIELD, P. GUPTA, M. M. RUHMANN, *W. D. TODD;
Zoology and Physiology, Program in Neurosci., Univ. of Wyoming, Laramie, WY

Abstract: Daily rhythms in pain intensity have been observed clinically for years, but only recently have these rhythms and their underlying biology begun to be explored in depth. Additionally, there is some evidence that persistent pain disrupts both central and peripheral circadian rhythms, however this topic has been studied considerably less and the circuit basis for such disruption has never been examined. The signaling pathways for pain heavily utilize the superficial lamina of the spinal dorsal horn and the lateral parabrachial nucleus (LPB), however whether these pathways directly project to the circadian system is less clear. Using retrograde tracing from the subparaventricular zone (SPZ), a heterogenous structure with diverse roles in numerous circadian rhythms at both the behavioral and autonomic level, we have found that there is a considerable innervation coming from the LPB. The LPB cells that send this projection appear to be primarly composed of those expressing FoxP2 as well as dynorphin, a subset of these FoxP2 cells. Importantly, dynorphin LPB cells have been implicated in many aspects of pain including physiological, emotional, and learning responses. We also found that the SPZ shows strong c-fos reactivity at ZT2, ZT6, and ZT14 following formalin injection into the right hind paw, lending credence to the potential for pain-related sigals to disrupt entrained and intrinsic rhythms via this pathway. Indeed, using biotelemetry recordings we find clear evidence of such rhythm disruption following intrplantar injection of a long-lasting inflammatory agent, complete Freund's adjuvant (CFA), in the phase angle of entrainment for locomotor activity (LMA) and body temperature (Tb) for the week following these injections. Phase angle represents a measure of how the circadian system is synchronized to the light-dark cycle, by reflecting the amount of time after light offset that the acrophase occurs. We found that phase angles for both LMA and Tb are consistent across mice before CFA injection but then show profound variation between mice following CFA. Such a change was not seen in saline injected controls, as their phase angles stay consistent before and after injection. Interestingly, we saw a less dramatic change in mean hour-by-hour LMA and Tb, although there was a clear increase in both variables at the dark-to-light transition, again suggesting that pain-related signals may be interfering with the circadian system's ability to properly entrain to the light-dark rhythms as its primary zeitgeber.

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Poster

P632. Sleep and Rhythms: Cellular and Molecular Contributors I

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Program #/Poster #: P632.03

Topic: F.08. Biological Rhythms and Sleep

Support: R01NS092388
5U54GM104942-03

Title: Dim light at night post stroke increases infarct volume and alters neuroimmune activation

Authors: *J. A. LIU[1], W. H. WALKER, II[1], J. R. BUMGARNER[1], O. H. MELENDEZ-FERNANDEZ[1], N. ZHANG[1], J. C. WALTON[1], A. C. DEVRIES[2], R. J. NELSON[1];
[1]Dept. of Neurosci., [2]Dept. of Neuroscience, Medicine, and Div. of Oncology/Hematology, WVU Cancer Inst., West Virginia Univ., Morgantown, WV

Abstract: Cerebral ischemia (stroke) results from impaired blood flow to the brain causing neuronal damage and injury resulting in physical disability or death. Stroke patients are exposed to increased light at night due to increased care requirements, especially during the first few days post-injury. Previous studies identified that disrupted circadian rhythms by exposure to dim light at night (dLAN) alters several aspects of the adaptive and innate immune system including neuroinflammation; thus, we hypothesized that exposure to dim light at night post stroke dysregulates gene expression related to neuroimmune activation and increased infarct size. Twelve month old male CFW mice received a right transient intraluminal right middle cerebral artery occlusion (MCAO) for 30 minutes confirmed by laser doppler flowmetry at the end of the inactive period, then, were either returned to normal light dark (LD; 12 h 150 lux, 12 h 0 lux) conditions, or exposed to dLAN (5 lux). 12 h post stroke, mice received a sensorimotor general and focal neurological deficit score using Clark's Test, then whole cerebral hemispheres were separated into ipsilateral and contralateral hemispheres and were collected for RNA sequencing. A second cohort of mice received a right MCAO, then exposed to the corresponding lighting conditions, and brains were collected after 24 h for infarct visualization using 2,3,5-triphenyltetrazolium chloride (TTC). We observed that mice exposed to dLAN post MCAO had significantly greater infarct volumes and displayed increased general and focal neurological deficits compared to LD conditions. DLAN mice post stroke displayed increased spleen masses relative to LD. We also identified differentially expressed genes and found that positive regulation of inflammatory response genes including (*MIF, S100A8, S100A9, MMP8*) were upregulated compared to LD conditions, whereas heat-shock protein genes (*Hspd1, Hsp90aa1, Hspe1*) were significantly reduced. Together, these findings suggest exposure to dim light at night is especially detrimental in stroke outcome and recovery, and that reducing circadian disruption post cerebral injury could decrease infarct progression.

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Poster

P632. Sleep and Rhythms: Cellular and Molecular Contributors I

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Vanderbilt International Scholarship

Title: Real-time Clock Gene Waveform Dynamics in the SCN Neural Network Encoding Phase Shifts and Entrainment to Photic Input

Authors: *S. KIM[1], D. G. MCMAHON[2];
[1]Neurosci. Grad. Program, [2]Dept of Biol. Sci., Vanderbilt Univ., Nashville, TN

Abstract: The suprachiasmatic nucleus (SCN) is a central circadian clock that orchestrates mammalian physiology and behavior in tune with daily and seasonal light cycles. It is not fully understood how the SCN neurons actively encode phase shifts and entrainment to light input at the molecular and network levels. Here we present new methods to measure molecular circadian rhythms in the ex vivo SCN while it undergoes phase shifts and entrainment. Our approach combines red optogenetic manipulation of clock neuron electrical activity with real-time bioluminescent recording of the clock gene expression, providing precise timing, duration, and intensity of recurring input stimulation to the isolated SCN, and tracking SCN clock molecular rhythms at high spatiotemporal resolution for 3-5 weeks ex vivo. Optogenetically simulated light input transforms the waveform of the molecular clockworks from sinusoidal in free-running conditions to highly asymmetrical trajectories with accelerated synthetic (rising) and extended degradative (falling) phases marking clock advances and delays at simulated dawn and dusk. Daily waveform changes are sufficient to entrain the isolated SCN to non-24h periods, and to different photoperiods with a minimum tolerable night. SCN imaging further reveals that acute waveform shifts are greatest in the ventrolateral SCN, while period effects are greatest in the dorsomedial SCN. Thus, circadian entrainment is encoded by SCN clock gene waveform changes that arise from spatiotemporally distinct intrinsic responses of the SCN neural network.

Disclosures: S. Kim: None. **D.G. McMahon:** None.

Poster

P632. Sleep and Rhythms: Cellular and Molecular Contributors I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P632.05

Topic: F.08. Biological Rhythms and Sleep

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2021 Joint Research Project of Institutes of Science and Technology

Title: Shortened Circadian Period Associated with Enhanced Autophagy in Cereblon-null Mice.

Authors: *J. KIM, J. JUNG, J. KANG, S. LEE, C.-S. PARK, T. KIM;
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Abstract: The circadian rhythm plays a crucial role in many biological processes, such as the sleep-wake cycle, hormone secretion, cardiovascular health, glucose homeostasis, and body temperature regulation. Recently, the level of autophagy was suggested as a regulating factor of circadian rhythm by affecting protein trafficking. Cereblon is a major component of the E3 ubiquitin ligase complex, which mediates the ubiquitination and subsequent proteasomal degradation of its substrates. Interestingly, cereblon ubiquitinates beclin 1, a potent autophagy inducer. Accordingly, the changes in cereblon are likely to affect the circadian rhythm. However, how cereblon can regulate circadian rhythm is not understood yet. Therefore, we hypothesized that cereblon might be an important regulator of the circadian rhythm and sought to investigate the relationship. We obtained 3-month-old CRBN gene knockout (KO) and wild type (WT) mice (n=4 and 8, respectively). The mice were housed in the wheel-based activity recording system under the 12:12 light-dark (LD) condition for one week and constant darkness (DD) condition for the subsequent two weeks. Autophagy-related proteins, including beclin1, microtubule-associated protein light chain-3 (LC3), and p63, were measured by the western blot. Activity data were analyzed by ClockLab Analysis 6 (Actimetrics, USA). We found that the period of circadian rhythm decreased (23.7 vs. 23.4 hours), and the circadian amplitude was increased in CRBN KO mice. The latency from the light-off time to initiating activities in the dark period was shorter in CRBN KO mice and in WT mice. Western blotting revealed that beclin1, LC3, and p62 increased in CRBN KO mice, indicating increased autophagy. Our results demonstrated that CRBN knockout mice showed a shorter circadian period and reduced autophagy-related molecules. We interpret that the lack of cereblon resulted in reduced degradation of beclin1 that triggers autophagy. As per Beesley et al. (2020), an autophagy-induced decrease in cytoplasmic congestion facilitates the protein trafficking for the crucial components of the molecular circadian clock. The shorter circadian period in CRBN KO mice was primarily due to the enhanced autophagy, although its implication in general health is unclear. Taken together, we suggest that CRBN is associated with circadian rhythm via controlling autophagy. Further investigations to understand how circadian amplitude was increased and the activity latency during the dark period was shortened are needed.

Disclosures: J. Kim: None. **J. Jung:** None. **J. Kang:** None. **S. Lee:** None. **C. Park:** None. **T. Kim:** None.

Poster

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Title: Rhythmic transcriptome network and its connection with autism spectrum disorder in mouse brain

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Abstract: The debilitating effects on behavior and cognition caused by autism spectrum disorder (ASD) have had significant impacts on medical and social care world-wide, where its prevalence is 1 case per 68 children. ASD is known to be influenced by transcriptome changes and is also associated with circadian rhythms, the 24 hour cycles of behavioral and physiological changes controlled by the internal biological clock. However, the connection between ASD and circadian rhythms has not been clearly identified at the transcriptome level. We used RNA-seq from wild type mouse brain to first test the connection at the tissue level. Across eight brain tissues (e.g., Olfactory bulb (OB), frontal cortex (FC), striatum (ST), parietal cortex (PC), hippocampus (HI), temporal cortex (TC), occipital cortex (OC), and cerebellum (CB)), we found gene expression rhythms that shared a common pattern in overall phase distribution. Enrichment studies also showed that their rhythmic transcriptomes are shared across brain tissues and are significantly enriched for ASD-risk genes. However, multidimensional scaling studies identified differences in both phase and amplitude. For phase data, HI and FC were different, with their average phases uniquely within the circadian time (CT)19-1. OB and CB exhibited phase shifted genes that show shifts between day and night, which were found to be fear response genes in neuronal synapses. For amplitude data, the rhythmic transcriptomes in OB and OC exhibited small amplitudes, in contrast to FC that had the largest amplitude. Using the phase and amplitude data, circadian rhythm networks could be identified across the brain tissues. Interestingly, OB and CB showed weak connection with the other brain tissues that had strong interconnections in the circadian rhythm network. Notably, FC is involved in social, language, communication, cognitive functions, which are the most disrupted functions in ASD patients. Therefore, I further studied FC transcriptome at the single-cell level using single nucleus RNA-seq. I identified 47 cell clusters from FC and their cell numbers from high to low in the following the order: neuron, astrocyte, oligodendrocyte, microglia, and endothelium. Among them, neurons drive most of the rhythmic transcriptome in FC tissue. Neuronal transcriptomes showed two clear, major peaks of phase that matched with tissue-level phase, and their amplitudes were smaller than non-neuronal transcriptomes. A gene set enrichment study showed that only neurons showed strong enrichment with ASD-risk genes. In conclusion, genome-wide transcriptome analyses showed a significant connection between the rhythmic transcriptome and ASD in neurons.

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Poster

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Program #/Poster #: P632.07

Topic: F.08. Biological Rhythms and Sleep

Title: Circadian clock and stem cell gene expression in quiescent glioma spheroid cultures

Authors: V. P. SHARMA[1], A. DE[2], ***M. E. GEUSZ**[3];
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Abstract: Understanding how cancer cells of the nervous system are regulated by circadian rhythms can indicate daily phases when cancers are most vulnerable to treatments. Cancer stem cells within tumors also need to be further characterized because they resist some cancer treatments by remaining mostly quiescent and then producing tumor recurrence by differentiating into rapidly dividing cancer cells. Glioblastomas contain cells of varying states of differentiation including glioma stem cells (GSCs) according to studies of tumorsphere cultures that serve as models of tumor organization. Tumorspheres made from rat C6 glioma cells express circadian rhythms of core clock gene Per2 that is often suppressed in cancer cells, indicating a tumor cell heterogeneity that depends on the culture microenvironment. Furthermore, C6 tumorspheres maintained in culture media lacking added growth factors remain mostly quiescent and produce large amplitude circadian rhythms. Because arrested cell division is typical of GSCs in many tumors we tested whether tumorspheres from a human glioma could also be maintained in this quiescent state. We found that tumorspheres from U251 human astrocytoma cells survive for weeks under quiescent culture conditions. Stem cell marker proteins SOX2, CD133, and MSI1 were expressed in different tumorsphere regions, and their corresponding genes were moderately expressed in published RNA-Seq data from U251 cell cultures where GSCs are sparse, contrasting with their expression in tumorspheres. High HIF1A expression in tumorspheres indicated a hypoxic microenvironment selective for GSCs. Tumorspheres induced to differentiate into cells expressing astrocyte (GFAP) and neuronal (MAP2) markers validated the multipotency of quiescent tumorsphere GSCs. We used RNA-Seq databases to compare astrocytes with U251 cells, which revealed increased expression of the TGFB1/SMAD3 stem cell pathway and NFkappaB signaling (RELA) in U251 cells. Expression of core circadian clock gene CLOCK and circadian-related gene ARNTL2 (BMAL2) was also elevated along with SERPINE1, a gene target of these two transcription factors that facilitates cancer cell migration and metastasis. Other circadian clock gene expression was lower in U251 cells, and to test functioning of the partially repressed genes we knocked down expression of core clock gene Arntl (BMAL1) in C6 cells. The Arntl antisense oligonucleotide inhibited epithelial-mesenchymal transition into viable quiescent cells suggesting a role for BMAL1 in producing or

maintaining quiescent cancer cells and tumorspheres. These results identified potential target genes for suppressing glioblastomas.

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Poster

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Program #/Poster #: P632.08

Topic: F.08. Biological Rhythms and Sleep

Support: DA039841

Title: High-throughput measurement of molecular rhythms reveals genetic heritability and quantitative trait loci of circadian phenotypes and their correlation with cocaine addiction in genetically diverse mouse populations.

Authors: *S.-M. KIM[1], V. M. PHILIP[2], L. H. GAGNON[2], P. E. DICKSON[2], S. SCHOENROCK[3], L. M. TARANTINO[3], E. J. CHESLER[2], R. W. LOGAN[1], C. A. MCCLUNG[1];
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Abstract: Circadian clocks coordinate many brain processes including reward-related pathways, and circadian disruption is commonly observed in nearly all individuals with substance use disorders. Accumulated evidence suggests that substance use alters endogenous clock machineries and circadian disruption leads to increased vulnerability for addiction, but the genetic mechanisms underlying this bidirectional interaction is not fully understood. Diversity Outbred (DO) and Collaborative Cross (CC) mice are powerful animal models for investigating the genetics of complex traits because genetic combinations of their eight founder strains including five common-inbred and three wild-derived strains provide high genetic and phenotypic diversity compared to conventional inbred mouse strains. To understand genetics of circadian rhythms and their relationship with drug addiction, our study performed a high-throughput assay to phenotype circadian rhythms and cocaine addiction-related behaviors using DO, CC and their founder strains. Among the founder strains, we found strong heritability and significant strain by sex effects for period and amplitude in molecular rhythms. Extreme period differences in molecular and behavioral rhythm were observed between the inbred A/J, which have the longest period, and the wild-derived CAST/EiJ, which have the shortest period. Our preliminary correlation analysis in the founders revealed that longer periods were associated with more relapse-like behaviors while reduced amplitude was associated with drug-seeking behaviors in cocaine self-administration (IVSA). In CC strains, CC004/TauUnc displayed

significantly higher addiction-related behaviors in cocaine-sensitization and IVSA relative to CC041/TauUnc. CC004/TauUnc also exhibited a longer circadian period and lower amplitude in molecular and behavioral rhythms relative to CC041/TauUnc mice. Furthermore, we characterized molecular rhythms in 329 DO mice for genetic mapping. They displayed more circadian phenotypic variability than the founders: 80% of founders compared to only 25% of DO mice had periods of ~24 h. Genetic mapping identified several quantitative trait loci for molecular rhythms in period and amplitude. Collectively, our findings demonstrate that genetic diversity contributes to phenotypic variability in circadian rhythms and prove that cell-based high-throughput screening of fibroblast rhythms in DO mice is a tractable system for high-precision genetic mapping.

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Poster

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Title: Mosquito Cryptochrome is a Well-Conserved Key Photoreceptor for Light-Mediated Behaviors in Flies

Authors: *D. D. AU, A. J. FODEN, S. PARK, M. TRAN, O. JAIME, T. C. HOLMES; Univ. of California, Irvine, Irvine, CA

Abstract: Light-evoked excitation of the ventral lateral subset of circadian neurons in Drosophila is mediated by the cell autonomous short-wavelength light-sensitive photoreceptor Cryptochrome (CRY), and to a lesser extent, cell autonomously expressed Rhodopsin 7 and photic inputs from the eyes. CRY also regulates the circadian molecular clock via light-induced degradation of the Timeless (TIM) clock protein, as well as avoidant/attractant behavioral responses to strong UV light. We explored the behavioral and electrophysiological light-responsiveness of CRY1s expressed by day-biting Aedes aegypti (Ae) and night-biting Anopheles gambiae (Ag) transgenically expressed in a cry-null Drosophila background. AeCRY1 and AgCRY1 partially rescue the UV (365 nm, 250 µW/cm2) light-excitatory electrophysiological responses of large lateral ventral arousal neurons (lLNvs). AgCRY1 further phenocopies dCRY's neuronal excitatory light-responses shown by blue (450 nm) and red (630 nm) light-evoked electrophysiological responses in lLNv. Our immunocytochemistry

experiments show under 12:12 LD that the phase of TIM cycling for dCRY, AgCRY1, and AeCRY1 are similar. However, the pattern of locomotor behavior under 12:12 LD in AeCRY1 expressing flies is similar to cry-null, while AgCRY1 expressing flies showed similar LD locomotor behavior compared to dCRY expressing flies. Furthermore, attraction/avoidance behaviors to UV light during the day track the distinct differences between daytime and nighttime active species' CRY; dCRY or day active AeCRY1 expressing flies are attracted to daytime UV light, while night active AgCRY1 expressing flies show strong daytime avoidance of UV light. These findings highlight CRY as a key photoreceptor that mediate light-mediated behaviors in a species specific fashion. This may useful for developing more efficient light-based insect control methods.

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Poster

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Title: The regulation of vasoactive intestinal peptide expression by ventral anterior homeobox 1 results in altered circadian output from the suprachiasmatic nucleus

Authors: *B. DEVRIES[1], J. BREUER[3], A. YAW[1], B. JACKSON[2], D. NGUYEN[1], M. GORMAN[4], L. SEMPERE[2], E. V. HO[5], P. L. MELLON[5], H. M. HOFFMANN[1,5];
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Abstract: The suprachiasmatic nucleus (SCN) is a brain structure in the hypothalamus important for circadian timekeeping and relaying environmental day-night information to the body. Ventral anterior homeobox 1 (Vax1) is a homeodomain transcription factor that is required for SCN development and function. Previous studies have shown that balanced expression of vasoactive intestinal polypeptide (VIP) and arginine vasopressin (AVP) is required for correct SCN period.

As VAX1 has an established role as an activator of VIP expression and 100% of postnatal day 2 VIP neurons were found to express Vax1, we hypothesized that Vax1 is required for VIP neuron maturation, which may play a role in SCN circadian function. To understand the role of VAX1 in VIP neurons, we conditionally deleted Vax1 in post-natal VIP neurons, generating Vax1-flox/flox:Vip-Cre+ (VaxVIP) mice. To examine whether VaxVIP mice had impaired SCN output, we evaluated wheel running behaviors in constant darkness, which reveals intrinsic SCN rhythms. VaxVIP male and female mice displayed a shortening of the endogenous free-running period (Tau) of the SCN, showing shortened circadian output from the SCN. This shortened behavioral rhythm was reflected in Per2-luciferase ex vivo tissue recordings. Loss of VIP in the SCN is known to shorten Tau, leading us to examine Vip levels. Indeed, VaxVIP mice had reduced VIP mRNA and protein in the SCN. Interestingly, VaxVIP mice had increased Avp expression, which could contribute to the shortened behavioral period. In addition to regulating SCN period through imbalance of VIP and AVP, our data also identified a direct effect of Vax1 on the expression of the molecular clock genes Bmal1 and Per2. Together, these data show a dual role of Vax1 in VIP neurons, where it is required for VIP expression, and impacts VIP neuronal connections to target neurons, leading to an increase in Avp expression. In addition, VAX1 also regulates molecular clock function through a direct action on the Bmal1 and Per2 promoters. In conclusion, we have identified a novel role for VAX1 in maturing SCN VIP neurons, where it is required for both Vip expression and molecular time keeping function.

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Poster

P636. Sleep: Regulation

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Title: Genetic and neuronal substrates of melatonin signaling in zebrafish sleep

Authors: *A. HILL, D. PROBER;
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Abstract: Sleep appears to be conserved across the animal kingdom, and adequate sleep is essential for human health. However, despite its ubiquity and importance, surprisingly little is known about the genetic, molecular, and neuronal mechanisms that govern sleep. Exogenous

melatonin (MT) is recognized as a somnogen in humans, but endogenous MT, whose synthesis is controlled by the circadian clock, has only recently been shown to be required for normal sleep levels. Using the diurnal vertebrate zebrafish, our lab demonstrated that loss of an enzyme essential for MT synthesis causes a dramatic decrease in sleep at night. We also found that loss of MT has no effect on circadian rhythms but abolishes the circadian regulation of sleep, indicating that MT acts downstream of the clock to promote sleep. We are now investigating the mechanisms by which MT promotes sleep. The human genome encodes two MT-binding receptors, MT1 and MT2, whose effects on sleep are not well-understood. The zebrafish genome encodes six MT receptors: 3 that are orthologous to mammalian MT1, 2 that are orthologous to MT2, and an additional receptor not present in mammals. We created zebrafish lines harboring mutations in each of the six receptors, as well as double and triple mutant combinations. We found that zebrafish larvae lacking all three MT1-type receptor paralogs sleep less at night, similar to MT-deficient animals, and are nearly totally resistant to sleep induced by exogenous MT. Next, using *in situ* hybridization chain reaction (HCR), we found that the MT1 receptors are primarily expressed in the periventricular grey zone of the optic tectum, a structure homologous to the superior colliculus of mammals, and in a discrete cluster of cells in the hindbrain. We are currently optimizing HCR to better ascertain the location of these receptors, which we anticipate will point to critical sleep-regulating parts of the brain. Last, we are using 2-photon selective plane illumination microscopy (2P-SPIM) to image the entire brain of larval zebrafish expressing the calcium sensor GCaMP6f pan-neuronally. We hypothesize that animals treated with MT will exhibit specific changes in neuronal activity, which may represent a subset of neuronal changes observed during normal sleep, and that these MT-induced changes are not observed in larvae lacking the relevant melatonin receptors.

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Poster

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Title: Quantitative EEG during REM sleep in patients with frontal brain tumor

Authors: *L. ORTEGA-LEONARD, Y. DEL RÍO-PORTILLA;
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Abstract: The frontal lobes previously have been associated in the regulation of eye movements and with the cognitive changes during the dreams in REM sleep stage. This can be explained by the changes in the electroencephalographic bands and in the temporal coupling of the frontal areas with other brain regions. Likewise, electrophysiological sleep patterns can be modified because of a brain pathology, such as tumors, which allows us to know in depth what happens when these lesions affect a particular area of the brain, such as the frontal lobes, and which are scant studies about it. Whereby, ten patients with frontal brain tumor underwent polysomnographic study. They had a mean age of 43.4 years (SD=11, three women and three in the left hemisphere). Patients were registered in the absence of treatment (surgery, chemotherapy or radiotherapy) and without neurological, psychiatric or autoimmune comorbidity. REM sleep stages were selected, inspected for artifacts and analyzed for the first episode from the second half of the night. For the analyses quantitative EEG (qEEG), Fast Fourier Transformation (FFT) algorithm was used to obtain the power spectral for each participant, channel and the following frequency bands: Delta2 (1-3.5 Hz), Theta (3.5-8 Hz), Alfa (8-12 Hz), Sigma1 (12-14 Hz), Beta1 (16-24 Hz), Beta2 (24-32 Hz) and Gamma (32-48 Hz). According to the results, higher spectral power densities for REM sleep, for all electrodes and for each of the bands; can be observed by patients compared to the normative values (Hertenstein et al., 2018). No significant differences by sex, laterality of the tumor, specific location of the frontal lobe (medial, lateral and orbital) and size of the lesion (volume and edema) in the spectral power. Also, there were significant differences in the spectral power in the delta band ($p = 0.003$) and in the total spectral power ($p = 0.02$) between the anterior and posterior electrodes. The changes observed in the spectral power in patients with frontal brain tumor can be considered as a plasticity effect (compensatory reaction) in the sample recorded in this study. Finally, the presence of this type of pathology, which causes structural and functional changes, allows us to understand the organization of the brain during this state.

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Poster

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Title: Basal forebrain astrocytes promote sustained wakefulness through G_q-protein-coupled pathways

Authors: *A. M. INGIOSI, C. R. HAYWORTH, M. G. FRANK;
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Abstract: Sleep disorders continue to be a prevalent health issue, but the cellular and molecular substrates of sleep regulation are incompletely understood. Understanding how sleep is regulated is necessary to better understand the underlying causes and consequences of abnormal sleep and sleep disorders. Accumulating evidence indicates astrocytes contribute to sleep regulation and behavior via intracellular calcium signaling. We recently showed that astroglial calcium activity 1) changes dynamically across arousal states, 2) increases with sleep need, and 3) plays a role in compensatory responses to sleep loss. We are now determining which astroglial signaling pathways act upstream of calcium activity to influence sleep-wake behavior. The latter are likely to involve neurotransmitters like noradrenaline which signal through G-protein-coupled pathways to increase astroglial calcium activity. To test this hypothesis, we used a chemogenetic approach to activate G_q signaling in astrocytes in the basal forebrain and measured sleep-wake behavior and electroencephalographic (EEG) activity. G_q DREADDs (i.e., Designer Receptors Exclusively Activated by Designer Drugs) were expressed selectively in astrocytes by crossing CAG-LSL-G_q-DREADD mice with Aldh1l1-cre mice. Male and female adult (10 - 14 weeks-old) mutant mice (n = 9) and wildtype littermates (n = 7) were stereotaxically implanted with electrodes for EEG and electromyographic (EMG) determination of sleep-wake behavior as well as bilateral guide cannulae in the basal forebrain for local delivery of DREADD ligands. We used a within subject, counterbalanced design to assess the impact of G_q DREADD activation via the DREADD ligand clozapine N-oxide (CNO) versus vehicle. CNO (or vehicle) was injected at light onset and mice were then allowed to sleep undisturbed for 24 h. We found that CNO activation of G_q DREADDs in basal forebrain astrocytes induced sustained wakefulness that could exceed 6 hours—an effect not observed after vehicle treatment or in control mice. JHU37160, a CNO alternative, also induced sustained wakefulness in G_q DREADD-expressing mice. Ongoing analyses and experiments will determine the impact of this sustained wakefulness on EEG spectra, sex differences, and which neuronal populations are responsive to this astroglial-induced wakefulness. Overall, we present preliminary evidence that basal forebrain astrocytes play a role in mediating sleep and wakefulness through G_q signaling pathways.

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Poster

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JSPS KAKENHI 18968064

Title: Perturbation in Hdac4 signaling leads to dysregulated NREMS

Authors: *S. J. KIM[1], N. HOTTA-HIRASHIMA[1], N. ASAMA[1], T. MATSUOKA[1], T. FUJIYAMA[1], A. IKKYU[1], M. KAKIZAKI[1], S. KANNO[1], S. MIZUNO[2], S. TAKAHASHI[2], C. MIYOSHI[1], H. FUNATO[1,4], M. YANAGISAWA[1,5,3];
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Abstract: Sleep is observed universally across various species from nematodes to mammals and the properly maintained sleep is directly linked to the general well-being of the organism. Much advances has been made in the identification of the distinct groups of neuronal population that are involved in the transitions between sleep and wake states. These networks of neurons can drive vigilance stage switches and changes in neuronal activity related to sleep and wakefulness. Despite the growing interest and findings in the maintenance of sleep/wakefulness, the intracellular regulatory mechanism of the response to sleep need changes remains largely unknown.
Our EEG/EMG-based forward genetics approach in mouse sleep/wake behavior has successfully identified several sleep regulatory genes. *Sleepy2* is one of such gene locus with a single nucleotide change in the splice acceptor site that leads to out-of-frame mutation. The heterozygous mutant mice with the resulting splice variants showed increase in both daily non-REM sleep (NREMS) time and slow-wave activity during NREMS. Interestingly, a phospho-deficient mutant type mice showed a complete opposite trend in sleep/wake behavior. These mice showed increase in daily wake time and significant decrease in the slow-wave activity during NREMS. These results suggest canonical signaling of *Sleepy2* gene is involved in the regulation of NREMS.
For further understanding of specific neuronal network where the identified sleep regulators may function, we employed a neuron type-specific manipulation engineered by Cre-loxP recombination. From the comparison of various *Cre* reporter lines, distinct neuronal types were identified to regulate the NREMS time and the slow-wave activity. It is plausible *Sleepy2* signaling modulates the amount and quality of sleep discretely neuronal network in type- and region-specific manner.

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Poster

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Title: Molecular hydrogen alters NREM delta oscillations during the recovery from sleep loss and BMAL1 in skeletal muscle

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Abstract: Sleeping species have an intrinsic need for sleep that is distinct from the circadian clock. Sleep need is determined by a system which interprets wake- and sleep-experience and generates sleep pressure to maintain sleep-wake homeostasis. The mechanisms underlying this homeostatic process are enigmatic, but we can measure sleep pressure through electroencephalography (EEG) changes in the delta band (0.5-4.0Hz) during non-rapid eye movement (non-REM) sleep in mammals. Acute sleep loss modulates pro-inflammatory mediators, impairs memory, and decreases insulin sensitivity. Behavioral interventions for improving sleep are often inadequate to resolve environmental- or disorder-driven sleep disturbances. Pharmaceutical hypnotics are typically accompanied by undesirable side-effects and long-term use can lead to dependence. Molecular hydrogen is a promising therapeutic with a wide range of potential benefits, including the regulation of pro-inflammatory mediators, and the modulation of insulin sensitivity. Molecular hydrogen has no known toxicity and is widely accepted to have no deleterious side effects. Here, male and female C57BL/6 mice were implanted with head-mounts for polysomnography containing two electrodes for EEG (frontal and parietal) and two electrodes for electromyography (nuchal). At 12-16 weeks, a 24-hour recording of baseline/spontaneous sleep was collected, immediately followed by 6 hours of total sleep deprivation and an 18-hour recovery period. One week later, baseline, sleep deprivation and recovery were evaluated again following 7 days of ad libitum access to hydrogen-rich water (HRW) at 0.7 -1.4 mM during the active phase. Two-way ANOVA revealed no significant differences in undisturbed or recovery sleep amounts, nor in delta power during spontaneous sleep. Significant differences were found in relative delta power during recovery sleep, suggesting that HRW may be sufficient to alter the response to the challenge of sleep loss. Oral ingestion of HRW does not increase hydrogen in the brain, but it does increase hydrogen in femoral arteries. Brain and Muscle-ARNT-like factor (Bmal1/BMAL1) in skeletal muscle is sufficient to regulate numerous aspects of sleep in mice. Hydrogen increases the phosphorylation of muscle glycogen synthase kinase 3 beta (GSK-3β), which is critical for preparing BMAL1 for degradation. We show here that HRW administration by oral gavage (0.25ml, 1.4mM) 30

minutes prior to tissue collection is sufficient to significantly increase BMAL1 protein levels in gastrocnemius of mice, providing a plausible mechanism through which HRW might influence sleep.

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Poster

P636. Sleep: Regulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P636.06

Topic: F.08. Biological Rhythms and Sleep

Support: JSPS Grant 21K07538
JSPS Grant 19K08037
AMED Grant JP20dm0107162

Title: Exploring the function of orexin and neurotensin as NREM and REM sleep modulators by the measurement of neuropeptides in the cerebral spinal fluid of hypersomnolence patients

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Abstract: While recent studies reveal an orexin-mediated wake-promoting system, molecular mechanisms regulating non-rapid eye movement (NREM) and REM sleep are less clear. We uncovered that specific neurons in the brainstem are crucial for the switching between NREM and REM sleep in mice (Hayashi et al., 2015). Further, neurotensin produced by a subset of these neurons appears to contribute to inhibiting REM sleep (Kashiwagi et al., 2020). In contrast to cumulative evidence from animal studies, only a few clinical observations demonstrate the direct relationship between sleep and neuropeptides. Here, we analyzed targeted neuropeptides in the cerebral spinal fluid (CSF) of hypersomnolence patients and compared their levels with reference to patients' sleep phenotypes. In this study, the concentration of orexin, neurotensin, melanin-concentrating hormone (MCH), and corticotropin-releasing hormone was measured with

radioimmunoassay or enzyme-linked immunosorbent assay in the CSF samples from more than 30 patients. CSF collections were conducted according to the protocols approved by the ethics review committee at Akita University and University of Tsukuba. The minimum measurable CSF volume for each neuropeptide was 0.1 mL for orexin, neurotensin, and CRH, or 0.25mL for MCH. Based on the absolute concentration, we found a positive correlation between the levels of orexin and neurotensin, both of which are known to suppress REM sleep in rodents. However, no significant correlation between other neuropeptides was detected from the patients' CSF samples. The results indicate that lowered levels of both orexin and neurotensin might be responsible for frequent occurrences of REM sleep seen in the patients. We further explore the role of these neuropeptides in sleep regulation, analyzing any relationship between the amount of NREM sleep or REM sleep and their concentrations.

Disclosures: S. Miyazaki: None. **K. Yoshizawa:** None. **T. Kodama:** None. **H. Ishido:** None. **A. Imanishi:** None. **M. Kashiwagi:** None. **T. Sasajima:** None. **S. Shimizu:** None. **S. Chiba:** None. **H. Kondo:** None. **Y. Hayashi:** None. **T. Kanbayashi:** None. **M. Kimura:** None.

Poster

P636. Sleep: Regulation

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Program #/Poster #: P636.07

Topic: F.08. Biological Rhythms and Sleep

Title: Brain waveforms during nrem sleep are altered in a mouse model for dup15q syndrome

Authors: *Y. G. GONZALEZ[1], K. N. PAUL[1], I. S. NICHOLS[2], S. M. VINCENT[1], S. WIMMER[1], D. TROTTER[1], V. SARAVANAPANDIAN[3], P. GOLSHANI[4];
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Abstract: Sleep problems are highly prevalent in autism spectrum disorders (ASD). Duplications of 15q11.2-13.1 (Dup15q syndrome) are highly penetrant for ASD, intellectual disability, hypotonia and epilepsy. In clinical overnight sleep EEG recordings, children with Dup15q syndrome demonstrate elevated beta band oscillations and abnormal NREM sleep physiology. In this study, we use a maternally inherited 15q duplication mouse model (Takumi model) that recapitulates many of the behavioral phenotypes observed in individuals with this condition, yet sleep phenotypes have not been characterized. Male and female adult Takumi mice (12-20 weeks) and wild-type controls were implanted with EEG/EMG recording electrodes in the frontal (EEG1) and parietal (EEG2) cortex for polysomnographic analysis of sleep and wake states during 24 hour of spontaneous sleep, and again after six hours of total sleep loss. Two-way ANOVA revealed no significant differences in NREM or REM sleep amount between Takumi mutant mice and wild-types during baseline (spontaneous) recording. However,

significant differences were found in relative delta and alpha brain waves in the parietal cortex during NREM sleep with respect to genotype, and a significant interaction of time and genotype in beta waves in the parietal cortex. The frontal cortex did not show any significant effects. These results suggest that NREM sleep features may be altered in this Dup15q mouse model.

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Poster

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Program #/Poster #: P636.08

Topic: F.08. Biological Rhythms and Sleep

Support: NIH T32 AR065972

Title: Molecular hydrogen decreases spontaneous sleep latency in mice

Authors: *K. GOLDEN[1], S. M. VINCENT[1], M. MADANI[1], C. JACKSON[1], T. LEBARON[3], A. TARNAVA[4], I. S. NICHOLS[2], K. N. PAUL[1];
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Abstract: There are numerous consequences of sleep loss, and it is important to find therapies which attune its negative effects- impaired memory, increased stress, and inflammation among others. Molecular hydrogen (H2) has been studied to determine if it acts as a therapy for several disorders. Several reports have demonstrated that H2 suspended in water (hydrogen rich water, HRW) regulates proinflammatory mediators (LeBaron et al. 2019). Additionally, HRW appears to have a neuroprotective effect for dopaminergic cells in a rodent model of Parkinson's disease (Yoshii et al. 2017). We investigated how the administration of HRW changes basic sleep architecture in baseline conditions and in response to sleep loss using a mice model. All mice were implanted with intracranial electroencephalography (EEG) and intra-nuchal electromyography (EMG) electrodes. After surgery and acclimation to a reverse light/dark cycle, a 24-hour baseline recording was taken, and mice were then sleep deprived during the first 6 hours of their inactive phase via gentle handling and were given 18 hours for recovery. After recovery from their untreated baseline, sleep deprivation and recovery periods, mice had ad libitum access to HRW for seven days. Another 24-hour baseline recording was taken, followed by 6 hours of sleep deprivation via gentle handling and another 18-hour recovery period. All files were scored manually by sleep experts utilizing Sirenia Sleep Pro, and each 10 second epoch was scored as wake, NREM, REM or artifact. Paired t-tests revealed no significant difference in NREM or REM sleep amount during baseline or recovery sleep. However, we did observe a

significant decrease in baseline sleep latency following 7 days of HRW, suggesting HRW may be a useful therapeutic for improving sleep onset.

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Poster

P644. Energy Metabolism II

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P644.01

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: BBRF NARSAD 2019 Young Investigator Award
NIH R01DK114279

Title: A novel MC4R neurocircuit for integrative control feeding and emotional status

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Abstract: Maladaptation in form of exaggerated feeding responses to risk indicators causes eating disorders that characterized by severe disturbances in both eating behaviors and emotional changes. Despite the well-recognized association between controlling emotional states and feeding behaviors, the brain neuropathway that underlies the connection between these two behavioral programs is still unclear. Lateral septum (LS) is a subcortical forebrain region known to regulate anxiety and stress. Our recent data found that ventral LS (LSv) receives abundant monosynaptic glutamatergic projections from Sim1-expressing neurons of paraventricular nucleus of hypothalamus (PVH), the brain region known extremely important for homeostatic regulation, including feeding and physiological adaptions for survival. We further revealed that MC4R, a key gene for feeding regulation and most frequent monogenic cause of human obesity, but not other subtypes of PVH Sim1 neurons (i.e. oxytocin, AVP, or CRH neurons) project to the LSv (denoted as PVH^{MC4R}→LSv neurocircuit). Optogenetic activation of the PVH^{MC4R}→LSv neurocircuit resulted in strong negative valences of anxiety-related responses, including increased self-grooming and real time place aversion, and produced significant inhibition to hunger-driven feeding in fasting mice. Of note, when the food was paired with laser stimulation, aversion induced by photostimulating PVH^{MC4R}→LSv projections was able to significantly abolish the food seeking motivation in fasting mice. Strikingly, chronic inhibition of PVH-projected LSv neurons led to robust obesity, but chronic activation of these neurons showed the resistance to high-fat-diet induced body weight gain. Furthermore, we found that specific

disruption of glutamate or MC4R neurotransmission from LSv-projecting PVH neurons resulted in the high susceptibility to obesity development. Altogether, our findings show that LSv is a novel extra-hypothalamic center essential for energy homeostasis, and the PVH^{MC4R}→LSv neurocircuit is a novel shared neural pathway for controlling feeding and anxiety-related emotional states.

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Poster

P644. Energy Metabolism II

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P644.02

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Title: The Role of the Paraventricular Hypothalamus in the Rhythmic Regulation of Feeding and Metabolism

Authors: R. VAN DRUNEN[1], B. FEKRY[1], R. BRAVO SANTOS[1], X. CHEN[3], H. WEI[2], Y. DAI[4], Z. ZHAO[3], J. WU[2], Q. TONG[1], K. MAHAN[1];
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Abstract: Obesity has reached epic proportions, however, current therapeutics are limited in efficacy and the underlying mechanisms of obesity are not fully understood. Recently, circadian rhythms have become inextricably connected to obesity. Particularly interesting for its circadian involvement, is the paraventricular nucleus (PVN) of the hypothalamus which functions as both an integrator and regulator of energy homeostasis. We discovered that PVN-specific deletion of the core clock gene, BMAL1 (Brain and Muscle ARNT-Like1), elicits arrhythmic patterns of feeding, metabolism and activity along with obesity and insulin resistance in the *Bmal1*-PVN knockout (KO) mice. We hypothesize molecular mechanisms driven by BMAL1 activity regulates rhythmicity of the PVN to mediate diurnal energy balance. Using bilateral injections of adenovirus-mediated (AAV)-Cre-GFP delivered to the PVN of $Bmal1^{fl/fl}$ mice, we produced *Bmal1*-PVN KO mice. Under infrared sensors to measure locomotion, *Bmal1*-PVN KO mice showed a loss of diurnal rhythms in activity even under entrained conditions. Furthermore, metabolic cage analysis revealed a loss of rhythmicity in energy intake. To test whether arrhythmic energy intake is the primary driver of disrupted circadian activity in these mice, we used an 8-hr time restricted feeding (TRF) paradigm in attempt to restore rhythmicity in the *Bmal1*-PVN KO mice. Two weeks of TRF successfully restored behavioral rhythmicity of the *Bmal1*-PVN KO mice, suggesting that altered energy intake, and not rhythmicity defects in the

master clock known as the suprachiasmatic nucleus (SCN), were the primary drivers of the arrhythmic phenotype. Post-mortem analysis of the tissues from TRF and non-TRF *Bmal1*-PVN KO mice demonstrated a rescue of rhythmic gene expression in mice undergoing TRF, and normal expression of BMAL1 in the SCN of both groups. To understand mechanisms by which BMAL1 activity controls rhythmic PVN function, we conducted chromatin immunoprecipitation sequencing (ChIP-seq) on PVNs harvested from C57/bL6/J mice at the peak of BMAL1 binding for several tissues analyzed to date (i.e. *Zeitgeber* time 8) using an antibody to BMAL1. Interesting targets relevant to synaptic plasticity and neuronal function were validated using ChIP-qPCR. ChIP-seq of PVN BMAL1 targets revealed 832 significant genes involved in several neuronal processes, including synaptogenesis, mitochondrial energetics, and cellular senescence. Our data support BMAL1 in the PVN as essential for diurnal energy balance independent of the SCN. We have identified multiple potential candidate genes which may serve as targets for future obesity treatments and therapeutics.

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Poster

P644. Energy Metabolism II

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P644.03

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIH Grant R01DK114279
Russell and Diana Hawkins Family Foundation Discovery Fellowship

Title: Role of VTA local neurocircuits in regulating obesity resistance

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Abstract: Currently, over 40% of US population have been diagnosed with obesity, which leads to severe metabolic results and poor quality of life. Yet, there is no effective therapy to cure obesity. Thus, identification of potential therapeutic targets is urgent. The Ventral Tegmental Area (VTA) is a brain region known to regulate various cognitive and emotional processes as well as reward consumption. Neuronal signaling in the VTA is implicated in obesity development in humans and manipulating VTA neuronal activities in rodents impacts eating behaviors. VTA GABAergic neurons accounts for 30% VTA neurons and send abundant inhibitory inputs to other local neurons. Whether and how VTA local GABAergic circuits are

involved in body weight regulation is unclear. To understand the function of VTA GABAergic neurons, a mouse model with chronic inhibition of VTA GABAergic neurons has been established by expressing mutant inward-rectifier potassium ion channels (Kir2.1) in those neurons using stereotaxic injections. These mice had lower body weight under chow diet and were resistant to high fat diet (60% fat diet) induced obesity. The decreased body weight was mainly due to increased energy expenditure and increased locomotion as measured by metabolic cages. The immunostaining results showed that putative local glutamatergic neurons were disinhibited. In addition, a model with chronic activation of VTA glutamatergic neurons was established by expressing bacterial sodium channels (NaChBac) in VTA glutamatergic neurons. Chronic activation of VTA glutamatergic neurons led to reduced body weight. Consistent with these results, acute chemogenetic inhibition of VTA GABAergic neurons mediated by designer receptors exclusively activated by designer drugs (DREADD) reduced fasting-refeeding food intake and increased locomotor activity. Meanwhile, chemogenetic activation of VTA glutamatergic neurons increased fasting refeeding food intake. Thus, it appears that chronic inhibition of VTA GABAergic neurons reduces body weight potentially through disinhibiting local glutamatergic neurons. Further experiments are required to verify the anatomical and functions connections. In sum, our current findings support a novel function of VTA GABAergic neurons and local circuits in regulating body weight, especially in regard to resistance to diet-induced obesity. Given the current obesity epidemic, this study will likely have a significant impact on research aiming to identify strategies to reverse obesity development.

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Poster

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Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: Pilot and Feasibility Grant, Sex Differences in Metabolic Syndrome SCORE, Iris Cantor – UCLA Women's Health Center
NSF GRFP

Title: Integration of Metabolic and Reproductive Cues in a Novel Hypothalamic Node of the Feeding Circuit

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Abstract: Recent research has identified somatostatin-expressing neurons within the tuberal nucleus of the hypothalamus (SSTtn) as modulators of food intake in mice. As feeding varies between females and males, we hypothesized that SSTtn neurons exhibit functional differences across sex. To test this hypothesis, we used the Cre-lox system and viral stereotaxic injections to selectively manipulate SSTtn neurons of 8-16-week-old *Sst*-Cre mice. Chemogenetic activation of SSTtn neurons via Gq-coupled DREADDs induced food intake in female and male mice. In contrast, selective ablation of SSTtn neurons via virally delivered Caspase-3 decreased food intake in females only. Ablation also revealed an effect of estrous stage, with food intake only decreasing during the night of proestrus when ovarian hormones like estrogen are rising. Surprisingly, this effect of estrous stage was dependent on body weight. While weight inversely correlated with food intake during the night of proestrus, ablation eliminated the weight-dependency of this effect. This indicates that SSTtn neurons are necessary for the integration of body weight and reproductive cues, promoting feeding in animals with low body weight despite hormonal conditions promoting mate-seeking. Furthermore, fluorescent labeling, flow sorting, and transcriptional profiling of SSTtn neurons revealed many sex differences in gene expression, including the gene for estrogen receptor alpha, *Esr1*. Together, these data point to a mechanism whereby SSTtn neurons sense rising ovarian hormone levels and counteract their anorexigenic effects when energy reserves are low. Thus, sex differential recruitment and/or activation of SSTtn neurons may work to mitigate the effects of sex steroids on behavioral feeding output. This research illustrates how gonadal steroid modulation of neuronal circuits can be context-dependent and gated by other physiological signals.

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Poster

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Program #/Poster #: P644.05

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: AFOSR MURI Grant FA9550-18-1-0051
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Title: Characterizing Cristae Lamellarity in Mouse Cerebellum Mitochondria

Authors: *R. N. MENDELSOHN[1], G. C. GARCIA[1], T. M. BARTOL[1], C. T. LEE[2], P. KHANDELWAL[1], E. LIU[1], D. J. SPENCER[1], A. HUSAR[1], E. A. BUSHONG[3], S. PHAN[3], G. PERKINS[3], M. H. ELLISMAN[3], A. SKUPIN[3,5], T. J. SEJNOWSKI[1,4], P. RANGAMANI[2];

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Abstract: The structure of a mitochodrion is highly compartmentalized and changes in response to the energy demands of its cell. The compartments within the inner membrane (known as cristae) undergo structural changes at the nanometer scale that have been connected to significant differences in ATP production rate. It is therefore beneficial to quantify the characteristics of the cristae on real mitochondria morphologies, especially in the dynamic energetic landscape of a neuron. To this end, the dual membrane structure of nine axonal mitochondria in the mouse cerebellum were digitally rendered in three dimensions, retaining dimensional accuracy from TEM tomography taken to an isotropic voxel size of 1.6 nanometers. Geometric analysis of the the cristae membrane, including its curvature, is employed to quantitatively describe the complex structure of the cristae, providing valuable insights into attributes like lamellarity. Derived from “thin plate” in Latin, the word lamellarity has been used interchangeably with cristae shape factor (the surface area to volume ratio of the cristae compartments). The degree of lamellarity, as determined by shape factor, was found to directly correlate with the percentage of cristae surface area having first principle curvature above a given threshold. This connection between lamellarity and high curvature (associated with supercomplex assembly and ATP synthase function) supports the theory that lamellarity increases energy production. Computational models have shown that high cristae shape factor also induces a high proton motive force. It has been speculated that the tradeoff between the efficiency offered by high cristae shape factor and the high overall capacity for ATP production associated with large surface area explains the different use cases of the mitochondria’s two main energetic states (orthodox and condensed). In these nine crisae geometries, however, the lack of an inverse relationship between surface area and surface area to volume ratio suggests that the variation of cristae architecture prevents a decrease in shape factor as the area increases. This optimization of the cristae structure for ATP production is investigated using geometric analysis, identifying common building blocks across these cristae structures and illuminating the role of lamellarity, surface area, and membrane curvature.

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Poster

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Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: Israel Science Foundation grant no. 1781/16
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Title: Exercise reverses histone regulation of high-fat diet related gene expression in the hypothalamus

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Abstract: Objective: Obesity affects many physiological systems. Recent research has shown that exercise, even in the absence of diet change, can enhance physiological health and reduce symptoms of metabolic syndrome (MeS) that are associated with overweight. However, the mechanisms by which these improvements occur are not clearly understood. Here, we aim to uncover the beneficial effect of exercise on epigenetic mechanisms and genes expressed in the hypothalamus of obese and lean rats. **Methods**: Adult female Wistar chow- or high fat-fed rats were provided with voluntary running wheels or remained sedentary for 4-weeks in computerized exercise chambers, while maintaining their diet. At the end of treatment, body composition was assessed, and the hypothalamic Arcuate nucleus (ARC) was extracted for molecular analysis. RNA-sequencing was utilized to uncover whole-genome transcriptional alterations between groups. Since changes in gene expression are involved, changes in candidate histone modifiers, revealed by the RNA-seq, were validated by RT-qPCR and their role in gene regulation was tested via chromatin immunoprecipitation (ChIP) assays on the promoter of selected differentially expressed genes (DEGs). **Results**: Active obese animals gained less weight and had lower body fat composition compared to non-active animals. Two unique functional families of genes, one associated with inflammation, and the other governing insulin sensitivity were impaired by obesity. These families of genes were rescued by moderate physical activity, even though rats maintained a high-fat diet. To understand the molecular mechanisms that govern transcriptional recovery, we examined major transcription factors (TFs) and epigenetic modulators and pinpointed two distinct modifications that play a key role in their transcriptional rebound. The decrease in expression of inflammatory related genes (such as *Zap70* and *Cd74*) is moderated by a decrease of H3K4me3 markers. Concurrently, the increase in expression of genes related to insulin sensitivity (such as *Synpr* and *Igf2bp2*) is modulated by reduction of the strong repressor, H3K27me3. **Conclusion**: Physical activity can partially reverse high-fat-diet related elevation in hypothalamic expression of inflammatory and insulin-sensitivity related genes. This process is mediated by specific epigenetic histone modifications, which might improve health status.

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Poster

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Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Title: High-fat, high-sugar diet down regulates activity of oxytocin neurons in female rats

Authors: S. BLYTHE[1], J. KIM[2], K. ATTAL[2], *N. TOPORIKOVA[2];
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Abstract: Estradiol potently suppresses food intake. Indeed, female rats eat less during the pre-ovulatory phase of the estrous cycle, and ovariectomy (OVX) produces hyperphagia and weight gain. The neurohormone oxytocin (OT) has also been shown to inhibit food intake and consumption of palatable sucrose solutions. Interestingly, chronic sugar consumption reduces activity of OT neurons in the hypothalamic paraventricular nucleus (PVN). Recently, sex differences and estrous cycle influences on oxytocin-stimulated food intake have been reported. However, the relationship between estrogen level and OT activity have not been explored in obese animals. The objective of this study is to compare OT neuron number and activation in cycling, OVX, and OVX + estradiol rats fed either a control diet or high-fat, high-sugar (HFHS) diet.
Our study utilized female Sprague-Dawley rats fed either a HFHS diet or control chow for 15 weeks from weaning. The HFHS animals received food containing 60% fat and a 32% liquid sucrose solution. To assess the central effect of diet on OT neuronal activity, we utilized following three groups (i) cycling females on Diestrus (ii) ovariectomized (OVX) females and (iii) ovariectomized females with slow-release estrogen pellet (OVXE). Two weeks following OVX surgeries, animals were sacrificed. Upon termination, HFHS animals weighed significantly more than their control counterparts and exhibited greater fat pad mass. Similarly, OVX animals weighed significantly more than OVXE animals. We used immunocytochemistry to double stain brain sections with OT and cFOS antibodies. Analysis revealed that OVX reduced number of OT neurons in PVN. In addition, we found that the fraction of cFOS-positive OT cells in PVN was smaller in HFHS group. These observations indicate HFHS diet reduces activity of OT neurons in estradiol-dependent manner. Given the fact that OT has been suggested as a treatment for obesity, these findings are clinically relevant.

Disclosures: S. Blythe: None. **J. Kim:** None. **K. Attal:** None. **N. Toporikova:** None.

Poster

P644. Energy Metabolism II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P644.08

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: WRII

Title: Lateral Hypothalamic Leptin Receptor Neurons Encode Motivation for Food Approach

Authors: K. KIM, Y. LEE, Y. KIM, H. SONG, M. JANG, D. HA, J. PARK, S. JUNG, J. LEE, S. KIM, H. CHOI;
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Abstract: <META NAME="author" CONTENT="김규식">Motivation elicited from physiological need drives adaptive behavior for homeostasis. The lateral hypothalamus (LH) in the brain has been known as a motivation center. Past studies have shown that stimulation of the LH region have effect on reinforcing motivation for feeding. Recently, in vivo calcium recordings of individual GABA neurons using miniature microscope in mice have shown diversity in neuronal population during feeding behavior. However, which neural subpopulation compute motivation during multi phasic feeding behavior remains a mystery. Here, we examined neural populations in the LH that encode motivation for feeding using photometry, optogenetics, chemogenetics, and behavioral quantification. Photometry results show that LH LepR neurons start to activate on food accessibility, increase during food consumption. We also show that LH LepR neurons start to activate before voluntary approach to food. Furthermore, LepR neuronal activity decreased when food inaccessibility was perceived. Optogenetic results show that LH LepR neurons induce food approach. Optogenetic activation of LH LepR neurons did not induce food approach when food could not be targeted. However, when food could be targeted, LH LepR neuronal activation induced food approach. Furthermore, cue approach behavior increased when mice entered food zone. Optogenetic deactivation of LH LepR neurons decreased food consumption in hungry mouse. Chemogenetic activation results also showed similar results, increasing approach behavior to food but not increasing consumption behavior. Overall our results show that LH LepR neurons encode motivation for food approach when food can be targeted by mouse. This shows that LH LepR neurons take role in the approach to food but not consumption during the multi phasic feeding behavior of animals.

Disclosures: K. Kim: None. **Y. Lee:** None. **Y. Kim:** None. **H. Song:** None. **M. Jang:** None. **D. Ha:** None. **J. Park:** None. **S. Jung:** None. **J. Lee:** None. **S. Kim:** None. **H. Choi:** None.

Poster

P644. Energy Metabolism II

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P644.09

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: CIHR MOP119298

Title: Effect of ketogenic diet and beta-hydroxybutyrate on socio-cognitive deficits and seizures in mouse models of GRIN disorder

Authors: *Y. YAN[1], T. LIPINA[2], D. J. STEVENS[2], L. PEPERA[2], A. J. RAMSEY[1];
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Abstract: NMDA receptors are encoded by *GRIN* genes *GRIN1*, *GRIN2A-D*, and *GRIN3A-B*. Rare *de novo* mutations in the *GRIN* genes cause GRIN disorder, a neurodevelopmental disorder with symptoms of profound intellectual disability, epilepsy, and visual cortical impairment. GRIN disorder can be modeled with *Grin1*^{KD} mice, which have a 90% reduction in GluN1 expression leading to NMDAR hypofunction. Cognitive deficits, reduced social motivation, impaired sensorimotor gating and seizures has been reported in *Grin1*^{KD} mouse, making it a promising model for GRIN disorder. Bioenergetic deficits in *Grin1*^{KD} mouse have been reported, suggesting deficient glucose uptake to meet high energy demand. We asked whether ketone bodies could serve as an alternative energy source in *Grin1*^{KD} mice, particularly since a ketogenic diet is an effective therapy for childhood-onset epilepsy. An emerging hypothesis suggests that elevation in beta-hydroxybutyrate (BHB) is key to the therapeutic effect of a ketogenic diet. BHB can be consumed as a supplement in water, making it a preferred substitute. We hypothesized that both a ketogenic diet and BHB can improve the behavioural consequences in *Grin1*^{KD} mice. Long-term exposure to BHB-supplemented water (NPN 80085843; Natural Health Product, USA; 6mg/ml; from 3 to 14 weeks of age) improved the hyperlocomotion and sensorimotor gating activity in *Grin1*^{KD} mice. However, deficits of social behaviour and spatial working memory (assessed by Y-maze) in *Grin1*^{KD} mice were not improved by BHB. We also tested the effect of a ketogenic diet (TD.07797, Envigo Canada; from 5 to 14 weeks of age) on behavioural impairments of *Grin1*^{KD} mice. Preliminary data suggested minimal effect of a ketogenic diet on social behaviour and spatial working memory function in *Grin1*^{KD} mice. To better understand GRIN disorder and develop treatment for *GRIN1* patients, our lab has generated mouse lines possessing same variants in *GRIN1* patients, including the p.Tyr647Ser (Y647S) variant (*Grin1*$^{+/Y647S}$ mice). We observed an increased number of spontaneous seizures after handling induced convulsion (HIC) test in *Grin1*$^{+/Y647S}$ mice. BHB treatment reduced the severity of seizures induced by HIC test and delayed the onset of spontaneous seizures after HIC. Our results suggested modest effect of BHB on cognitive function and a protective effect of BHB on seizures in two mouse models of GRIN disorder. Preliminary data suggested that a ketogenic diet is not superior to BHB. In fact, it showed unwanted side effects including inadequate weight gain. Our study supports BHB as a preferred substitute over a ketogenic diet because BHB produces comparable beneficial effects with fewer adverse effects.

Disclosures: Y. Yan: None. **T. Lipina:** None. **D.J. Stevens:** None. **L. Pepera:** None. **A.J. Ramsey:** None.

Poster

P644. Energy Metabolism II

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Program #/Poster #: P644.10

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: AdamFest Research Fund
Pilot Grant, University of Iowa Psychiatry Department

Title: Bipolar disorder and redox imbalance: probing the link between metabolism and psychiatric illness

Authors: *K. KRUTH[1], J. MEHR[1], A. J. WILLIAMS[2];
[2]Psychiatry, [1]Univ. of Iowa, Iowa City, IA

Abstract: Bipolar disorder (BD) remains one of the most difficult psychiatric illnesses to treat, and a lack of understanding behind the molecular mechanism of the disorder has hindered efforts to produce effective pharmacotherapies. Several studies have suggested that BD manifests with mitochondrial and metabolic dysfunction such as mitochondrial hyperactivity and elevated markers of oxidative stress. Additionally, SNPs in multiple mitochondrial genes have been identified as risk SNPs for BD, further indicating that mitochondrial dysfunction may play a role in BD etiology. Here, we investigate the role of reduction-oxidation (redox) imbalance in human fibroblasts, induced pluripotent stem cells, and induced neurons derived from bipolar and healthy control subjects to further elucidate the source of these mitochondrial phenotypes. We have measured key redox ratios including NAD^+/NADH, $NADP^+$/NADPH, and GSSG/GSH to determine whether these cell lines display oxidative or reductive stress. Further, we have assessed reactive oxygen species (ROS) production in these lines both under baseline and mitochondrial stress conditions to probe for differences between BD and control cell lines in their ability to cope with reduced mitochondrial efficiency. Our results suggest that manipulation of metabolism through supplementation with metabolic intermediates may prove therapeutic in BD by restoring redox balance, thereby ameliorating the accumulation of damaging ROS that contributes to neuronal dysfunction.

Disclosures: K. Kruth: None. **J. Mehr:** None. **A.J. Williams:** None.

Poster

P658. Threat/Fear: Neural Circuits III

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P658.01

Topic: G.01. Fear and Aversive Learning and Memory

Support: R01 DK109934
DAMD W81XWH-19-1-0428

Title: Genetic dissection of a basal forebrain-to-lateral habenula circuit that drives reflexive aversion and modulates feeding behavior

Authors: *J. SWANSON[1], J. ORTIZ-GUZMAN[1], J. M. PATEL[2], E. HANSON[1], P. HUNT[1], S. DOOLING[1], B. R. ARENKIEL[1];
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Abstract: Feeding circuitry in the brain is regulated by much more than energy state. Environmental cues, mood, and reward/aversion all drive non-homeostatic feeding. While the hypothalamus has been extensively studied regarding homeostatic feeding, it remains to be determined the neural circuits driving more complex, non-homeostatic feeding mechanisms. Recently, we identified excitatory, glutamatergic neurons of the basal forebrain (BF) potently drive appetite suppression. Notably, acute activation of these neurons induces aversive behaviors, as well as aversion to food-associated cues. While this behavior is in part due to projections to the lateral hypothalamic area (LHA), acute activation of these projections doesn't fully re-capitulate the BF activation phenotype, indicating additional targets must be responsible for this appetite suppression and aversion. One prominent projection of the glutamatergic BF that we've uncovered is the lateral habenula (LHb). The LHb is a particularly interesting candidate for feeding regulation, as it's a critical node in aversion, reward, and addiction, inhibiting reward centers of the brain. Thus, *we hypothesize that glutamatergic BF projections to the LHb decrease food intake due to aversion.* Using viral tracing and channel-rhodopsin assisted circuit mapping, we have determined that the BF and LHb are anatomically and functionally connected. Optogenetic activation of BF-to-LHb circuitry drives a remarkable aversive phenotype, and this aversion overrides the drive to eat in a fasted state. Interestingly, this aversion cannot become associated with contextual markers, nor does it promote release of stress or fight-or-flight hormones. Thus, we have concluded this is reflex-like aversion, bypassing learning and memory or any stress response, to modulate feeding behavior. These data reveal new insights into the complex neural circuitry governing aversive and appetitive behaviors, which may be at play in conditions such as eating disorders like anorexia nervosa and bulimia, as well as behavioral causes of obesity.

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Poster

P658. Threat/Fear: Neural Circuits III

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Program #/Poster #: P658.02

Topic: G.01. Fear and Aversive Learning and Memory

Support: Japan Society for the Promotion of Science Overseas Research Fellowships
NIH Grant R01NS108740

Title: Distinct roles of dopamine receptor type-1 and type-2 medium spiny neurons in the tail of the striatum in avoidance of potential threats in a semi-naturalistic foraging task

Authors: *I. TSUTSUI-KIMURA, N. UCHIDA, M. WATABE-UCHIDA;
Harvard Univ., Cambridge, MA

Abstract: In natural environments, it is critical to properly assess potential threats for appropriate behavioral choices. Although it is important to predict threats without experiencing an outcome such as pain, previous studies have often used painful stimuli to model avoidance. Here we used a semi-naturalistic foraging paradigm to model avoidance of potential threats. In this paradigm, a thirsty mouse freely goes out of a homing area and enters a foraging arena to obtain a water reward. In test sessions, a predator-like big object ("monster") surges toward it while a mouse approaches to a reward. On initial encounters with a big monster, most mice advanced close to the reward location but failed to obtain it, and fled into the homing area. In subsequent trials, mice returned home earlier before obtaining reward even before the monster charged, indicating that mice learned to predict a potential threat even before the monster charges. In contrast, mice did not avoid a big monster which remained stationary, or a smaller monster that charged, indicating that the movement and the size are important factors to make an object threatening. Over days of sessions with a moving big monster, mice gradually succeeded more to acquire reward, suggesting that they learned to overcome the threat (habituation). We have previously shown that dopamine neurons projecting to the tail of the striatum (TS) play an essential role in avoiding a threatening stimulus (e.g. air puff and a novel object) (Menegas et al., 2018). In this study, we found that increasing the extracellular dopamine level in TS with dopamine transporter inhibitor (GBR12909, bilateral) induced avoidance of a small monster, while ablation of TS-projecting dopamine neurons with 6-hydroxidopamine (bilateral) decreased avoidance of a big monster. We next examined whether dopamine receptor type-1 or type-2 expressing medium spiny neurons (D1- or D2-MSNs) in TS are involved in avoidance of a monster. D1- and D2-MSNs were ablated by cre-dependent expression of diphtheria toxin in TS of Tac1-cre and Adora2a-cre mice, respectively. We found that mice with D1-MSNs ablation did not avoid the big monster. In contrast, mice with D2-MSNs ablation avoided the big monster and failed to overcome a big monster in subsequent sessions over days. These results suggest a crucial role of dopamine and D1-MSNs in TS in avoidance and prediction of a potential threat, and a contrasting role of D2-MSNs for threat management (habituation). Together, these results

provide not only the mechanism for threat assessment, but also a basic principle of how D1- and D2-MSNs regulate behaviors under the control of dopamine over the course of learning and habituation.

Disclosures: I. Tsutsui-Kimura: None. **N. Uchida:** None. **M. Watabe-Uchida:** None.

Poster

P658. Threat/Fear: Neural Circuits III

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Program #/Poster #: P658.03

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH Grant R01MH065961
NIH Grant R01MH117852

Title: Basolateral amygdala mediates retrieval of both recent and remote fear memories

Authors: *J. LIU, M. S. TOTTY, L. MALISSARI, S. MAREN;
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Abstract: Considerable evidence indicates that the basolateral amygdala (BLA) has a critical role in the acquisition and expression of Pavlovian fear conditioning in both rats and humans. For example, BLA lesions made up to one year after fear conditioning in rats prevent the expression of conditioned freezing. However, selective optogenetic silencing of BLA principal neurons disrupts freezing only hours, but not days after conditioning. These data suggest a time-limited role for the BLA in fear memory retrieval. To clarify this issue, we reexamined the role of the BLA in fear memory retrieval using optogenetic, electrophysiological, and calcium imaging methods in rats. Importantly, we used a behavioral design that permitted a comparison of recent and remote retrieval in the same subjects. First, we found that the retrieval of recent (1 day) or remote (2 weeks) fear memories yielded similar levels of c-Fos expression in the BLA. Extracellular recordings of BLA neuronal activity in behaving rats revealed similar numbers of CS-responsive neurons during the retrieval of recent or remote fear memories. Moreover, individual BLA neurons exhibited similar increases in CS-evoked spike firing during both the recent and remote retrieval tests. This outcome was also observed in fiber photometric recordings of BLA principal neuron activity (AAV-CaMKII-GCaMP6f), suggesting an enduring role for BLA principal cells in the expression of long-term fear memory. Consistent with these results, continuous optogenetic inhibition (AAV-CaMKII-Jaws) of BLA principal neurons produced robust impairments in conditioned freezing to recent and remote CSs. Similar outcomes were observed when BLA activity was inhibited only during the CS, though these effects were weaker than with continuous inhibition. Lastly, optogenetic activation of BLA interneurons (AAV-mDlx-ChR2) suppressed conditioned freezing to both the recent and remote CS. Collectively,

these results indicate that BLA neurons are indispensable for both recent and remote fear memory retrieval. We conclude the BLA is a long-term storage site for emotional memories acquired during fear conditioning.

Disclosures: J. Liu: None. **M.S. Totty:** None. **L. Malissari:** None. **S. Maren:** None.

Poster

P658. Threat/Fear: Neural Circuits III

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Program #/Poster #: P658.04

Topic: G.01. Fear and Aversive Learning and Memory

Support: NSF 2014862
NIH MH119517

Title: Redefining the Sensory Inputs to the Lateral Amygdala

Authors: *J. OREA[1], L. OSTROFF[2], E. PARRISH[2];
[1]Univ. of Connecticut, Storrs, CT; [2]Physiol. and Neurobio., Univ. of Connecticut, Storrs Mansfield, CT

Abstract: The lateral amygdala (LA) has a central role in processing the emotional content of sensory stimuli, and synaptic plasticity in the LA is a well-established mechanism of memory storage for auditory Pavlovian conditioning. LA neurons receive sensory input from both the cortex and thalamus, but how these inputs are organized is not fully understood. According to the canonical literature, the predominant auditory inputs to the LA are thalamic projections from the medial division of the medial geniculate (MGm) and adjacent posterior interlaminar nucleus (PIN), and cortical projections from association area TE3. A projection from the multimodal agranular posterior insular cortex (AIP) has also been reported. The LA may offer unique insights into how neurons integrate converging sensory information, but the classic neuroanatomy literature lacks fine-scale data on the input regions and targets. To better delineate the input regions and map their convergence in the LA, and to determine whether the circuits defined in males are identical in females, we began with small injections of the retrograde tracer Fluorogold in the dorsolateral LA of Sprague-Dawley rats of both sexes. Retrogradely labeled neurons were found in TE3, PIN and the dorsal division of MG (MGd), as previously reported, but there was no labeling in the insular cortex or in MGm. To visualize the boundaries between MGm, PIN, and ventral MG (MGv), we used parvalbumin immunostaining to reveal the MGm-specific inputs from the cochlear nucleus. It is likely that earlier studies reporting MGm projections to LA used less stringent boundaries between PIN and MGm, accounting for the discrepancy. Previous studies reporting AIP projections used bidirectional, transsynaptic tracers and thus may have included anterograde or polysynaptic connections. MGd labeling has been

observed in previous retrograde tracing studies of LA but the inputs have not been mapped. To explore this projection, we performed small injections of an adenoviral tracer expressing a GFP reporter in MGd, which revealed very heavy projections that were restricted to the caudal extent of the dorsolateral LA and validated retrograde observations. Further anterograde injections confirmed that the MGd and TE3 projections have identical projection patterns to dorsolateral LA, which are denser and more restricted than those of PIN. Overall, our results confirm the traditional model of converging inputs to LA from auditory thalamus and cortex, but revise the predominant source of thalamic inputs from MGm/PIN to MGd and PIN.

Disclosures: J. Orea: None. **L. Ostroff:** None. **E. Parrish:** None.

Poster

P658. Threat/Fear: Neural Circuits III

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Topic: G.01. Fear and Aversive Learning and Memory

Support: NRF grant funded by the Korea government (No.2020R1A2C2014830

Title: Sex differences in the expression of defensive behavior of rats under chasing threat paradigm

Authors: *G. PYEON, J. LEE, J.-S. CHOI;
Korea Univ., Seoul, Korea, Republic of

Abstract: Women are more vulnerable to anxiety- and fear-related disorders. However, most of the relevant basic studies have been conducted in male animals. Here we investigated sex differences in anxiety level in the elevated plus-maze and in the defensive response under chasing threat and in Pavlovian threat conditioning. The current study developed a novel fear paradigm called “chasing threat” in which rats are chased by a fast-moving robot in an inescapable maze, which mimics a predatory threat in nature. In Experiment 1, anxiety levels in naturally cycling female and male rats in the elevated plus-maze were measured. The proestrus female rats entered and spent more time in the open arms compared to the male and diestrus female rats. In Experiment 2, female and male rats were exposed to chasing threat and their threat memory was assessed in a retention test 3 days later. The female rats exhibited an active defensive response known as darting, whereas male rats dominantly displayed freezing response, and the darting response in females was maintained until the retention test. In Experiment 3, a new group of rats was used to investigate sex differences in defensive response during Pavlovian threat conditioning. Similar to the chasing threat, female rats displayed a darting response, whereas male rats showed mostly freezing. However, the darting responses in female rats were not maintained until the retention test. Taken together, differential responses to chasing threat

and to the Pavlovian conditioned stimulus demonstrated sex differences in defensive response in which the male rats exhibited passive freezing responses dominantly whereas the female rats displayed active darting responses. However, the chasing threat with high ecological validity showed the more long-lasting effect of sex differences in active defensive responses compared to the Pavlovian situation.

Disclosures: G. Pyeon: None. **J. Lee:** None. **J. Choi:** None.

Poster

P658. Threat/Fear: Neural Circuits III

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Topic: G.01. Fear and Aversive Learning and Memory

Support: EU 896825
FONDECYT 3170497
EMERGENCES Paris

Title: Prefrontal cortex neuronal ensembles process anxiety and fear behavior

Authors: *J. P. CASANOVA[1], C. POUGET[2], I. AGARWAL[2], G. VETERE[2];
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Abstract: Anxiety and fear are distinct but overlapping emotions that express different levels ofrisk assessment and applied behaviour. In this work we studied the dorsomedial prefrontal cortex (dmPFC) neuronal activity underlying the behavioral expression offear and anxiety-like behavior. The neural calcium dynamics from hundreds of dmPFC cells was visualized using a miniaturized fluorescence microscope. Mice were subjected to 3 behavioral tests, in an increasing anxiety- and fear-inducing potential way. Single dmPFC cell activity is preferentially modulated by either anxiety or fear-like behaviour and by electric footshocks. In behavioural tasks where risk assessment is requested, we observed that the dmPFC population cell activity switch between states. Moreover, population cell activity recorder during anxiety-like behaviour can predict freezing behaviour, but not the opposite. However, in tasks where a threatening stimulus elicit strong fear, dmPFC population cell activity stabilizes to a single state over the entire period. Our results further support that dmPFC activity is engaged during anxiety-like behavior and fear at both, single neuron and population level, and that dmPFC has a key role in assessing the risk in uncertain situations.

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Poster

P658. Threat/Fear: Neural Circuits III

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Program #/Poster #: P658.07

Topic: G.01. Fear and Aversive Learning and Memory

Support: PAPIIT -DGAPA 201420
CONACYT 1085633
Vera-Rivera, G.
Rangel-Hernández, J. A.

Title: Effects of lateral habenula lesions during conditioned sugar aversion

Authors: *J. LOMELI-CASTILLO[1], M. I. MIRANDA[2];
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Abstract: The habenula is a phylogenetically conserved structure present in all vertebrate animals and it is located in the epithalamus, together with the pineal gland. This structure is anatomically divided into medial habenula and lateral habenula (LHb). LHb presents broader connections towards the limbic system and basal ganglia, as well as several structures that transmit information of the internal state, like the insular cortex. Accordingly, LHb connects with some areas of the brain that are crucial during rewarded learning. In this regard, evidence indicates that LHb plays a critical role in aversion-mediated learning since it responds to signals that predict adverse stimuli with aversive outcomes. On experimental animal models, LHb neurons' pharmacological inactivation produces behaviors of "indifference" to the consequences of outstanding events. For example, rats that received deep brain stimulation in the LHb significantly reduced the levels of sucrose self-administration; in contrast, LHb lesion increased sucrose-seeking behavior, delaying extinction response to the substitution of sucrose for water. Thus recently, this structure has related to taste recognition memory as well as during signaling relevance for appetitive stimuli such as sweet tastes. Thus, indirect evidence suggests the involvement of LHb during conditioned taste aversion (CTA), where animals learn to avoid a novel taste associated with visceral discomfort. Hence, it is important to evaluate the participation of the habenula during CTA of a highly appetitive stimulus such as sugar. Therefore, our objective was to assess the function of the LHb through LHb lesions induced by a high concentration of n-methyl-d- aspartate (NMDA) injected before CTA. Adult male Wistar rats with an initial weight of 250-310 g were liquid deprived and trained to consume liquid in a single daily presentation (20min/day). Thirty minutes before CTA acquisition were bilaterally LHb-injected with NMDA (10 μg/μl). Throughout the acquisition, memory retrieval, and three aversive memory extinction sessions, sugar consumptions (10% solution) were measured. The results show that NMDA injection does not affect sensory, motivational, or motor processes during acquisition, and at the dose used, aversive sugar formation was not affected. Furthermore,

no changes in aversive memory retrieval nor the extinction process were observed. In conclusion, the lesion of NMDA-responsive cells in LHb does not alter conditioned sugar aversion, suggesting that during new aversive learning, LHb has not significant involvement.

Disclosures: J. Lomeli-Castillo: None. **M.I. Miranda:** None.

Poster

P658. Threat/Fear: Neural Circuits III

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Program #/Poster #: P658.08

Topic: G.01. Fear and Aversive Learning and Memory

Support: Center on Compulsive Behaviors

Title: Gabaergic and glutamatergic neurons of the medial septal area respond differently to punishment and cues predicting punishment

Authors: *C. CALVA, J. COUDRIET, S. IKEMOTO;
Natl. Inst. on Drug Abuse, Baltimore, MD

Abstract: Uncertainty in the environment prompts decision-making through the evaluation of potential risks/rewards. Decision-making is fundamental to an animal's ability to survive in constantly changing environments. Therefore, it is critical to understand the neural mechanisms that underlie uncertainty-driven behaviors. Medial septum (MS) neurons have been shown to play a role in motivated behaviors and respond to salient stimuli and cues related to uncertain rewards and punishments. Prior unit activity studies in primates demonstrated that MS neurons respond to certain and uncertain punishments and cues predicting these punishments. Whether similar neural activities can be found in mice and the specific phenotypic neurons responsible remains unknown. Here we examined whether MS glutamatergic (vGlut2) neurons and GABAergic (vGAT) neurons respond to punishment and cues predicting uncertain and certain punishment. AAV9-syn-FLEX-jGCaMP7f-WPRE was injected and a probe implanted into the MS of vGlut2-cre and vGAT-cre mice. We then performed a Pavlovian conditioning paradigm in which three different tones were paired with a 100%, 50%, or 0% chance of a 0.45mA foot shock, respectively. Behavioral procedures were conducted in tandem with fiber photometry to determine if vGlut2 and vGAT neurons in the MS respond to punishment and cues predicting uncertain and certain punishment. Our preliminary evidence suggests that MS vGlut2 neurons and vGAT neurons display divergent responses. Whereas MS vGlut2 neurons did not respond to the 0% tone, vGlut2 neurons located in the dorsal portion of the MS (n=3) displayed ramping activities upon tones signaling 50% or 100% shock. Moreover, these vGlut2 neurons displayed a response to the foot shock. Interestingly, vGlut2 neurons in the ventral portions of the MS (n=4) responded little to any of the conditioned tones or to the shock. These data suggest that dorsal

MS vGlut2 neurons respond to both the anticipation of punishment and the punishment itself. It is unclear whether MS vGlut2 neurons respond differently between certain and uncertain conditions. By contrast, MS vGAT neurons (n=5) did not clearly respond to the cues predicting shocks but markedly responded to shocks. In general, the presentation of tones had no clear effect on MS vGAT neuron activity. The results suggest that MS vGAT and MS vGlut2 neurons distinctly respond to conditioned stimuli and punishments. MS vGlut2 neurons may respond to both the anticipation of punishment and punishment, whereas MS vGAT neurons may respond to punishment. NIDA-IRP and the Center on Compulsive Behaviors supported this work.

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Poster

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Rackham Merit Fellowship

Title: Activation of Ventral Subicular Neurons Decreases Open Field Exploration in Mice

Authors: *J. GEWARGES[1,4], J. STOKES[5], C. JOHNSTON[2], J. SPENCER-SEGAL[3];
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Abstract: Hypervigilance, or decreased risk-taking behavior, is a core feature of anxiety disorders in humans. Previous work identified the ventral hippocampus as a neural structure, in both humans and rodents, that mediates risk-taking behavior, although the directionality of this control is not consistent across studies. However, little is known about the projections emanating from the ventral hippocampus and how their activation drives risk-taking behaviors in response to a perceived threat. One candidate structure downstream of the ventral hippocampus is the ventral subiculum (vSUB), which receives its major input from the ventral hippocampus. Previously, vSUB-lesioned rats showed lower risk-taking behavior, spending less time in the center of a novel open field, and vSUB pyramidal neurons were activated by acute psychological stress. Given this previous work, we hypothesized that vSUB pyramidal cell activity drives risk-taking behavior. Specifically, we expected that chemogenetic activation of vSUB neurons in the vSUB pyramidal cell layer would increase exploration of the center in a large, brightly lit open

field. In 12 male and 12 female 12-15 week-old C57Bl/6 mice, we injected AAV5-hSyn-hM3D(Gq)-mCherry into the vSUB pyramidal cell layer. After allowing adequate time for viral expression (3 weeks), mice were injected with clozapine-N-oxide (to activate the hM3D(Gq)) (N = 12) or Vehicle (N = 12) 20 minutes prior to being placed in a large (72 cm square), brightly-lit open field for 10 minutes. CNO and Vehicle-injected mice showed similar total exploration of the open field (mean distance traveled, CNO: 3194 cm +/- 2139 cm, Veh: 3678 cm +/- 766 cm, $p = 0.4682$), but CNO mice showed less exploration of the center (mean distance traveled in the center, CNO: 162 cm +/- 127 cm, Veh: 352 cm +/- 145 cm, $p = 0.0026$). CNO-injected mice also made fewer entries into the center (CNO: 23 +/- 17, Veh: 42 +/- 12, $p = 0.0054$), but there was no difference in time spent in the center (CNO: 80 s +/- 93 s, Veh: 78 s +/- 33 s, $p = 0.941$). There were no sex differences. These results show that chemogenetic activation of vSUB pyramidal layer neurons decreases exploration of the center of a brightly-lit open field. They suggest that vSUB neuronal activity decreases risk-taking behavior, which is contrary to our hypothesis informed by previous lesion studies. These findings may further our understanding of the neural circuitry behind individual differences in the behavioral response to a perceived threat. Future studies aim to further understand the neural circuitry downstream of the vSUB mediating its role in risk-taking behavior.

Disclosures: J. Gewarges: None. **J. Stokes:** None. **C. Johnston:** None. **J. Spencer-Segal:** None.

Poster

P658. Threat/Fear: Neural Circuits III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P658.10

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH 5R01MH106617
NARSAD/BBRF

Title: Local prefrontal circuits in fear modulation

Authors: *T. W. BAILEY, J. H. SPEIGEL, III, J. MAYER, J. PASTORE, E. KORZUS; Dept. of Psychology and Neurosci. Program, Univ. of California Riverside, Riverside, CA

Abstract: When contextual information is associated with an aversive stimulus, contextual fear memories are created. Post-traumatic stress disorder (PTSD) is characterized by the overgeneralized fear response evoked by trauma reminders that resemble cues from the original traumatic experience. We investigated the hypothesis that fear memory specificity is achieved through an mPFC-dependent mechanism involving the reduction of fear responses to the harmless, overgeneralized context in mice. Fear behavior has been shown to be differentially

modulated by the mPFC subdivisions of prelimbic (PL) and infralimbic (IL) cortex, which enhance fear responses and reduce fear responses, respectively. This differential function of mPFC subdivisions may be due to differential connectivity with the amygdala and its nuclei. cAMP response element binding protein (CREB)- binding protein (CBP) is a coactivator of transcription and histone acetyltransferase, and has been shown to be required for long-term memory consolidation. Using CBP mutants and viral gene-delivery neurogenetic tools, we have developed a system for targeting neuronal hypofunction to specific cortical populations by interfering with long-term memory consolidation. When CBPΔHAT is expressed in IL neurons, severe deficits in contextual fear discrimination learning, but not conditioning, are observed. Also, expression of CBPΔHAT in PL neurons results in profound deficits in fear discrimination learning. While observed deficits in selected prefrontal populations can be rescued by enhanced training protocols, the hypofunction targeted to excitatory neurons throughout the mPFC region resulted in a more severe deficit in differential fear conditioning. In addition, we have identified specific prefrontal projections to amygdala which are not engaged during fear acquisition. These findings provide strong evidence for our hypothesis that mPFC-dependent mechanisms drive modulation of fear responses during discrimination learning. The deficit in fear discrimination learning caused by impaired prefrontal/amygdala circuits is likely to be relevant to understanding and treating neuropsychiatric disorders such as PTSD and anxiety disorders.

Disclosures: T.W. Bailey: None. **J.H. Speigel:** None. **J. Mayer:** None. **J. Pastore:** None. **E. Korzus:** None.

Poster

P670. Neural Systems I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P670.01

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIH Grant EY019049
NIH Grant MH116990
NIH Grant DC008983

Title: A Bottom-up Reward Pathway Mediated by Somatostatin Neurons in Medial Septum Underlying Appetitive Learning

Authors: *L. SHEN[1], G. ZHANG[1], C. TAO[1], M. B. SEO[1], L. I. ZHANG[2], H. TAO[3]; [2]Physiol. & Neurosci., [1]USC, Los Angeles, CA; [3]USC Keck Sch. Med., Los Angeles, CA

Abstract: Valence detection and processing are essential for the survival of animals and their life quality in complex environments. Neural circuits underlying the transformation of external sensory signals into positive valence coding to generate appropriate behavioral responses remain

not well-studied. Here, we report that somatostatin (SOM) subtype of GABAergic neurons in mouse medial septum (MS), but not parvalbumin subtype or glutamatergic neurons, specifically encode reward signals and positive valence. Through an ascending pathway from the nucleus of solitary tract (NST) and then parabrachial nucleus (PBN), MS SOM neurons receive innately rewarding taste signals and suppress the lateral habenula (LHb). They contribute essentially to appetitive associative learning via their projections to LHb: learning enhances their responses to conditioned sensory cues, and suppressing their responses to either conditioned or unconditioned stimulus impairs acquisition of reward learning. Thus, MS serves as a critical hub for transforming bottom-up sensory signals to mediate appetitive behaviors.

Disclosures: L. Shen: None. **G. Zhang:** None. **C. Tao:** None. **M.B. Seo:** None. **L.I. Zhang:** None. **H. Tao:** None.

Poster

P670. Neural Systems I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P670.02

Topic: G.02. Reward and Appetitive Learning and Memory

Support: 3DP2MH119427-01S1

Title: Dopaminergic plasticity underlying bonding and loss in monogamous prairie voles

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Abstract: While healthy social relationships positively contribute to our health and wellbeing, their loss is detrimental for our mental and physical health. In monogamous prairie voles, dopamine (DA) binding in the nucleus accumbens (NAc) is necessary to induce a preference for a mating partner. After a bond as formed, DA binding is no longer necessary for demonstrating a partner preference but is necessary for displaying aggression towards non-partners, which is accompanied by an increase in DA receptors in the NAc. An increase in DA receptors alone is insufficient to explain the shift in the role of DA from affiliative to aggressive. One hypothesis is that in a long-term bond, interaction with a non-partner induces a larger bolus of DA compared to partner interaction, which may illicit aggression towards non-partners. It remains unknown how rapid temporal DA release dynamics change as bonds mature and can mediate the change in the behavioral function of DA. Additionally, there is no research available investigating how DA release dynamics adapt upon partner separation (loss). To test the hypothesis that bonding and loss results in changes in DA release dynamics, our work leverages $GRAB_{DA}$, a fluorescent DA

sensor with millisecond resolution for measuring extracellular DA levels in the NAc of behaving voles. We combined $GRAB_{DA}$ with fiber photometry to test whether bonding results in alterations in socially-evoked release during unrestrained social interaction with a partner or novel vole. We also examined changes in DA release during appetitive versus consummatory social behaviors using an operant task. Finally, we asked whether bonding and loss can change DA release capacity using optical stimulation. In all paradigms, we used a within-animal approach to assess changes in DA release prior to bonding, at short- and long-term bonding timepoints, and following partner separation. Thus far we found that bond formation decreased optically-evoked DA release compared to an unbonded, naïve state and bond loss decreased DA release relative to a long-term bond. Compellingly, we also found that voles with the greatest change in DA release 30 days after partner separation spent the most time with their partner upon reunion. This suggests that plasticity in DA release dynamics may contribute to individual differences in response to partner loss, which can inform treatments for pathological forms of social attachment and grief.

Disclosures: A.F. Pierce: None. **E.E. Culter:** None. **Z.R. Donaldson:** None.

Poster

P670. Neural Systems I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P670.03

Topic: G.02. Reward and Appetitive Learning and Memory

Support: JSPS KAKENHI Grant Numbers JP19K11423

Title: Rhythmic tactile stroking induces 50-kHz ultrasonic vocalizations in rats via the accumbal dopaminergic system

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Abstract: Tactile stimulations, such as rhythmic stroking, elicit 50-kHz ultrasonic vocalizations (USVs) in rats, which are thought to reflect positive affective states. We have previously shown that rhythmic stroking increases dopamine release in the nucleus accumbens (NAc). However, it remains unknown whether the mesolimbic accumbal dopaminergic system is involved in the emission of 50-kHz USVs during rhythmic stroking and whether this system is associated with certain call subtypes and call parameters (such as peak frequency and duration). The present study aimed to investigate the effects of dopamine (DA) antagonism in the NAc on 50-kHz USVs induced by stroking, on call rates, and on subtype profiles. Eight male Wistar/ST rats (8 weeks old) were used in this study. For rhythmic hand stroking procedures, each rat was gently held by the back skin in a vertical position and the whole palm was used to gently stroke the rat

repeatedly in one direction for 30 s. Each rat received one dose of either a vehicle or DA antagonist solution once daily. Stroking stimulation was applied for 30 s at 10 minutes before drug/vehicle injection (control condition) and at 5, 15, and 30 min after injection. USVs were recorded in an UltraSoundGate 116H audio device having a CM16/CMA microphone. For acoustic analyses, recordings were transferred to SASLab Pro. The 50-kHz calls were analyzed manually according to the 14-subtype scheme described by Wright et al. (2010). However, calls with overtones that could not be classified were considered as "predominantly flat calls with harmonics." The influence of drug and time (pre- and post-treatment) on USV parameters was assessed using two-way repeated-measures analysis of variance and post hoc Bonferroni comparisons, as appropriate. Bilateral injection of D1 receptor antagonist SCH 23390 (500 ng/side) plus D2/D3 receptor antagonist raclopride (25 μg/side) significantly reduced the number of "predominantly flat calls with harmonics" during stimulation and the number of frequency-modulated (FM) calls after stimulation. In contrast, no substantial changes were observed in the mean peak frequencies and call durations of total and categorized 50-kHz USVs. These results suggest that the emission of different subtypes of 50-kHz USVs may be differently regulated by dopaminergic transmission. The 50-kHz harmonics and FM USVs induced by rhythmic stroking may be useful behavioral markers for tactile reward in rats.

Disclosures: R. Shimoju: None.

Poster

P670. Neural Systems I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P670.04

Topic: G.02. Reward and Appetitive Learning and Memory

Support: Academy of Finland Grant 1278174
Academy of Finland Grant 1317399

Title: Somatostatin-expressing neurons from ventral tegmental area have long-range projections.

Authors: *E. NAGAEVA, L. ELSILÄ, A.-M. LINDEN, E. R. KORPI;
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Abstract: Somatostatin-containing (Sst) neurons make up a well-known interneuron subtype within the inhibitory gamma-aminobutyric acid (GABA)-releasing neurons in mammalian forebrain. Interestingly, these neurons can also project far from their original locations, as confirmed in cortex, hippocampus, amygdala and other brain regions (for review, Viollet et al., 2008). Major part of the GABAergic long-range projections in the brain express Sst neuropeptide and might be involved in synchronization of oscillatory activity of distant brain regions. Our recent research (Nagaeva et al., 2020; *eLife*) describes the Sst population in mouse VTA for the

first time. These neurons appear to be very heterogeneous by having three different electrophysiological subtypes and expressing both GABAergic inhibitory and/or glutamatergic excitatory markers, and even dopaminergic (DA) markers. We have also shown that physiologically, several Sst neurons can converge on single VTA dopamine neuron and inhibit it, acting as interneurons. Here we report that the mouse VTA Sst neurons also send their projections far outside the midbrain. We injected anterograde CRE-dependent viral tracer into VTA of Sst-CRE mice and found fluorescent Sst axons to end up in the lateral hypothalamus, particularly in its peduncular part and dorsomedial nucleus (n=4); in the central amygdala (CeA), particularly in its medial part (n=5); bilaterally in the paraventricular thalamic nucleus (PVT), particularly in its central and posterior parts (n=4); in the bed nucleus of stria terminalis (BNST), particularly in its lateral division (n=5). Next, we injected retrograde CRE-dependent viral tracer in each found target region (n=3) and confirmed that VTA Sst projections end up there and are not just passing by. Our results demonstrate that Sst neurons in VTA, as in several other brain regions, can be both interneurons and projection neurons. In addition, this study suggests the VTA Sst neurons are part of the global somatostatinergic system in the brain.

Disclosures: E. Nagaeva: None. **L. Elsilä:** None. **A. Linden:** None. **E.R. Korpi:** None.

Poster

P670. Neural Systems I

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Program #/Poster #: P670.05

Topic: G.02. Reward and Appetitive Learning and Memory

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NIH Grant DA042739
NIH Grant DA005010
NIH Grant GM008042
NSF NeuroNex Award 1707408

Title: Combining electrophysiology and fluorescent dopamine sensing to assess neuromodulatory effects on nucleus accumbens reward activity

Authors: *C. LONG[1], K. LEE[2], L. YANG[1], T. DAFALIAS[1], S. MASMANIDIS[1];
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Abstract: Despite significant progress, we have an imperfect understanding of the rapid modulatory effect of dopamine (DA) neurons on downstream brain activity. Here we focus on one of the main projection sites of midbrain DA neurons: the nucleus accumbens, and a prominent type of signal in this pathway: unexpected rewards. DA neurons, as well as DA

release in the nucleus accumbens, strongly signal unexpected reward events, which are thought to play a critical role in reinforcement learning. Neural firing rates in the nucleus accumbens are also similarly modulated by rewards. The temporal coincidence of reward-evoked DA signals with nucleus accumbens spiking has led to the view that rapid DA signaling is a key driver of reward activity in nucleus accumbens neurons. However, this hypothesis has not been rigorously tested. Thus, it is unknown the degree to which DA release is capable of directly altering neural activity on rapid (subsecond) timescales. Furthermore, it is unclear how varying amounts of DA differentially impact nucleus accumbens firing. To facilitate a systematic study of these questions, here we present an approach combining electrophysiology, photometry, and optogenetics. We use a 256 electrode silicon microprobe to record spiking activity from dozens of neurons, and an integrated optical fiber to concurrently monitor local DA signaling with the fluorescent sensor dLight1.2. In parallel, we perform optogenetic manipulations of VTA DA neurons in DAT-Cre male and female mice, to transiently raise or reduce DA levels in the nucleus accumbens. We present data comparing spiking activity during DA release triggered by either caloric rewards or optogenetic stimulation. We find that optogenetically evoked DA release is sufficient to alter the firing rate of a subset of neurons; however, this effect is only evident when the level of DA release exceeds the level corresponding to reward delivery. Taken together, this combined approach enables rigorous evaluation of the neuromodulatory effects of DA in the nucleus accumbens under both physiological and supraphysiological DA regimes.

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Poster

P670. Neural Systems I

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Program #/Poster #: P670.06

Topic: G.02. Reward and Appetitive Learning and Memory

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JSPS Grants in Aid for Scientific Research 20K15926

Title: Stimulation of the rat medial forebrain bundles enhances gamma oscillations in the primary motor cortex.

Authors: *A. YOSHIMOTO, Y. SHIBATA, M. KUDARA, N. MATSUMOTO, Y. IKEGAYA;
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Abstract: Stimulation of the medial forebrain bundle (MFB), which includes axonal fibers of dopaminergic neurons of the ventral tegmental area projecting to the nucleus accumbens, is used as a neural reward for operant conditioning of animals. Rewarding brain stimulation is a powerful motivator, and animals rapidly learn to perform a variety of operant tasks to obtain the reward. Although stimulation of the MFB modulates integration of reward and pleasure, it remains unknown how central and peripheral functions are affected by MFB stimulation. To address this question, we simultaneously recorded electrocorticograms (ECoGs) of the cerebral cortices (primary somatosensory cortices (S1) and primary motor cortices (M1)) and the olfactory bulbs in behaving rats while the MFB was stimulated. We first implanted multiple screw electrodes into the cerebral cortices and the olfactory bulbs as well as a stimulating electrode into the MFB or its nearby region. After recovery, we trained each rat to reproducibly perform a nose-poke test, in which poking its nose into a hole was associated with MFB stimulation. Using the nose-poke test, we optimized the current pulse frequency and amplitude for each rat to demonstrate consistent and sustainable self-stimulation. After the three-day nose-poke test, we observed a significant increase in spontaneous locomotion when stimulating the MFB compared to the neighboring region, suggesting the reward-seeking behavior. We also found that MFB stimulation at short intervals increased sniffing rates, monitored by olfactory bulb ECoGs. Moreover, the MFB stimulation at shorter intervals more increased the power of gamma components of M1 ECoGs. Our simultaneous recordings of central and peripheral activity from awake animals with MFB stimulation have demonstrated that the respiration and neural activity is affected by the changes in interstimulus intervals.

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Poster

P670. Neural Systems I

Location: Virtual Only

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Program #/Poster #: P670.07

Topic: G.02. Reward and Appetitive Learning and Memory

Support: DP2 MH119425-01
RF1 MH117055-01

Title: Synaptic controllability of dopamine as a mechanism for adaptive reward behaviors

Authors: *S. C. V. BURWELL[1], H. YAN[1], B. C. SHIELDS[2], M. R. TADROSS[2,1];
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Abstract: Dopamine neurons in the ventral tegmental area (VTA_{DA} neurons) associate sensory cues with outcomes in a manner consistent with Reward Prediction Error (RPE) theory. Namely,

unexpected reward causes a *phasic burst* in VTA_{DA} neurons, whereas unexpected omission of reward produces a *phasic pause* in these cells. Though RPE is an accepted description of VTA_{DA} dynamics, it remains unresolved whether bursts and pauses are necessary 'teaching signals' for adaptive learning behaviors. Optogenetics has established that exogenous bursts and pauses are sufficient to drive behavioral conditioning and extinction, respectively. However, endogenous bursts and pauses are more heterogeneous than those produced by optical methods, leaving unresolved whether naturally occurring VTA_{DA} dynamics have a causal role in adaptive behaviors. Here, we propose a "synaptic controllability" conceptual framework, reasoning that modifying the strength of synapses that control VTA_{DA} activity would enable causal interrogation of burst and pause dynamics along physiologically meaningful dimensions. Exploration of this question is enabled by an emerging technology, DART (drugs acutely restricted by tethering), which delivers clinical drugs to genetically defined cells. We combine this approach with a custom multiplexed behavioral task to quantify the rate of conditioning (associating a sensory cue with reward) and extinction (learning that a cue is no longer rewarded) simultaneously within a single mouse. Our key finding is that acute $GABA_A$ antagonism of VTA_{DA} neurons with gabazine-DART accelerates extinction with no impact on conditioning. This confirms the hypothesis that $GABA_A$ receptors are critical for negative RPE teaching signals. However, the direction of the effect is surprising, being opposite to the canonical prediction that an antagonist of VTA_{DA} inhibition should prevent phasic pauses and thus block extinction. We are examining this behavioral result with *in vivo* neural recordings, and performing a converse experiment using diazepam-DART, which is a positive allosteric modulator of $GABA_A$. This framework is a starting point for examining how the synaptic controllability of dopamine by the rest of the brain impacts reward prediction error coding in conditioning and extinction behaviors, with fundamental relevance to our understanding of reward circuitry in adaptive and maladaptive behaviors.

Disclosures: S.C.V. Burwell: None. **H. Yan:** None. **B.C. Shields:** None. **M.R. Tadross:** None.

Poster

P674. Motivation: Subcortical Neurocircuitry I

Location: Virtual Only

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Program #/Poster #: P674.01

Topic: G.03. Motivation

Support: Swiss National Science Foundation 31003A_182010

Title: Changes in nucleus accumbens dopamine signals accompanying reward-based learning of a goal-directed sensorimotor transformation

Authors: *J. HUANG[1], C. SANDI[2], C. C. PETERSEN[3];
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Mind Institute, EPFL, Lausanne, Switzerland; [3]École Polytechnique Fédérale de Lausanne (EPFL), Lausanne, Switzerland

Abstract: Dopamine signals are thought to be important for reward-based learning and appear to play important roles in regulating synaptic plasticity in the nucleus accumbens and the striatum. However, the precise neuronal circuit mechanisms underlying the learning of even the simplest goal-directed sensorimotor transformations remain to be precisely defined. Here, we measured dopamine signals with fiber photometry using dLight expressed in the nucleus accumbens of head-restrained mice across reward-based sensorimotor task learning. Thirsty mice were first trained in a free-licking task, during which the mouse learned to lick a spout, for which they were sometimes rewarded with water delivery. In free licking sessions, reward triggered a positive dopamine signal, consistent with previous findings, while unrewarded licks evoked a negative dopamine signal. The amplitude of the reward-triggered dopamine response decreased across most individual sessions, likely reflecting the gradual reduction in thirst across each session with accumulated reward. Subsequently, the same mice were trained over days in a whisker-detection task, in which mice learned to lick the reward spout in response to a single brief whisker deflection. Reward delivery appeared to evoke a consistent dLight response across learning days. Whisker detection task learning was accompanied by an increase in a fast sensory-evoked dopamine signal, consistent with a large body of literature indicating dopaminergic reward prediction error signals. These dopamine reward-related signals could causally contribute to task-learning, for example through regulating synaptic plasticity at corticostriatal synapses. Interestingly, dopamine signal dynamics in individual mice during the free-licking task appeared to be predictive of performance on the first day of learning of the whisker detection task. Our results are thus consistent with a role of dopaminergic signalling in reward-based learning and further suggest that inter-individual differences in dopamine dynamics may be an early predictor of future learning performance. As a next step, it will be important to carry out pharmacological and optogenetic manipulations to test these hypotheses in further detail.

Disclosures: J. Huang: None. **C. Sandi:** None. **C.C. Petersen:** None.

Poster

P674. Motivation: Subcortical Neurocircuitry I

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Program #/Poster #: P674.02

Topic: G.03. Motivation

Support: NIDA R00 045765

Title: Binge-like eating promotes behaviors reminiscent of ‘food addiction’ in obese female rats: Role for the orexin/hypocretin system

Authors: *K. FERNANDOPULLE[1,2], S. LIU[1,2], J. B. MEHR[1,2], S. L. O'CONNOR[1,2], A. ARMANIOUS[1,2], M. H. JAMES[1];
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Abstract: Introduction: Significant recent attention has been given to the notion that compulsive overeating, often seen in binge eating disorder and obesity, reflects 'addictive' properties of foods, notably those high in sugar and fat content. Rats with a history of overconsuming palatable food exhibit several addiction-relevant behaviors, including compulsive-like food seeking and enhanced food motivation. The orexin/hypocretin neuropeptide system has been demonstrated to mediate various drug behaviors, however its role in 'food addiction' outcomes has not been examined. Here we examined addiction-relevant behaviors in obese female rats with a history of binge-like eating, and tested whether the orexin system is important for their development and expression. **Methods**: Female Long Evans rats (n=18) were maintained on a high fat diet (45% fat, ad libitum) for 8w before being given intermittent access (30 min, twice/week, 4w) to sweetened fat (vegetable shortening/10% sucrose) in a distinct environmental context ('binge' group). A control ('no binge') group was treated identically but were given an empty food tray during 'binge' sessions. Rats were then trained to lever press for sucrose pellets before being tested for several addiction-relevant behaviors, including food seeking during signaled non-availability periods, 'compulsive' (punished) food seeking, food motivation, and food demand intensity/elasticity. In a second group (n=12), we studied the recruitment of orexin cells after exposure to the binge environment. In a third group (n=16), we tested the effects of a selective orexin-1 receptor antagonist (SB334867; SB) and a dual orexin receptor antagonist (suvorexant) on addiction-relevant behaviors. **Results:** Across all behavioral tests, binge rats showed stronger addiction-like behaviors compared to no-binge controls. Exposure to the binge context was associated with robust recruitment of orexin neurons; orexin receptor antagonism reduced the expression of addiction-like behaviors. **Conclusions**: These data indicate that orexin signaling contributes to the expression of 'food addiction-like' behaviors in obese rats with past binge experience, thus highlighting the orexin system as a potential therapeutic target for compulsive overeating.

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Poster

P674. Motivation: Subcortical Neurocircuitry I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P674.03

Topic: G.03. Motivation

Title: Hunger drives perseveration by dampening a nucleus accumbens circuit

Authors: *N. K. SMITH[1], J. M. PLOTKIN[1], B. A. GRUETER[2];
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Abstract: Obesity and eating disorders are characterized by rigid food seeking behaviors that are resistant to change. Food seeking behaviors are directed by higher-order motivated behaviors that rely on circuits centered on the nucleus accumbens (NAc). Synaptic alterations within these circuits underlie the powerful changes in behavior across different motivational states. Here we show that hunger enhances food-seeking perseveration by dampening excitatory transmission in a NAc to lateral hypothalamus (LH) circuit. NAc GPR171 receptor signaling allows hunger to enhance perseveration, but not persistence, of food-seeking behavior by suppressing presynaptic release probability of glutamate on to a select population of NAc neurons that project to the LH. This presynaptic depression proceeds through a circuit driven mechanism involving acetylcholine signaling. We find that chemogenetic upregulation of the NAc to LH circuit is able to reduce perseveration in food deprived animals. These results describe how hunger-driven neuromodulation targets a distinct dimension of motivated behavior by shaping information flow through anatomically defined circuit elements, and provide a novel target for the rigid food seeking behaviors seen in obesity and eating disorders.

Disclosures: N.K. Smith: None. **J.M. Plotkin:** None. **B.A. Grueter:** None.

Poster

P674. Motivation: Subcortical Neurocircuitry I

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Program #/Poster #: P674.04

Topic: G.03. Motivation

Support: NIH Grant 5R01DA044925

Title: Prenatal THC exposure promotes transcriptional network changes in the ventral tegmental area and promotes reward seeking and phasic dopamine terminal release in the adult rat offspring

Authors: *M. A. LUJAN[1], C. CALARCO[1], B. MAHASWETA[1], S. ARONI[2], S. AMENT[1], G. KUMAR[1], M. MELIS[2], M. K. LOBO[1], J. F. CHEER[1];
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Abstract: Marijuana is the most common illicit drug used by pregnant women and is associated with offspring's attention and learning deficits from early childhood until later in life. However, the impact of prenatal cannabis exposure (PCE) on maladaptive gene expression changes and its consequences on accumbal dopaminergic encoding of reward-seeking behavior have not been

specifically addressed in the adult progeny. In order to address this, we conducted transcriptomic analyses (RNA-*seq*) in VTA brain samples of PCE adult rats. Weighted Gene Co-expression Network (WGCNA), Differential Gene Expression (DGE) and Gene Ontology (GO) analyses all identified single-gene and network-wide alterations of cytoskeletal, mitochondrial, and synaptic plasticity processes related to PCE. Furthermore, the addition of sex as a co-variate revealed transcriptome-wide homeostatic adaptations in female but not in male rats, which failed to cope with the prenatal THC insult. A separate cohort of PCE rats underwent operant training to obtain palatable sucrose pellets under different reinforcement schedules. Congruent with our transcriptome findings and with our previously published work, adult male PCE rats showed sex-specific alterations in reward-seeking behaviors and exhibited exacerbated mesoaccumbal dopamine release ($GRAB_{DA}$) in response to reward-associated cues. Our results suggest that a series of complex, long-lasting transcriptomic maladaptations to *in utero* THC exposure were accompanied by alterations in dopaminergic encoding of natural rewards and their associated environmental cues.

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Poster

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Topic: G.03. Motivation

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Title: Ventral pallidum GABA neurons bi-directionally control remifentanil relapse following voluntary or imposed abstinence in rats

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Abstract: Opioid-addicted individuals often cease their drug use due to mounting life consequences, only to succumb to temptation when encountering cues and contexts previously associated with drug use. Such self-imposed abstinence, despite its prevalence in the addiction cycle, is rarely modeled in rodent addiction experiments, though prior studies have shown that distinct neural circuits are involved in reinstatement of drug seeking after voluntary versus experimenter-imposed abstinence. We and others have shown that ventral pallidum (VP), and

specifically VP GABAergic (VP^{GABA}) neurons, mediate highly motivated reward seeking, as is characteristic of addiction. Here, we tested the functions of these neurons in a model of relapse to opioid seeking after 1) punishment-induced suppression of drug seeking or 2) extinction-mediated suppression of drug seeking. Male and female GAD1:Cre rats and wildtype littermates were implanted with jugular catheters, and infused bilaterally into VP with Cre-dependent inhibitory Gi-coupled DREADDs (n = 13), excitatory Gq-coupled DREADDs (n = 25), or a control vector (n = 31). Rats were trained to lever press for i.v. infusions of the short-acting opioid agonist remifentanil (1.9 μg/50 μL) and a contingent light/tone cue, daily for 14 days in Context A. Next, rats were moved to a distinct Context B, where presses yielded 1) remifentanil+cues, but also a 50% chance of co-administered footshock, or 2) no reward/cue (i.e. extinction training). After achieving abstinence criteria in Context B, we asked how VP^{GABA} DREADD inhibition or activation affected remifentanil seeking in the following tests: Context A (safe context) *with* or *without* response contingent cues, Punishment Context B *with* or *without* response contingent cues, or Extinction Context B *with* or *without* response contingent cues. We found that relative to vehicle day tests, both chemogenetic inhibition and stimulation with CNO (5mg/kg) bi-directionally affected reinstatement following either self- or experimenter-imposed abstinence, in a manner dependent upon discrete drug-paired cues. These results provide new insights into the roles for VP^{GABA} neurons in opioid relapse-like behavior, and demonstrate that these neurons play a key role in relapse to opioid drug use regardless of the experimental model employed.

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Poster

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Topic: G.03. Motivation

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Title: Feeding and exercise essential values examined in cannabinoid type-1 (CB_1) receptor mutant mice exposed to closed economy conditions

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Abstract: Body homeostasis is regulated by the balance between feeding and activity. Dysregulations in this balance, as observed in obesity or anorexia nervosa, find their roots at the motivation level. This led us to develop an operant conditioning protocol wherein wheel-running motivation, palatable food motivation and preference for either reward in a choice task can be assessed. The use of this protocol, combined with pharmacological and genetic (i.e. constitutive and conditional mutant mice for cannabinoid type-1 (CB_1) receptors) approaches, has revealed the involvement of CB_1 receptors located on GABAergic neurons in the choice between palatable feeding and wheel-running (Muguruza et al., JCI Insight 2019). Our protocol however suffers two main limitations which are respectively related to the facts that (i) mice were daily exposed to the operant chambers, albeit for a limited time (1h), and (ii) home cages were provided with (unlimited) food but not with a running wheel. Thus, the drives for palatable food and wheel-running might have been respectively underestimated and overestimated. Accordingly, we next investigated the role of CB_1 receptors in a closed-economy choice setting wherein mice living in the operant chambers were able to feed a standard diet and to run, doing so under fixed ratio (FR) reinforcement schedules (FR1, FR3, FR10 and FR30). Demand curves for food and wheel running allowed the calculations of their respective essential values. We found that CB_1 receptors located on GABAergic neurons in adult male/female mice (n=5-7 mice/genotype/sex) positively control the essential value for wheel-running. As opposed to the role exerted by CB_1 receptors on palatable feeding motivation, the full deletion of CB_1 receptors proved inefficient on standard food motivation, even after successive changes in the price of food. Using conditional male/female mice expressing CB_1 receptors exclusively in GABAergic neurons, we next asked if the essential value for running was rescued in this group. Although such a CB_1 receptor re-expression allowed an increase in the essential value for running, its level did not fully reach that measured in wild-type mice. This finding indicates that CB_1 receptors on GABAergic neurons are necessary, but only partly sufficient, for running motivation under closed economy conditions. Overall, this model provides a new framework for studying genetic and environmental causes of obesity and anorexia nervosa.

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Poster

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Topic: G.03. Motivation

Title: Dorsal raphe provides a glutamatergic input to lateral hypothalamic GABA neurons that plays a role in reward

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Abstract: The dorsal raphe (DR) nucleus is known for containing serotonin neurons that innervate many brain structures. However, we previously demonstrated that a subset of DR neurons expressing the vesicular glutamate transporter 3 (VGluT3) provides a major glutamatergic input to ventral tegmental area, and also identified the Lateral Hypothalamus (LH) as a major target of DR-VGluT3 neurons. Here, we applied anatomical, electrophysiological and behavioral approaches to characterize these DR-VGluT3 inputs to LH. By LH application of the retrograde tracer fluorogold (FG), followed by *in situ* hybridization, we found that within the total population of FG neurons, as many as 60% expressed only VGluT3 mRNA, 10% percent co-expressed VGluT3 with tryptophan hydroxylase (for synthesis of serotonin), and 3% co-expressed VGluT3 and glutamic acid decarboxylase (for the synthesis of GABA). To determine the extent to which DR-VGluT3 terminals form synapses on LH-GABA (VGAT) or glutamate (VGluT2) neurons, we tagged DR neurons by local injection of AAV-CamKII-ChR2-mCherry and LH neurons by local injections of AAV-DIO-eYFP in VGAT::Cre or VGluT2::Cre mice. Within the LH, we observed axons co-expressing mCherry and VGluT3 intermixed with both VGAT and VGluT2 neurons. By immuno-electron microscopy, we found that within the LH, DR-VGluT3 neurons made asymmetric (putative excitatory) synapses mostly on GABA neurons (≈ 80 %) and to a less extent on glutamate neurons. To further confirm the excitatory synapses established by DR-VGluT3 terminals on LH neurons, we prepared LH slices from VGluT3::Cre mice expressing ChR2-eYFP in fibers from DR-VGluT3 neurons. By *in vitro* recording, we detected AMPA-mediated excitatory postsynaptic currents evoked by photoactivation of LH VGluT3 fibers. Next, by in vivo tests, we found that activation of DR-VGluT3 fibers in the LH induced c-Fos expression in local LH-GABA neurons, reinforced instrumental behavior, and produced a conditioned place preference. Both photoactivation-induced behaviors were diminished by intra LH application of glutamate receptor antagonists, but not by serotonin or GABA receptor antagonists. We also found that LH photoactivation of VGluT3 inputs increases sucrose reinforcement behaviors without changing the latency to eat. In a mouse model of depression, we detected attenuation of the activity of DR-VGluT3 neurons innervating the LH to a sucrose reward, suggesting that these DR-VGluT3 neurons play a role in reward and may mediate behavioral symptoms of anhedonia. In conclusion, we demonstrated that DR-VGluT3 neurons provide an excitatory input to LH-GABA neurons that play a role in reward.

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Poster

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Title: Hierarchical control of mesolimbic dopamine and striatal encoding of reward-paired cues governs behavioral flexibility

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Abstract: The ability to resolve uncertainty surrounding reward-associated cues is essential for the proper organization and generation of reward-seeking. Dopamine neurons are reported to be sensitive to the likelihood of reinforcement yet how dopamine neurons, dopamine release, and the activity of their striatal targets are related to the coding and resolution of uncertainty is unclear. Here we make use of pharmacology, *in vivo* electrophysiology, calcium imaging, and optogenetics to determine the contributions of the mesolimbic dopamine system to the hierarchical organization of reward-seeking. Long-evans rats were trained to discriminate when a conditioned stimulus would be followed by sucrose reward by exploiting the prior and non-overlapping presentation of a separate discrete stimulus, an occasion setter. Only when the occasion setter's presentation preceded the conditioned stimulus did the conditioned stimulus predict sucrose delivery. We found that either reversible inactivation or dopamine antagonism within the nucleus accumbens prevented rats from properly estimating when the conditioned stimulus would be rewarded. We recorded single neurons within the nucleus accumbens (n=295) and observed that the magnitude of conditioned-stimulus evoked inhibitions were greater when the conditioned stimulus would be followed by reward, than when it would not. Moreover, we observed a population of neurons in the nucleus accumbens that dynamically altered their firing to the conditioned stimulus, being excited when this cue was not predictive and inhibited when it would predict reward. We monitored dopamine release in the nucleus accumbens making use of fiber photometry and the fluorescent dopamine sensor dLight 1.3b. Dopamine release tracked the presentation of the occasion setter and dynamically scaled when the conditioned stimulus would or would not predict reward predicated on the prior presence or absence of the occasion setting cue. Finally, we find that optogenetic inhibition of dopamine neurons in the ventral tegmental area disrupts the ability of rats to use occasion setters to flexibly scale their reward-seeking. Together these results reveal a mechanism for dopamine neurons and the effects of dopamine release in the nucleus accumbens to dynamically control striatal encoding of ambiguous reward-predictive cues and appropriately generate reward-seeking.

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Poster

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Title: Ventral arkypallidal neurons inhibit accumbal firing to promote reward consumption

Authors: *Y. M. VACHEZ[1], J. R. TOOLEY[1], K. ABIRAMAN[1], B. MATIKAINEN-ANKNEY[3], E. CASEY[2], T. EARNEST[2], L. M. RAMOS[4], H. SILBERBERG[4], E. GODYNYUK[1], O. UDDIN[5], L. MARCONI[6], C. E. LE PICHON[4], M. C. CREED[1];
[1]Anesthesiol., [2]Psychiatry, Washington Univ. Sch. of medicine in St. Louis, Saint Louis, MO; [3]Psychiatry, Bridget Matikainen-Ankney, Saint Louis, MO; [4]Eunice Kennedy Shriver Natl. Inst. of Child Hlth. and Human Development, Natl. Inst. of Health., Bethesda,, MD; [5]Univ. of Maryland Baltimore, Baltimore, MD; [6]Perelman Sch. of Med. at Univ. of Pennsylvania, Philapdephia, PA

Abstract: The nucleus accumbens shell (NAcSh) and the ventral pallidum (VP) are critical for reward processing, although the question of how coordinated activity within these nuclei orchestrates reward valuation and consumption remains unclear. Inhibition of NAcSh firing is necessary for reward consumption, but the source of this inhibition remains unknown. Here, we report that a subpopulation of VP neurons, the ventral arkypallidal (vArky) neurons, project back to the NAcSh, where they inhibit NAcSh neurons in vivo in mice. Consistent with this pathway driving reward consumption via inhibition of the NAcSh, calcium activity of vArky neurons scaled with reward palatability (which was dissociable from reward seeking) and predicted the subsequent drinking behavior during a free-access paradigm. Activation of the VP-NAcSh pathway increased ongoing reward consumption while amplifying hedonic reactions to reward. These results establish a pivotal role for vArky neurons in the promotion of reward consumption through modulation of NAcSh firing in a value-dependent manner.

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Poster

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Topic: G.03. Motivation

Support: WRII

Title: Distinct neuronal dynamics of lateral hypothalamus encode approach and consumption behavior

Authors: *Y. KIM, Y. LEE, H. SONG, K. KIM, M. JANG, J. LEE, D.-S. HA, S. JUNG, J. PARK, S. KIM, H. CHOI;
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Abstract: Lateral hypothalamus (LH) has been known as a prime candidate for the motivation center in feeding. However, recent studies demonstrated that GABAergic neurons in LH are responsible for behaviors outside feeding contexts; gnawing and chewing non-food objects. In addition, LH GABAergic neurons exhibit heterogeneity, while located widely throughout unspecified areas. Thus, we aimed to identify the food-specific role of LH GABAergic neurons and which subpopulation neurons encode the motivation for food. Micro-endoscope results from LH GABAergic neurons showed that there are food-specific and non-specific neurons in these neurons. Especially, food-specific LH GABAergic neurons responded consistently to food-related situations. Followed by these experiments, we conducted photometry experiments and found that neural activity of LH LepR neurons, which are the subpopulation of LH GABAergic neurons, significantly increases during feeding behavior, but did not change when mice contacted non-food or drank water. More interestingly, micro-endoscope results further demonstrated that LH LepR neurons can be divided into two distinct subpopulations: those that activate at the initiation of food-approach behavior, and those that activate at the initiation of food-consummatory behavior. To investigate the upstream regulator of LH LepR neurons, we tested the role of NPY, which is known to be the AGRP neuron neuropeptide driving sustained feeding behavior. Brain slice study results showed that the activity of LH LepR neurons increases significantly after the application of NPY. In conclusion, by using fiber photometry, micro-endoscope, and brain slice study, we suggest that LH LepR neurons activate only in food-specific contexts and can be further classified into two groups based on feeding phases.

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Poster

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Topic: G.02. Reward and Appetitive Learning and Memory

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Title: Effective connectivity of reward networks in humorous and monetary rewards

Authors: *Y.-C. CHAN[1], Y.-C. CHEN[1], W.-C. HSU[2], J.-Y. YANG[1], C.-C. YEH[1], C.-H. YEH[1]; [1]Natl. Tsing Hua Univ., Hsinchu, Taiwan; [2]Natl. Taiwan Univ. of Sci. and Technol., Taipei, Taiwan

Abstract: Most of this research has focused on brain regions and functional connectivity and less is known about patterns of effective connectivity for humorous and monetary rewards. Thirty right-handed with no history of neurological or psychiatric disorders participated in this study. The fMRI study used the 32 humor stimuli, along with 32 monetary stimuli with an image of coins and 32 non-reward stimuli with a gray scrambled image were used. Each trial started with a central fixation. One of the visual reward cues appeared, followed by an anticipatory delay period. Next, participants responded a numerical judgment task. Participants were then notified of whether or not they had earned a reward on the trial and provided a subjective pleasantness rating. Imaging was performed using a 3 T Siemens Magnetom Prisma scanner using a standard 20-channel head coil. The images were pre-processed and analyzed using Statistical Parametric Mapping (SPM12) implemented in Matlab. DCM with PEB analysis was conducted to evaluate effective connectivity for the different reward. Effective connectivity during the 'liking' or outcome phase was then conducted with the subjective pleasantness ratings included as covariates. The modulatory effects for humor rewards involved stronger connections from the VTA to the amygdala and from the vmPFC to the amygdala. Additionally, the modulatory effect of pleasure from humor showed that NAc activity causes a decrease in the rate of amygdala activity. Pleasure from monetary gains activated a network from the VTA to the NAc and from the amygdala to the NAc. The modulatory effects of pleasure from monetary gains were also associated with stronger connections from the VTA to the vmPFC and the lOFC to theVTA. In sum, the VTA seems to play a crucial role in receiving rewards. Amusement from humor increased the modulation effect from the VTA to the amygdala, while the VTA to NAc connection was found to be active during consumption of monetary rewards.

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Poster

P694. Positive and Negative Emotional States

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Title: Amygdalostriatal transition zone neurons encode sustained valence to direct conditioned behaviors

Authors: *F. MILLS[1], C. R. LEE[1,2], J. R. HOWE[2], S. SHAO[3], M. E. LEMIEUX[1], M. N. KEISLER[1], R. R. PATEL[1], H. S. CHEN[4], A. L. GROSS[6], F. H. TASCHBACH[1,2], K. BATRA[1,2], K. M. TYE[5];
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Abstract: The ability to respond appropriately to stimuli that predict rewards or punishments lies at the core of evolutionary fitness, and is disrupted in a number of neuropsychiatric disease states. The amygdalostriatal transition zone (ASt) is anatomically poised to provide a shortcut between corticolimbic and basal ganglia circuitry, and, in parallel with the amygdala, mediate associative learning and behavioral responses to salient stimuli. Like the amygdala, the ASt receives converging sensory input from the thalamic and cortical pathways. However, the downstream projections of the ASt are distinct from canonical outputs of the amygdala complex, and are integrated with striatal circuits involved in action selection. Despite this intriguing circuit connectivity, the function of the ASt is almost completely unknown. Here, we characterized the transcriptomic profile of the ASt in comparison to neighboring amygdalar or striatal nuclei, and collect cellular resolution recordings of genetically-defined neurons during a valence discrimination task to interrogate the functional role of ASt circuitry. We first used single-nucleus RNA sequencing to generate a comprehensive profile of gene expression in ASt neurons, and find that the ASt is a genetically distinct structure from adjacent GABAergic brain regions

(n=18-25 mice, >15000 cells per brain region, >50k unique molecular identifiers per cell). RNAscope labelling also showed a greater proportion of neurons expressing dopamine receptor 2 (D2+) than dopamine receptor 1 (D1+) in the ASt compared to other regions of the striatum (n=8 mice, >8 sections per group, Chi-square test, p=0.0032). Using in vivo electrophysiology we found that that ASt neuron responses to a shock-predicting cue were significantly greater following fear conditioning in 'paired group' mice compared with 'unpaired group' controls (n=8 mice, 30 neurons Paired, n=5 mice, 27 neurons Unpaired, p=0.028, RM ANOVA). We also identify distinct groups of ASt neurons that encode opposing responses to cues predicting reward and punishment, including sustained responses to negative valence cues (n=9 mice, n=41 neurons). Preliminary calcium imaging data also suggests that D2+ ASt neurons show increased conditioned cue responses following fear conditioning. Finally, we find that optogenetic inhibition of D2+ ASt neurons causes a striking reduction in conditioned fear responses to a shock-predicting cue (43% decreased freezing, p=0.0145, paired t-test, n=8 mice NpHR, n=9 mice eYFP). Consequently, our findings suggest that the ASt may be an overlooked and critical structure of the amygdala complex that contributes to conditioned responding.

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Poster

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Title: Downstream targets of DRN dopamine neurons represent distinct facets of the experience of social isolation

Authors: *C. R. LEE[1,2], G. A. MATTHEWS[2,3], M. E. LEMIEUX[2,3], G. SCHNEIDER[2], E. M. BREWER[3], M. R. BORIO[2,3], R. MIRANDA[2,3], L. KEYES[2], E. PERONI[3], G. S. PEREIRA[3], A. LOPEZ MORAGA[3], A. PALLÉ[3], E. Y. KIMCHI[3], N. PADILLA-COREANO[3,2], R. WICHMANN[3,2], K. M. TYE[4];

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Abstract: Affiliative social connections facilitate well-being and survival in numerous species. Engaging in social interactions requires positive and negative motivational drive, elicited through coordinated activity across neural circuits. However, the identity, interconnectivity, and functional encoding of social information within motivational neural circuits remains poorly understood. Here, we focused on the downstream projections of dorsal raphe nucleus (DRN) dopamine neurons using dopamine transporter (DAT)-Cre transgenic male mice (DRN_{DAT}), allowing us to target this subpopulation of cells which we previously implicated in isolation-induced social motivation (Matthews et al., 2016). We wondered whether dopaminergic innervation from the DRN could transmit information about social isolation to be represented in downstream targets. Using optogenetics, anatomical tracing, ex vivo electrophysiology, and in vivo cellular resolution calcium imaging, we explored how DRN_{DAT} innervation affected social motivation and neural dynamics of the different downstream targets. We investigated four prominent DRN_{DAT} downstream targets - the bed nucleus of the stria terminalis (BNST), central amygdala (CeA), posterior basolateral amygdala (BLP) and medial prefrontal cortex (mPFC) to probe how each facet of this ethological behaviors promoted by DRN_{DAT} photostimulation was implemented by each circuit component. Photoactivation of the DRN_{DAT}-CeA projection promoted social behavior (n=21 mice; p=0.027) and photostimulation of the DRN_{DAT} -BNST projection promoted exploratory behavior (n=22 mice; p=0.0298), while the DRN_{DAT} -BLP projection supported place avoidance (n=12 mice; p=0.0455), suggesting a negative affective state. Given that DRN_{DAT}-CeA projections promoted social behavior, we performed calcium imaging using GCaMP7f and miniature microendoscopes during social interaction before and after acute (24 hours) social isolation. While we were able to weakly decode if animals were group housed or isolated prior to social interaction based on CeA activity (p = 0.0054, effect size = 12.32% of mean), we observed more robust decoding of housing status from mPFC activity (p = 0.0035, effect size 31.68% of mean). Our findings support a role for DRN_{DAT} projections in promoting distinct features of the response to novel social stimuli - orchestrating a coordinated, flexible response through recruitment of specific downstream circuits. Uncovering the neural circuit mechanisms which govern sociability and revealing the neural representation of a loneliness-like-state are keys to understanding the fundamental need for social connection.

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Poster

P694. Positive and Negative Emotional States

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Title: A neuropeptidergic mechanism for governing valence assignment

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Abstract: The ability to assign positive or negative valence to environmental cues is paramount for survival. Basolateral amygdala (BLA) circuits have been implicated in associative learning, and BLA projections to the nucleus accumbens (NAc) predominantly mediate positive valence (reward), while BLA projections to the centromedial nucleus (CeM) of the central amygdala (CeA) predominantly mediate negative valence (punishment). However, the neural mechanisms initially directing signals to the appropriate pathway during learning remain mysterious. Here, we demonstrate the neuropeptide neurotensin (NT) plays a critical role in valence assignment during the acquisition of learned associations. BLA-NAc and BLA-CeM neurons express the neurotensin receptor 1 (NTSR1) at different levels (***P=0.0005), and NT dose-dependently altered glutamatergic transmission in an opposing manner for BLA-NAc (facilitation; *P=0.0425) and BLA-CeM (suppression; **P=0.0039) neurons. Among inputs providing NT innervation to the BLA, we identified the paraventricular thalamus (PVT) as a functional source of NT that mediates learning. Photostimulation of the PVT-BLA:NT projection enhanced reward learning (*P=0.018) and impaired punishment learning (*P=0.027). However, we also found that PVT:NT neurons corelease glutamate into the BLA. To selectively isolate the contribution of NT, while leaving glutamate transmission unaffected, we used a CRISPR-Cas9 mediated *Nt* gene conditional knockout (cKO) and found an impairment in reward learning (**P=0.0052), and an enhancement in punishment learning (*P=0.039). By developing, validating, and applying a genetically-encoded fluorescent NT sensor to isolate NT signaling from coreleased glutamate,

we observed that NT concentration in the BLA increased during reward conditioning (*P=0.047) and decreased during punishment conditioning (*P=0.003). Finally, to reveal the impact of PVT-BLA:NT on the ensemble dynamics during valence processing, we recorded the activity of BLA neurons (n=683) during a valence-discrimination task in mice with PVT-BLA:*Nt* CRISPR-cKO relative to scrambled guide controls. We found that *Nt* cKO blunted responses to both reward- and punishment-predictive cues, and that valence-coding properties of BLA-NAc and BLA-CeA were disrupted, even when controlling for behavioral states on a trial-by-trial basis. Taken together, we have identified a neuropeptide that signals valence in the BLA, and show that NT is a critical modulator of valence processing, distinctly gating synaptic transmission and orchestrating ongoing activity of amygdala projection neurons mediating positive and negative valence.

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Poster

P694. Positive and Negative Emotional States

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P694.04

Topic: G.04. Emotion

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Title: Cortical ensembles decode social rank and promote dominance behavior via hypothalamic projections

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Abstract: How do we know our social rank? Most social species, from insects to humans, self-organize into social dominance hierarchies. The establishment of social ranks serves to decrease aggression, conserve energy, and maximize survival for the entire group. Despite dominance behaviors being critical for successful interactions and ultimately, survival, we have only begun to learn how the brain represents social rank and guides behavior based on this representation. The medial prefrontal cortex (mPFC) has been implicated in the expression of social dominance in rodents, and in social rank learning in humans. Yet precisely how the mPFC encodes relative social rank and which circuits mediate this computation is not known. We developed a trial-based social competition assay in which mice compete for rewards, as well as a computer vision tool to track multiple, unmarked animals. With the development of our open source deep learning computer vision tool (AlphaTracker) and wireless electrophysiology recording devices, we have established a novel platform to facilitate quantitative examination of how the brain gives rise to social behaviors. We describe nine behaviors during social competition that were accurately decoded from mPFC ensemble activity using a hidden Markov model combined with generalized linear models (HMM-GLM) (sign test p=0.004 HMM-GLM to chance model; n=9 behaviors). Population dynamics in the mPFC were predictive of relative and absolute social rank and competitive success (p=0.00018 using Wilcoxon rank sum test on auROC curves when compared to shuffled data, for both decoders; n=10 folds/1410 trials). This population-level rank representation translated into differences in the individual cell responses to task-relevant events across ranks that were not present when the mice were alone (p<0.001 using Fisher's exact test for 3 out of 6 events). Finally, we demonstrate that mPFC cells that project to the lateral hypothalamus promote dominance behavior during the reward competition (p=0.03 using 2-way RM ANOVA ChR2 and eYFP). Thus, we reveal a cortico-hypothalamic circuit by which mPFC exerts top-down modulation of social dominance.

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Poster

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Title: Alphatracker2.0: a platform for behavioral estimation of multiple animals, enabling real-time, closed-loop manipulations

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Abstract: To understand how neural dynamics give rise to complex, ethological behaviors, neuroscience must move towards a comparable temporal resolution of measuring behavior as exists for measuring neural activity. Recent advances in deep learning have opened the door for automated behavioral scoring. State-of-the-art tools allow users to capture pose estimates of behaving animals and classify them into distinct behavioral patterns, but this approach has limitations. First, labeling both pose and behavior can be time-consuming. Second, users must label body parts that will be important for detecting behaviors, but such selection can be difficult and subjective. Finally, other existing methods directly classify frames of a video into behaviors; however, these approaches are unable to deal with cases of more than one animal or behavior, such as social interaction studies, where multiple animals often perform distinct independent actions in addition to interactions with each other. To provide solutions to these challenges, we present AlphaTracker2.0 (AT2.0) - a supervised, deep learning-based, behavioral extraction method that directly estimates social and non-social behaviors of multiple animals from images. AT2.0 provides the benefits of 1) greatly reducing the effort expended on labeling by requiring only behavioral labels without body parts, 2) fast inference times by using a single, lightweight neural network, making real-time, closed-loop manipulations possible, and 3) allowing investigations of groups of interacting animals performing various actions by detecting the precise location of behaviors in images. To achieve this, we conceptualize behavioral identification as an object detection problem. We utilize “keypoint” labels common to pose estimation methods, but instead use them to directly label behaviors on images, thereby bypassing body part detection and increasing the rapidity of this process. For example, to classify face-to-face interactions in mice, a “keypoint” can be placed at the location of intersection of the noses of the two mice.
We demonstrate AT2.0’s efficacy by classifying 5 behaviors (chasing, face-to-face, flank attack, rearing, and grooming) in up to four interacting mice, and show high accuracy with over 0.9 mean average precision values across all behaviors. Additionally, AT2.0’s one-step approach results in rapid inference speeds (9.24 ± 0.00852 milliseconds per frame), increasing the feasibility of closed-loop experiments. Overall, we show that AT2.0 functions as an accurate behavior detector capable of running in real-time, which can greatly facilitate neuroscientific studies of behaviors.

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Poster

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Title: Divergent circuits of the cortical amygdala mediate innate olfactory valence

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Abstract: Innate behaviors are observed across the animal kingdom, wherein individuals display stereotyped responses to specific evolutionarily relevant stimuli in the absence of prior learning or experience, suggesting developmental or genetic origins. Our past research has demonstrated that the cortical amygdala (CoA) is a key brain region mediating innate motivational valence to olfactory stimuli. However, little is known about the pathways linking these stimuli to valence, nor the genetic basis for such circuits. Here, we identify and characterize interlinked molecular organization, projection motifs, and subregions underlying the cortical amygdala's control of innate olfactory aversion and attraction. First, we found evidence suggesting divergent contributions of the anterior (aCoA) and posterior cortical amygdala (pCoA) to negative and positive valence behaviors, respectively. Next, we performed single-nucleus RNA sequencing and single nucleus ATAC-seq separately on aCoA and pCoA, revealing correlated multi-omic gradients in gene expression and chromatin accessibility along the anterior-posterior CoA axis with distinct populations of divergent cell types across the two sub-regions. Further, using anterograde and retrograde circuit tracing, we were able to identify two circuits, where the CoA projects to multiple valence-related subcortical structures, but primarily to the medial amygdala (MeA) and to the nucleus accumbens (NAc). When interrogating these circuits separately, we found that the CoA-MeA projection mediates innate olfactory aversion, and the CoA-NAc projection mediates innate olfactory attraction. Together, these results illustrate the previously unknown molecular, circuit, and anatomical mechanisms involved in innate olfactory valence.

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Poster

P694. Positive and Negative Emotional States

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Title: Heart rate change of rhesus monkeys in anticipation of appetitive rewards

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Abstract: Autonomic response measures are often used to infer internal states of subjects who cannot do self-reporting. Although the autonomic nervous system is mainly involved in homeostasis, it has been shown in many studies that autonomic activity reflects cognitive and affective states in humans and animals. As a step to develop a method to study affect and mood regulation, we measured heart rates of rhesus monkeys while they performed tasks in which different fluid reward amounts were cued. For the convenience of cardiac activity measurement, a pulse oximetry (PO) sensor was used. We first validated the consistency between electrocardiography (ECG) and PO by recording both signals simultaneously and comparing them. In our data, the peaks in the ECG signals preceded those in the PO signals by about 300 ms and the correlation of heart rates calculated from both signals was high ($r = 0.95$, $p < 1.0 \times 10^{-15}$). Next the monkeys were trained on a choice task in which different visual cues signaled different magnitudes of fluid reward. A fixed set of 7 cues was associated with fixed amounts of fluid, ranging from 0.15 ml to 1.05 ml. A trial began when the monkey pressed a central button for 1.5 s. Then two visual cues, pseudo-randomly selected from the set, were presented for 1.5 s, one on each side of the monitor. The monkey could choose the cue on the left or right side of the screen by selecting and pressing either the left or right button, respectively, after which a 1.5-s waiting period ensued. At the end of the waiting period, the fluid reward amount associated with the chosen cue was delivered. The temporal separation of the cue period and reward delivery ensured that any change in heart rate during the cue period could not be attributed to breath holding or other reward consumption activities. We found that the heart rates estimated by PO increased as the reward amount became larger. The changes in the heart rate began to occur approximately 500 ms after the cue onset. The heart rates during the cue period were analyzed with a regression model which included variables such as the location of the larger reward, choice direction, reaction time and the amount of the larger reward. The heart rate during the cue period was significantly related to the amount of the larger reward (t-test; $p < 0.01$), but not to the reaction time ($p > 0.05$). It has been suggested that heart rate may reflect a temporally extended affective state (10-50 seconds) in nonhuman primates, because of its long latency. However, our results show that heart rate can be modulated fast enough to reflect trial-by-trial changes of attention, arousal or motivation.

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Poster

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Title: Involvement the lateral habenul-dorsal raphe nucleus-ventral tegmental area projection in social instigation-heightened aggression in male mice.

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Abstract: A brief encounter with a potential rival induces aggressive arousal which increases subsequent aggressive behavior, phenomena referred to as social instigation or aggression priming. Previously, we found that the level of glutamate input into the dorsal raphe nucleus (DRN) was increased when animal was engaging instigation-heightened aggression. The DRN receives glutamatergic projections from the lateral habenula (LHb), and c-Fos analysis showed an activation of DRN-projecting LHb neurons (LHb-DRN projection) during social instigation-heightened aggression. Both chemogenetic and optogenetic inhibition of the LHb-DRN projection blocked aggression-heightening effect of social instigation in male mice. Optogenetic activation of the LHb-DRN projection caused an increase of aggressive behavior, indicating that the activation of this pathway is sufficient to induce escalated aggression. We then examined

which DRN neurons are responsible for the aggression-promoting effect of the LHb-DRN glutamatergic input. Although the DRN is a major serotonin (5-HT) nucleus, optogenetic inhibition of DRN 5-HT neurons did not block instigation-heightened aggression. Anatomical analysis identified that LHb glutamate projection innervate non-serotonergic DRN neurons that project to the ventral tegmental area (VTA). Indeed, optogenetic activation of DRN-VTA projection increased aggressive behaviors. Our results indicate that the LHb-DRN-VTA projection is involved in the escalation of aggression induced by social instigation.

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Poster

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Title: Internal state and behavior dependent responses of Central Amygdala PKC-δ+ neurons to food related stimuli

Authors: *M. SCHMIT[1], M. RIVERA[2], K. VO[2], T. HASNEEN[2], G. OZTURK[2], H. CAI[1]; [1]Neurosci., [2]Univ. of Arizona, Tucson, AZ

Abstract: The central amygdala (CeA) has been shown to play a role in a variety of functions including arousal, anxiety, fear responses, and feeding, among other motivated behaviors. How this confluence in the CeA influences our perception and experience of different emotional and motivational states is a key unresolved question. Our lab focuses on a subset of these CeA neurons, marked by the expression of protein kinase C-delta (PKC-δ+). Although optogenetic and chemogenetic studies suggest these neurons play a role in feeding inhibition, it is unknown how the neural activity dynamics line up with their function. Our work fills this gap, employing in-vivo calcium imaging - expressing Cre-dependent GCaMP6s in *prckd-cre* mice - and observing the neural activity through a head mounted miniature microendoscope during various feeding-related behaviors. Our data show that the baseline activity of PKC- δ+ neurons is increased after i.p. injection of CCK, consistent with previous *cfos* studies. However, we also show that PKC-δ+ neurons respond transiently during the approach to food. Unlike baseline activity, food approach response is enhanced in both fasted and sated conditions. Continuous optogenetic PKC-δ+ neuron activation in a fasted state inhibits food intake, but our data suggests

that activity paired with food approach may serve a different purpose or possibly be parsed differently downstream. We suggest that the homeostatic need of the mouse is represented in the baseline activity of PKC-δ+ neurons, while the salience of the food stimulus is reflected in their transient response during approach. Understanding how these two aspects of feeding are represented in the same neurons in the central amygdala may help us understand how multimodal pieces of information are integrated in this structure.

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Poster

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Title: Amygdala-cortical circuit determinants of social isolation-induced alcohol consumption

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Abstract: An individual's standing in a social hierarchy is inversely related to alcohol consumption in rodents and non-human primates as well as problematic drinking in humans, highlighting the conserved impact of subordination on motivation for alcohol. Social rank also influences how individuals respond to challenges such as social isolation, a particularly profound stressor with increasing human relevance. However, the neural circuit mechanisms by which social stress experiences engender aberrant alcohol drinking remain largely unknown. In this study, we found that social rank of adult, male mice correlates with alcohol intake (r^2 = 0.25 by Pearson's correlation; **p = 0.01; n = 26), and social isolation increases alcohol intake (***p < 0.001 by paired t-test; n = 14). Notably, we identified a previously unknown relationship between social rank and social isolation-induced escalated alcohol drinking, where subordinates

display a greater magnitude increase in drinking compared to dominants (**$p < 0.01$ by unpaired t-test; n=3/group). These data suggest behavioral factors, emerging from dominance hierarchies, can predict vulnerability to social isolation-induced escalated alcohol drinking. Using *ex vivo* whole-cell patch-clamp electrophysiology, we found that social isolation increases basolateral amygdala (BLA) excitability (*$p < 0.05$ by two-way ANOVA; n= 13-14 cells), highlighting the vulnerability of the BLA to isolation stress. Unilateral photoactivation of ChR2-expressing BLA terminals in the mPFC increased alcohol (YFP: 138.5 licks and ChR2: 392.5 licks; ***$p < 0.001$ by two-way ANOVA post hoc; n = 6/group), but not sucrose or water, drinking, suggesting that social isolation may escalate alcohol drinking through BLA projections to the mPFC. Indeed, using single-cell calcium imaging, we found that the BLA encodes alcohol (n = 180-109 cells), and the percentage of alcohol-encoding neurons is increased following social isolation (from 9.5% to 18.3%). Using a SVM classifier, we found that population-level BLA activity preceding a drinking bout accurately (>70%) decodes whether alcohol or water was consumed, further supporting a role of the BLA in encoding alcohol. Together, these findings suggest that low social rank may be a potent risk factor for increased alcohol drinking following a social isolation challenge, which may be mediated by amygdala-cortical circuits.

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Poster

P715. Other Psychiatric Disorders Including Schizoaffective and Eating Disorders

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Topic: F.03. Neuroendocrine Processes

Title: Suvn-2300191, a 5-hydroxytryptamine 1a receptor partial agonist for the treatment of depressive disorders

Authors: *V. BENADE, H. TALAKANTI, R. ABRAHAM, N. MUDDANA, L. GUTTIKONDA, R. SUBRAMANIAN, S. PETLU, N. NATH, V. TELIDEVARAPALLI, P. AMIDALA, A. MOHAMMED, R. NIROGI;
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Abstract: Depression is a mental condition affecting more than 300 million people globally, and is considered to be a leading cause of disability worldwide. The treatments available for depression are still inadequate because a good proportion of patients (60%) do not respond to existing antidepressants and majority of patients experience relapse. Serotonin (5-hydroxytryptamine, 5-HT) is one of the major neurotransmitter systems implicated in several neuropsychiatric and neurological disorders. Among all the 5-HT receptors, the potential role of serotonin 5-HT_{1A} subtype is implicated in the pathophysiology of depression and in the

mechanism of action of antidepressants. SUVN-2300191 is a potent and selective 5-HT_{1A} receptor partial agonist. It showed good oral bioavailability and excellent brain penetration in rodents. Dose-dependent occupancy was observed in rat brain for 5-HT_{1A} receptors using non-radiolabelled tracer. In the forced swim assay, it showed antidepressant like effects by significantly reducing the immobility in both rats and mice. SUVN-2300191 produced dose-dependent increase in dopamine and norepinephrine levels in prefrontal cortex. SUVN-2300191 is currently being profiled as a new therapeutic option for the treatment of depression.

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Poster

P715. Other Psychiatric Disorders Including Schizoaffective and Eating Disorders

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Topic: G.08. Other Psychiatric Disorders

Support: HRZZ (Croatian Science Foundation) IP-2018-01-9424
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Title: Aggregation of CRMP1, DISC1 and NPAS3 in the brains of victims of suicide

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Abstract: Chronic mental illnesses (CMIs), such as schizophrenia and major depression, are complex conditions with hereditary and non-genetic components. As a result of overlapping symptoms between neurodegenerative disorders and CMIs, we and others have proposed protein aggregation as a possible mechanism. Several proteins have so far been implicated as aggregating in CMIs, including Collapsin Response Mediator Protein 1 (CRMP1), Disrupted in

Schizophrenia 1 (DISC1), Neuronal PAS domain-containing protein (NPAS3), and TRIO and F-actin-binding protein (TRIOBP-1). Most of these have been detected in multiple CMIs, suggesting aggregation to cross diagnostic boundaries. In this study, we looked at suicide as an alternative, discrete non-subjective measure that also spans diagnoses. Studies were performed on brains samples from 14 victims of suicide and 14 control individuals. 6 samples each from patients with depression and Alzheimer's disease were also included for context. Brains were donated as part of the Hungary-wide Lenhossék Program, after granting of family or a legal permission. Samples of the insular cortex were isolated, and flash-frozen after a post-mortem interval of 4.60±0.28 hours. Ethical approval for each stage of this work was obtained from the ethical committees and boards from the Ministry of Health of Hungary, the Semmelweis University, and/or the University of Rijeka, as appropriate. Brain samples were homogenized, and the insoluble (including aggregating) protein fraction was purified with a series of solubilization buffers and ultracentrifugation. Both the original homogenates and this purified insoluble fraction were analyzed with Western Blot. All procedures were performed under blinded conditions. Strikingly, specific species of NPAS3 (n=4), DISC1 (n=2) and CRMP1 (n=3) were found only in the insoluble fractions of suicide victims, and not in any control samples. Three of these samples showed multiple aggregated proteins present. Additionally, levels of CRMP1 were seen to vary in the whole sample between depression patients and control individuals or victims of suicide (in both cases $p<0.05$ by one-way ANOVA). Neither sex nor age showed any significant effect on the results. TRIOBP-1, previously only seen to aggregate in schizophrenia, was not seen as insoluble in suicide victims or depression patients. Based on this small-scale study, it, therefore, appears that species of CRMP1, DISC1, and NPAS3 may aggregate in the brains of a subset of victims of studies. Replication studies, as well as confirmation in additional brain regions are now required to examine this phenomenon in more detail and understand its pathological relevance.

Disclosures: B. Samardzija: None. **M. Palkovits:** None. **N.J. Bradshaw:** None.

Poster

P715. Other Psychiatric Disorders Including Schizoaffective and Eating Disorders

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P715.03

Topic: G.08. Other Psychiatric Disorders

Support: HRZZ (Croatian Science Foundation) IP-2018-01-9424
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European Regional Development Fund RC.2.2.06.0001

Title: Determination of the domain structure of DISC1 in mammalian cells identifies a region crucial for its aggregation in mental illness

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Abstract: To date, the pathophysiology of psychiatric disorders remains poorly understood. While traditionally investigated using genetics, disrupted protein homeostasis has been suggested as a biological basis for these diseases in at least a subset of patients. Several proteins previously implicated by genetic studies have been shown to form insoluble aggregates in such illnesses, including Disrupted in Schizophrenia 1 (DISC1). DISC1 is a multi-functional scaffolding protein of significant importance for neurodevelopment. However, delineation of the domain structure of DISC1 has only recently been attempted, using high-throughput analysis in *E. coli*. This approach identified four distinct stable domains named D, I, S and C. Here, we used this knowledge to confirm and refine the structure of DISC1 in mammalian cells, as well as to identify the region of DISC1 responsible for its aggregation. Firstly, we attempted to refine the borders of DISC1 by combining previously published bioinformatics data on DISC1 domains with the recent empirical data. Constructs encoding variants of domains D, I and C were cloned, with borders modified based on theoretical predictions. These were transfected into HEK293 cells and tested for stability via a proteasome inhibition assay. Results obtained by immunoblotting showed that modified versions of D and C domains show improved stability over their empirical counterparts, with evidence that the I region may not represent a stable folded domain by itself. The D, S and C domains were further shown to be functional in isolation, based on interaction with known protein binding partners. In order to investigate which structural region(s) might be responsible for its aggregation, fragments of DISC1 were expressed in SH-SY5Y neuroblastoma cells and their localization patterns viewed by fluorescent microscopy. Single domains exhibited clear cytoplasmic localization with no aggregate formation. In contrast, the combination of domains D and I showed clear signs of aggregation, with full length DISC1 used as a positive control. Based on this, we hypothesized that the unstructured region between these two domains, D and I, is responsible for DISC1 aggregation propensity, which was verified using further truncation constructs. With this work, we confirmed and refined the domain structure of DISC1, a known risk factor for mental disorders. Additionally, these results provided an insight into its aggregation as an important emerging aspect of DISC1 pathology. Together, our data will aid in generation of high-quality animal models to study the mechanism and behavioral consequences of DISC1 aggregation and its relevance for major mental illness.

Disclosures: B. Zaharija: None. **L. Sanchez-Pulido:** None. **N.J. Bradshaw:** None.

Poster

P715. Other Psychiatric Disorders Including Schizoaffective and Eating Disorders

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Program #/Poster #: P715.04

Topic: G.08. Other Psychiatric Disorders

Title: Understanding the Therapeutic Potential of Several Muscarinic M4 Selective Activators by Evaluating Their In Vivo Activity Across Different CNS Models

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Abstract: The M4 muscarinic acetylcholine receptor is a Gi-coupled G-protein coupled receptor (GPCR) preferentially expressed in the CNS, particularly in the striatum with more modest levels seen in the cortex and hippocampus. M4 receptors are expressed both presynaptically and postsynaptically. Presynaptic M4 receptors have been linked with negative modulation of various neurotransmitters including acetylcholine, glutamate and dopamine. The unique brain expression profile of the M4 receptor makes it a compelling target for addressing the symptoms associated with a number of different psychiatric diseases, specifically, recent clinical data have suggested significant potential in the treatment of psychosis associated with schizophrenia or Alzheimer's disease. The key hypothesis underlying the apparent antipsychotic effects of M4 receptors is a reduction in the striatal dopaminergic hyperactivity believed to drive the positive symptoms observed in both schizophrenia and potentially Alzheimer's disease. However, the current tool compounds have been limited with a lack of M4 selective molecules available. Here, we present in vivo data in several different behavioral and physiological assays comparing selective positive allosteric modulators, full agonists and partial agonists of the M4 receptor. All molecules were active in reducing amphetamine-stimulated hyperlocomor activity. However, only the full agonists showed a robust effect in conditioned avoidance responding. The results suggest that each mechanism of activation may provide a unique profile that could make it better suited to the treatment of the symptoms associated with a particular CNS disorder.

Disclosures: P. Iredale: None. **H.N. Nguyen:** None. **S. Chakilam:** None. **G. Suidan:** None.

Poster

P715. Other Psychiatric Disorders Including Schizoaffective and Eating Disorders

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Program #/Poster #: P715.05

Topic: G.08. Other Psychiatric Disorders

Title: Suvn-4404091, a multi-target compound for the treatment of schizophrenia and psychotic disorders

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Abstract: Schizophrenia is a chronic and severe mental disorder affecting 20 million people worldwide. It is characterized by distortions in thinking, perception, emotions, language, sense of self and behavior. Common experiences include hallucinations (hearing voices or seeing things that are not there) and delusions (fixed false beliefs). Schizophrenia is associated with considerable disability and may affect educational and occupational performance. Currently available treatments are marred with moderate efficacy and side effects. Approximately 20 to 33% of patients do not have a therapeutic response to existing treatments, and others have residual psychotic symptoms. Lack of therapeutic effects is also attributed to poor compliance due to side effects like extrapyramidal symptoms, sedation, weight gain, metabolic disturbances, and hyperprolactinemia. SUVN-4404091 is one of our lead compounds which targets dopaminergic pathways of schizophrenia via the D_2 receptors. Affinity for serotonergic receptors such as 5-HT_{2A} and 5-HT_{1A} were observed as well. SUVN-4404091 was found to be orally bioavailable with adequate brain penetration. SUVN-4404091 showed dose dependent occupancy at 5-HT_{1A}, 5-HT_{2A} and D_2 receptors with ED_{50} values of approximately 1 mg/kg, *p.o*. Dose dependent increase in cortical dopamine levels was observed in rats when tested at the therapeutically active doses. In an open field study, SUVN-4404091 attenuated amphetamine induced hyperlocomotion. No significant effect was observed on QTcF or blood pressure parameters (systolic, diastolic and mean arterial blood pressure) in beagle dogs. Wide separation was observed between the efficacy and extrapyramidal side effects. There were no off-target effects in preliminary toxicity studies in rats. SUVN-4404091 could be a promising treatment for the management of schizophrenia and psychotic disorders.

Disclosures: J. Thentu: A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **S. Petlu:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **V. Mekala:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **R. Subramanian:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **R. Abraham:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **N. Muddana:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **N. Guduru:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **A. Shinde:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **K. Bojja:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **P. Achanta:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd. **R. Nirogi:** A. Employment/Salary (full or part-time):; Suven Life Sciences Ltd..

Poster

P715. Other Psychiatric Disorders Including Schizoaffective and Eating Disorders

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P715.06

Topic: H.04. Executive Functions

Title: Upregulation of cyclophilin D contributes to mitochondrial dysfunction and oxidative stress in parvalbumin expressing interneurons in schizophrenia

Authors: *J. T. O'BRIEN[1], A. J. PHENSY[2], S. KROENER[1];
[1]Sch. of Behavioral and Brain Sci., Univ. of Texas at Dallas, Richardson, TX; [2]Dept. of Psychiatry, Univ. of California San Francisco, San Francisco, CA

Abstract: The cognitive symptoms of schizophrenia have been linked to functional deficits in parvalbumin expressing interneurons (PVI) in the frontal cortex and hippocampus. PVI are fast spiking GABAergic neurons which modulate large networks of pyramidal cells and serve as crucial regulators of the excitatory/inhibitory balance (E/I) in the prefrontal cortex (PFC). Reductions in parvalbumin (PV) expression represent a core finding in human postmortem studies and PVI impairments have been linked to dysfunctions in the corticolimbic system in schizophrenia. Aberrant PVI activity disrupts the brake on excitatory signaling, shifting the cortical E/I balance in favor on increased basal neural activity, leading to changes in PFC networks required for cognitive control.
While the precise etiology of these changes remains uncertain, redox dysregulation and oxidative stress may be a final common pathway in the pathophysiology of schizophrenia, causing dysfunction of GABAergic interneurons and loss of PV. Mitochondria are the major source of reactive oxygen species (ROS) in neurons, and they are both a pivotal contributor and amplifier of intracellular oxidative stress in pathological states. The mitochondrial matrix protein cyclophilin D (CypD) modulates the formation of the mitochondrial permeability transition pore (mPTP), which causes release of mitochondrial ROS, initiating a feed-forward cycle of ROS generation and release, metabolic changes, and eventual loss of cell function.
To test if CypD is upregulated in schizophrenia and may contribute to the loss of PV expression in the frontal cortex of patients with schizophrenia, we obtained human brain tissue from postmortem schizophrenic patients and healthy subjects via the NIH NeuroBioBank. We used multilabel immunofluorescent microscopy, paired with an unbiased stereological approach to quantify the expression of PV and CypD within PVI across layers 2/3 of Broadman Areas 9/46. Our results show an upregulation of CypD within PVI, suggesting that CypD-mediated oxidative stress contributes to PVI dysfunction in schizophrenia.
Upregulated levels of CypD and mitochondrial redox dysregulation may thus cause a loss of PVI function which contributes to frontal cortical dysfunction in schizophrenia. These results suggest new mechanisms for the pathophysiology of schizophrenia, as well as potential new therapeutic interventions that target CypD.

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Poster

P715. Other Psychiatric Disorders Including Schizoaffective and Eating Disorders

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Program #/Poster #: P715.07

Topic: G.08. Other Psychiatric Disorders

Support: Funded by Transpharmation Ltd., UK

Title: Efficacy of low doses neuroleptic drugs on aggressive behaviour and corticosterone levels in CD-1 male mice

Authors: *E. SOKOLOWSKA[1], E. STOJEK[2], D. KOZAREVA[2], J. PRENDERVILLE[2]; [1]Transpharmation Poland Sp. z o.o, Olsztyn, Poland; [2]Transpharmation Ireland Ltd., Dublin, Ireland

Abstract: Several psychiatric disorders are associated with increased aggression resulting in impairment in social skills. Current treatments which target aggressive behaviour can induce sedation, affect mobility and induce idleness. Therefore, a better understanding of the behavioural and neurobiological aspects of aggression is necessary. Here we investigate the effects of two low doses of antipsychotic drugs: risperidone and S15535 on aggressive behaviour in the CD-1 male mice. Risperidone is an atypical antipsychotic used to treat schizophrenia acting at several 5-HT (serotonin) receptor subtypes. S15535 is a highly selective 5-HT1A receptor ligand that acts as an agonist and antagonist at the pre- and post-synaptic 5-HT1A receptors, respectively. In addition, we are investigating if either drug affects levels of corticosterone (CORT), a hormone previously associated with aggression. Briefly, 10-13week old CD-1 mice underwent 3 days of aggression screening by introducing 6-7 weeks old C57BL/6J male intruder to their home cage for a maximum of 180s and latency to the first attack was recorded. Following, CD-1 aggression phenotype was evaluated, leading to selecting only the most aggressive subjects. The aggressive CD-1 mice were then treated with vehicle, risperidone (0.05, 0.5 and 3 mg/kg i.p.) or S15535 (0.5 and 2.5 mg/kg s.c.). Thirty minutes post-treatment mice underwent a single aggression screen followed by locomotor assessment in the open field. Corticosterone levels in the trunk blood were analysed in aggressive and non-aggressive mice using ELISA kit (Enzo, USA). In addition, the prefrontal cortex, amygdala and hypothalamus were collected for future molecular analyses. Only the lowest (0.05 mg/kg) dose of risperidone resulted in a significant reduction ($p<0.005$) of aggressive behaviour without affecting locomotion. The high (2.5 mg/kg) dose of S15535 significantly reduced aggression ($p<0.005$) with 50% of CD-1 mice abstaining from attack initiation and engaging in normal locomotor activity. Risperidone, but not S15535 or vehicle significantly decreased corticosterone levels ($p<0.05$) in the plasma of aggressive mice. CD-1 mice exhibiting non-aggressive phenotype presented the highest levels of CORT, suggesting that social interaction with another mouse during the screening paradigm can induce stress. Taken together, this study successfully demonstrates the aggression inhibiting the activity of both drugs, confirming that, modulation of the 5-HT1A receptors may represent a therapeutic target for aggressive behaviour.

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Poster

P715. Other Psychiatric Disorders Including Schizoaffective and Eating Disorders

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Program #/Poster #: P715.08

Topic: G.08. Other Psychiatric Disorders

Support: NIDA P30 DA048742-01A1

Title: Phenotypic subtyping in binge eating and associated neural correlates

Authors: *L. R. BRUCAR[1,5], E. RAWLS[1], G. DROSSEL[2], C. B. PETERSON[1], A. F. HAYNOS[1], T. HENDRICKSON[3], A. ZILVERSTAND[1,4];
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Abstract: Standard treatments for binge-type eating disorders are only moderately effective. To improve efficacy, a better understanding of the mechanisms contributing to the maintenance of binge eating (BE) is critical. We applied a data-driven dimensionality reduction approach to a community sample to develop an empirical model of BE behaviors, identify phenotypic subtypes and investigate their neural correlates. We hypothesized that three domains would be impaired in BE and revealed in the subtyping: reward-related processing, negative emotionality, and executive function. We applied exploratory factor analysis to extract phenotypic latent factors from the extensive measures in the enhanced NKI-Rockland Sample (N = 644; 66% F). Subjects were categorized into a BE (N=130, 64% F) or non-BE group (N = 423, 67% F) based on frequency of objective binge episodes (OBE, BE: >=1 OBE/28days; non-BE: 0 OBE). Within the BE group, subtypes were determined using latent profile analysis on the latent factors identified in the full sample. We investigated resting-state functional connectivity related to OBE within each subtype (nodal global and local efficiency, betweenness centrality; Brain Connectivity Toolbox; covariates: age, gender, race, BMI). Twelve latent factors were uncovered that grouped into the hypothesized functional domains, with the BE group showing impairments on all three domains. Subtyping within the BE group revealed a: (I) ‘Internalizing’ type (High Internalizing/General Psychiatric/Negative Affect/Risk Perception), (II) ‘Impulsive’ type (High Urgency, Lack of Cognitive Control, Low Risk Perception), and (III) ‘Risk-Averse’ type (High Risk Perception, Low Norm-Breaking, High Cognitive Control). The Internalizing Subtype exhibited abnormal connectivity in the frontoparietal network (mSPL). The Impulsive Subtype displayed abnormal connectivity in the frontoparietal (mSPL), default mode (sPCC), and visual 2 (V7/l-IPS) networks. The Risk-Averse Subtype demonstrated abnormal connectivity in the frontoparietal (dlPFC, FPC, aIPS), cingulo-opercular (dACC), default mode (mPFC), dorsal-attention (pIPS), somatomotor (aSPL), and ventral-attention (aIFS) networks, and the thalamus. All regions were significant at p-FDR < 0.05.These findings provide evidence for a three-domain model of BE and confirm the existence of substantial heterogeneity in the neurobehavioral

impairments underlying binge eating. Subtyping on functional domains will be important for modelling the vast heterogeneity seen in BE and investigating their clinical relevance and utility in developing more effective, targeted treatments.

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Poster

P715. Other Psychiatric Disorders Including Schizoaffective and Eating Disorders

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Program #/Poster #: P715.09

Topic: G.08. Other Psychiatric Disorders

Title: Clinical features correlated with comethylation modules in mexican teenagers with eating disorders

Authors: G. NOLASCO-ROSALES[1], J. MARTÍNEZ-MAGAÑA[2], ***I. JUAREZ-ROJOP**[3], C.-Z. TOVILLA-ZARATE[4], G. VILLAR-JUÁREZ[5], A. GENIS-MENDOZA[2], H. NICOLINI[2]; [1]Univ. Juarez Autonoma de Tabasco, Villahermosa, Tabasco, Mexico; [2]Inst. Nacional de Medicina Genómica (INMEGEN),, Ciudad de Mexico, Mexico; [3]Univ. Juárez Autonoma De Tabasco, Villahermosa, Tabasco, Mexico; [4]Univ. Juarez Autonoma de Tabasco. DAMC, Comalcalco, Tabasco, Mexico; [5]Escuela de Medicina. Univ. Anáhuac Querétaro, Querétaro, Querétaro, Mexico

Abstract: Eating disorders (ED) are psychiatric disorders characterized by disturbed eating behavior. They have a complex etiology, where genetic and environmental factors are interacting. Comethylation modules analysis could detect small effects of epigenetic interactions, reflecting environmental factors influences. The objective of this exploratory study was to create comethylation modules from children and teenagers' DNA methylation samples with an ED, and to correlate these modules with clinical features and genetic variability. We used MethylationEPIC microarray to obtain methylation levels from 63 teenagers with eating disorders. Evaluated features were age, gender, BMI zscore, psychiatric comorbidities and eating behaviours. This study followed principles from the Declaration of Helsinki. We created comethylation modules in WGCNA (Weighted Gene Correlation Network Analysis) and correlated them with clinical features in our subjects. Also, we performed enrichment analysis of CpG sites from comethylation modules. We observed two groups of comethylation modules given the methylation level from beta values. Meanwhile, one group had partially methylated values (turquoise, blue and purple); the other group had hypermethylated values (brown, yellow, green, red, black, pink and magenta). Seven clinical features and comorbidities correlated with different comethylation modules. The yellow comethylation module correlated with body mass index (BMI) zscore (R^2 = 0.47, p = 0.0006), conduct disorder (R^2 = -0.41, p = 0.0030), and

psychotic disorder (R^2 = -0.45, p = 0.0010). Meanwhile, purple comethylation module correlated with gender (R^2 = -1, p < 1e-50), suicide behavior (R^2 = 0.41, p = 0.0030), and attention-deficit/hyperactivity disorder (R^2 = -0.59, p = 6e-06); finally, black module correlated with height (R^2 = 0.4, p = 0.0040). We found significant enrichment pathways from the genes annotated on turquoise and blue modules. These modules were enriched for pathways that could be influenced by development status of the sampled individuals. Partial methylation of these modules suggests transcriptional activation of these pathways. Black module was non-significant enriched in morphine addiction synaptic vesicle cycle, apoptosis, and mTOR signaling pathway. Meanwhile, yellow module was non-significant enriched in alpha-Linolenic acid metabolism, VEGF signaling pathway, and hepatitis C. These data suggest that perturbations in DNA methylation could correlate with somatometric measures and comorbidities in individuals with ED.

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Poster

P715. Other Psychiatric Disorders Including Schizoaffective and Eating Disorders

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Program #/Poster #: P715.10

Topic: G.08. Other Psychiatric Disorders

Support: BPEndure Vulnerable Brain Project

Title: Is exercise addictive for patients diagnosed with anorexia nervosa: An anatomical exploration of the role of mu opioid receptors in the release of dopamine in nucleus accumbent of mice acclimated to the wheel with and without food restriction

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Abstract: Anorexia nervosa (AN) is one of the deadliest of all mental illnesses, even beating out depression. Characterized by a distorted self-image, food restriction and, in most cases, excessive exercise, these three features are the reasons why AN's morbidity rate, especially among adolescent girls, is so high. But how is it that we are seeing patients compulsively exercise when science and their physicians advise them to curtail exercise? This is the question that drives the current research. Previous research suggests that aspects of AN, like food restriction (FR) and exercise (EX), may be addictive but the studies are incomplete. We asked how the putatively addictive nature of EX and FR might be represented in the brain of individuals diagnosed with

AN. We hypothesized that addiction to EX among AN patient may be due to the activation of mu opioid receptors (MOR), causing inhibition of the GABAergic neurons in the nucleus accumbens (NAcb), a key brain structure affected in addiction. This would enhance the release of dopamine by the process of disinhibition that enhances firing of the dopaminergic cell bodies in the ventral tegmental area. Alternatively, MOR might modulate dopamine release more directly through their expression on dopaminergic axons in NAcb. We have begun to use the mouse model of AN, called activity-based anorexia (ABA), to examine neuroanatomical substrates supporting these modes of action of MOR while also asking whether animals exhibiting greater EX express greater prevalence of MOR and if so, whether this is specific to animals undergoing ABA or might also be a trend observed among mice acclimated to the wheel without ABA induction (EX only). To this end, we have generated four groups of mice: food restricted and exercising (ABA), EX only, FR only, and control (no EX and no FR). Electron microscopic immunocytochemistry (EM-ICC) was employed to detect MOR and tyrosine hydroxylase, the rate-limiting synthetic enzyme present in dopaminergic axons in NAcb. Correlational analyses between the behaviors of EX and FR and the anatomical features MOR expression relative to dopaminergic neurons promise to provide us with an insight about the addictive nature of excessive EX and FR in ABA and pharmacological treatments to ameliorate these putatively addictive behaviors in AN. Preliminary data (N=2, >100 dopaminergic axon terminals) indicate that MOR are rarely expressed on dopaminergic axons in NAcb shell, favoring the alternative mechanism involving MOR's role in disinhibition.

Disclosures: I. Pat-Osagie: None. **C. Aoki:** None.

Poster

P720. Addiction: Genetics, Translational, and Clinical Studies

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Program #/Poster #: P720.01

Topic: G.09. Drugs of Abuse and Addiction

Support: Internal grants from CNU

Title: Ethanol behaviors were unaffected by heterozygous deletion of Cdc42bpb in mice

Authors: K. E. KITCHEN, A. T. LIMAYE, T. N. FOREMAN, *J. BOGENPOHL;
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Abstract: *Cdc42bpb* encodes a kinase (MRCKβ) that regulates cytoskeleton reorganization and cell migration, which functions downstream of the important Rho GTPase, CDC42. In humans, heterozygous loss of function mutations in *Cdc42bpb* result in a neurodevelopmental disorder reminiscent of the CDC42-associated Takenouchi-Kosaki syndrome. Importantly, *Cdc42bpb* was also recently discovered to be involved in the genomic response to chronic ethanol exposure in

the prefrontal cortex of monkeys and mice. As such, we have conducted behavioral experiments in *Cdc42bpb*+/- mice and wildtype littermates, aimed at characterizing the ethanol phenotypes produced by the deletion. Experimental mice underwent three ethanol-related tests, including the loss of righting reflex test, a light-dark box test, and a two-bottle choice drinking study. Results showed that heterozygous deletion of *Cdc42bpb* did not alter ethanol behaviors, as comparted to wildtype littermates. Other phenotypes included mild hyperactivity and an increased incidence of prolapsed penis, in line with genitourinary defects reported in *Cdc42bpb*+/- humans.

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Poster

P720. Addiction: Genetics, Translational, and Clinical Studies

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Topic: G.09. Drugs of Abuse and Addiction

Support: U01 DA039168
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T32 GM008541

Title: A reduced complexity cross between BALB/c substrains identifies Zhx2 is a candidate gene underlying oxycodone metabolite brain concentration and state-dependent learning of opioid reward.

Authors: *J. BEIERLE[1], E. YAO[1], S. GOLDSTEIN[1], J. SCOTELLARO[1], O. AVERIN[3], D. MOODY[3], A. EMILI[2], G. PELTZ[4], M. FERRIS[5], C. D. BRYANT[1];
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Abstract: Understanding the pharmacokinetic profile of an opioid drug is vital to therapeutic success, and mutations in human PK genes can alter therapeutic efficacy as well as addiction liability. Oxycodone is a semisynthetic opioid metabolized into noroxycodone and oxymorphone by CYP450 enzymes. We observed that 30 min post-oxycodone administration (1.25 mg/kg, i.p.) BALB/cJ mice, in particular females, showed higher whole brain concentrations of oxycodone, noroxycodone, and oxymorphone compared to BALB/cByJ that could explain increased oxycodone state-dependent conditioned place preference in BALB/cJ mice. To potentially link behavioral differences with PK differences, we conducted quantitative trait locus (**QTL**) mapping of whole brain oxycodone and metabolite concentrations in a reduced complexity cross

(**RCC**). Because BALB/cJ and BALB/cByJ substrains differ by ~10,000 SNPs/indels, large genetic loci identified in an F2 cross are offset by dramatic reduction in potentially causal variants. QTL mapping in 133 BALB/cJ x BALB/cByJ F2 mice (68F, 65M) revealed a single QTL on chromosome 15 associated with brain [oxymorphone]. Oxymorphone is a full agonist at the mu opioid receptor, with 8x the potency of oxycodone, and likely contributes to oxycodone's addictive properties. Within our interval, hippocampal and striatal cis-eQTL analysis revealed differential expression of Zhx2, a transcriptional inhibitor known to harbor a private BALB/cJ retroviral insertion that dramatically reduces protein expression and subsequent sex specific dysregulation of CYP450 genes within the liver. Whole brain proteomics in parental strain mice corroborated these RNA findings. We hypothesize disruption in Zhx2 expression alters CYP450 expression, and therefore alters whole brain oxymorphone and oxycodone-induced behaviors.

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Poster

P720. Addiction: Genetics, Translational, and Clinical Studies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P720.03

Topic: G.09. Drugs of Abuse and Addiction

Support: NIDA Grant 5DP1DA046587 (JF)

Title: Effects of cocaine use on three-dimensional chromatin organization in medium spiny neurons of the mouse nucleus accumbens

Authors: *J. M. CHITAMAN[1,2], R. S. JASROTIA[1,2], V. MALYSHEVA[3], H. XU[1,2], Y. LI[1,2], M. SPIVAKOV[3], P. FRASER[1], J. FENG[1,2];
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Abstract: Drugs of abuse alter the epigenetic landscape in the nucleus accumbens (NAc), a key region in the brain reward pathway, which underlies aberrant gene expression associated with maladaptive synaptic plasticity in drug addiction. In the eukaryotic nucleus, two meters of linear DNA require extensive three-dimensional (3D) folding to fit a micrometer-sized nuclear space. This 3D folding of chromosomes is highly organized and provides higher-order layers of epigenetic regulation that bring distal (i.e., up to a megabase away) regulatory elements within close spatial proximity of target genes. However, such 3D chromatin organization and its potential impact on gene expression remain obscure in drug addiction. In this study, we characterize 3D chromatin interactions in D1- and D2-type medium spiny neurons (MSNs),

respectively, of adult male mice after intravenous cocaine self-administration. D1- and D2-MSNs account for ~95% of neurons within the NAc, but are known to play distinct roles in drug addiction. Using next-generation sequencing approaches including genome-wide and promoter capture Hi-C, we identify hierarchical chromatin structures such as active/inactive compartments, topologically associated domains (TADs), subTADs, and DNA loops. We observe dysregulation of chromatin architecture by cocaine use, particularly in the promoter interactome. Furthermore, we find that cocaine-altered 3D interactions predominantly occur at the submegabase level and contain promoter regions of numerous genes that are differentially expressed in cocaine addiction. Therefore, our study elucidates features of 3D chromatin organization at various scales in NAc neuronal subtypes in cocaine addiction. Our research also provides a framework to explore novel molecular mechanisms of cocaine action in the brain that have the potential to serve as improved diagnostic and therapeutic targets in the future.

Disclosures: J.M. Chitaman: None. **R.S. Jasrotia:** None. **V. Malysheva:** None. **H. Xu:** None. **Y. Li:** None. **M. Spivakov:** None. **P. Fraser:** None. **J. Feng:** None.

Poster

P720. Addiction: Genetics, Translational, and Clinical Studies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P720.04

Topic: G.09. Drugs of Abuse and Addiction

Support: R01 AA023489

Title: Brain-specific inhibition of mTORC1 by a dual drug strategy: implications for the treatment of alcohol use disorder

Authors: *Y. EHINGER[1], Z. ZHANG[2], K. PHAMLUONG[1], D. SONEJA[1], K. SHOKAT[2], D. RON[1];
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Abstract: Brain-specific inhibition of mTORC1 by a dual drug strategy: mplications for the treatment of alcohol use disorder Yann Ehinger[1], Ziyang Zhang[2], Khanhky Phamluong[1], Drishti Soneja[1], Kevan Shokat[2], Dorit Ron[11]Department of Neurology, [2]Department of Cellular and Molecular Pharmacology, University of California San Francisco
The mechanistic target of rapamycin complex 1 (mTORC1) plays an important role in mechanisms underlying numerous alcohol-dependent adverse phenotypes such as excessive alcohol use, alcohol seeking, habit, and relapse (Ron and Barak, 2016; Morisot et al., 2019). Therefore, mTORC1 represents a promising therapeutic target for the treatment of alcohol use disorder (AUD). mTORC1 inhibitors are used in the clinic to prevent organ rejection after transplantation. However, chronic inhibition of mTORC1 in the periphery results in undesirable

side effects which reduce the potential use of the inhibitors for the treatment of AUD. To overcome these limitations, we developed a novel approach to selectively inhibit mTORC1 in the brain but not in the periphery (Zhang Z. et al., 2020 bioRxiv). Specifically, we developed a small molecule (Rapablock) which protects mTORC1 activity in the periphery when administered together with the mTORC1 inhibitor Rapalink-1. We demonstrate that a combined treatment of Rapalink-1 and Rapablock in mice enables the selective inhibition of mTORC1 in the brain, while protecting mTORC1 activity in the liver. We further show that whereas chronic inhibition of mice with Rapalink-1 produces sides effects such as body weight loss, glucose intolerance and liver toxicity, co-administration of the two drugs attenuated these adverse effects. Furthermore, we report that Rapalink-1 inhibited alcohol-dependent activation of mTORC1 in the nucleus accumbens (NAc) in the presence of Rapablock. Finally, we show that the co-administration of Rapablock and Rapalink-1 moderates
alcohol drinking and seeking in mice. Together, these data put forward a promising new approach for the treatment of AUD.
ReferencesRon D. and Barak S. *Nat Reviews Neurosci*. 2016.Morisot N. et al. *Elife,* 2019.Zhang Z. et al., *bioRxiv* 2020
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Poster

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Program #/Poster #: P720.05

Topic: G.09. Drugs of Abuse and Addiction

Support: NIDA R00 DA043573
Dunavant Professorship

Title: Strain- and sex-dependent effects of isolation housing relative to environmental enrichment on operant sensation seeking in mice

Authors: *P. E. DICKSON[1], G. MITTLEMAN[2];
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Abstract: Sensation seeking is a multidimensional phenotype that predicts the development of drug addiction in humans and addiction-like drug seeking in rodents. Several lines of evidence suggest that chronic stress increases sensation seeking and addiction-like drug seeking through common genetic mechanisms. Discovery and characterization of these mechanisms would reveal

how chronic stress interacts with the genome to influence sensation seeking and how drugs of abuse hijack these fundamental reward mechanisms to drive addiction. To this end, we tested the hypothesis that chronic isolation housing stress (relative to environmental enrichment) influences operant sensation seeking as a function of strain, sex, or their interaction. To determine if the BXD recombinant inbred panel could be used to identify genetic mechanisms underlying any identified gene-by-environment interactions, we used mice from the two BXD founder strains. Following 10 weeks of differential housing, we assessed operant sensation seeking using several reinforcement schedules. The primary finding from this study was that DBA/2J but not C57BL/6J mice were significantly vulnerable to an isolation-induced increase in sensation seeking during extinction when the sensory reward was no longer available; this effect was significantly more robust in females. These data reveal a previously unknown isolation-induced effect on extinction of operant sensation seeking that is sex-dependent, addiction-relevant, and that can be dissected using the BXD recombinant inbred panel.

Disclosures: P.E. Dickson: None. **G. Mittleman:** None.

Poster

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Program #/Poster #: P720.06

Topic: G.09. Drugs of Abuse and Addiction

Support: AA13484
AA10760

Title: Effects of Selection for Ethanol Preference on Gene Co-Splicing Networks across Multiple Brain Regions of Heterogeneous Stock Mice

Authors: A. COLVILLE[1], O. IANCU[1], ***D. LOCKWOOD**[2], P. DARAKJIAN[1], S. MCWEENEY[1], R. SEARLES[1], C. ZHENG[1], R. J. HITZEMANN[3];
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Abstract: Colville et al (2017) observed that selection for ethanol preference from HS-CC founders had marked effects on alternative splicing in the nucleus accumbens shell (SH); GTPase related genes were especially affected. The current study expands this observation by including larger sample sizes and two additional components of the addiction network, the central nucleus of the amygdala (CeA) and the prelimbic cortex (PL). The data illustrate that from the perspective of CoSplicEx networks (Iancu et al. 2015), preference selection had its greatest effect on the CeA. Across regions, significant effects were detected for genes associated with GTPase activity and synaptic genes. Some of the synaptic genes were hub nodes in the

CoSplicEx network that had been previously identified as associated with chronic ethanol consumption and included *Nbea* and *Sv2a,* genes previously identified in non-human primates.

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Poster

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Program #/Poster #: P720.07

Topic: G.09. Drugs of Abuse and Addiction

Support: R21DA052101

Title: Epigenome editing of nucleus accumbens cell subtype transcripts regulated by fentanyl abstinence

Authors: *M. TURNER[1,2], E. CHOI[1,2], R. CHANDRA[3,1], S. A. AMENT[4,5], M. E. FOX[1,2], M. LOBO[1,2];
[2]Anat. and Neurobio., [3]Virus Vector Core, [4]Inst. for Genome Sci., [5]Dept. of Psychiatry, [1]Univ. of Maryland Sch. of Med., Baltimore, MD

Abstract: In recent years, the opioid use epidemic has significantly impacted the financial and societal health burdens around the world, particularly the United States. This is in part is due to the ease at which synthetic opioids like fentanyl are obtained resulting in an increase in opioid overdoses. Many studies indicate that dysfunctionality in the mesocorticolimbic brain areas are central to understanding opioid use, dependence, and addiction. The nucleus accumbens (NAc) is a critical brain hub for altered molecular processes mediating behavioral responses to opioids and other used substances. Therefore, understanding the molecular and transcriptional programs of the neuronal subtypes within the NAc deserve considerable and critical attention. Our lab has performed cell subtype specific translatome profiling in the two NAc projection neuron subtypes- dopamine receptor 1 and 2 expressing medium spiny neurons (D1- and D2-MSNs) after prolonged abstinence from repeated fentanyl exposure. We identified distinct MSN subtype gene expression networks that are altered during fentanyl abstinence including hub genes genes, which are key drivers of gene expression alterations in MSN subtype specific modules. To build on this, we are developing CRISPR epigenome editing tools that will target specific hub genes in MSN subtype modules that are significantly regulated by fentanyl abstinence- 5 days 10 μg/ml fentanyl followed by 10 days of abstinence. Since most genes are down-regulated we use CRISPRa (activation) to upregulate expression of hub genes in MSN subtypes. Using a two vector adenoassociated virus (AAV) system we designed single gRNA or multiplex gRNAs, the latter targeting multiple hub genes within one module. gRNAs targeting hub genes or a lacZ

control gRNA are cloned into an AAV-gRNA-nlsGFP, which allows expression of nuclear GFP. The second vector is an AAV-DIO-dCas9-VP64 containing the nuclease dead Cas9 fused the VP64 transcriptional activation domain. We have transfected these vectors along with Cre into Neuro2A cells and demonstrate upregulation of MSN subtype fentanyl abstinence regulated hub genes. We are currently packaging these vectors into AAVs to target NAc MSN subtypes, using D1-Cre and A2A-Cre mice, during opioid exposure and abstinence to determine if upregulating hub genes can influence MSN subtype/fentanyl abstinence gene expression networks.

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Poster

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National Science Foundation Graduate Student Research Fellowship DGE1745016
Carnegie Mellon Brainhub Presidential Fellowship
Carnegie Mellon Computational Biology Department Lane Postdoctoral Fellowship

Title: Addiction-associated genetic variants implicate brain cell type- and region-specific cis-regulatory elements in addiction neurobiology

Authors: *C. SRINIVASAN[1], B. N. PHAN[1,4], A. J. LAWLER[2], E. RAMAMURTHY[1], M. KLEYMAN[1], A. R. BROWN[1], I. M. KAPLOW[1,3], M. E. WIRTHLIN[1,3], A. R. PFENNING[1,2,3]; [1]Computat. Biol. Dept., [2]Dept. of Biol. Sci., [3]Neurosci. Inst., Carnegie Mellon Univ., Pittsburgh, PA; [4]Med. Scientist Training Program, Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Recent large genome-wide association studies (GWAS) have identified multiple confident risk loci linked to addiction-associated behavioral traits. Most genetic variants linked to addiction-associated traits lie in non-coding regions of the genome, likely disrupting cis-regulatory element (CRE) function. CREs tend to be highly cell type-specific and may contribute to the functional development of the neural circuits underlying addiction. Yet, a systematic

approach for predicting the impact of risk variants on the CREs of specific cell populations is lacking. To dissect the cell types and brain regions underlying addiction-associated traits, we applied stratified LD score regression to compare GWAS to genomic regions collected from human and mouse assays for open chromatin, which is associated with CRE activity. We found enrichment of addiction-associated variants in putative CREs marked by open chromatin in neuronal (NeuN+) nuclei collected from multiple prefrontal cortical areas and striatal regions known to play major roles in reward and addiction. To further dissect the cell type-specific basis of addiction-associated traits, we also identified enrichments in human orthologs of open chromatin regions of female and male mouse neuronal subtypes: cortical excitatory, D1, D2, and PV. Lastly, we developed machine learning models to predict mouse cell type-specific open chromatin, enabling us to further categorize human NeuN+ open chromatin regions into cortical excitatory or striatal D1 and D2 neurons and predict the functional impact of addiction-associated genetic variants. Our results suggest that different neuronal subtypes within the reward system play distinct roles in the variety of traits that contribute to addiction.

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Poster

P720. Addiction: Genetics, Translational, and Clinical Studies

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Program #/Poster #: P720.09

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH 1ZIAAA000301-22

Title: COMT val^{158}met (rs4680) genotype influences gray matter, caudate, and thalamic volume and affects Addictions Neuroclinical Assessment incentive salience factor scores

Authors: *D. G. SUCICH[1,4], C. A. HODGKINSON[1], Q. YUAN[1], P.-H. SHEN[1], M. KERICH[2], R. MOMENAN[2,3], D. GOLDMAN[1,3];
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Abstract: The addictions are genetically heterogeneous, chronic relapsing disorders. Ignoring the diversity of etiology within DSM diagnoses for addictions has hindered progress determining the full contribution made by genetics to their development. Directly addressing this limitation, the Addictions Neuroclinical Assessment (ANA) utilizes a variety of quantitative measurements that have been constructed into neurofunctional domains believed to underlie the three stages of the addiction cycle—incentive salience (binge intoxication), negative emotionality (withdrawal/negative affect), and executive function (anticipation/preoccupation). The common human COMT Val158Met variant results in altered rates of monoamine catabolism, a risk factor for substance use disorders. In this exploratory study, genotypic effects of the COMT val158met (rs4680) polymorphism were assessed on the quantitative ANA neurofunctional domain factor scores and brain structure using MRI in 233 individuals (male=134, female=99). Only the volumes of the left side of brain regions were investigated in this preliminary study. Given previous evidence of the importance of COMT in cognitional tasks, such as attention and emotional processing, it was surprising to not see genotypic effects on executive function or negative emotionality factor scores. Individuals homozygous for the lower activity Met allele showed higher incentive salience factor scores, indicative of maladapted reward system function [$F(2,230) = 3.336$, $p = 0.0373$]. Met/Met carriers also presented with higher brain volumes of gray matter [$F(2,230) = 5.629$, $p = 0.0041$], left caudate [$F(2,230) = 4.08$, $p = 0.0181$], and left thalamus [$F(2, 230) = 3.739$, $p = 0.0252$]. High innervation from monoaminergic regions to the paraventricular thalamus (PVT), known to mediate attention to salient stimuli, may explain the effect of altered monoaminergic catabolism by the COMT Val158Met genotype on thalamic volume. Further investigation into monoamine catabolism in the thalamus, sex dependent effects, population admixture, as well as interactions with childhood trauma are warranted.

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Poster

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Topic: G.09. Drugs of Abuse and Addiction

Support: NIH NIDA Grant DP1DA046585
NIH NIDA Grant F30DA053020
NIH NIMH Grant UG3MH120094

Title: Fine-mapping candidate addiction-associated regulatory variants using cell type-aware comparative genomics across mammalian genomes

Authors: *B. N. PHAN[1,3], A. LAWLER[1,2], J. HE[4], I. M. KAPLOW[1], D. SCHAFFER[1], A. BROWN[1], W. R. STAUFFER[4], A. R. PFENNING[1];
[1]Computat. Biol., [2]Biol., Carnegie Mellon Univ., Pittsburgh, PA; [3]Med. Scientist Training Program, [4]Neurobio., Univ. of Pittsburgh, Pittsburgh, PA

Abstract: Hundreds of loci with thousands of genetic variants have been associated with addiction-associated behaviors, which are largely thought to act at distal regulatory elements and in tissue- and cell type-specific contexts. Measures of genome conservation have proven to be invaluable tools to prioritize which candidate causal genetic variants. Although powerful, these measures currently don't account for the highly cell type-specific nature of regulatory elements nor propose how to test causal effects of genetic variants in closely and distantly related model organisms. Here, we leverage large cross-mammalian genome constraint measurements and machine learning predictions of neural cell type-specific regulatory elements activity to improve the prioritization of genetic variants associated with addiction-associated and other neuropsychiatric traits. We matched 8 orthologous cell types in the caudate nucleus across human, rhesus macaque, and mouse with single nucleus or cre-specific Sun1Gfp nuclear-anchored independent labeling (cSNAIL) open chromatin assays. We trained convolutional neural network models on active open chromatin in 3 species. We apply these models, the Cell Type-Activity Conservation Inference Toolkit (CellTACIT), to predict the open chromatin activity of orthologous conserved DNA of 240 mammals. We interpreted a wide set of human genetic variants across 62 genome-wide association studies spanning addiction-associated traits, other neuropsychiatric, and non-brain traits. We show that chromatin regions that can be mapped to increasingly distant mammals (e.g. primates, mouse, bats) and are predicted by CellTACIT to remain active show increasing heritability enrichment, up to 2- and 10-fold above human open chromatin and mammalian conservation, respectively. Furthermore, we fine-mapped loci dense with genetic variants for addiction- and sleep-associated traits to identify candidate causal variants overlapping human orthologs of D1 and D2 medium spiny open chromatin regions measured in macaque and mouse and predicted active across mammalia. Lastly, we report evidence of mammalian trait diveristy (e.g. brain size) that correlate with predicted cell type-specific open chromatin activity in regions with fine-mapped human genetic variants of related traits. Mapping the conserved cell-type regulatory patterns in the human genome refines the list of candidate causal variants in regions associated with addiction-associated traits beyond conservation or human cell type-specific open chromatin alone, illustrating the power of combining epigenomic and evolutionary information in human polygenic trait studies.

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Poster

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Support: Korea Healthcare Technology R&D Project, Ministry for Health and Welfare, Republic of Korea (HM14C2603)
the National Research Foundation of Korea (Grant No. 2014M3C7A1062894)

Title: Relationship between harm avoidance and problematic smartphone use in children and adolescents: role of stress from the longitudinal perspectives

Authors: *C.-H. CHOI[1], S. CHUNG[2], S.-Y. BHANG[3], J.-W. KIM[4], Y.-S. KWEON[5], J.-S. CHOI[1];
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Abstract: The use of smartphones in children and adolescents might increase the level of stress and problematic smartphone use (PSU). In this study, the relationships between harm avoidance (HA) personality trait, stress, and PSU in children and adolescents was investigated from the perspective of longitudinal changes. At baseline, all participants (184 children and adolescents, 46 girls; mean age 13.15 years) completed screening questionnaires on PSU using the smartphone addiction scale-short version (SAS-SV) and the Korean smartphone addiction proneness scale (SAPS). The daily hassles questionnaire (DHQ) and junior temperament and character inventory (JTCI) were also administered to evaluate stress levels and personality traits. SAS-SV, SAPS, and DHQ were reassessed at 3 and 6 months. Pearson correlation analysis was used to examine the relationships between variables. Further, mediation effects were assessed using the PROCESS macro with 5,000 bias-corrected bootstrap samples. Among the temperament dimensions of JTCI, only HA displayed consistent positive correlations with SAS-SV, SAPS, and DHQ at all time points. Mediation effects of baseline stress on the relationship between HA and PSU were observed at baseline, 3, and 6 months ($\beta = 0.08$, $\beta = 0.07$ at baseline, $\beta = 0.06$, $\beta = 0.04$ at 3 months, $\beta = 0.09$, $\beta = 0.08$ at 6 months in SAS-SV and SAPS, respectively). Stress levels at 3 and 6 months also showed mediation effects on the relationship between HA and PSU at each time point. Our results suggest that managing stress may reduce the risk of PSU in children and adolescents with high HA.

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Poster

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Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant P30 DA048742-01A1
NIH Grant 5T32DA007234-29

Title: Subtypes in Addiction and their Neurobehavioral Impairments in Approach Behavior, Negative Emotionality and Executive Function

Authors: *G. L. DROSSEL[1], L. R. BRUCAR[2], E. L. RAWLS[2], T. HENDRICKSON[3], A. ZILVERSTAND[2,4,5];
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Abstract: Background: We propose that impairments on three independent functional domains are linked to addiction: 1) approach-related behavior, 2) negative emotionality and 3) executive function. While it is generally accepted that these three mechanisms are linked to substance use disorder (SUD), it has not been investigated if they are independent, which would warrant a subtyping and personalized medicine approach. We hypothesized the existence of at least three distinct subtypes: "reward users" with deviant approach-related behavior, "relief users" with high negative emotionality, and "low functioning users" with impaired executive function.
Methods: To describe the complete phenotypic space, we derived latent factors from the psychosocial and behavioral data from the NKI-Rockland Enhanced sample (*N = 644, 66% F; 54% F within SUD*). Subtypes were determined using clustering via latent profile analysis on these latent factors within those with past SUD (*N = 173, 55% F*). Graph theory-based resting-state connectivity analyses (Brain Connectivity Toolbox) were conducted for each subtype in a subsample with complete fMRI and NIDA Quick Screen data (*past SUD N = 104, 58.6% F; controls N = 302, 73.5% F*). Differences in nodal global efficiency, local efficiency, and betweenness were measured for each subtype compared to matched controls using regression analysis.
Results: The latent factor analysis revealed the hypothesized phenotypic domains. Three subtypes existed among those with past SUD: 1) Reward Users (high sensation seeking/social risk taking/norm-breaking), 2) Relief Users (high internalizing/general psychiatric symptoms/negative affect, lack of perseverance), and 3) Lower Functioning Users (low openness/sensitivity, high general psychiatric symptoms/sensation-seeking). Reward users demonstrated decreased connectivity (betweenness) for the supplementary motor network (SCEF) compared to controls, when predicting on lifetime number of drugs used while covarying for past 6-month tobacco use, and comorbidities ($p_{FDR} < 0.05$). Relief users had increased connectivity (betweenness) for the attention network (7P, VIP, MIP, 6a) compared to controls, when predicting on current number of drugs used ($p_{FDR} < 0.05$). This remained significant after covarying for comorbidities.
Conclusion: These empirical findings provide evidence for the three-domain model of addiction

and the existence of at least three distinct “addiction subtypes” with differential neurobehavioral profiles. This warrants future research into the clinical relevance of these subtypes and their utility in developing personalized medicine approaches.

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Poster

P768. Attention: Human Studies

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Program #/Poster #: P768.01

Topic: H.01. Attention

Support: National Institute of General Medical Sciences, (NIGMS), P20 GM103429 from the National Institutes of Health.

Title: Effect of transcutaneous vagus nerve stimulation on EEG alpha power.

Authors: J. MARTINEZ[1], R. STERN[1], C. BRIGGLER[1], M. S. MENNEMEIER[2], ***R. ROOSEVELT**[1];
[1]Arkansas Tech. Univ., Russellville, AR; [2]Neurobio. and Developmental Sci., Univ. of Arkansas for Med. Sci., Little Rock, AR

Abstract: Invasive vagus nerve stimulation (VNS) is an FDA treatment for refractory epilepsy and depression with potential application for the treatment of a host of CNS disorders including Alzheimer Disease, attention deficit, stroke, traumatic brain injury. However, the usefulness of VNS is limited to its highly invasive nature. More recently, the observation that auricle branch of the vagus may be stimulated transcutanously (TVNS) without invasive procedures has led to increased interest in vagal stimulation with multiple published studies of cognitive effects. Despite enthusiasm for TVNS, the effectiveness of TVNS has not been sufficiently demonstrated, nor have agreed upon measures of effective stimulation been established. The noradrenergic hypothesis of VNS posits that many of the desired effects of VNS are the result of the release of norepinephrine in the terminal fibers of Locus Coeruleus in the cortex. Considerable evidence supports this view including direct measurement of NE increases following VNS in the rat. We have previously reported here that TVNS in humans alters heart rate variability, increases salivary alpha amylase, and alters pupil dilation. Here we report tentative findings that TVNS suppresses alpha power in humans, a finding consistent with increased release of NE. EEG recordings were obtained from **twelve** participants for analysis. A 10 minutes baseline period and 10 minutes of TVNS were recorded. The TVNS block consisted to 14 seconds of stimulation each minute (0.5mA, 30 HZ). EEG for each block was then decomposed into power bands using FFT and percent change scores between baseline and

stimulation computed. RESULTS: We observed decreased alpha power associated with TVNS consistent with increased NE release at the terminal synapses in the cortex. Overall all decrease in alpha across all channels was 10%. 85% of channels displayed decreased alpha and 15% increased alpha. Thus, these findings support the hypothesis that TVNS is sufficient to alter CNS activity and that TVNS may exert effect in accordance with the noradrenergic hypothesis. Importantly, these findings also suggest that alpha power may serve as a biomarker of effective TVNS

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Poster

P768. Attention: Human Studies

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Topic: H.01. Attention

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Title: Brain network causal interactions underlying task-evoked pupil response: a simultaneous pupillometry, EEG, fMRI study

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Abstract: Pupil size not only changes in response to lighting conditions, but also can be modulated by cognition and mental processes, such as effort, arousal, attention, fluid intelligence, and cognitive states. Studies have shown that pupil changes are related to the locus coeruleus (LC) norepinephrine (NE) system, but the cortical-subcortical interactions and the interactions between large-scale brain networks underlying the pupil fluctuations are not well studied. In this study, we used simultaneously acquired pupillometry, EEG and fMRI and an auditory oddball task to study the networks' causal interactions underlying task-evoked pupil response (PR). After data preprocessing, we first employed a single-trial EEG analysis and a general linear model EEG-informed fMRI analysis, which reveals the neural cascade underlying the salient stimuli processing. With the identified neural correlates, the peak voxels of the statistically significant clusters were used to define network nodes, and these regions were fed into an EEG-based state-space model to infer the effective connectivity (EC) between them. With the fMRI-based functional connectivity, we observed an intrinsic organization (early,

middle and late time brain networks) of these nodes. Then, we summarized the EC patterns by the sum of all positive connections within or between networks based on this functional organization of the grouped nodes. We observed a significant ($r = 0.6352$; $p < 0.0035$) correlation between oddball-evoked PR and oddball-modulated late-to-early EC strength. This result aligns with the network-reset theory of LC. The LC phasic activity (indexed by PR) seems to be related to a late-to-early feedback signal from the late network to the early target network (subsystem of dorsal attention network, DAN), which reflects the reconfiguration of DAN to disengage from the task and adapt into the new environment after the response. Our results suggest that LC might modulate pupil changes through large-scale brain networks of the cortex, reflecting a NE-dependent brain-wide network reorganization.

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Poster

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Program #/Poster #: P768.03

Topic: H.01. Attention

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Title: The effect of task-irrelevant semantic information during salient attentional capture

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Abstract: Attention is essential for efficient focusing of cognitive resources in order to accomplish tasks at hand. While it has been shown that attention is constrained by task-relevant factors, recently it has been shown that attention is also influenced by a variety of task-irrelevant factors. A previous study shows that semantic relationships, even when task-irrelevant, bias attention during visual search (Malcolm et al., 2016). In order to elucidate the constraints on task-irrelevant semantic contribution to attentional guidance, we asked a question of whether semantic information influences voluntary shifts of attention as well as automatic (capture) shifts of attention. In the current human behavioral experiment adapted from the Posner cueing paradigm, participants viewed four objects presented around a central fixation, two of which formed a semantic pair (e.g., a cucumber and a carrot). After a variable object exposure delay, a bright circle, serving as a salient cue, flashed around one object of the semantic pair for 100 ms. After either a short (100 ms) or long (750 ms) delay, a target (T or L) and three distractors (T/L hybrids) appeared each on the center of the four objects. The task was to discriminate the target

as T or L. The location of the target was coded as Valid (same object as the cued object), SR (different but semantically-related object), and NR (non-related object). We analyzed participants' reaction time (RT) and accuracy. A validity effect was observed for both short and long post-cue delay conditions, with faster RTs to targets that appeared in the validly cued locations. In addition, when the post-cue delay was short, RT on SR trials was shorter than that on NR trials, showing that semantic information biased attention towards the related object. Most interestingly, this semantic bias disappeared when the post-cue delay was long. We propose that this lack of semantic effect during long delay may be a result of inhibition of return. The results indicated that semantic information influences attention allocation together with other attentional factors, contributing to the same spatial priority maps. The fact that sematic effect was sensitive to delay while validity effect did not, indicates that semantic effect and salience effect on attention might have different temporal profiles.

Disclosures: X. Zhang: None. **S.S. Shomstein:** None.

Poster

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Title: Stimulus Invariant Attentional Responses In The Human Temporal Cortex

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Abstract: The role of parietal and frontal cortices in the control of visual attention in humans and macaques is well-known. Recent studies in macaques identified a temporal cortical region involved in the control of visual attention independent of the type of visual stimulation. This temporal cortical region is part of a network including the superior colliculus (SC) in the control of visual attention. In humans, a homolog of this attention-related temporal cortical region has not been identified until now. We set out to locate such an attention-related region in the human temporal cortex using visual stimuli that identified the respective region in macaques, i.e., motion (MS) and dynamic white-noise stimuli (WNS).
In 6 participants (median age: 29 std: 8.6, 2 female), we acquired functional MRI at 9.4T (voxel size 1.5mm isotropic, TR = 1s, TE=21ms) while the participants performed a covert attention task with MS and WNS. The MS was a circular patch of dots with motion direction 30° above horizontal, while the WNS was a dynamic white-noise patch. Two stimuli were presented

symmetrically with 4.5° eccentricity relative to a central fixation and 3.5° above horizontal. The covert attention task had three experimental conditions: Attend Left (AL), Attend Right (AR) and Ignore (IG). In AL, the subjects attend to the stimulus in the left visual field and press a button when the attended stimulus changed direction (MS) or a second-order orientation appeared (WNS). In AR, subjects reported on the stimulus in the right visual field. In IG, the subjects ignored the peripheral stimuli and report dimming of the central fixation spot with a button press.
We find a consistent, lateralized activation in MT, MST, FST and PH across motion and non-motion stimuli when contrasting AR vs AL conditions. Posterior-dorsal sub-regions of the TPOJ and medio-dorsal parahippocampal regions also show attentional activation across tasks without lateralization. These findings add to the growing evidence supporting the involvement of sub-regions of the temporal cortex in visual attentional processing. Further analyses will address the interplay between attention-related regions of temporal cortex and subcortical structures.

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Poster

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Institute of Information & Communications Technology Planning & Evaluation (IITP) grant funded by the Korea government (No. 2017-0-00451)

Title: A neurofeedback game for attention improvement and its evaluation

Authors: *H. KIM[1], K. WON[2], S. JUN[2], M. AHN[1];
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Abstract: Recently, there has been an increasing interest in digital healthcare, where technologies are used to manage and improve one's health. Neurofeedback is a training technique that helps a user to self-regulate the brain state. This technology is used for treatment of various disease [1]. In addition, it is reported that neurofeedback is effective in improving cognitive function in healthy people [2]. In this study, we introduce a neurofeedback-based car racing game that measures a player’s attention level which is quantified from the ratio between beta and theta power in electroencephalogram (EEG) and applies the information to control the speed of a car in the game. To evaluate the effectiveness of the game, we conducted online EEG

experiments (acquired by DSI VR300, Wearable Sensing Inc.) with four subjects (approved by Institutional Review Board at Handong Global University). Each subject played five training blocks; each block takes 5-minute car racing. For measuring the performance of different cognitive functions, three behavioral tasks (Color Word Stroop Task (CWST), Psychomotor Vigilance Test (PVT), and 2-back test) were introduced. Thus, each subject completed the tasks and resting state EEG were measured before and after the neurofeedback training, respectively. The game and experiment were implemented in Unity3D and in Python and OpenViBE. As a result, we found that all subjects consistently show beta increase from pre-training (4.53±2.19) to post-training (5.6±1.78) and decreased Reaction time (RT) in 2-back task (post/pre; CWST: 0.02/0.01, PVT: 0.28/0.27, 2-back: 0.65/0.6). But there were no significant results in accuracies of tasks. In conclusion, we introduced a neurofeedback game which is proven to increase beta rhythm in the brain and also shorten the reaction time in 2-back task. In the future, we will further confirm the results with more subjects and compare with placebo group.
Reference [1] Sitaram, R., Ros, T., Stoeckel, L., Haller, S., Scharnowski, F., Lewis-Peacock, J., ... and Sulzer, J., ”Closed-loop brain training: the science of neurofeedback,“ *Nature Reviews Neuroscience*, Vol. 18, No. 2, pp. 86-100, 2016, doi:10.1038/nrn.2016.164. [2] Wang, J.-R., and Hsieh, S., “Neurofeedback training improves attention and working memory performance,” *Clinical Neurophysiology*, Vol. 124, no. 12, pp. 2406-2420, 2013, doi:10.1016/j.clinph.2013.05.020.

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Poster

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Title: Neural decoding of concurrent speech: lessons from selective and divided attention

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Abstract: It is widely assumed that when individuals apply selective attention to a single speaker in a Cocktail Party setting, other so-called “task-irrelevant” speakers are ignored. However, the extent to which task-irrelevant speech is processed and the nature of its neural representation remains a highly debated topic. This ambiguity is further extended when considering how attention might operate under naturalistic conditions, where it might not always be beneficial to fully tune out task-irrelevant speech. Some have suggested that natural listening in multi-speaker

contexts might involve dynamic attention-switching between relevant and irrelevant speakers. However, this hypothesis is extremely difficult to test experimentally, given the limited access to the listener's internal state in selective attention paradigms. To circumvent this methodological challenge, here we explicitly manipulated the task-relevance of concurrently presented speech and examined how this affects its neural encoding in the brain. Specifically, we compared neural speech tracking of two natural speech streams when only one was task-relevant (Selective Attention) vs. when both were task-relevant (Divided Attention). We recorded the magnetoencephalographic (MEG) response from 27 Hebrew native speakers, while they listened to two concurrent speakers telling short narratives. Before each trial, participants were either instructed to attend to one speaker (Selective Attention condition) or to both speakers (Divided Attention condition). After each trial, participants answered comprehension questions either about the pre-designated attended narrative (Selective) or about both narratives (Divided). Subjects performed significantly better on the Selective Attention task (~89%) compared to the Divided Attention task (~72%), indicating that there is a behavioral cost to dividing attention. Speech-tracking analysis of the neural response to the two speakers replicated previous results of Selective Attention, with reduced responses to the task-irrelevant speaker vs. the task-relevant speaker in auditory regions. Interestingly, the neural representation of the two speakers in the Divided Attention condition was comparable to that of the task-irrelevant speaker in the Selective Attention condition. This suggests that the partial encoding of task-irrelevant speech is akin to the encoding applied when attempting to divide attention among two speakers. By contrasting these two opposing attention regimes, our results broaden the ongoing conversation about the system's capacity and limitations for processing two speech streams in ecologically-valid settings.

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Title: Does the visual cortex play a causal role in endogenous covert attention?

Authors: *A. FERNANDEZ, M. CARRASCO;
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Abstract: Covert endogenous (voluntary) spatial attention improves performance in many visual tasks and modulates neural activity at different visual and higher-level areas. The frontal-eye fields and intraparietal sulcus are considered necessary for this improvement to occur, but it is unknown whether early visual areas (V1-V2) play a causal role. We used transcranial magnetic stimulation (TMS) to briefly alter ongoing activity around the occipital pole while observers performed an orientation discrimination task and attention was manipulated via central valid, neutral, and invalid cues. On every trial, observers (n=10) received double pulse TMS at an individually predetermined phosphene region (stimulated region) around the occipital pole. Two cortically magnified gratings were presented simultaneously, one in the stimulated region and another in the symmetric (non-stimulated) region in the opposite hemifield. In every trial the same region was stimulated, in half of the trials the stimulus processed by that region was the target and in the remainder trials was the distractor. Grating contrast was varied to measure contrast response functions for all combinations of attention (valid, invalid, neutral) and stimulation (target, distractor) conditions. Attention affected contrast responses similarly regardless of whether the target or distractor were stimulated. These findings indicate that early visual areas are not necessary for endogenous attention to modulate performance. Additionally, we compare the current results to our previous study where TMS extinguished the effect of exogenous (involuntary) attention on performance (Curr Bio, 2020), and provide competing computational models for the effect of TMS on attention, which are extensions of the Reynolds-Heeger (2009) normalization model of attention (NMA): (1) NMATMS: TMS modifies the strength and spatial spread of attention by scaling the attention field; (2) RGTMS (response-gain): TMS modifies the stimulus drive and attention exerts its effects after normalization; (3) tunedTMS: TMS modifies baseline suppression by changing the bandwidth and strength of the suppressive field. The tuned-NMA model best captured both the endogenous and exogenous attention results. In both cases TMS decreased the spatial spread of suppression, but total suppression was weaker with endogenous than exogenous attention. In conclusion, unlike exogenous attention, early visual areas are not necessary for endogenous attention to modulate performance. Additionally, endogenous and exogenous attention results can be captured by assuming that TMS alters both suppression strength and bandwidth.

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Poster

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Topic: H.01. Attention

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Title: Attentional modulation of category-selective regions in ventral occipitotemporal cortex

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Abstract: The rapid recognition of a face in a crowd or the words on this page involves a dynamic interplay between visual input and the direction of limited attentional resources required for rapid identification of these inputs. Visual categorization of words and other visual stimuli is known to involve category-selective regions of the ventral occipitotemporal cortex (vOTC) as well as interaction with a broader language and attentional network. Previous functional imaging work demonstrates a topographic map of category selectivity in vOTC. Recent evidence suggests that the boundaries of category-selective regions of the vOTC may shift based on selective attention. Given limited temporal resolution of these methods, the dynamics of these interactions with language and attentional networks remain unclear. To probe these dynamics, we utilize the high spatiotemporal resolution of intracranial EEG recordings during a task where visual attention is directly modulated.
To explore the influence of attentional modulation on category-selective regions of vOTC, we performed intracranial recordings in 18 patients (2,000 + electrodes) undergoing electrode implantation for seizure localization in intractable epilepsy. All participants performed a category localizer task in which visual stimuli of different categories (Faces, Words, Scenes, or Animals) were presented. Patients responded based on a color-changing fixation point, repetition of a presented stimuli (i.e., a one-back task), or when target stimuli within a specific category appeared (e.g., respond to fruit words like “apple”). We analyzed broadband high gamma activity (BGA, 70 - 150 Hz) following each visual stimulus onset, contrasting between stimulus category and attentional task conditions.
High BGA and strong category-selectivity was seen across vOTC and inferior frontal gyrus (IFG). There was enhancement of BGA when the stimuli were attended, compared to BGA activation for fixation. We observed early attentional modulation of BGA activation in IFG and vOTC. Additionally, selective attention of individual categories enhanced category selectivity for the attended category. Research into the localization and modulation of cortical regions selective for different categories of visual stimuli will yield insight into the organizational principles and functionality of these regions. Beyond a purely structural analysis, this research explores how attention dynamically modulates intrinsic selectivity of neural populations in category selective vOTC.

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Poster

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Title: Brain state dynamics reflect generalizable cognitive and attentional state dynamics

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Abstract: Understanding relationships between cognitive and neural state dynamics is a pivotal goal of cognitive neuroscience. Recent work suggests that the dynamics of large-scale brain states—characterized by distinct patterns of activity and functional interactions—represent time-varying cognitive and attentional states (Shine et al. 2019; Vidaurre et al. 2018), for example, capturing behavioral performance in cognitive tasks (Yamashita et al. 2021; Cornblath et al. 2020) or attentional engagement during movie watching (Van der Meer et al. 2020). However, the underlying characteristics of these brain state dynamics, the nature of the cognitive and attentional states that they reflect, and their consequences for behavior have not been studied in diverse controlled and naturalistic contexts. To ask these questions, we collected multi-session fMRI data (N=27) as participants underwent a variety of paradigms—sustained attention tasks, movie watching, and rest—and measured individuals' moment-to-moment attentional states. Hidden Markov model analyses revealed four latent brain states spanning canonical gradient axes of functional brain organization (Margulies et al. 2016), with a brain state situated at the center of the gradient axes acting as a hub in state transitions. Transient de-synchronization in global cortico-cortical and cortico-subcortical connections (Esfahlani et al. 2020) preceded neural state transitions. To elucidate the functional relevance of latent brain states, we related their occurrences to ongoing measures of attention. We found that brain state dynamics were synchronized across participants and time-locked to narrative event boundaries when subjectively engaging movies (sitcom episodes) were viewed. However, in a less engaging movie (an educational documentary), brain state dynamics were less synchronized across people and more similar to dynamics observed during controlled tasks and rest than to dynamics observed during engaging movie watching. Furthermore, we found that brain state dynamics reflected fluctuations in attention task performance and self-reported engagement during movies. Notably, a common brain state indicated suboptimal attention during tasks and movies, whereas different brain states indicated engaged attention in different contexts. Our results not only generalized across fMRI runs but also across data collected from three independent research groups (Chen et al. 2017; Rosenberg et al. 2016; Chang et al. 2021). Together, these results demonstrate the functional significance of large-scale neural state dynamics for ongoing cognition and attention.

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Poster

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Title: Attention operates differently across eccentricities

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Abstract: In primates, foveal and peripheral vision have distinct neural architectures and functions. However, it has been debated if selective attention operates via the same or different neural mechanisms across these different eccentric locations. We hypothesized that the discrepancies between past results arose in part from the differences in neural signals used to index attentional modulations and the differences in task difficulty across eccentricities. Here, we measured three EEG indices of visual processing, while male and female human subjects performed a visual attention task, where they either attended to the fixation or to the visual

stimuli presented at different eccentricities. These EEG indices include the amplitudes of the steady-state visually evoked potentials (SSVEPs), the sustained negative deflection (SND), and the alpha band activity. Critically, we equated task difficulty across the different attention conditions and eccentricities. Overall, the amplitudes of these neural markers reduce as a function of eccentricity, reflecting changes in cortical magnification. However, they expressed totally different patterns of attentional modulations across eccentricities. The SSVEP exhibited prominent attentional gain modulations at the peripheral locations, but attentional modulations were significantly reduced at the parafoveal locations. In contrast, the SND showed relatively similar levels of attentional modulations across the parafoveal and the peripheral locations. On the other hand, alpha band activity exhibited significant attentional modulations only at the peripheral locations, which relied primarily on the relative differences between the contralateral and ipsilateral signals. Taken together, these results suggest that sustained visual attention comprises multiple biophysical processes that differentially impact sensory information processing across eccentricities.

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Poster

P798. Human Functional Connectivity and Network Activity

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Support: NINDS R01 NS057156
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Title: A new tripartite landmark in human posterior cingulate cortex

Authors: *E. H. WILLBRAND[1], B. J. PARKER[1], T. HALLOCK[1], W. I. VOORHIES[1], L. APONIK-GREMILLION[2], J. A. MILLER[1], S. A. BUNGE[1], B. L. FOSTER[3], K. S. WEINER[1]; [1]Univ. of California, Berkeley, Berkeley, CA; [2]Baylor Col. of Med., Houston, TX; [3]Univ. of Pennsylvania, Philadelphia, PA

Abstract: Human neuroimaging shows that the posterior cingulate cortex (PCC) is an anatomically and physiologically unique brain region, being a central network hub of neocortical connectivity and displaying one of the highest cortical metabolic rates. Nevertheless, PCC function remains poorly understood. For example, while the PCC is typically viewed as a key

node of the default mode network (DN), additional human neuroimaging studies suggest a role of PCC in episodic memory retrieval and cognitive control. Central to this confusion is the varied and inconsistent anatomy/nomenclature used to define the PCC. Across human studies, PCC is used interchangeably with precuneus (PrC) and/or retrosplenial cortex (RSC). Therefore, establishing consistent anatomical landmarks and nomenclature is foundational to progress in understanding PCC function. With this goal in mind, we conducted a multimodal investigation of the sulcal morphology of the posteromedial cortex, which includes PCC, RSC and PrC. Manual identification of 4319 sulci across 552 hemispheres, spanning the human and chimpanzee lifespan, revealed the presence of a novel and shallow sulcus, which we termed the inframarginal sulcus (*ifrms*). Subsequent analyses revealed five main findings regarding the *ifrms*. First, the *ifrms* is a macroanatomical and microanatomical landmark: the *ifrms* identifies a focal point of high cortical thickness and light myelination in PCC with the largest thickness/myelination ratio of PCC sulci. Second, the *ifrms* is a functional landmark, co-localizing with, and predicting the location of, a functional sub-region of the cognitive control network (CCN). Third, the *ifrms* is identifiable in juvenile and healthy older adults, as well as in 48% of chimpanzee hemispheres examined. Fourth, the characteristic morphological features of the *ifrms* change across age groups and between species: the *ifrms* is deeper in older adults and juveniles than in young adults across species, as well as is deeper in humans compared to chimpanzees. Fifth, the *ifrms* gray matter thins across age groups in humans and chimpanzees. Altogether, our results reveal that the *ifrms* is a new tripartite landmark in human PCC that is developmentally and evolutionarily relevant. These findings help reconcile prior discrepancies regarding where the DN ends and the CCN begins in PCC, as well as support a classic, yet largely untested theory that tertiary sulci may serve as landmarks in association cortices, laying the groundwork for future investigations of PCC functional neuroanatomy.

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Poster

P798. Human Functional Connectivity and Network Activity

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Program #/Poster #: P798.02

Topic: H.04. Executive Functions

Title: Transcriptomic contributions to the cytoarchitectonic parcellation of the human cerebral cortex.

Authors: *L. KING[1], J. GOMEZ[3], Z. ZHEN[4], K. WEINER[2];
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Abstract: *Introduction*: Transcriptomic contributions to the functional and anatomical parcellation of the human cerebral cortex has become a major interest in human brain mapping in the last decade. Recent research shows that active transcription of a small set of genes contributes to large-scale gradients and functional hierarchies in the human cerebral cortex. The combination of previous findings has also shown that different sets of genes contribute to the positioning of cortical areas within functional hierarchies and the organization of functional maps within cortical areas. Complementing these previous micro- and meso-scale approaches, here, we implemented a whole brain, macroscale approach to examine transcriptomic contributions to the cytoarchitectonic parcellation of the human cerebral cortex. *Methods*: This investigation makes novel use of three publicly available datasets: the AHBA transcriptomic dataset, the JuBrain cytoarchitectonic atlas, and the von Economo-Koskinas atlas. This is done by overlaying all three datasets in the common MNI-152 space followed by the application of data-driven methods. This includes the use of PCA to determine the genes most differentially expressed across 87 cytoarchitectonic ROIs in which these ROIs were then clustered based on their similarity in gene expression. Furthermore, using variables such as cellular density and cell width, we correlated microanatomical features to the transcriptomic profiles of these cytoarchitectonic ROIs. *Results*: We show that a novel set of genes contribute to the cytoarchitectonic parcellation of the human cerebral cortex beyond those genes previously identified that contribute to either the parcellation of functional hierarchies or maps within cortical areas. Additionally, the clustering of the cortical ROIs via their transcriptomic profile reveals areas that are more similar in function rather than macroanatomical location: areas between networks are often more transcriptomically similar than areas within the same network. *Conclusion*: These results support a novel hypothesis that there is a nested spatial relationship among transcriptomic, anatomical, and functional parcellations of the human cerebral cortex at the level of maps (mm; areal), gradients and hierarchies (cm; within lobes), and networks (several cm; across lobes). We conclude by showing that a microanatomical substrate supporting this hypothesis across spatial scales is cell density.

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P798. Human Functional Connectivity and Network Activity

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P798.03

Topic: H.04. Executive Functions

Support: NIH R01MH120080

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Yale University Kavli Institute for Neuroscience Postdoctoral Fellowship for Academic Diversity

Title: Anatomical properties of individual-specific areal-level parcellations predict cognition and behaviour

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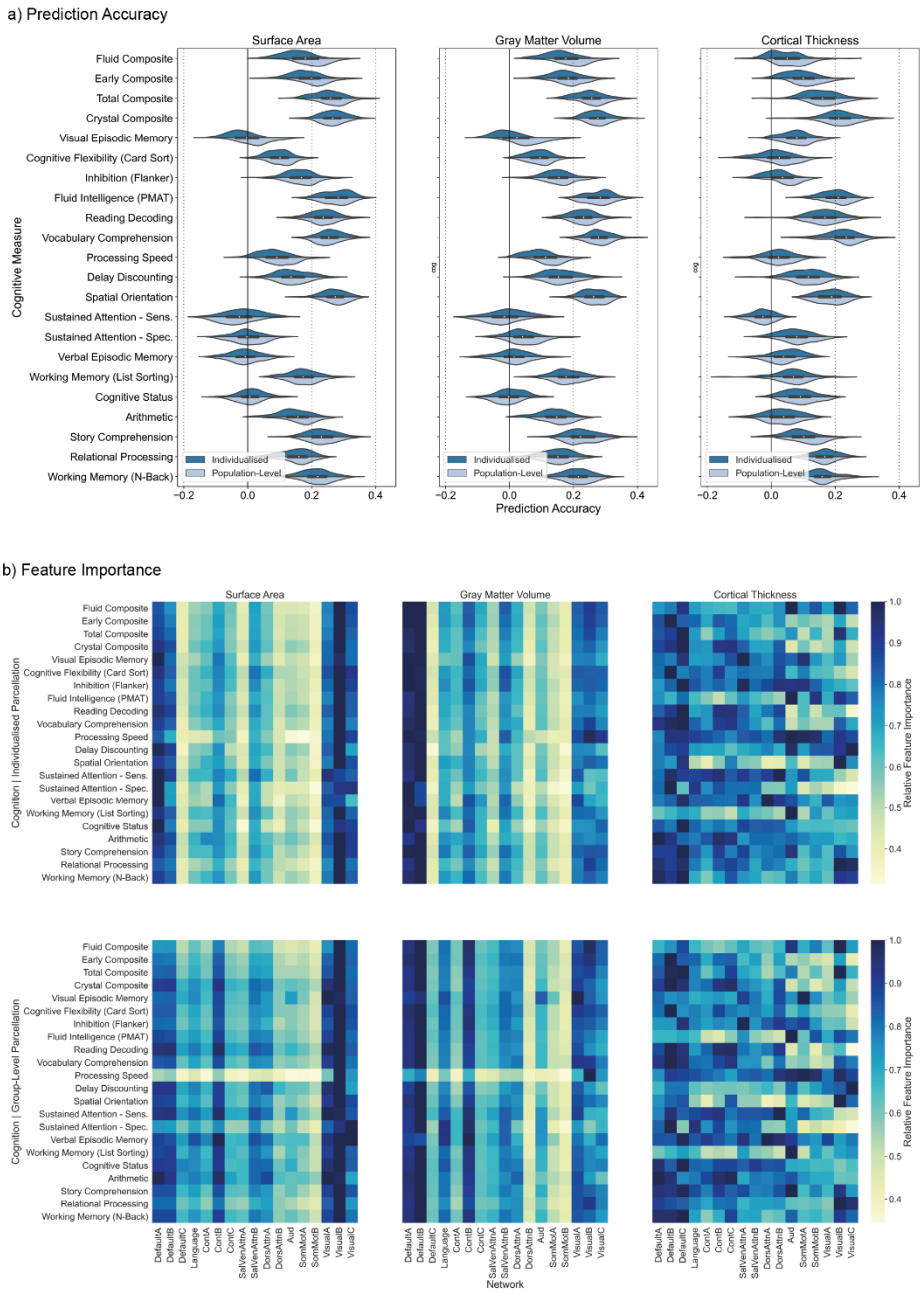


Figure 1: Model Performance for the Prediction of Cognitive Measures.

a) Prediction accuracy (correlation between true and predicted values) violin plots for models using surface area (left), gray matter volume (middle) and cortical thickness (right) computed using individualised (dark blue) and group-level (light blue) Schaeffer parcellations to predict cognitive measures. Average prediction accuracy across all cognitive measures using the individualised parcellations are r = 0.145 ± 0.043 (mean ± standard deviation) for surface area, r = 0.150 ± 0.043 for gray matter volume, and r = 0.097 ± 0.045 for thickness. Average prediction accuracy across all cognitive measures using the individualised parcellations are r = 0.158 ± 0.042 (mean ± standard deviation) for surface area, r = 0.163 ± 0.043 for gray matter volume, and r = 0.112 ± 0.045 for thickness.

b) Network-level mean felative feature importance for models using surface area (left), gray matter volume (middle), and cortical thickness (right) computed using individualised (top) and group-level (bottom) Schaeffer predict cognitive measures. Absolute areal-level feature weights were Haufe-inverted (Haufe et al. 2014), summarised to a network level (Yeo et al. 2011), and averaged over the 100 splits for each model.

Abstract:

Functional and structural properties of the brain underlie cognition. Prior work has shown that individual differences in the spatial topography and size of functional networks predict a wide

range of behaviours (Kong 2018), while variation in regional patterns of cortical thickness and morphometric similarity respectively link with aspects of psychopathology (Parkes 2021) and cognition (Seidlitz 2018). Moreover, functional connectivity measures based on individualised parcellations improve predictions of behaviour (Kong 2021) beyond those achieved with population-level parcellations. Here, we investigate whether areal-level cortical surface area, gray matter volume, and thickness derived from individualised or population-level functional parcellations more accurately capture neural biomarkers underlying cognition. We examined resting-state functional MRI surface data from 967 healthy young adults (ages 22-37; 453 males) from the Human Connectome Project (Van Essen 2013). We used the population-level Schaeffer parcellation (n=400; Schaeffer 2018) and derived individualised parcellations using the gradient-infused multi-session hierarchical Bayesian model (Kong 2021). We then estimated surface area, gray matter volume, and thickness of each parcel, and used those independently to predict cognitive measures with linear ridge regression models with k-fold group-matched cross-validation and 100 distinct train/test (2:1) splits. Prediction accuracy distributions are comparable across individualised and population-level parcellations (Fig 1a). Surface area and gray matter volume in the Default Mode and Visual networks strongly contributed to predictions across cognitive measures, while a diffuse profile of parcels contributed to predictions using thickness (Fig 1b). These results demonstrate that areal-level properties derived using individualised and population-level parcellations comparably capture relationships with cognitive functioning, and these relationships are driven by the Default Mode and Visual networks.

Disclosures: E. Dhamala: None. **R. Kong:** None. **K.M. Anderson:** None. **L. Ooi:** None. **J. Chen:** None. **B.T.T. Yeo:** None. **A.J. Holmes:** None.

Poster

P798. Human Functional Connectivity and Network Activity

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scholarship from Knud Højgaards Fond
scholarship from Reinholdt W. Jorck og Hustrus Fond
scholarship from Viet-Jacobsens Fond

Title: Uncertainty Modulates Visual Maps During Non-Instrumental Information Demand

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Abstract: Humans and animals seek to obtain information that reduces uncertainty even when the information cannot be utilized to increase reward gain. Neurons in the basal ganglia and medial prefrontal cortex encode the motivation to reduce uncertainty, but it is unknown how these global motivational signals are translated into concrete information sampling policies. To examine this question, we investigated the impact of uncertainty on visual representations in the monkey lateral intraparietal area (LIP) that is implicated in attention and eye movement control. Monkeys performed trials with 0%, 50% or 100% reward probabilities, with the 50% condition uniquely inducing reward outcome uncertainty, and had a free viewing period when they could shift gaze to a mask to uncover a cue that predicted the outcome (reward or lack of reward). Importantly, revealing the additional cue was not incentivized and did not alter the trial's reward. Nevertheless, monkeys were intrinsically motivated to reveal the cue, and were more likely to do so at 100% relative to 0% probability (indicating a sensitivity to reward probability) and at 50% relative to 100% probability (indicating a desire to resolve uncertainty). Consistent with this behavioral pattern, LIP neurons showed distinct modulations by reward probability and uncertainty. A time-resolved linear model showed that reward uncertainty signals arose at longer latencies relative to those of reward probability and enhanced firing rates in all epochs of the neurons' response, including anticipatory and visual pre-saccadic responses and post-saccadic responses to the masked or revealed cue. Uncertainty modulations did not encode immediate saccade plans but correlated with behavior on longer time-scales - i.e., predicted variations in the sensitivity to uncertainty in the monkeys' daily revealing behavior. Moreover, in a passive task with no opportunity to sample information, reward modulations were unchanged while uncertainty modulations became negative (suppressing rather than enhancing the visual response). Our results suggest that reward and uncertainty are two independent motivational signals that converge on spatially tuned LIP cells and, by modulating visual priority, affect the likelihood that monkeys will select and process informative visual cues.

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Poster

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Topic: H.04. Executive Functions

Support: NSF Graduate Research Fellowship in Cognitive Neuroscience
Beckman Institute Graduate Research Fellowship

Title: Networks underlying the maintenance of endogenous cognitive control

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Abstract: Cognitive control comprises a multitude of functions that are difficult to dissociate neurobiologically. These functions are commonly tested in the context of external events that modulate control demands (e.g. cues), informing about externally-driven control but providing no insights into intrinsically maintained cognitive control processes. Here, we designed an fMRI task to investigate the role of internally-driven cognitive control and its relationship with intrinsic functional connectivity networks implicated in cognitive control (dorsal attention network (DAN), fronto-parietal network (FPN), and cingulo-opercular network (CON)). 25 human subjects (13 female) underwent fMRI. Each trial consisted of a pair of sequential stimuli with the subject responding if the stimuli were identical or not. The design had two factors: "Cueing" manipulated the presence of an auditory cue prior to the first stimulus, and "Content" manipulated whether the stimulus category was known (only faces) or unknown (randomly selected from 4 visual and 4 auditory). Factors were analyzed for two time periods, the "Intertrial" period (1.5-10s), and the "Trial" period - the time from cue onset (if present) to response. Interblock intervals (15s) provided implicit baseline. Using a general linear model (GLM), we found that for the contrast of No Cue > Cue significant activations were found in both time periods. During the Intertrial period (endogenous phasic control) activations were found in areas overlapping with DAN. During the Trial period (exogenous phasic control) activations were found in areas overlapping with FPN. To understand how DAN maintains endogenous control during the intertrial period, we investigated the task-related modulation of DAN connectivity using psychophysiological interactions (PPIs). Using the significant voxels within the DAN during the No Cue > Cue contrast as the region of interest (ROI) mask and the same contrast as the context weights, PPI analyses showed significant connectivity changes between the DAN and default mode network (DMN) during the Intertrial period. Finally, we performed an adapted region-by-region PPI analysis using all individual regions of DAN, FP, DAT, and DMN (39 ROIs total). Using network-based statistics we found significant clusters of connections between the DAN and DMN whose connectivity strength increased in the Intertrial period when cues were unavailable. We conclude that the DAN is more engaged during periods of endogenous phasic control, the FPN is more engaged during exogenous phasic control, and there is a significant relationship between the DAN and DMN that is contextually dependent on the presence of a cue.

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Poster

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Program #/Poster #: P798.06

Topic: H.04. Executive Functions

Support: NARSAD 5103660

Title: Dynamic neuronal networks supporting working memory shift with emotion

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Abstract: Brain activity supporting executive functions is dynamic, adapting and accessing flexible neuronal networks to meet task demands. Theta (4-8Hz) and alpha (8-12Hz) band activity support executive functions, yet their network activity has been less clear. The current study investigated these networks in two affective working memory tasks: one for updating and one for inhibition. Thirty participants completed the affective executive tasks while EEG was recorded with a 64-channel BioSemi Active2 system. In the n-back updating task, participants identified emotional images (from the International Affective Picture System) that were presented 1 or 2 items ago in the series. In the affective go/no-go task participants withheld a pre-potent response to a particular affective image category. For each task and affective category (positive, neutral, or negative) we extracted the evoked power and used the power correlations as the bases for the network. We then assessed network features (centrality, clustering) and dynamic network features using a multilayered network. Within this multilayered network we detected underlying communities. The dynamic network communities clearly aligned with stages of information processing, and a greater number of communities were detected for the tasks with negative information. These results further support the differential nature of negative information and highlight how this information shifts neuronal networks supporting executive functions.

Disclosures: A. Campbell: None. **A.S. Ferguson:** None. **P.J. Mucha:** None.

Poster

P798. Human Functional Connectivity and Network Activity

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Program #/Poster #: P798.07

Topic: H.04. Executive Functions

Title: Common brain regions support emotion regulation and cognitive control tasks

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Abstract: Cognitive control is the ability to organize behavior in flexible ways in accordance with internal goals. Cognitive control processes can be applied in situations requiring the regulation of emotions. In this case, the cognitive control task demands include monitoring, evaluating, and modulating reactions. The ability to regulate emotion is thought to be critical for maintaining and regaining mental health. Therefore, emotion regulation is related to a variety of mental disorders, such as depression, anxiety, and addiction. Although cognitive control and emotion regulation are conceptually related, the empirical relationship between these processes is largely unknown. Recent data has suggested that the flexible hub properties of the frontoparietal control network (FPN) contribute to cognitive control by being able to update brain network connections in a flexible and rapid manner according to current task demands. I hypothesize the same flexible hub mechanisms that contribute to non-emotional cognitive control tasks extend to emotion regulation. Recent research has suggested that different emotion regulation strategies may be more effective and better associated with outcome measures. The purpose of this study is to examine the relationship between non-emotional cognitive control and emotion regulation, identify a common neural mechanism and assess the efficacy of two different reappraisal strategies (distancing and reinterpretation). I tested these hypotheses in a sample of participants (n=21, 14 females) that completed a non-emotional cognitive control task (n-back) and an emotion regulation task. Participants completed the tasks in the fMRI scanner. Consistent with my hypotheses, the non-emotional cognitive control task and emotion regulation engaged overlapping brain regions. This overlap was apparent in the frontoparietal and cingulo-opercular networks. Both networks have been previously identified as having a role in cognitive control and emotion regulation. Despite a large degree in overlap between cognitive control and emotion regulation activity, vlPFC was preferentially engaged by emotion regulation. This supports the hypothesis that the vlPFC serves an inhibitory function on negative emotion when participants use reappraisal strategies. Finally, we found that activity in the dlPFC was greater when reinterpretation was used relative to distancing-based reappraisal strategies. This is consistent with the hypothesis that reinterpretation may require more effort and resources than distance-based strategies because distance-based approaches are not bound by specific sensory features.

Disclosures: D.H. Schultz: None.

Poster

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Topic: H.04. Executive Functions

Support: NIH/NIA R21AG048170
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Title: Is brain modularity a biomarker of exercise intervention-related plasticity?

Authors: *S. JAIN[1], M. A. PIPOLY[2], M. SODOMA[1], V. MAGNOTTA[3], G. PIERCE[4], M. VOSS[1];
[1]Dept. of Psychological and Brain Sci., [2]Neurosci., [3]Dept. of Radiology, [4]Dept. of Hlth. and Human Physiol., Univ. of Iowa, Iowa City, IA

Abstract: Research supports promise for physical and cognitive interventions to promote cognitive plasticity. However, there is wide variability in benefits across individuals. Brain modularity, a network-based measure quantifying the extent a brain has non-overlapping subnetworks, has been proposed as a biomarker of individual variability related to training gains including on tasks of working memory. However, no studies have tested whether this relationship would also be seen following a single acute intervention session. We tested the predictive strength of modularity on exercise-induced changes in working memory after an acute and long-term physical exercise intervention. We collected resting state scans and performance data from a face N-Back task (1-Back, 2-Back) from cognitively normal older adults (N = 29, $M_{age} = 67 \pm 4.21$) at the beginning of the study and after both acute and long-term intervention. As part of the acute intervention, each participant performed two physical exercise sessions for 20 mins each on a stationary bike at moderate intensity (increased their heart-rate (HR) to 65% of their HRmax) as well as light intensity (minimal effect on HR). These sessions were performed on different days and their order was counterbalanced. This was followed by a three-month intervention where 62% (N=18) participants trained at moderate intensity and the remaining at primarily light intensity with short moderate intensity intervals. N-back accuracy improved more following moderate compared to light intensity exercise at the acute session; training related behavioral changes after the acute session were predictive of long-term intervention gains for all participants.We tested the predictive ability of a) pre-training modularity and b) modularity after acute interventions on post-training performance. We used weighted undirected functional matrices and the Schaefer 400 parcellation. Modules were defined in a data-driven manner and modularity calculated using the Community Louvain function. Regression analysis showed pre-training modularity did not predict acute or long-term intervention-related accuracy gains. However, modularity calculated after moderate-intensity acute exercise was a significant predictor of working memory accuracy gains after training. Identifying a reliable biomarker of intervention-related cognitive plasticity is not just theoretically important but has wide ranging practical significance. Brain modularity might be a valid biomarker of potential for training-induced plasticity, though our results suggest acute manipulations may enhance sensitivity for predicting exercise-induced benefits on cognition.

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Poster

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Topic: H.04. Executive Functions

Support: U.S. Army Medical Research Acquisition Award No. W81XWH-17-1-0457

Title: The low glutamate diet improves working memory performance and alters functional connectivity within pertinent networks in veterans with Gulf War Illness

Authors: *A. E. KIRKLAND[1], M. T. LANGAN[2], V. C. MUCCIARONE[3], L. C. RICE[3], J. W. VANMETER[4], K. F. HOLTON[3], *A. KIRKLAND[5], *A. KIRKLAND[5];
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Abstract: Gulf War Illness (GWI) is a chronic multi-symptom disorder with severe cognitive impairments thought to be related to glutamatergic excitotoxicity and central nervous system (CNS) dysfunction. The low glutamate diet has been proposed as a comprehensive intervention for GWI because it removes dietary amino acids that can act as excitotoxins in the CNS, which may affect glutamatergic neurotransmission and symptoms of GWI. The objective of this study was to assess the effects of the low glutamate diet on verbal working memory and functional connectivity (FC) in veterans with GWI. Twenty-four participants (age= 53.3 (5.9) yrs, 75% male) completed a fMRI verbal N-back task at baseline and after the one-month low glutamate diet. A data-driven multi-voxel pattern analysis (MVPA) approach was used to identify regions with significant differences in whole-brain FC during the 2-back block of the N-back task (highest cognitive load) before and after the diet. Subsequent seed-to-voxel analyses were used to assess differences in whole-brain task-based FC patterns associated with the MVPA-identified seeds. All analyses were conducted in the CONN toolbox (Version 20b) using a peak level threshold of $p<0.001$ and a cluster defining threshold of $p<0.05$ FDR correction ($k>75$). Accuracy during the working memory 2-back improved after the one-month low glutamate diet (+13%; $p=0.006$). MVPA analysis resulted in 3 clusters with significant differences: part of the frontoparietal network (FPN) was represented in cluster 1 (left anterior and posterior supramarginal gyrus, planum temporale, parietal operculum) and cluster 2 (left inferior and middle frontal gyrus, precentral gyrus), while cluster 3 (right anterior supramarginal gyrus, postcentral gyrus) was part of the ventral attention network (VAN). The seed-to-voxel analysis identified significant FC changes for clusters 1 and 2 after the one-month low glutamate diet. Relative to baseline, cluster 1 and cluster 2 had decreased FC with regions comprising the VAN and somatomotor network. Cluster 2 also had increased FC with regions of the default mode network (DMN; lateral occipital cortex) and the FPN (cerebellum Crus I, II, and VI). Combined, these results suggest that the low glutamate diet reduces potential interference from sensory/attention reorienting networks while strengthening connectivity between the two networks central to working memory (FPN and DMN), which could underlie the working memory improvement. Future studies should replicate these findings in a larger sample of veterans with GWI and explore the effects of the low glutamate diet on performance and functional connectivity within other cognitive domains.

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Poster

P798. Human Functional Connectivity and Network Activity

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Program #/Poster #: P798.10

Topic: H.04. Executive Functions

Title: Cognitive Implications of Brain Structural Connectivity Alterations Between Nodes of the Default Mode Network Related to Cannabis Use and HIV Infection

Authors: *P. VIERA PEREZ[1], J. S. FLANNERY[1], R. GONZALEZ, Jr[2], A. R. LAIRD[3], M. T. SUTHERLAND[1];
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Abstract: To alleviate HIV-related symptoms people living with HIV (PLWH) often report using cannabis (CB). Most PLWH demonstrate neurocognitive impairments showing a need to clarify manifestations of impairments. Emerging evidence suggests that HIV and CB use are both linked with alterations in default mode network (DMN) functional connectivity. Prior studies have demonstrated that resting-state connectivity networks correlate with underlying structural connections between functional nodes. Elucidating the relationship between chronic CB use, HIV, and the structural connectivity of the DMN is important for understanding the possible compounding effects of both CB and HIV on neurocognitive functions. Using diffusion-weighted imaging (DWI) data, we characterized structural connectivity profiles of DMN regions among 100 participants stratified into four groups based on HIV status and CB use history. Structural connectivity profiles across the DMN were established using a deterministic fiber tractography algorithm to track bilateral cingulum tracts (CT), the bilateral superior fronto-occipital fasciculus (FOF), and the genu of the corpus callosum (CC). Participants completed a battery of well-validated tasks to probe behavioral measures of inhibitory control, error processing, and general cognitive control. To explore the influence of HIV infection and CB use on structural connectivity, an ANOVA analysis was conducted on individual white matter tracts. For tracts that showed significant group-level differences, a mixed linear regression was conducted. The ANOVA analysis identified significant group differences in the number of tracts of the right CT between HIV groups, tract volume of the left CT between CB groups, and the average FA of the right superior FOF between CB groups. Posthoc tests revealed that HIV+ groups had more right cingulum tracts, and CB+ groups had higher FA in the right superior FOF and greater tract volume in the left cingulum. The mixed linear regression analyses identified a significant interaction between CB and FA in the left cingulum, such that CB+ groups were associated with more preservative errors than the CB- groups. It was shown that left cingulum

tract volume was positively associated with general processing speed. The left cingulum tract connects areas commonly linked with both memory and emotional regulation. Our results suggest that the left cingulum tract may be more susceptible to CB-related structural connectivity alterations regardless of HIV serostatus. These results suggest alterations to structural connectivity between nodes of the DMN contribute to more preservative errors.

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Poster

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Topic: H.04. Executive Functions

Support: STCSM Grant 19JC1410100

Title: Individual differences in children's neural state segmentation performance are related to intelligence

Authors: *X. XU, S. MA, X. LIU, Y. LI, K. ZHANG, H. WANG, Y. HU, L. LIN, Q. CAI; East China Normal Univ., Shanghai, China

Abstract: To understand the ever-changing world, humans need to maintain a stable representation of the current context, and segregate the contextual information when it is no longer relevant. Such ability is of developmental significance. Using naturalistic materials and fMRI multi-voxel neural state segmentation analyses, studies have found that neural state boundaries co-occur with movie event boundaries, suggesting it might be possible to neurally index state segmentation ability. However, it remains unknown whether neural state segmentation performance might be indicative of individual differences in general cognitive abilities. Here, we collected an fMRI dataset of children (n=61, age: 6-7) watching a customized animation in which cognitive tasks and narrative events were interleaved, resulting in movie state boundaries. We found that neural state boundaries in several cortical networks align with movie state boundaries. To index individual differences in neural state segmentation, we calculated a neural pattern differentiation score to represent how well children's neural patterns differentiate between consecutive movie states. We found that the differentiation scores were reliable across two runs, and higher differentiation scores were associated with higher intelligence scores, with the highest correlation observed in the control network ($r = 0.30$). Further analyses showed that between-consecutive-state neural pattern dissimilarities were more associated with intelligence than within state neural pattern similarities. Moreover, this differentiation score based on neural pattern information outperformed univariate task-related neural response magnitude in predicting

intelligence. Together, these results showed that there are meaningful individual differences in children's neural state segmentation performance, which might be indicative of general cognitive development.

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Poster

P798. Human Functional Connectivity and Network Activity

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Program #/Poster #: P798.12

Topic: H.04. Executive Functions

Title: Hemispheric Asymmetries in Individual Differences in Functional Connectivity

Authors: *D. C. PEREZ, A. DWORETSKY, C. GRATTON;
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Abstract: Resting-state functional connectivity (rs-FC) research has produced highly validated group representations of the brain's network architecture. However, cortical network architecture is known to vary across individuals. "Precision" fMRI approaches allow us to examine functional organization at the individual level with high reliability by obtaining extended amounts of fMRI data for each subject. Individualized network maps derived using precision fMRI have revealed the existence of localized regions that exhibit divergent network organization in individuals compared to the group average. These regions have been termed "network variants" because their rs-FC pattern diverges from that of the canonical network associated with its location. Network variants have been shown to be present in all individuals, stable across time, and related to functional variation in tasks. However, the potential differences across hemispheres in the properties of variants have not been examined in detail. Here, we use data from the Human Connectome Project (HCP; n = 384 unrelated individuals) to investigate hemispheric differences in the properties of network variants. For each subject, we identified regions where the pattern of functional connectivity differs strongly from the group average. We found that the left and right hemispheres differ significantly in spatial location of network variants ($p < 0.001$), with more variants in the inferior frontal gyrus, angular gyrus and dorsomedial prefrontal cortex in the right hemisphere and in the supramarginal gyrus and posterior temporal gyrus in the left hemisphere. This result was replicated with an independent sample from the Midnight Scan Club dataset ($n = 9$; $p < 0.001$). Additional analyses using the HCP dataset show hemispheric differences in average variant size, where variants tend to be bigger in right compared to left hemisphere ($p(FDR) < 0.05$). Moreover, the number of variants associated with the frontoparietal and default mode networks was significantly greater in the right hemisphere compared to left hemisphere ($p(FDR) < 0.01$). Interestingly, the opposite

pattern was observed for network variants associated with the language network (p(FDR) < 0.01). Understanding the properties of network variants may help elucidate the relationship of variability in rs-FC to individual differences in behavior.

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Poster

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Topic: H.04. Executive Functions

Support: Simons SFARI Pilot Award
NJ ACTS Fellowship TL1TR003019

Title: Unique sampling of cortex-wide neural dynamics underlies individual behavioral phenotypes

Authors: *C. MACDOWELL, K. S. MILLER, M. LENZI, D.-L. FERGUSON, T. J. BUSCHMAN;
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Abstract: Behavior differs across individuals, lying along a spectrum that can range from typical to atypical phenotypes. Previous work has shown individual differences in behavior are related to differences in neural structure and/or function. Here, we show that individual differences in behavior are related to differences in the sampling of cortex-wide neural dynamics. Using a battery of behavioral tests, we quantified the spectrum of behavioral phenotypes ranging from 'typical' wildtype mice to mice with an induced 'atypical' phenotype. Then, using cortex-wide calcium imaging, we quantified cortical neural activity into a set of 16 'motifs'. These motifs captured repeated spatio-temporal patterns of neural activity that propagated across cortical areas. The same set of motifs were seen in all animals. However, an individual's phenotype was correlated with the likelihood of observing specific motifs. This relationship was seen both when animals were alone, and at rest, and when they were engaged in social interactions. Furthermore, animals' motif activity correlated with traditional measures of zero-lag functional connectivity across cortical areas. Since multiple motifs can engage the same brain areas, this suggests that differences in zero-lag functional connectivity reflects cortical activity dominated by specific spatiotemporal patterns in information flow. Together, our results relate cortical neural dynamics to behavioral phenotypes and suggest that individual differences may be driven by biases in animals' sampling of cortical dynamics.

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Poster

P811. Neural Mechanisms of Social Cognition: Higher Mammals

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Title: The impact of clutter on the spatial sensitivity of face selective populations in the macaque brain.

Authors: *J. TAUBERT, S. G. WARDLE, C. T. TARDIFF, A. PATTERSON, C. I. BAKER; The Natl. Inst. of Mental Hlth., Bethesda, MD

Abstract: Representations of stimuli in the ventral visual pathway are thought to be organized according to two principles: retinotopic space (i.e., where something is) and category (i.e., what something is). Although these principles were originally thought to be reflected in different sectors of the ventral visual pathway, recent fMRI studies have reported that category-selective regions in the human brain are also spatially biased, with some support coming from macaque single unit recordings. Our goal was to investigate the spatial sensitivity of face-selective populations in the macaque when preferred stimuli are presented under more naturalistic circumstances, i.e., in cluttered visual displays. We tested a large sample size of macaques (N=7) using contrast-enhanced, high resolution (4.7T) functional imaging. This whole-brain approach circumvents the sampling limitations associated with electrophysiological studies and facilitates comparisons across species. In an initial experiment, we presented two subjects with three objects at a time (i.e., a face, an object, and a scene), with only one of these objects occupying the central screen location (i.e., at fixation). For example, in the *face* condition, faces were presented behind the central fixation point while objects and scenes were presented in the two peripheral locations to the left and right of the face. Fixation on the central stimulus was reinforced with juice, blocks were removed from the analysis if the subject was not fixating more than 95% of the time. We found that face-selective populations responded selectively to their preferred stimulus category when it was presented in the center of the cluttered stimulus array. However, there was no evidence of this selective response when the preferred stimulus was moved to the more peripheral locations within the stimulus array. Next, in a series of three experiments (n = 5) we interrogated the spatial sensitivity of face-selective populations to faces that were either presented alone or within cluttered displays. We found that face-selective populations are more sensitive to peripheral shifts when the subject is viewing a cluttered stimulus array than when faces are presented in isolation. However, multivariate analyses

revealed that information about the peripheral location of faces within cluttered arrays is encoded by face-selective populations in ventral inferior temporal cortex, raising interesting questions about the time course of these effects. Overall, these findings demonstrate that visual clutter, which more closely simulates the processing demands of natural environments, alters the response profile of face-selective regions.

Disclosures: J. Taubert: None. **S.G. Wardle:** None. **C.T. Tardiff:** None. **A. Patterson:** None. **C.I. Baker:** None.

Poster

P811. Neural Mechanisms of Social Cognition: Higher Mammals

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P811.02

Topic: H.06. Social Cognition

Support: NIH Grant T32GM007739

Title: Visual, Social, Reward, and Eye Movement Activity in Macaque Area F7

Authors: *F. ABOHARB[1], J. SLIWA[2], S. SERENE[1], W. FREIWALD[1];
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Abstract: Primate medial prefrontal cortex is assumed to play a central role in social cognition. For example, a recent fMRI study (Sliwa & Freiwald, 2017) found selectivity for social interactions in this region simply during the observation of videos. Other studies demonstrated reward related activity, and classical work on the control of eye movements found saccade selectivity in the nearby supplemental eye fields. Yet, how these very different properties are encoded within the same population of cells has not been investigated before. To this end, we recorded in two awake, behaving macaque monkeys while they viewed clips falling into 8 categories: socially interacting conspecifics engaging in ethologically relevant behaviors, such as chasing, grooming, mounting, and fighting, or controls, which included conspecifics engaging in non-social behaviors, such as idling and goal directed behavior, such as eating, foraging, and manipulating a toy, as well scenes and moving objects. Recordings were performed using linear S-probes and targeted to area F7. Of 1392 recorded multi-unit sites, 978 (70%) show modulation during one of the three visual epochs (pre-video fixation period, stimulus presentation, and reward). Additionally, sites responded to different task elements: one fifth (297, 21%) were modulated by the appearance of the fixation dot and half the sites (696, 50%) were modulated activity in response to reward delivery. Using a sliding-window encoding method, we found 527 (38%) of sites encode the category of stimulus being presented. Unexpectedly, the neural population also carried signatures of more specific visual events, in particular the occurrence of particularly salient events which could drive responses across categories. To examine this effect

quantitatively, we compared firing rates in the periods following labeled events which included head turns, body turns, and eye contact. Across the population, 273 (20%) responded to either a left or right head turn, 84 (6%) responded to body turns, and 72 (5%) responded to eye contact. We found that 292 sites (21%) modulated activity around subject-generated saccades. Analyzing joint tuning, we found that neurons carried multiple of these very different selectivities. Together, these results show that an area in medial prefrontal cortex, F7, encodes visual information at both a fine-grain and a categorical level, motor, and reward information in a mixed format, thought to maximize information representations in neural populations. Thus functionality in F7 goes beyond a simple label like social interaction selectivity.

Disclosures: F. Aboharb: None. **J. Sliwa:** None. **S. Serene:** None. **W. Freiwald:** None.

Poster

P811. Neural Mechanisms of Social Cognition: Higher Mammals

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Program #/Poster #: P811.03

Topic: H.06. Social Cognition

Support: R01 NS109294
R01 DC012087

Title: Cross modal representation of identity in marmoset hippocampus

Authors: *M. METKE[1], T. TYREE[1], C. T. MILLER[2];
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Abstract: Human and nonhuman primate communication are dominated by visual and acoustic social signals (i.e. faces and voices). Not only are these communicative signals integrated perceptually (i.e. McGurk), categorical information about the individual of the signaler converge to form highly-selective, cohesive cross-modal representations of identity in human hippocampus (i.e. concept cells). Such schema-based representations of identity are likely crucial for the rapid individual recognition needed to effectively navigate human societies. Although human and nonhuman primates enjoy broad similarities in their social cognitive faculties, whether a similar schema-based representation of individual identity would also be evident in the hippocampus of our simian cousins is not yet clear. Here we sought to test this question by presenting awake marmoset monkeys stimuli with faces and vocalizations of multiple conspecifics while recording the single-unit activity across numerous regions of hippocampal. We presented subjects with these social signals either as unimodal or cross-modal stimulus trials. During the latter cross-modal stimulus trials, the identity of the individual face image and vocalization stimulus was either matched or mismatched. We recorded the activity of ~2500 single neurons in the hippocampus of four adult marmosets in these test conditions. Preliminary

analyses of this population revealed three key results. First, while hippocampal neurons were responsive to faces and voices as unimodal stimuli, a population of neurons also exhibited cross-modal integration for these social signals. Second, neurons in marmoset hippocampus exhibited significant changes in the neural firing rate between identity match and mismatch trials suggesting that this population of neurons was sensitive to the individual identity of multiple conspecifics when integrating between visual and auditory modalities. Third, a population of hippocampal neurons was also highly selective to specific individual animals suggesting a potential analog in marmosets to human concept cells. Ongoing analyses are focused on explicating the relationship between these two parallel cross-modal representations of identity in marmoset hippocampus.

Disclosures: M. Metke: None. **T. Tyree:** None. **C.T. Miller:** None.

Poster

P811. Neural Mechanisms of Social Cognition: Higher Mammals

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P811.04

Topic: H.06. Social Cognition

Title: Large scale ECoG recording during social scene watching in awake marmosets

Authors: *H. XU, Y. SU, J. SHARMA, W. MENEGAS, C. TRIMMER, R. LANDMAN, M. SUR, R. SAXE, G. FENG, R. DESIMONE;
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Abstract: Humans can understand social scenes at a glance. Abilities such as these are thought to rely on a network of brain regions often referred to as the social brain. Although people have found cortical patches selective for social content including faces, bodies and social interactions, it is mostly unknown how these domains work collectively dynamically during perception. People with autism spectrum disorders typically have deficits in some aspects of social cognition, but there are disagreements over which brain regions are responsible. Here we “map” the regions of cortex important for social perception and cognition in wild type marmosets which is a new world monkey that is highly social. We showed images and videos with marmoset faces and/or marmosets engaged in several forms of social behavior, together with matched control images and videos. We mapped brain regions responsive to the visual stimuli with micro-electrocorticogram (ECoG) recordings over the posterior and middle temporal cortex and lateral prefrontal cortex in animals seated in a primate chair with head fixed and gaze monitored. Stimulus selectivity was evident in the high gamma band. We localized several face/body/object patches in temporal and frontal lobe, which appear to be in similar locations to areas mapped with fMRI scanning. We also identified patches that appear to be selective for social interactions. Timing data suggests a first posterior to anterior feedforward wave of activation, followed by a

second feedback wave. Moreover, we also recorded with the same ECoGs but with untethered, wireless, recordings in the same animal, during social behaviors in the home cage. With these recordings, we hope to compare patterns of activation during the perception of social stimuli with patterns activated during performance of social behaviors.

Disclosures: H. Xu: None. **Y. Su:** None. **J. Sharma:** None. **W. Menegas:** None. **C. Trimmer:** None. **R. Landman:** None. **M. Sur:** None. **R. Saxe:** None. **G. Feng:** None. **R. Desimone:** None.

Poster

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Topic: H.06. Social Cognition

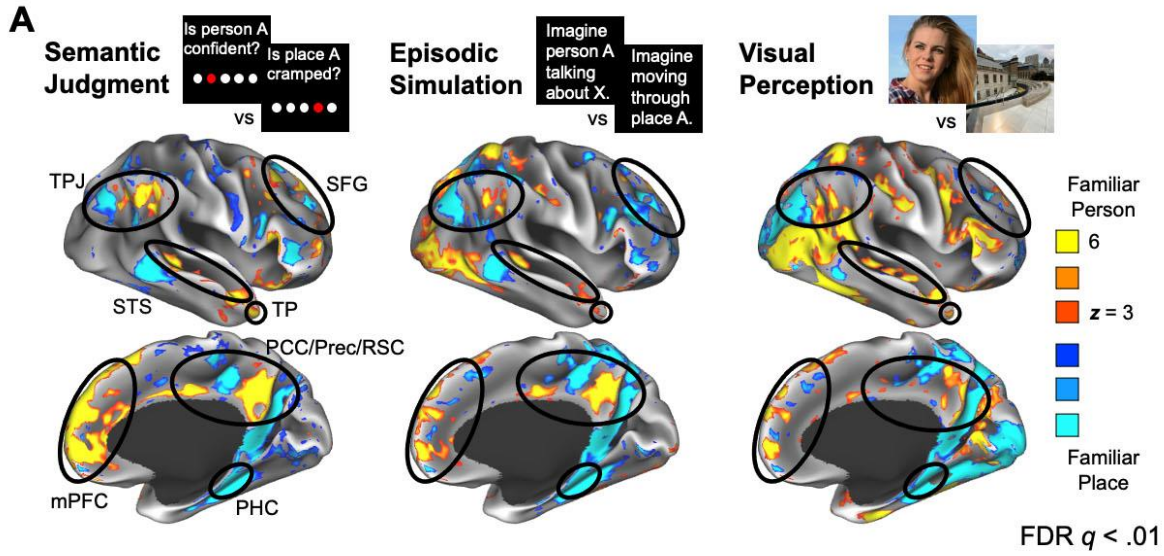
Support: NSF CCF- 1231216

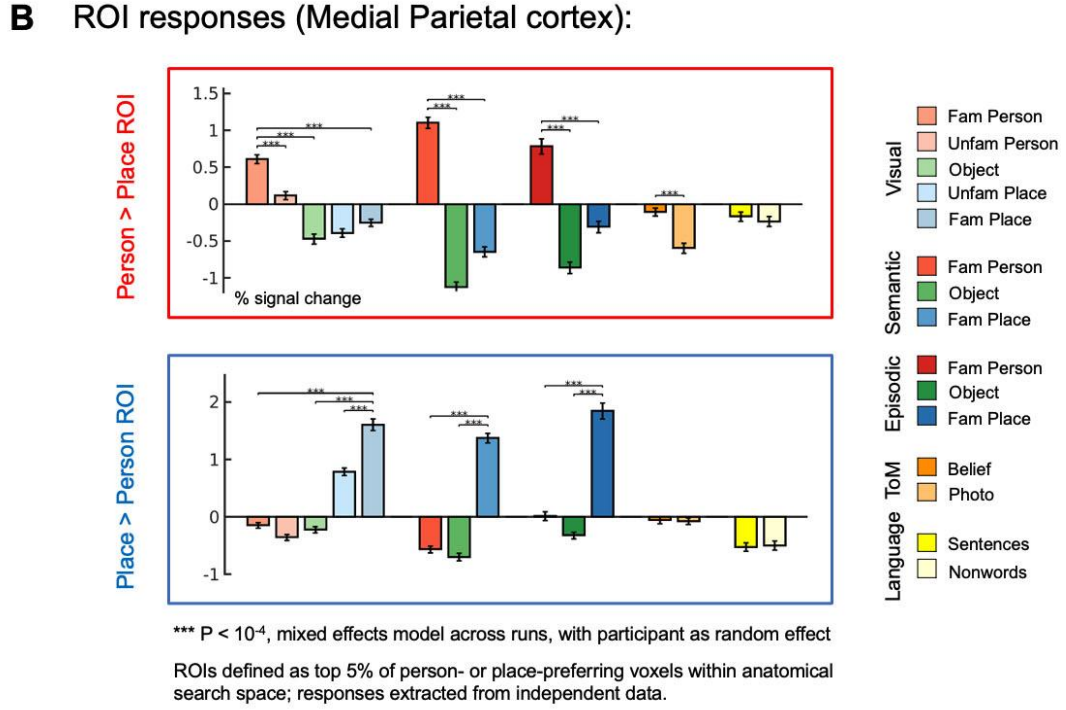
Title: Parallel systems for social and spatial reasoning within the brain's apex network

Authors: *B. DEEN, W. FREIWALD;
Rockefeller Univ., New York, NY

Abstract: In the primate visual system, there is extensive evidence for domain-specificity: brain regions specifically involved in processing certain stimulus categories, such as faces or scenes. To what extent are higher-order brain regions, and cognitive processes, domain-specific? Here we consider areas within the default mode or apex network, argued to be anatomically positioned at the top of the cortical hierarchy, and implicated in a range of abstract cognitive processes, including social cognition, spatial navigation, and episodic recall. To precisely characterize functional organization in individual human brains, we scanned $N = 10$ participants using fMRI on a range of perceptual and cognitive tasks, across three scan sessions (7.5 hours per participant). Tasks involved visual perception, semantic judgment, and episodic simulation of close familiar people and places, and everyday objects. We find evidence for parallel subnetworks within the apex network that respond specifically during conditions involving familiar people and places, across visual, semantic, and episodic tasks. Person- and place-specific areas are identified across multiple zones of association cortex, including medial prefrontal cortex, medial parietal cortex, the superior frontal gyrus, and the temporal-parietal junction (FSL's improved linear model, $q < .01$). Within each zone, we observe a pattern of interdigitation between person- and place-specific areas. Across zones, regions with a given category preference are specifically functionally connected with one another in resting-state data, while regions with differing category preferences are not functionally connected. These two parallel, functionally specialized apex subnetworks were observed reliably across all participants.

These results demonstrate that category-specificity is not limited to perceptual systems, but is pervasive across brain regions positioned at the top of the cortical hierarchy. They suggest that our ability to reason about other people and places rely on parallel cognitive and neural mechanisms.





Disclosures: B. Deen: None. **W. Freiwald:** None.

Poster

P811. Neural Mechanisms of Social Cognition: Higher Mammals

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NIH R01MH111629
NIH R37HD090153
CSC 201906140133
ECNU YBNLTS2019-025

Title: Face mechanisms differ between gaze at live faces and gaze at webcam faces: Comparisons of eye-tracking, electroencephalography, neuroimaging, and cross-brain coherence between dyads

Authors: *N. ZHAO[1], X. ZHANG[2], J. A. NOAH[2], M. TIEDE[5], J. HIRSCH[2,3,4,6,5];
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Abstract: A common experience secondary to the COVID-19 lockdown has been the reliance on web-cam platforms such as Zoom for conducting business and maintaining interpersonal relationships. Experience tells us that webcam faces and interactions are not the same as real face-to-face interactions, even though the faces and the people are the same. Why? We test the hypothesis that variations in visual sensing are related to interactive face processing and cross brain synchrony. A multimodal methodological approach was applied to investigate dyads in two conditions: face-to-face gaze and webcam gaze using functional near-infrared spectroscopy (fNIRS), with simultaneous electroencephalography (EEG) and eye-tracking. Stranger dyads (n=28 participants, 14 dyads) engaged in live mutual face-watching (real face and same-face on webcam) using a previously described[1] face-watching paradigm. Eye-tracking showed longer ($p < 0.001$) dwell times and reduced ($p<0.001$) horizontal positional variation for face-to-face relative to the webcam condition consistent with variations in visual sensing mechanisms. Additionally, the N170 event related potential showed a greater negative amplitude for real faces than for web faces consistent with an early advantage for the real face stimulus. Spectrum power in the theta band was also higher in the face-to-face condition consistent with oscillatory specificity for real-face processing. Neuroimaging findings for [face-to-face > gaze at webcam face] included bilateral and dorsal temporal regions, whereas activity for the [gaze at webcam face > face-to-face gaze] showed robust left activity in ventral regions of temporal, occipital, and parietal areas ($p<0.05$) consistent with regional mechanistic specificity for the real face condition. Inter-brain synchrony was also greater ($p = 0.05$) for the real-face dyadic condition for signals between angular gyrus, occipital/temporal gyrus, and somatosensory cortex consistent

with neural coupling effects specific to the real faces. Further, brain-to-brain synchrony for the somatosensory regions was positively correlated with visual sensing (dwell time) (r = 0.66) for face-to-face gaze and not for web cam gaze. Findings support the hypothesis that visual sensing mechanisms, neuro electrical oscillations, and visual face-processing pathways play an important role in live face-to-face interactions. Together, these converging lines of simultaneous multimodal evidence reveal early to late neurobiological processes that differentiate face watching pathways based on live face-to-face or web cam presentations.
[1]Noah, et al, 2020.

Disclosures: N. Zhao: None. **X. Zhang:** None. **J.A. Noah:** None. **M. Tiede:** None. **J. Hirsch:** None.

Poster

P811. Neural Mechanisms of Social Cognition: Higher Mammals

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Program #/Poster #: P811.07

Topic: H.06. Social Cognition

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NIH R01MH11629
NIH R01MH111629
NIH R37HD090153
NIH NIMH 1F 30MH116626

Title: Simultaneous fNIRS-EEG reveals theta band differences spatially localized to the supramarginal gyrus during Face-to-face gaze at a human as compared to a robot-control

Authors: *M. KELLEY[1], J. A. NOAH[2], X. ZHANG[2], T. PARKER[1], J. HIRSCH[2,3,4,5,6];
[1]Interdepartmental Neurosci. Program, [2]Psychiatry, [3]Neurosci., [4]Comparative Med., Yale Sch. of Med., New Haven, CT; [5]Med. Physics and Biomed. Engin., Univ. Col. London, London, United Kingdom; [6]Haskins Labs., New Haven, CT

Abstract: While the neural and hemodynamic processing mechanisms of face-to-face interaction have previously been explored[1], the related temporal dynamics of these mechanisms are less well understood. Here, functional Near Infrared Spectroscopy (fNIRS) and electroencephalography (EEG) were acquired simultaneously to assess the temporal features of neural responses during eye-contact with another person and a robot control. **Methods.** Subjects (n=11) performed a 3-minute eye-contact task twice with two partners: a real human and a robot bust with eyes that blink and move but which otherwise lacks naturalistic features or behaviors. The task consisted of alternating 15-second (s) blocks of eye-contact with partner and of rest. Eye-contact was either allowed or prevented via a “smart glass” divider which separated participants from their

partner, and which had electronically controlled transparency or opacity. Eye-contact blocks were divided into 850ms EEG events with randomized no-eye-contact intertrial intervals of 200-400ms. Hemodynamic data were acquired via a 124-channel Shimadzu LABNIRS system and electromagnetic data were acquired in a modified 10-10 layout via a 32-channel g.USBamp system. **Data analysis and Results. fNIRS:** A general linear model (GLM) of task and rest blocks was convolved with the canonical hemodynamic response function to identify regions which differed between human and robot eye-contact. Past findings[1] of differences in a supramarginal gyrus and angular gyrus cluster (SMG/AG) were replicated (peak T=4.49, $p<0.05$). **EEG:** Data were filtered to 0.01-40 Hz, epoched to -100—850 ms, and decomposed via an independent component analysis. A normal mesh model boundary element method was used for component source localization and the GLM-defined SMG/AG cluster was used to constrain solutions. Components with <50% residual variance for the GLM-informed source were identified, clustered by scalp topography, and limited to one component per subject. Group time-frequency wavelet analysis of the SMG/AG component cluster was compared across partners. Significance was assessed via permutation testing. The SMG/AG cluster showed equivalent theta band oscillations (4-8hz) early in eye-contact epochs for both partners, but differences emerged later in the epoch (540ms-740ms). This preliminary data suggests that temporal differences in theta band activity in the SMG are involved in processing live face stimuli as social partners.
[1]Kelley et al, *Fronts in Robotics.* (2020)

Disclosures: M. Kelley: None. **J.A. Noah:** None. **X. Zhang:** None. **T. Parker:** None. **J. Hirsch:** None.

Poster

P811. Neural Mechanisms of Social Cognition: Higher Mammals

Location: Virtual Only

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Program #/Poster #: P811.08

Topic: H.06. Social Cognition

Support: NIDA grant

Title: The hippocampus represents abstract social place: encoding and decoding evidence in independent samples

Authors: *M. SCHAFER[1], P. KAMILAR-BRITT[1], V. SAHANI[1], S. J. MOELLER[2], V. GABBAY[3], Y. HURD[1], K. BACHI[1], D. SCHILLER[1];
[1]Mount Sinai Sch. of Med., New York, NY; [2]Stony Brook Univ., Stony Brook, NY; [3]Albert Einstein Col. of Med., New York, NY

Abstract: Background: Humans adeptly navigate complex social landscapes. **T**his capacity may depend upon social mapping: a hippocampal centered network may organize information

about social relationships into a multidimensional map, similar to that of physical space. In this framework, other people can be thought of as occupying abstract social places based on their locations along social dimensions, such as power and affiliation.
Methods: To test for such a brain representation, functional magnetic resonance imaging (fMRI) was collected in two samples of healthy individuals (n=27, 12 female and n=16, 6 female) while they completed a choose-your-own-adventure game where they interacted with fictional characters. Participants' relationships (i.e., decisions) with each character were modeled as sequences of locations in an abstract two-dimensional social space of power and affiliation. These locations were then compared to brain patterns.
Results: Hippocampal fMRI signals consistent with a social place representation were observed in a representational similarity regression with both region-of-interest (ROI) and whole-brain searchlight approaches, as well as an ROI-based decoding probability analysis. This effect was present in both samples, and was not explained by other measures of task behavior or other kinds of categorical or continuous task-based social information (e.g., character identity or familiarity), or by demographic variables (i.e., age and sex).
Conclusions: Our findings suggest that other people's social places are represented in the human hippocampus. Future work will ask how social place representations are combined into maps and used for flexible behavior - so-called "social navigation" - and how alterations in these representations may be implicated in disorders that feature both hippocampal and social dysfunction.

Disclosures: M. Schafer: None. **P. Kamilar-Britt:** None. **V. Sahani:** None. **S.J. Moeller:** None. **V. Gabbay:** None. **Y. Hurd:** None. **K. Bachi:** None. **D. Schiller:** None.

Poster

P828. Striatal and Corticostriatal Circuits

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P828.01

Topic: H.08. Learning and Memory

Support: MOST 110-2811-B-182-503
MOST 109-2320-B-182-004

Title: Micu1 deficiency leads to cognitive impairment and anxiety-like behavior

Authors: S.-M. DENG, G.-J. HUANG;
Grad. Inst. of Biomed. Sci., Chung Gung Univ., Taoyuan, Taiwan

Abstract: The regulation of calcium by mitochondria is an essential event for maintaining cellular physiology. Studies have provided evidence that the mitochondrial calcium uniporter (MCU) regulates calcium uptake into mitochondria matrix. Mitochondrial calcium uptake

protein 1 (Micu1) has been demonstrated to play a critical role in regulation of MCU activity. Human patients carrying loss-of-function mutations in Micu1 suffer from muscle weakness, ataxia, and learning difficulties. Despite of numerous cell culture studies, there is limited information from knockout mice models. In this study, we used Cre-loxp system to generate Micu1 conditional KO mice, and various behavioral tests and electrophysiology will be accessed to measure behavioral phenotypes and long term potentiation (LTP). In order to understanding the morphology and functions of mitochondria, the electron microscopy imaging and mitochondrial oxygen consumption rate had been detected. Our pilot results showed mice without Micu1 in the brain exhibiting weaker motor function, higher level of anxiety, and poor performance in learning, which is in line with the symptoms of human patients. To answer the mechanism of phenotype alteration, we will use inducible knockout mice model and primary culture of neuron and glia cell, to investigate the role of Micu1 in both cell type-specific and time dependent manner. To better understating whether restoration of Micu1 expression can rescue the anxiety and learning behavior, we will inject AAV viral vector to overexpress Micu1 in the specific brain regions and measure mice behavior. This analysis shall give us a better understanding of how Micu1 influences the downstream expression profile and provides potential treatment of Micu1 deficiency.

Disclosures: S. Deng: None. **G. Huang:** None.

Poster

P828. Striatal and Corticostriatal Circuits

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P828.02

Topic: H.08. Learning and Memory

Support: CONACYT fellowship 620183 to A-L H.
Ciencia Básica CONACyT Grant 220412
Fronteras de la Ciencia CONACyT Grants 2022 and 154039
DGAPA-PAPIIT-UNAM Grants IA200815, IN226517, IN203420
Moshinsky fellowship to F.T

Title: The striatal neurons contribute to specific behavioral updates in a classical conditioning task

Authors: *H. ALATRISTE-LEON, J. O. RAMÍREZ-JARQUÍN, F. TECUAPETLA;
Univ. Nacional Autonoma de Mexico, Mexico City, Mexico

Abstract: The striatum, the main input to the basal ganglia, is implicated in the control of behavioral flexibility, movements, and learning mediated by reinforces. The spiny projections neurons (SPNs) and cholinergic interneurons (ChIs) have been suggested to control the

behavior´s update of animals to new contingencies in instrumental conditioning, but little is known about how these neurons participate to adapt behaviors to changes in classical conditioning. Therefore, this study aims to evaluate the contribution of SPNs and ChIs activity on the behavioral flexibility of mice in a classical conditioning task. Preliminary results: We standardized a head-fixed task with four different auditory stimuli paired to four different outcomes (stim 1 to big, stim 2 to small, stim 3 to neutral, stim 4 to air puff), and after verifying the proper acquisition of the tones (monitoring anticipatory responses), we switch the tones-outcomes in a reverse mode. As the first experiment and to evaluate the contribution of striatal activity on the behavior update, we optogenetically inhibited the striatum, using AAV-Syn-Arch, during the switch of the tones. Later as a second experiment and to evaluate the ChIs contribution on the behavior update, we investigated the optogenetic inhibition of ChIs, using AAV-DIO-Arch in ChAT-Cre mice. Lastly, to investigate how the dynamic of striatal projection cells may read the cholinergic modulation, we are recording the striatal activity by expressing the genetically encoded calcium indicator (GCaMP6f) and monitoring the striatal activity by micro-endoscopes in the striatum of either D1-Cre or A2a-Cre mice. Our preliminary conclusions suggest that the SPNs and ChIs contribute to specific updates, in a classical conditioning task, depending on the values of the outcome's changes.

Disclosures: H. Alatriste-Leon: None. **J.O. Ramírez-Jarquín:** None. **F. Tecuapetla:** None.

Poster

P828. Striatal and Corticostriatal Circuits

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P828.03

Topic: H.08. Learning and Memory

Support: Foundation for OCD Research, "Defining a molecular-circuit pathophysiology of OCD for targeted intervention"

Title: Investigating amygdala striatal projections during positive and negative reinforcement regimes.

Authors: *E. DIAZ-HERNANDEZ[1], E. ILIAKIS[2], J. GALANAUGH[2], K. CHOI[3], M. V. FUCCILLO[4];
[1]Pennsylvania State Univ., Philadelphia, PA; [2]Upenn, Medicine School, PA; [4]Neurosci., [3]Univ. of Pennsylvania, Philadelphia, PA

Abstract: The selection of current actions is under control of both positive and negative reinforcement. Alterations in the balance between these reinforcement systems can create conflicts and lead to compulsive, maladaptive behavioral responding. While dorsomedial striatal (DMS) neurons play a central role during action selection, it is unclear how this structure

differentially contributes to positive and negative reinforcement behavior. One DMS afferent population strongly related to emotional value is the amygdala which exhibits robust modulation to positive and negative stimuli. Here we examine amygdala-DMS circuits and their role during positive and negative reinforcement regimes. We start by mapping relevant circuits using two parallel approaches: (1) retro-cre AAV injected in DMS and AAV-Flex-Synaptophysin-mRuby injected into amygdala to assess how amygdala-DMS projections are distributed across striatum; (2) retrograde tracing using cholera toxin (CTB) in different DMS regions to assess how amygdalar afferents distributed by anterior-posterior striatal target. In order to determine the in vivo contributions of DMS-targeting amygdala populations, we designed an operant task that elicits positive and negative reinforcement within the same session. Using tone discrimination, the animal is directed to perform either positive operant reinforcement (lever press for chocolate) or negative reinforcement (lever press to avoid shock). Using in vivo electrophysiology single-unit activity and opto-tagging in freely moving mice, we recorded striatal-targeting amygdala neurons to identify how this population is differentially modulated by positive and negative reinforcement strategies. Preliminary anatomical results reveal a specific region where amygdala-DMS projections are concentrated while preliminary in vivo recordings exhibit divergent neural coding for amygdalar-DMS neurons depending on reinforcement type. This work will provide insight into potential circuit mechanisms that balance positive and negative reinforcement strategies.

Disclosures: E. Diaz-Hernandez: None. **E. Iliakis:** None. **J. Galanaugh:** None. **K. Choi:** None. **M.V. Fuccillo:** None.

Poster

P828. Striatal and Corticostriatal Circuits

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Support: NIH DA044297
NIH 5T32NS096050-24
Brain and Behavior Foundation (NARSAD)
Emory University Research Council
Children's Healthcare of Atlanta
Silvio O Conte Center

Title: Melanocortin-4 receptor control of striatal-dependent action selection

Authors: *E. C. HEATON[1], G. TRINOSKEY-RICE[2], S. L. GOURLEY[3];
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Abstract: Goal-directed action refers to behaviors that are dynamic, sensitive to unexpected events, and require the dorsomedial striatum (DMS). Meanwhile, habitual behaviors are reflexive and unchanging and rely on the dorsolateral striatum (DLS). Molecular factors underlying an organism's ability to flexibly shift between goal-directed and habitual behavior are incompletely understood. We recently discovered that striatal melanocortin-4 receptor (*Mc4r)* expression correlates with this behavioral flexibility in adult male and female mice. To identify functional consequences, we used viral-mediated gene silencing to reduce *Mc4r*. *Mc4r* knockdown in the DMS enhanced the ability of mice to select actions based on reward likelihood and value, while reduction in the DLS facilitated habit formation. Thus, MC4R *presence* serves as a brake on the functions of distinct striatal subregions in decision making. Changes in *Mc4r* expression modulates the activity of medium spiny neurons, leading to the hypothesis that chemogenetic manipulation of *Mc4r+* DMS neurons would impact expression of behavioral flexibility. Stimulation of *Mc4r+* DMS neurons via Gq-coupled Designer Receptors Exclusively Activated by Designer Drugs (DREADDs) facilitated animals' ability to select actions based on reward likelihood. Meanwhile, inhibition via Gi-DREADDs rendered animals insensitive to changes in reward likelihood, promoting habits. These results reveal that striatal MC4R may be a key factor in sustaining *versus* "breaking" habits, and thus could serve as a target for treating maladaptive habits that contribute to neuropsychiatric disease.

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Poster

P828. Striatal and Corticostriatal Circuits

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Title: Accumbal D1 and D2 medium spiny neurons control distinct learning parameters in complex behavior

Authors: *J. E. ZACHRY[1], M. G. KUTLU[4], P. MELUGIN[2], S. KELLY[3], S. CAJIGAS[2], A. U. ISIKTAS[6], E. S. CALIPARI[5];
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Abstract: Value-based decision-making is at the core of nearly all motivated behaviors and requires the ability to associate outcomes with specific actions and make adaptive decisions about future behavioral action. At the core of value-based decision-making and reinforcement is the nucleus accumbens (NAc), which is integrally involved in learning, selecting, and executing goal-oriented behaviors. The NAc is a heterogeneous population primarily composed of D1 and D2 medium spiny projection (MSN) neurons that are thought to have opposing roles in behavior, in which D1 MSNs promote reward and D2 MSNs promote aversion. By expressing channelrhodopsin selectively in D1- and D2- populations (using D1-Cre and A2A-Cre mice) in the NAc core, we show that mice will nose poke for optical self-stimulation of both cell types, suggesting D2-MSN activity is not inherently aversive. While optogenetic approaches give some information about how cellular activation can modulate behavior, they eliminate the temporally specific neural activity patterns that encode information in behaving animals. To understand how real-time activity in these populations is linked to behavioral execution, we expressed the genetically encoded calcium indicator (GCaMP6f) within D1 and D2 MSNs coupled with in vivo fiber photometry to record from these cell populations in awake and behaving animals during pavlovian and operant conditioning tasks. Utilizing complex reinforcement schedules that allow dissociation of stimulus value, outcome, cue learning, and action, we show that D1 MSNs respond to the presence and intensity of unconditioned stimuli – regardless of value. Conversely, D2 MSNs respond to a mismatch between what is expected and what is received and thus encode errors in prediction in a value-independent fashion. To establish a causal link between the information encoded within these neuronal populations and behavioral output, we next expressed an inhibitory opsin (halorhodopsin) selectively in either D1- or D2-MSNs to inhibit these cells at specific time points during a fear conditioning task. We show that inhibition at select time points, either by inhibiting D2-MSN activity during the predictive cue or D1-MSN activity during the shock outcome, disrupts learning of the association. We thus provide foundational evidence for the discrete aspects of information that are encoded within these cellular populations.

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Poster

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Title: Dopamine release in the nucleus accumbens core signals valence-free perceived saliency

Authors: *M. KUTLU[1], J. E. ZACHRY[1], P. MELUGIN[1], S. CAJIGAS[1], M. CHEVEE[1], S. KELLY[1], L. TIAN[3], C. SICILIANO[1], E. S. CALIPARI[2];
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Abstract: Significant work has aimed to define the precise information encoded by dopaminergic projections innervating the nucleus accumbens. Prevailing models are based on reward prediction error (RPE) where dopamine encodes associations between rewards and predictive cues and responds to perceived errors in predictions to update behavior. However, RPE cannot describe multiple phenomena to which dopamine is inextricably linked, such as behavior driven by aversive and neutral stimuli. We combined complex reinforcement tasks with direct, subsecond dopamine monitoring in the nucleus accumbens (NAc) of mice, machine learning, and computational modeling to describe behavior across multiple contingencies reinforced by differentially valanced outcomes. We show that dopamine release only conforms to RPE predictions in a subset of learning scenarios but fits valence-independent perceived saliency encoding across conditions. Together, we provide an extended, comprehensive framework for accumbal dopamine release in valence-independent behavioral control.

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Poster

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Topic: H.08. Learning and Memory

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Title: The impact of oxytocin on dopamine signaling in the dorsal striatum of anesthetized rats

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Abstract: Oxytocin (OXT) is involved in many aspects of brain function. Current research suggests that, due to the possible modulatory effect of OXT on learning and memory circuitry, OXT treatment for depression and addiction could be beneficial. Defining the neural mechanisms of OXT is crucial to understanding how OXT may be therapeutic for certain psychological disorders. Dopamine (DA), a neurotransmitter highly implicated in learning and memory, may be modulated by OXT. In general, OXT and DA receptors are located within the same brain regions. More specifically, there is overlap in the expression of OXT and DA receptors in the dorsal and ventral striatum. Furthermore, OXT fibers innervate DA neurons. The neuroanatomical association between the dopaminergic and oxytonergic systems provides a neurological mechanism by which OXT could potentially alter DA neurotransmission. METHODS: Male Sprague-Dawley rats were anesthetized, immobilized in a stereotaxic frame (Stoelting Co. Wood Dale, IL, USA), skin/fascia was cleared, and small holes drilled in the skull for placement of electrodes. A reference electrode (Ag/AgCl) was placed just below dura and a fast-scan cyclic voltammetry (FSCV) electrode was placed in the dorsal striatum (+1.0 AP, +2.0 ML, -5.0 DV). Subsequently, a twisted bipolar stimulating electrode (Plastics One, Roanoke, VA) with ~1 mm tip separation was placed just above the medial forebrain bundle (-4.6 AP, +1.4 ML, -7.0 DV). Constant current, biphasic stimulus pulses (60 Hz, 60 pulses, 300 µA) were applied (NL 800, Neurolog, Medical Systems, Great Neck, NY) and the stimulating electrode was incrementally lowered until a robust DA signal was evoked. Once clear and consistent DA signals were observed, intranasal OXT (1.0 µg/kg or an equivalent volume of saline for controls) was administered. RESULTS: Intranasal OXT increased DA release in the dorsal striatum, relative to controls (saline-treated rats). CONCLUSIONS: OXT modulates DA neurotransmission in the dorsal striatum. These results suggest OXT is potentially therapeutic for psychological disorders via, in part, the modulation of DA signaling. FUNDING: Eastern Washington University Department of Biology and David Daberkow Faculty Research Grant (EWU FGRCW 2018-19).

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Poster

P828. Striatal and Corticostriatal Circuits

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Program #/Poster #: P828.08

Topic: H.08. Learning and Memory

Title: The computational role of the basal ganglia in human cognition: A comparison of cognitive architectures through dynamic causal modeling

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Abstract: Regulatory input from the basal ganglia (BG) to the prefrontal cortex (PFC) performs an important role in cognition. Theoretical models of this input, however, disagree on its computational properties, characterizing the majority of BG input as either direct (directly influencing PFC activity), or modulatory (indirectly influencing PFC activity through the gating of signals from other cortical areas). To determine the computational nature of these inputs in cognition, three alternate configurations of BG connectivity were tested using fMRI data from 200 individuals in the Human Connectome Project acquired during seven representative cognitive tasks and two resting state sessions. The structure of each model configuration (Direct, Modulatory, and Mixed (both Direct and Modulatory)) was based on a high-level cognitive architecture known as the Common Model of Cognition (CMC). The CMC seeks to explain abstract computations underlying cognition in terms of five interacting brain modules. Connectivity between these brain modules, specifically those containing the BG and the PFC, were modified to reflect these three models of BG connectivity. The Direct model had a direct, bidirectional connection between BG and PFC modules (basic CMC configuration). The Modulatory model lost this bidirectional connection, and instead contained modulatory connections from the BG to other cortical modules in this architecture. The Mixed model contained both direct and modulatory connections. Model configurations were then tested using Dynamical Causal Modeling, a mathematical modeling procedure that takes the structural and functional framework provided by cognitive architectures to measure if activity in certain brain regions is affecting another. Here, we show that for each of the seven cognitive tasks and the resting state, the Mixed model consistently outperformed Direct and Modulatory models. The models that performed second to the Mixed model depended on the specific cognitive task, suggesting an ability for the BG to flexibly adapt to various task demands. Taken together, the current data provide evidence for a likely set of core computations that the BG uses to differentially regulate cortical activity, and are present during task and task-free behavior.

Disclosures: H. Hake: None.

Poster

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Title: Distinct Roles of Dopamine D1 and D2 Receptor-Expressing Neurons in the Nucleus Accumbens for a Strategy Dependent Decision Making

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Abstract: To optimize decision making, animals need to execute not only a strategy to choose a good option but sometimes also one to avoid a bad option. A psychological study indicates that positive and negative information is processed in a different manner in the brain. The nucleus accumbens (NAc) contains two different types of neurons, dopamine D1 and D2 receptor-expressing neurons which are implicated in reward-based decision making and aversive learning. However, little is known about the neural mechanisms by which D1 or D2 receptor-expressing neurons in the NAc contribute to the execution of the strategy to choose a good option or one to avoid a bad option under decision making. Here, we have developed two novel visual discrimination tasks for mice to assess the strategy to choose a good option and one to avoid a bad option. By chemogenetically suppressing the subpopulation of the NAc neurons, we have shown that dopamine D2 receptor-expressing neurons in the NAc selectively contribute to the strategy to avoid a bad option under reward-based decision making. Furthermore, our optogenetic and calcium imaging experiments indicate that dopamine D2 receptor-expressing neurons are activated by error choices and the activation following an error plays an important role in optimizing the strategy in the next trial. Our findings suggest that the activation of D2 receptor-expressing neurons by error choices through learning enables animals to execute the appropriate strategy.

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Poster

P845. The Role of Oscillations

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Program #/Poster #: P845.01

Topic: H.08. Learning and Memory

Title: Oscillations regulate attractor dynamics within neural circuits

Authors: *A. LIBBY[1], T. BUSCHMAN[2];
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Abstract: Previous knowledge improves perceptual accuracy by overcoming sensory noise. This is thought to be due to attractor dynamics within sensory cortex that pull noisy stimuli into a sensible percept (Burak and Fiete 2012; Litwin-Kumar and Doiron 2012). However, when a neural representation falls into an attractor, it can become 'stuck', limiting the the network's ability to respond to changes in the environment. These two needs are in tension - strong attractor dynamics facilitate perception, but weak attractor dynamics allow the network to be sensitive to new sensory inputs. It is not clear how the brain balances (or switches) between having strong and weak attractors. Here, we propose oscillations may allow the brain to sample between these two dynamical regimes. To test this hypothesis, we embedded strong attractor dynamics within the excitatory connections of a recurrent neural network. These attractors facilitated 'perception' of noisy/weak sensory drive. However, once in an attractor, the network required extra sensory drive to escape the attractor. Attractors could be weakened by increasing inhibition (Wong and Wang 2006); however, this then weakened attractor dynamics. We found that oscillations in inhibition could balance between these two regimes. At the peak of oscillations (when inhibition is high), a weak sensory drive can move the network between attractors. In contrast, at the trough of oscillations (when inhibition is low), attractors are strong (facilitating perception), but transitions between attractors only occur for very large sensory inputs. In this way, oscillations allow the network to 'breathe' between a dynamical regime that emphasizes attractor dynamics and a regime that is sensitive to external inputs. In this way, the model captures previous experimental results that found sensory processing (Mathewson et al. 2011; Dugué, Marque, and VanRullen 2011), visual attention (Busch and VanRullen 2010; Buschman and Miller 2009), short-term memory (Siegel, Warden, and Miller 2009), and memory retrieval (Pfeiffer and Foster 2015) occur at specific phases of oscillations. In addition, our results provide a mechanistic reason for oscillations, suggesting they alter the dynamic landscape of recurrent networks, allowing them to sample between a strong attractor regime exploiting prior knowledge and a flat regime exploring new states.

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Poster

P845. The Role of Oscillations

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Program #/Poster #: P845.02

Topic: H.08. Learning and Memory

Title: Neural oscillations in fronto-parietal network encode learning of new rules

Authors: *S. TAFAZOLI[1], C. I. JAHN[2], N. T. MARKOV[4], C. J. MACDOWELL[1], T. BUSCHMAN[3];

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Abstract: To act intelligently in a situation, we must learn what rules to follow for that context. Then, as the situation changes, we must learn new rules. To understand the neural mechanisms of learning rules, we trained monkeys on a task that required them to learn a new 'rule' on each block of trials. On each trial, the monkeys chose between three colored squares (with the color randomly drawn from the full color wheel). The 'rule' for that block of trials guided the animals response -- their goal was to choose the square with the closest color to a 'target' color. Importantly, the target color was not instructed. Instead, the animals had to learn this new rule through trial and error. Once the animals learned the target color, it changed abruptly, requiring the animal to detect the change and learn a new rule. Consistent with animals using feedback to learn the rule, a Q-learning model predicted the monkeys' choices. Previous work has found that rule representations are distributed across multiple brain regions including prefrontal cortex (PFC) and posterior parietal cortex (PPC). To assess how this representation emerges during learning of a new rule, we used large-scale simultaneous recordings across the fronto-parietal network and found that oscillations in local field potentials (LFPs) in frontal and parietal areas change with learning. Gamma-band oscillations (35-55 Hz) were increased in PFC immediately after a change in rule, consistent with gamma oscillations increasing the bottom-up flow of sensory information when learning the rule. In addition, the power of alpha-band oscillations (10 Hz) was correlated with the animal's uncertainty about the current rule. Alpha oscillations have been associated with suppression of a rule, suggesting the current rule was inhibited when the animal was uncertain. Our results build on previous work showing oscillations support rule representations and argue that these representations can develop during learning.

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Poster

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Title: Perineuronal nets in the dorsomedial striatum contribute to behavioral dysfunction in mouse models of excessive repetitive behaviors

Authors: *B. A. BRIONES, M. N. PITCHER, W. T. FLEMING, A. LIBBY, E. J. DIETHORN, A. E. HAYE, C. J. MACDOWELL, A. D. ZYCH, R. C. WATERS, T. J. BUSCHMAN, I. B. WITTEN, E. GOULD;
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Abstract: Excessive repetitive behavior is a debilitating symptom of several neuropsychiatric disorders. Parvalbumin-positive (PV+) inhibitory interneurons in the dorsal striatum have been linked to repetitive behavior, and most of these cells are surrounded by perineuronal nets (PNNs), specialized extracellular matrix structures. We examined four mouse models of excessive repetitive behavior (BTBR, Cntnap2, Shank3, prenatal valproate treatment) and found a greater percentage of PV+ interneurons with PNNs in the dorsomedial striatum (DMS) compared to controls. In BTBR mice, we found that excessive repetitive behavior was associated with low numbers of dendritic spines on medium spiny neurons (targets of PV+ interneurons), and differences in neuronal oscillations as well as inhibitory postsynaptic potentials (IPSPs). Reduction of DMS PNNs to control levels normalized repetitive behavior and altered dendritic spine density and IPSPs. These findings suggest that cellular abnormalities in the DMS may contribute to maladaptive repetitive behaviors and that manipulating PNNs can restore typical structure and function.

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Poster

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Title: Low frequency spike-field coupling for error monitoring in medial frontal cortex: Empirical findings and biophysical modeling

Authors: *B. HERRERA[1], A. SAJAD[2], S. P. ERRINGTON[3], J. D. SCHALL[4,3], J. J. RIERA[1]; [1]Biomed. Engin., Florida Intl. Univ., Miami, FL; [2]Psychological Sci., [3]Psychology, Vanderbilt Univ., Nashville, TN; [4]Biol., York Univ., Toronto, ON, Canada

Abstract: The medial frontal theta rhythm has been linked to cognitive control and error monitoring (Cohen, *Trends Neurosci*, 2014). However, its mechanisms and functions remain uncertain. Here, we bring new insights combining computational simulations with neurophysiological data. First, we simulated 100 layer 5 pyramidal cells (L5PCs) receiving background Poisson-like spike trains on their apical dendrites. We found that low frequency oscillations in the membrane potentials corresponding to the theta and low-alpha range can inherently arise from such a random stimulation, even with these L5PCs completely disconnected. However, differences in phase make this oscillation unobservable within local field potentials (LFPs). Short (~10 ms) interruption of these inputs resulted in a phase-reset, creating broad frequency band spectral changes of the LFPs. Resetting was coincident with a transient elevated spike rate across all simulated neurons. Our previous work has reported similar spectral changes in the theta-alpha frequency band and increases in putative PCs spiking in neurophysiological data recorded from two monkeys performing a saccade stop-signal task. In this task, subjects were required to generate a saccade to a peripheral target, but to inhibit this planned saccade when a stop-signal appeared. Errors occurred when monkeys generated saccade despite the appearance of stop-signal. Motivated by the theoretical results, we quantified the strength of spike-field phase synchronization of 293 neurons recorded across 16 penetrations sampling neural activity across all layers of the Supplementary Eye Field (SEF) around the period in which a saccade was generated. Spike-field phase-synchronization, indexed by pairwise-consistency measure (Vinck et al., *J Comput Neurosci*, 2012) was calculated for the spiking activity of each neuron relative to its corresponding channel LFP. Overall, spike-field phase-synchronization of putative pyramidal neurons was highest for the theta and low-alpha bands (5-10 Hz) compared to other frequency bands. Furthermore, we observed a relative increase of this phase-synchronization on error compared to correct trials. Preliminary results found this relationship for neurons in all layers, while our simulations suggest the possibility of an enhanced input at the level of the L5PC basal dendrites when an error is made. With this, ongoing work aims to investigate whether increases in phase-synchronization may reflect further input to the deep cortical layers involved in error processing.

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Poster

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Title: Transient beta and gamma bursts in simulations and the mouse basolateral amygdala during the open field test

Authors: *S. CHO, J. CHOI;
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Abstract: Neural oscillations reflect the phenomenon by which the individual neurons dynamically interconnect to rhythmically shift their overall activity as a neuronal ensemble. The spectral components of these oscillations have been traditionally characterized as increased power at specific frequency bands over sustained periods of time. Recent studies, however, suggest that such sustained oscillations can be an accumulation of temporally transient and spectrally specific burst events observed at a single trial level. In showing the physiological relevance of beta and gamma bursts in sensorimotor and cognitive systems of different species, multiple algorithms were used to distinguish temporally precise and possibly causal correspondence between neural activities and behaviors. Yet, no rigorous comparison has been made for these algorithms to address degrees to which the methodological differences result in disparate analysis outcomes.

We here examine five burst detection algorithms, namely bandpass filtering (BP), envelope-based (ENV), single-tapered spectrogram (STP), multitaper spectrogram (MTP), and continuous wavelet transform (CWT), and present their efficacy in simulation and experimental data.

For simulations, the synthetic neural signal was constructed by superimposing beta (25 Hz) and gamma (40 Hz) bursts onto aperiodic noise processes. We first tested burst detectability of each algorithm under the effect of Gaussian noise and burst duration in the simulated signals. Next, the temporal accuracy of individual burst detections was observed by quantifying how much these detections shift in time compared to the simulated bursts. The algorithmic performances did not differ much in detecting long, salient bursts bur rather showed distinctions for short and transient events, for which CWT reported best detectability and temporal accuracy. The statistical differences in algorithms were further tested on the mouse LFP data (Thy1-ChR2-YFP; n=5) acquired from the basolateral amygdala (BLA) during the open field test. The statistical dependency of burst durations and burst occurrence rates on locations and algorithms was evaluated. Although no statistical significance was found between locations, the algorithms showed significant differences in their parameterized burst durations and rates.

Our results therefore demonstrate the necessity of concerning the distinct advantages of each method in relation to the signal properties (e.g., waveforms, spectral characteristics) and encourage the hybrid use of multiple methods to maximize temporal precisions of burst detection, especially when cognition and behaviors have to be correlated.

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Poster

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Topic: H.08. Learning and Memory

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Title: Phase-amplitude coupling of brain oscillations in verbal memory encoding

Authors: *A. V. MEDVEDEV;
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Abstract: Objective
Brain oscillations have been suggested to be involved in information transfer within and between neural networks by modulating neural excitability at different spatial and temporal scales. The interaction of oscillations across different time scales is revealed as cross-frequency coupling (CFC) and it represents a high-order structure in the functional organization of brain rhythms. There is growing evidence on the functional significance of CFC in various cognitive processes. In particular, phase-amplitude coupling (PAC) between gamma-band and low frequency oscillations have been implicated in working memory. To investigate the role of PAC in episodic memory, intracranial EEG (iEEG) records from patients with epilepsy during successful and unsuccessful verbal memory encoding (memorization of unrelated word lists) were analyzed in this study.
Methods
IEEG data were used from the open access repository (Restoring Active Memory, RAM). Time-frequency spectra in relation to each word presentation (1600 ms) were analyzed and 16 patients were selected for further analysis of PAC based on the availability of electrode locations within the four regions-of-interest (ROIs) of the temporal lobe (hippocampus, inferior, medial temporal and fusiform gyri). Modulation Index (MI; an amplitude-independent PAC measure proposed by Tort et al 2010) was then used to analyze the amplitude modulations of gamma-band frequencies in relation to the phase of low frequencies 4-16 Hz. The MI values were compared for subsequent successful vs. unsuccessful word retrieval.
Results
In each patient, several electrode sites showed significant increase in power of gamma-band activity during word encoding, and this increase was significantly larger for successfully recalled words ($p < 0.01$; the subsequent memory effect). For group analysis, the MI values were averaged across sites showing significant gamma response within each ROI and then across patients with the same ROI. The MI was significantly higher overall for successful encoding in the hippocampus, inferior and medial temporal gyri ($p < 0.01$). In addition, ROIs were characterized by regional specificity of coupling in terms of specific low and high frequencies showing the strongest association.

Conclusions
These results provide preliminary evidence that phase-amplitude coupling between gamma-band and low frequencies may play a functional role in memory encoding. It can be suggested that a more precise temporal structure of brain rhythmic processes results in a more efficient formation of transient neural assemblies which in turn determines the successful formation of a memory engram.

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Poster

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Title: Investigating implicit versus explicit motor sequence learning in an intra-subject EEG-study

Authors: *S. DYCK[1], C. KLAES[2];
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Abstract: Motor sequences play an essential part in our daily life since many everyday actions we perform have a sequential nature. Learning a motor sequence, for example a dance choreography, usually consists of two parts: Acquiring knowledge about the sequence elements and their temporal order (explicit component) and being able to combine these elements into a single, skilled behavior and to actually perform the sequence (implicit component). In this project, we investigate the neural oscillations that play a role in implicit and explicit motor sequence learning (MSL) using EEG. In our task design, participants concurrently learn an implicit and an explicit motor sequence of key presses in a modified version of the serial reaction time task (Nissen & Bullemer, 1987). The participants undergo five training sessions over the course of a week, while EEG recordings are performed at the first and last training session using a wet EEG with 64 electrodes. The two time points of measurement enable us to not only contrast implicit and explicit motor sequence learning in an intra-subject design, but to also investigate changes in neural oscillations over the course of learning. We will present our study design and preliminary data of 10 participants, including behavioral as well as EEG data. On a behavioral level, implicit and explicit sequence learning is reflected by significantly reduced reaction times, while implicit learning takes more time to be evident compared to explicit learning. A time-frequency analysis of the obtained EEG data shows that sequence learning is

related to alpha and beta suppression prior and during the key presses at motor-cortical electrodes.
References: Nissen MJ & Bullemer P (1987). Attentional requirements of learning: Evidence from performance measures. Cognitive Psychology, Volume 19, Issue 1, January 1987, Pages 1-32.

Disclosures: S. Dyck: None. **C. Klaes:** None.

Poster

P845. The Role of Oscillations

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P845.08

Topic: H.08. Learning and Memory

Support: NIH Grant 7R01MH073689-12

Title: Combining journeys into expected outcomes: orbitofrontal-hippocampal interactions during spatial reversal learning

Authors: *J. RICEBERG[1], A. SRINIVAS[3], M. SHAPIRO[2];
[2]Neurosci., [1]Icahn Inst. of Med. at Mount Sinai & Albany Med. Col., New York, NY; [3]Albany Med. Col., Albany, NY

Abstract: To adapt to changing circumstances, we often recall similar situations and the consequences of past choices. Memory for recent events requires the hippocampus; using memory to adapt to changing circumstances requires the prefrontal cortex. Spatial discrimination learning in the plus maze is impaired by hippocampal inactivation, whereas OFC inactivation leaves such learning intact but impairs reversals of well-established contingencies. The mechanisms required for hippocampus-OFC interactions that support learning are unclear. As rats perform spatial reversals in the plus maze, both CA1 and OFC units fire differentially across maze locations and predict pending choices. To investigate functional interactions, we simultaneously recorded ensembles OFC and CA1 units as rats learned spatial reversals. Neuronal activity recorded in the start arms revealed striking interactive dynamics as rats learned a novel reversal: (1) OFC units were reliably phase locked to hippocampal theta oscillations. (2) Mixed pairs of CA1 and OFC neurons fired together reliably within single theta cycles. (3) Though CA1 population vectors (PVs) predicted changes in OFC activity, OFC PVs did not predict CA1 dynamics. (4) Locations decoded from PVs in the two structures followed different dynamics: CA1 ensembles stably represented distinct spatial journeys, whereas OFC ensembles developed more generalized representations of journeys to different goal locations that held identical rewards. These activity dynamics occurred only during new learning, not during stable

performance or familiar reversals. The results indicate frontotemporal interaction mechanisms for computing cognitive maps that guide decision-making.

Disclosures: J. Riceberg: None. **A. Srinivas:** None. **M. Shapiro:** None.

Poster

P863. Spatial Navigation: Circuits

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P863.01

Topic: H.09. Spatial Navigation

Support: NIH Grant 1U01NS111695-01

Title: Vestibular and Eye-Movement Related Correlates in Rodent Nucleus Prepositus

Authors: *J. STOTT, M. A. VAN DER MEER;
Psychological & Brain Sci., Dartmouth Col., Hanover, NH

Abstract: The head direction (HD) system in rodents comprises a set of interconnected brain areas. While we know a great deal about the HD system in the cortex and thalamus, much less is known about its origins in the brainstem. Theories of how the HD system works assume that the core attractor network in the dorsal tegmental nucleus (DTN) and lateral mammilary nucleus (LMN) receives angular head velocity information (AHV) to update the HD signal. One of two major candidates to provide this signal to the DTN is the nucleus prepositus hypoglossi (NPH). However, based on anatomical and recording studies in the primate, NPH is thought of as an oculomotor nucleus. It remains unknown what information is carried by NPH in the rodent, and if it conveys an AHV signal to DTN required by current HD models. To evaluate the role of NPH in signaling AHV and/or eye-movement related signals, we recorded acutely from NPH neurons in male and female mice during passive (sinusoidal) rotation, under head-fixed conditions. In daily sessions, after the probe was lowered into NPH, the platform was manually rotated to generate a range of angular head velocities. Eye movements were recorded during the vestibular-ocular reflex (VOR). At the end of each recording, current was passed to mark the recording site. In our analyses, we asked two general questions. First, is coding of AHV and eye-movement related signals in NPH intermixed at the single cell level, or separate? Second, how does AHV tuning in NPH compare to that found in DTN? Greater than half of NPH cells showed significant modulation of activity just before the time of saccade onset. Also, a large majority of cells showed significant, linear tuning to the speed of the platform (AHV). These populations were largely overlapping. Among, AHV cells, tuning was almost entirely of the asymmetric type; that is, showing a linear increase or decrease in firing rate along the continuum of head velocities. This result stands in stark contrast to at least one previous report of AHV cells in the DTN of freely moving rats (Bassett & Taube, 2001), which showed a predominance of

symmetric (V-shaped) tuning. These findings raise interesting questions for models of the HD attractor. Purely vestibular tuning was rare in NPH; rather, most AHV cells showed some modulation by eye movements. How then, is estimated head orientation accurately updated, if eye movements influence NPH output? Additionally, how do we account for symmetric AHV tuning in DTN when the inputs from NPH are strictly asymmetric? Further experiments are in progress to record from pathway-defined NPH and supragenual nucleus neurons, and to characterize these same cells in freely moving animals with simultaneous eye tracking.

Disclosures: J. Stott: None. **M.A. van der Meer:** None.

Poster

P863. Spatial Navigation: Circuits

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P863.02

Topic: H.09. Spatial Navigation

Support: Wellcome Trust-DBT India Alliance (IA/S/16/2/502727)
Department of Biotechnology through the DBT-IISc partnership program
Revati & Satya Nadham Atluri Chair Professorship
Ministry of Education, Government of India

Title: HCN channels stabilize heterogeneous conductance-based continuous attractor networks

Authors: *D. MITTAL, R. NARAYANAN;
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Abstract: Continuous attractor network (CAN) models lend a powerful theoretical framework that has provided deep insights about grid cells and head-direction cells. Most models employed for understanding CAN dynamics rely on homogeneous rate-based or artificially spiking neurons with repeating motifs of structured synaptic connectivity. However, this formulation precludes detailed analyses of the impact of neural circuit components and associated heterogeneities on CAN dynamics. To address this caveat, we built multiple populations of conductance-based, physiologically constrained, one-dimensional CAN models consisting of distinct excitatory (E) and inhibitory (I) cell layers, endowed with different heterogeneities. CAN models built with homogeneous E & I neurons were tunable and scalable to exhibit “activity bump” movement across the neural lattice, irrespective of the relative excitability of the neurons. Next, we employed independent stochastic search algorithms to generate biophysically heterogeneous and physiologically validated populations of E (n=449) & I (n=930) neurons. We found I-layer, but not E-layer, heterogeneities to impact attractor dynamics. Specifically, none of 1000 randomized networks built with I-layer heterogeneities manifested activity bump propagation, whereas all 100 randomized networks with E-layer heterogeneities exhibited stable propagation. To address

the impact of synaptic heterogeneities on CAN dynamics, we picked distinct homogeneous networks and randomly scaled all synaptic strengths across multiple trials. We found ~65% of these networks to be stable, with no interdependencies or low-dimensional manifolds within the parametric space of stable networks. Finally, motivated by the ability of intrinsic neuronal resonance to stabilize rate-based CAN networks, we asked if modulation of HCN channels altered network stability. Strikingly, the proportion of stable models exhibiting activity propagation increased with increase in HCN-channel density, varying from ~60% to ~85% of the population. As HCN channels also contribute to reduction in excitability, we performed excitability-matched controls with fast HCN channels that do not introduce resonance. Fast HCN channels did not stabilize CAN model dynamics over a wide range of conductance values, suggesting resonance as a critical requirement for stabilization of network dynamics. Together, our conductance-based CAN models demonstrated the critical role of inhibitory neuronal excitability on CAN dynamics, and suggest intrinsic neuronal resonance as an efficacious mechanism to stabilize perturbation introduced by synaptic heterogeneities.

Disclosures: D. Mittal: None. **R. Narayanan:** None.

Poster

P863. Spatial Navigation: Circuits

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P863.03

Topic: H.09. Spatial Navigation

Support: ERC starting grant 639272
Research Council of Norway 274306
EEA grant RO-NO-2019-0504

Title: Characterizing the tuning properties of all major projections to a spatially tuned cortical area.

Authors: M. GIANATTI, ***A. GARVERT**, K. VERVAEKE;
Univ. of Oslo, Oslo, Norway

Abstract: Place cells likely contribute to spatial navigation and memory formation. However, while place cells were discovered in area CA1 of the hippocampus several decades ago, it remains unclear how networks generate spatially tuned cells. Many models assume that place cells receive input from similarly tuned cells. However, recordings from presynaptic neurons in the hippocampus have found neurons with a wide range of activity and spatial selectivity. An essential step towards addressing this question is to characterize the tuning properties of all major input pathways. However, in area CA1, this has not been done due to technical challenges. Recently, neurons with very similar spatial tuning properties as in area CA1 were discovered in

the retrosplenial cortex (RSC). Compared to the hippocampus, RSC has the benefit of being located at the brain's surface, which enables characterizing the tuning properties of all major input pathways with two-photon microscopy. To do this, we first determined the main input pathways to RSC using retrograde viral tracing methods. This identified the orbitofrontal cortex (OFC), the secondary motor cortex (M2), anterior cingulate cortex (ACC), posterior parietal cortex (PPC), secondary visual cortex (V2) and various thalamic nuclei as the major projections to RSC. Next, we functionally characterized these input pathways using two-photon microscopy in head-fixed mice that perform a foraging task on a linear track. We selectively expressed GCaMP6s in each of these identified areas and measured calcium signals from their axons in RSC. We quantified the tuning properties and spatial information of axonal signals using generalized linear-nonlinear models and Bayesian decoding. Surprisingly, we found that the tuning properties of all input pathways were similar to the spatial tuning found in RSC. This suggests that RSC is part of a widely distributed network of spatially tuned circuits from which it may inherit its spatial tuning.

Disclosures: M. Gianatti: None. **A. Garvert:** None. **K. Vervaeke:** None.

Poster

P863. Spatial Navigation: Circuits

Location: Virtual Only

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Program #/Poster #: P863.04

Topic: H.09. Spatial Navigation

Support: ERC starting grant (#639272)
Research Council of Norway (#274306)

Title: Tactile landmark encoding in the mouse retrosplenial cortex

Authors: *A. LANDE, K. VERVAEKE;
Univ. of Oslo, Oslo, Norway

Abstract: To orient ourselves, we use familiar landmarks such as buildings or mountains. However, landmark representations need to be embedded in a cognitive map of our environment to be useful for navigation. While decades of research have revealed how multiple connected brain regions can represent spatial maps, how neural circuits represent landmarks remains less clear. Work in humans and rodents have revealed that the retrosplenial cortex (RSC) is a key area for landmark computation. A recent report also found that RSC represents spatial maps, suggesting that this brain area may link landmarks to spatial maps. However, because sensory stimuli and motor outputs are difficult to quantify during natural behaviours, it remains unclear how RSC represent landmarks and how this interacts with its representation of space. To address these questions, we capitalize on the fact that mice, as nocturnal animals, often rely on tactile

stimuli for navigation. Therefore, we developed a whisker-based, tactile virtual reality system for head-fixed mice running on a treadmill. Rotation of the treadmill engaged the movement of an object across the whiskers in specific locations, with a speed that matched the running speed, thus mimicking the sensation of passing a gap. Head restriction enabled us to use high-speed cameras to detect whisker stimuli with great temporal precision allowing optimal control of sensorimotor variables. Furthermore, this enabled us to use two-photon imaging and genetically encoded Ca^{2+} indicators to densely sample neurons in RSC. We found that a population of excitatory neurons in RSC represent tactile stimuli along the track. Interestingly, when we omitted stimuli on random laps in well-trained mice, a subset of cells maintained their representation of the stimulus, suggesting a mnemonic function of landmark representation. Furthermore, a subset of touch-responsive cells became active before a touch happened, predicting the upcoming contact. These findings strengthen the idea that RSC serves a role in encoding the position of landmarks in familiar environments and can predict the location of known landmarks when they are absent.

Disclosures: A. Lande: None. **K. Vervaeke:** None.

Poster

P863. Spatial Navigation: Circuits

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Program #/Poster #: P863.05

Topic: H.09. Spatial Navigation

Support: Natural Sciences and Engineering Council of Canada
New Frontiers Research Fund
Alberta Innovates
Branchout Neurological Foundation
Beswick Fellowship
Canadian Institute of Health Research

Title: The psychedelic ibogaine disrupts cognitive maps and activity correlation structure in mouse retrosplenial cortex

Authors: *V. E. IVAN[1], D. T. CUESTA[1], D. CURIC[2], J. DAVIDSEN[2], A. J. GRUBER[1]; [1]Neurosci., Univ. of Lethbridge, Lethbridge, AB, Canada; [2]Physics and Astronomy, Univ. of Calgary, Calgary, AB, Canada

Abstract: Psychedelic compounds strongly modulate mental function, but their effects on cellular-level information processing are poorly understood. We tested recent theories of psychedelic effects on neural signaling and dynamics using neural recordings in the retrosplenial cortex (RSC) of behaving mice. The representation of position encoded by ensembles of

individual RSC neurons was destabilized by the non-classic psychedelic ibogaine when mice had to infer position between tactile landmarks. This corresponded with increased neural activity rates, loss of correlation structure, and increased responses to cues. Ibogaine had surprisingly little effect on the size-frequency distribution of network activity events, suggesting that signal propagation within RSC was largely unaffected, and that disruption of correlation & encoding arise from corrupted inputs, such as a predictive coding of events. These data support proposals that psychedelics increase the entropy of neural signaling due to impaired expectation-based constraint of activity. Furthermore, the loss of expected position encoding between landmarks recapitulated effects of hippocampal impairment, suggesting that disruption of cognitive maps or other hippocampal processing may be a contributing mechanism of such discoordinated neocortical activity.

Disclosures: V.E. Ivan: None. **D.T. Cuesta:** None. **D. Curic:** None. **J. Davidsen:** None. **A.J. Gruber:** None.

Poster

P863. Spatial Navigation: Circuits

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Program #/Poster #: P863.06

Topic: H.09. Spatial Navigation

Support: NIH Grant EY025613

Title: Spatial modulation of primary visual cortex in freely behaving rats

Authors: *B. CARY[1], G. TURRIGIANO[2];
[1]Brandeis Univ. Grad. Neurosci. Program, Waltham, MA; [2]Dept of Biol., Brandeis Univ., Waltham, MA

Abstract: Vision plays a crucial role in helping animals navigate and actively engage with their environment. Recent work from multiple labs has found that neuronal activity in primary visual cortex (V1) is modulated by spatial location, but whether this modulation is primarily driven by spatial cues or more broadly by other salient aspects of an animal's interaction with the environment is unclear. Here we address this by analyzing chronic extracellular recordings from V1 of freely behaving Long Evans rats. In this naturalistic context, we find many neurons in V1 with strong spatial modulation of firing. By following single neurons continuously for many days we are able to evaluate the stability and complexity of this spatial modulation during free behavior. Ongoing work focuses on analyzing the relationship between firing fields and the complex behavioral repertoires exhibited by these freely behaving animals, to investigate how broader aspects of the environment influence V1 representations.

Disclosures: B. Cary: None. **G. Turrigiano:** None.

Poster

P863. Spatial Navigation: Circuits

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P863.07

Topic: H.09. Spatial Navigation

Support: NIH Grant NS053907

Title: Projections to the lateral mammillary and dorsal tegmental nuclei arise from a small but overlapping population of neurons in the supragenual nucleus.

Authors: *J. A. GRAHAM, M. MCGUIER, M. S. CHANG, A. C. GUNDLACH, J. S. TAUBE;
Psychological & Brain Sci., Dartmouth Col., Hanover, NH

Abstract: Previous studies have shown that the site of the putative ring attractor for generating and stabilizing the head direction (HD) signal is thought to arise within the connections between two brainstem nuclei - the dorsal tegmental nucleus (DTN) and the lateral mammillary nucleus (LMN). Models differ on exact mechanisms, but all models result in a stable activity packet that represents the current HD. The activity packet is moved around the ring by vestibular inputs to accurately reflect the current HD as an organism moves through space. Models agree that the turning mechanism requires cells that fire in response to only one direction of movement (e.g. clockwise or counterclockwise turns). The supragenual nucleus (SGN) is an attractive candidate for such a mechanism, providing inputs to both DTN and LMN that are strongly lateralized (e.g. SGN output projects to the contralateral DTN and ipsilateral LMN). In addition, lesions of SGN result in the 'bursting' effect noted when lesioning upstream vestibular nuclei that is not present when lesioning LMN or DTN. This finding suggests that the SGN may provide the bulk of vestibular information to the HD system. While the SGN projects to both DTN and LMN, it is not known whether distinct or overlapping populations within SGN project to these areas, which has implications for how the HD signal is generated. To address this issue, we injected two different retrograde labels into the DTN and LMN of Long-Evans rats(n=18) and determined whether cells in SGN were single or double-labelled; double labeling would indicate that the projections to LMN and DTN arise from the same population of SGN cells. Tracer filled both targets in 14/18 animals. Results showed that the SGN à DTN projection is markedly stronger than the SGN to LMN projection, filling most cells within the central body of the SGN while cells projecting to LMN tended to be smaller, sparser and located on the periphery of the nucleus. A portion of cells projecting to LMN were double-labelled indicating that that they also projected to DTN. In contrast, there was a large number of single-labelled cells arising from the projections to DTN. These results suggest that there is a large population of neurons in SGN with

distinct location and morphology characteristics that project only to DTN. A smaller, more peripheral population projects to both DTN and LMN.

Disclosures: J.A. Graham: None. **M. McGuier:** None. **M.S. Chang:** None. **A.C. Gundlach:** None. **J.S. Taube:** None.

Poster

P863. Spatial Navigation: Circuits

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Program #/Poster #: P863.08

Topic: H.09. Spatial Navigation

Support: Zegar Family Foundation PG005813
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Howard Hughes Medical Institute (to ERK)
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Title: Mice need the cortical representation of proprioception in order to establish new visuospatial memories.

Authors: *O. RUTLER, S. PERSAUD, J. M. PARK, R. M. BRUNO, E. R. KANDEL, S. KOSMIDIS, *M. E. GOLDBERG;
Neurosci., Columbia Univ., New York, NY

Abstract: Light enters the brain via the retina, which then transmits a retinotopic map of visual space to higher centers. Because the eyes are constantly moving, an object in a given spatial location can appear at many different retinal locations. Therefore, the position of an object on a retinotopic map does not, by itself, describe the spatial position of the object. Instead, the brain must establish a spatially accurate map by transforming this retinotopic map into a stable map of space independent of eye and head position. Two great neuroscientists proposed different strategies for solving this problem. Helmholtz postulated that the brain uses a corollary discharge arising from the motor commands for eye and head movements to transform the retinotopic map, thereby making the transformed retinal location of an object in the world spatially accurate. This transformation can occur even before the movement. Sherrington postulated that the brain could measure the locations of the eyes in the orbit and the head on the shoulders, and calculate the position of the object in space, a calculation which must occur after the actual movement. Psychophysical evidence in humans (Poletteti et al.) suggests that a corollary discharge mechanism is dominant soon after the brief appearance of a remembered stimulus in the environment, but is later replaced by a proprioceptive mechanism for long-term spatial memory. There is a proprioceptive representation of eye position in Area 3a of monkey somatosensory

cortex (Wang et al.), which is necessary for long term spatial memory (Sun et al.). Here we asked if proprioception were also necessary for establishing a spatially accurate visual memory in the mouse. We trained 24 C57Bl/6 mice to find a marked platform in a Morris water maze. After 2 days of training, we put 4 large visual cues on the walls of the maze but removed the cue from the platform. The mice gradually learned to find the hidden platform as measured by swim path length and latency. We then made sham or aspiration lesions in the dysgranular zone of somatosensory cortex (S1DZ), the mouse equivalent of monkey Area 3a. The sham mice continued to improve their performance, but the lesioned mice did not. We then moved the platform to a different quadrant of the maze. The sham mice learned the new platform location, but not the lesioned mice. Both groups easily found a visibly marked platform in a new location. Air-righting and rotarod tests showed that both groups of mice had normal motor and vestibular function. These results suggest that proprioception is necessary for the establishment of long-term visual memory of a vanished target, but corollary discharge is adequate for on-line navigation to a visual target.

Disclosures: O. Rutler: None. **S. Persaud:** None. **J.M. Park:** None. **R.M. Bruno:** None. **E.R. Kandel:** None. **S. Kosmidis:** None. **M.E. Goldberg:** None.

Poster

P863. Spatial Navigation: Circuits

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Program #/Poster #: P863.09

Topic: H.09. Spatial Navigation

Support: NSF GRFP 1058262
NINDS F99NS108537
NEI F32EY031561
NIMH R01MH108729

Title: Attentional alpha oscillations identified in rat postrhinal cortex and hippocampus

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Abstract: Alpha rhythms (8-12 Hz) are the most prevalent oscillations in the primate brain, but their functional role remains controversial. Historically, alpha was thought of as a correlate of a “ground state” or “cortical idling” that occurs during the absence of sensory input (Haegens et al., 2011; Pfurtscheller et al., 1996). According to this view, alpha oscillations are a passive mechanism that emerge when the brain is disengaged, whereas during sensorimotor or cognitive challenges they lose amplitude and are replaced by other neural mechanisms. More recently,

however, alpha has been shown to be an active mechanism, becoming stronger when visual inputs are actively ignored, leading to a proposed top-down inhibitory role for alpha rhythms in attention (Klimesch et al., 1998; Worden et al., 2000; Jenson & Mazaheri, 2010; Jones et al., 2010; Sacchet et al., 2015; Sadaghiani & Kleinschmidt, 2016; Kizuk & Mathewson, 2017). Currently, oscillation bands described in the rodent brain include a very broad theta band of 4-12 Hz and do not include a distinct range of frequencies for the alpha band. In humans, theta is generally defined as 4-8 Hz and alpha is defined as 8-12 Hz (Uhlhaas & Singer, 2010). Here, we describe a phenomenon in the rodent brain recorded in the postrhinal cortex and the hippocampus that exhibits characteristics similar to alpha oscillations in the human brain. These oscillatory events are generally 2-5 second episodes of increased power in the 9-12 Hz frequency band, peaking at ~10 Hz. They are markedly different from Type 1 theta in that they occur only when the rat is nearly motionless, whereas Type 1 theta power positively correlates with running speed. They also differ from Type 2 theta, which has a lower frequency of 4-7 Hz. Behaviorally, these 10 Hz events occurred during three parts of a bi-conditional task trial. They were most robust immediately after stimulus onset and after object selection, but they also occurred between trials. The 10 Hz events emerged as rats moved from simple strategies in early trials to complex strategies in later trials. These data suggest the need to update the defined oscillation bands in the rodent brain to include this novel oscillation that is analogous to attentional alpha in the human brain.

Disclosures: V. Estela-Pro: None. **S.G. Trettel:** None. **R.D. Burwell:** None.

Poster

P863. Spatial Navigation: Circuits

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P863.10

Topic: H.09. Spatial Navigation

Support: NSF IOS-1656488
NIMH R01MH108729

Title: Pulvinar manipulation alters posterior parietal cortex activity during performance on a visuospatial attention task

Authors: *F.-C. YANG[1], R. D. BURWELL[1,2];
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Abstract: The posterior parietal cortex (PPC), which is important for visuospatial attention, is connected with the pulvinar (lateral posterior nucleus of thalamus in rats). In primates, the pulvinar differentially projects to two subdivisions of the PPC. One subdivision supports top-

down attention and the other supports bottom-up attention (Cabeza, 2008). We have shown that pulvinar and dorsal PPC activity correlates with multiple phases of a visuospatial attention (VSA) task, including onset of the visual stimuli, decision-making, task-relevant location, and behavioral outcome (Yang, Jacobson, & Burwell, 2017 ; Yang & Burwell, 2020). However, the rodent PPC can also be subdivided into dorsal and ventral subdivisions (dPPC and vPPC), and neuronal correlates in the dPPC and vPPC are consistent with roles in top-down and bottom-up attention, respectively (Yang, Dokovna, & Burwell, under revision). To further understand how the pulvinar modulates PPC activity, we optogenetically manipulated pulvinar activity while simultaneously recording neuronal activity in the pulvinar, dPPC, and vPPC in male Long Evans rats during performance on a visuospatial attention (VSA) task. Our VSA task was adapted from the five-choice serial reaction time task for a double-sided, bowtie shaped enclosure atop the Floor Projection Maze in which stimuli are back-projected to the floor of the maze (Jacobson, Ho, Kent, Yang & Burwell, 2014). In the VSA task, trials alternated from side to side. On each trial, rats were required to attend to three locations. In one of these locations, a target stimulus was randomly and briefly illuminated. An approach to the correct target location was followed by a liquid reward. For analysis, a trial was divided into behavioral epochs including stimulus onset, selection behavior, and reward. Trials were sorted by whether or not the pulvinar as inactivated. We recorded neuronal activity of six rats, and isolated 318 cells in the dPPC, 225 cells in the vPPC, and 474 cells in the pulvinar. Both pulvinar and PPC cells signaled stimulus onset and behavior consistent with the interpretation that the pulvinar and PPC are engaged in multiple aspects of visuospatial attention. Additionally, PPC ensembles were altered by pulvinar inactivation. Details will be presented.

Disclosures: F. Yang: None. **R.D. Burwell:** None.

Poster

P886. Neural Circuits and Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P886.01

Topic: H.11. Language

Support: NINDS BRAIN Initiative Grant U01NS098981

Title: The Neural Correlates of Speech Errors

Authors: *A. MEDVEDEVA[1], K. FORSETH[2], O. WOOLNOUGH[3], P. ROLLO[4], N. TANDON[5];
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Abstract: To investigate the neural correlates of error detection and correction, we recorded intracranial EEG during a tongue twisters task that reliable evokes overt speech errors in 17 focal epilepsy patients undergoing intracranial electrode implantation for seizure monitoring. Then we compared activity during errors vs during correct speech while patients read aloud and then recited each tongue twister from memory. As expected, phonological substitution errors were higher for stimuli with phonologically-similar compared to dissimilar onsets but only when the error was likely to form a word (similar word error condition), Bonferroni-corrected $t(16) = 2.04$, $p = .004$. Articulation during errors revealed potential error detection processes in frontal, parietal, and superior temporal regions, FDR-corrected $ps<.01$. The results suggest that a wide network contributes to planning speech and correcting and detecting errors to minimize errors in everyday speech.

Disclosures: A. Medvedeva: None. **K. Forseth:** None. **O. Woolnough:** None. **P. Rollo:** None. **N. Tandon:** None.

Poster

P886. Neural Circuits and Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P886.02

Topic: H.11. Language

Support: JSPS KAKENHI JP20K07951

Title: Neural fiber "conductivity" elucidates a mechanism of language lateralization

Authors: *K. MATSUO[1], N. YASUI-FURUKORI[2], K. SHIMODA[2], Y. KAJI[3], K. AKIYAMA[4];
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Abstract: Patients with mental illness often show a reduced left language lateralization. The mechanism may be that the neural “conductivity” would be reduced in the patients and the non-dominant hemisphere would become active to facilitate the real-time language processing. The supposed mechanism entails a rapidity hypothesis; a rapid communication between hemispheres would achieve a lateralization but a delayed one would effect a bilaterality. The corpus callosum (CC) would be responsible for this procedure. By contrast, the direction of the lateralization (usually, leftward) might be defined to the hemisphere with a faster language processing than the other, which would involve language fibers including the arcuate fasciculus (AF). If the rapidity hypothesis is correct, (1) people with a strong lateralization should have a higher “conductivity” in the CC than those with bilaterality, and (2) the stronger the left or right lateralization, the

higher the “conductivity” in the left or right AF, respectively. We investigated these by 73 healthy controls after obtaining the written informed consent. A 3-T MRI (Siemens Prisma) was used to conduct diffusion spectrum imaging [1] and compute the generalized fractional anisotropy (GFA) as the measure of “conductivity” (computed by [2]). AveLI [3-4] was applied as the lateralization index (LI) by task-fMRI with verb generation. Two types of masks were prepared: perisylvian language area (PSL) and area round Brodmann’s area 44 (area44) [5]. A regression analysis was performed using GFA volumes (transformed to the MNI space) with a regressor of either the LI or the absolute LI using SPM12 software. A regression analysis with the absolute LI by the PSL mask depicted a region in the CC around the anterior midbody. Moreover, a left superior temporal region around the AF was indicated by a positive regression using the LI by the PSL mask. The GFA in the CC was higher in people with a left lateralization (LI>0.2 by area44 mask; N=61) and those with a right lateralization (LI<-0.2; N=5) than those with a bilateral LI (N=7), whereas the GFA in the AF was higher in the order of left lateralization, bilateral, and right lateralization (no significance). The results were consistent with the rapidity hypothesis; the speed of neural communication plays a role in the language lateralization, which explains the reduced left lateralization in patients at least partly. References. [1] Wedeen et al., 2005, Magn Reson Med 54, 1377-1386. [2] AcroVis Inc., Taiwan. [3] Matsuo et al., 2012, J. Neurosci Methods 205, 119-129. [4] Matsuo et al., 2021, J. Neurolinguistics 57, 100943. [5] Glasser et al., 2016, Nature 536, 171-178.

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Poster

P886. Neural Circuits and Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P886.03

Topic: H.11. Language

Support: NIH (NINDS) R01 NS115929
NIH (NINDS) R01 NS062756
Procter Foundation

Title: Spatiotemporal Dynamics of Visual Naming

Authors: *B. ERVIN[1,2], J. BUROKER[1], A. BYARS[1], L. ROZHKOV[1], J. L. LEACH[1], P. S. HORN[1], C. SCHOLLE[1], F. T. MANGANO[1], H. M. GREINER[1], K. D. HOLLAND[1], R. ARYA[1]; [1]Cincinnati Children's Hosp. Med. Ctr., Cincinnati, OH; [2]Univ. of Cincinnati, Cincinnati, OH

Abstract: Objective: Spatiotemporal dynamics of visual naming were investigated in pediatric epilepsy patients undergoing stereo-electroencephalography (SEEG) monitoring.

Methods: Volumetric brain regions were linked to cognitive phases of visual naming by the Parcel-Activation-Resection-Symptom matching (PARS) approach, matching high-gamma (50-150 Hz) stereo-EEG power modulations (HGM) localized to neuroanatomic parcels during visual naming, with changes in neuropsychological metrics after resection or ablation of those parcels. Published clinical electrical stimulation observations were matched where resection data was sparse. Cortical parcels wherein at least half electrode contacts simultaneously showed significant HGM were aligned by temporal onset, to delineate spatiotemporal propagation of naming-related HGM.
Results: In 41 epilepsy patients, parcel-specific visual naming HGM showed sequential onset with extensive temporal overlap. Early activation was shown bilaterally in the occipital lobes, but HGM became increasingly left lateralized until vocalization, with propagation through limbic system. At response time, HGM was found in bilateral superior temporal, and right frontal activation was noted post-vocalization. Correlations between resected/ablated parcels, and post-surgical neuropsychological outcomes showed specific spatiotemporal groupings.
Conclusions: Functional roles of cortical parcels were inferred from changes in neuropsychological scores after resection/ablation and published clinical electrical stimulation observations. Convergence of this data with spatiotemporal propagation of HGM during visual naming supports a cognitive model with six subcomponents of visual naming having overlapping temporal profiles: Visual Decoding, Semantic Association, Lexical Access, Phonemic Representations and Pre-articulation, Vocalization, and Monitoring.
Significance: Cerebral substrates assisting in visual naming are bilaterally distributed. Relative hemispheric contribution varies with sub-task-specific cognitive demands. The PARS approach can be extended to study other cognitive and functional brain networks.

Disclosures: **B. Ervin:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Research Disclosure #D19-0026. **J. Buroker:** None. **A. Byars:** None. **L. Rozhkov:** None. **J.L. Leach:** None. **P.S. Horn:** None. **C. Scholle:** None. **F.T. Mangano:** None. **H.M. Greiner:** None. **K.D. Holland:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; NIH (NINDS) R01 NS062756. **R. Arya:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; NIH (NINDS) R01 NS115929, Procter Foundation. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Research Disclosure #D19-0026.

Poster

P886. Neural Circuits and Mechanisms I

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P886.04

Topic: H.11. Language

Support: NATS/NIH UL1 TR000445
U54 HD083211
NICHD/NIH R01 HD044073

Title: Text arousal mediates developmental language and salience network activity during reading

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Abstract: Emotional content is a critical yet understudied metric of literacy texts. Compared to emotionally neutral passages, texts with overall high arousal tend to activate affect- and linguistic-related regions. However, less is known about how phrase-by-phrase arousal impacts neuronal processing or how this relationship may be pertinent for children learning to focus on and comprehend passages. We hypothesized that more intense passage phrases would correspond with increased activation in the reading and language network (RLN) and the salience network. To investigate these associations, we examined a cohort of third-grade children (N = 35) while they read two passages during a functional magnetic resonance imaging (fMRI) session. Phrases were presented in grammatical units one at a time, and maximum arousal per phrase was calculated based on word-level ratings from the Warriner et al. (2013) affective norms. Using a psycho-physiological interaction analysis, we modeled how phrase-level arousal impacted the BOLD response during passage reading. Children's behavioral reading comprehension (RC) proficiency was measured using the Gates-MacGinitie Reading Test one year following fMRI data collection. During passage reading, we see expected activation of the RLN and disengagement of the default mode network. Higher arousal phrases were associated with increased activity in a critical RLN region, the left fusiform gyrus, but also decreased activity in affect processing brain regions, such as the anterior cingulate cortex and the ventromedial prefrontal cortex. Additionally, children who had increased activity in the posterior superior temporal sulcus, mediating phonological processing, during high arousal phrases had better RC scores the following year. These findings suggest that as children process more intense text, they upregulate linguistic processing and downregulate affect processing - contrary to adults. This study highlights the importance of emotion processing during text reading in childhood and how arousal may impact passage comprehension and intervention.

Disclosures: A.N. Burgess: None. **K.S. Aboud:** None. **L.E. Cutting:** None.

Poster

P886. Neural Circuits and Mechanisms I

Location: Virtual Only

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Program #/Poster #: P886.05

Topic: H.11. Language

Support: NIH/NIMH Grant 1-DP5-OD-021352-01
Perelman School of Medicine Translational Neuroscience Initiative Grant

Title: Network models evaluating semantic cognition theories show white matter tracts differentially mediate language production processes

Authors: *S. Y. PARCHURE[1,2], J. D. MEDAGLIA[4,3], D. Y. HARVEY[3], H. STOLL[4], A. KELKAR[4], J. P. ZIMMERMAN[3], D. S. BASSETT[2,3], R. H. HAMILTON[3];
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Abstract: Semantic cognition is critical to language production and comprehension. Patients with semantic impairment due to neural disease have language disabilities that profoundly disrupt daily living. However, the anatomical basis of semantic cognition is as yet poorly understood, with proposed theories implicating left-lateralized regions in the peri-sylvian fissure to a broader network comprising nearly the entire brain. To evaluate the various models of semantic cognition as predictors of language production performance, we used a network neuroscience approach along with behavioral tests. N=31 native English speaking healthy adults (Age= 25.6 +/- 6 yrs, 13 male) participated in the study. All subjects underwent Diffusion Spectrum Imaging (DSI) scans, with adjacency matrices computed from pairwise region to region streamlines among 234 brain regions reconstructed from tractography within each subject. Each participant completed 2 language tasks: Verb generation and Sentence completion. Networks corresponding to each language model were constructed for each subject: 1. Left inferior fronto-occipital fasciculus (IFOF), 2. Inferior longitudinal fasciculus (ILF), 3. Uncinate fasciculus (UF), 4. Peri-sylvian language network, and 5. Connections from language network to rest of brain. The mean of streamline edge weights, a measure of strength of anatomical network connections, was obtained for each modeled network. A stepwise linear mixed effects modeling approach using Ln(response times) as the behavioral outcome measure was used. For Verb generation, the IFOF ($p<0.001$) and ILF ($p=0.015$) significantly predicted responses. UF, Peri-sylvian language network, and edges from language network to rest of brain were not significant ($p>0.05$). For Sentence completion, models of UF ($p=0.0044$) and Peri-sylvian language network ($p<0.001$) were significant. ILF, IFOF, and edges from language network to rest of brain were not significant ($p>0.05$). This research represents a novel use of network neuroscience to evaluate theories of semantic cognition, identifying relationships between semantically demanding language tasks and specific tracts with connections across the brain. The results corroborate prior fMRI studies on the role of IFOF in semantic processing and retrieval, and the relevance of IFOF and ILF but not UF in Verb generation. Further, a double dissociation between networks predicting Verb generation (IFOF and ILF) and Sentence completion (UF and Peri-sylvian language network) was seen. This finding suggests a neurobiological basis of

differential recruitment of language network regions according to the specific semantic processing task.

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Poster

P886. Neural Circuits and Mechanisms I

Location: Virtual Only

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Program #/Poster #: P886.06

Topic: H.11. Language

Support: National Institute of Neurological Disorders and Stroke (NS098981)

Title: Intracranial recordings of reference to common objects

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Abstract: We isolated the mapping of sentence representations onto successful versus unsuccessful lexical referents. We used intracranial electrocorticography with high spatio-temporal resolution to address this issue. Patients (n=56) were implanted with either penetrating depth or surface grid electrodes (n=13,298) for the evaluation of medically refractory epilepsy. They were asked to articulate the name of common objects in response to orthographic descriptions (average: 6.5 words, range: 3-12 words), presented in rapid serial visual presentation (500ms per word). The final word in each sentence was either referential to a lexical item, or non-referential ("A person at the circus who makes you [laugh/commute]") (84 trials per condition). We analyzed broadband gamma (70-150Hz) activity at each electrode to index cognitive engagement. We found robust responses in medial parietal cortex, para-hippocampal cortex and lateral prefrontal cortex for the encoding of semantically coherent referential expressions. We identified greater gamma activity at the onset of the final word in referential trials across left inferior frontal regions (150ms post-onset), medial parietal cortex (250ms), para-hippocampal gyrus (400ms) and ventromedial prefrontal cortex (730ms). We also found greater gamma power for non-referential trials in pSTG, marking the violation effect. Medial parietal regions mediate action encoding, which possibly explains these effects since non-referential items often involved impossible/failed actions. These regions are also part of the default network, which shows activity increases during endogenous attentional tasks, with representational search likely being greater for referential relations. We also contrasted non-referential trials that were semantically coherent and contained only weak violations of lexico-semantic or syntactic rules (e.g., "Something you beat with grass") with those containing strong

violations (e.g., “A glass made of Wednesday”). We found increased gamma activity in anterior inferior frontal gyrus, angular gyrus and posterior middle temporal gyrus for semantically coherent non-referential trials. This points to a role for these regions in constructing semantically legal structures. Insights into this referential network will contribute to rehabilitative solutions and neuro-prosthetic designs for individuals with reading deficits, and in particular patients with anomic aphasia and alexia.

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Poster

P886. Neural Circuits and Mechanisms I

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Program #/Poster #: P886.07

Topic: H.11. Language

Support: NINDS-5U01-NS098969-03l
NINDS-5R01-NS091139-05

Title: Auditory cortex influences subthalamic nucleus activity during reading aloud

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Abstract: Speech production requires successful information transfer within a basal ganglia-thalamo-cortical loop and can be disrupted in a number of neurological diseases that involve the basal ganglia (BG). Subthalamic nucleus (STN) activity is modulated during speech, and deep brain stimulation (DBS) of the STN can decrease both semantic and phonological fluency in patients with PD. The functional significance of this STN activity, these deficits, and their relation to activity in cortical speech areas are not well understood. Indeed, many prominent models of speech production exclude the STN, possibly due to the difficulty in acquiring empirical data. Here, we sought to better understand the role of STN in speech production by studying its electrophysiological interactions with cortical speech areas, which was possible because of a unique opportunity to directly assess basal ganglia neuronal activity during DBS surgery.
Intracranial local field potentials (LFP) were recorded in four patients while they read aloud words and non-words during the surgical implantation of DBS electrodes for PD. Depth electrodes were implanted in the STN (the locations of which were altered over the course of

surgery), and electrocorticography (ECoG) strips were implanted over primary motor cortex (M1), primary somatosensory cortex (S1), and superior temporal gyrus (STG). To estimate the strength of neural interactions between STN and different cortical areas, we used event-related causality (ERC), a method based on the concept of Granger causality, to study task-related propagation of neural population activity indexed by high gamma (70-150Hz) power in local field potentials and ECoG. We aligned trials to spoken response onset prior to analyzing high gamma event-related activity and ERC.
During reading aloud, we observed sustained and reciprocal neural interactions between ventral sensorimotor cortex and STN. Moreover, we observed a directionally specific influence of posterior STG on STN, prior to speech onset. The strength of this influence was affected by the lexical status of the utterance, with increased propagation during word vs. non-word reading. The difference in network interactions manifested as significantly shorter reading latencies during word trials than during non-word trials.
These results indicate that the basal ganglia receive neural information from auditory cortex, suggesting a potential role for the STN in state feedback control of speech. Models of speech production that include basal ganglia may help explain and mitigate speech deficits from DBS and help develop treatments for speech impairments associated with movement disorders.

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Poster

P886. Neural Circuits and Mechanisms I

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Program #/Poster #: P886.08

Topic: H.11. Language

Support: CONACyT-scholarship to E. V. C. No. 755580
DGAPA-PAPIIT (IN 203818)
CONACyT-Fronteras No. 225-2015

Title: The neurocognition of social communication

Authors: *E. VALLES-CAPETILLO[1], M. GIORDANO[2];
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Abstract: Social communication is fundamental for successful social interactions, one of the most difficult communication forms is verbal irony[1]. Verbal irony is a statement that transmits the opposite meaning to its literal counterpart[2]. If social cues are nor interpreted correctly, the iirony can be interpreted as literal, white lie[3]; or unrelated. Mentalizing and holistic perceptual

processing have been associated with an adequate comprehension of verbal irony[4]. The brain areas that have been associated with verbal irony comprehension are the dmPFC and rmPFC, IFG, and STG[6], these results are based on 12 studies, many of them in Japanese. The purpose of the present study was to evaluate the cognitive and neural correlates of ironic comprehension in a Spanish speaking sample. Method: We created 14 contexts per condition (i.e. ironic, deceitful, literal and unrelated), and 14 statements. The linguistic proprieties, that allow us to distinguish between conditions, are comprehensibility, relevance, appropriateness, sincerity, and emotional valence. They were evaluated by 120 adults (mean age = 22). Then, 30 adults (15 M, age mean = 22) classified the intention of the statements according to the context. Individual differences in reading speed were controlled by recording the stimuli with one female and a male voice. Forty-eight adults (mean age: 26) participated in the MRI study. All were Spanish native speakers and right-handed. During the first session participants completed 18 tests that evaluated different processes associated with mentalizing, social abilities, executive functions, verbal abilities, and perceptual style; during a second session we acquired the functional brain images using magnetic resonance (MRI). Preprocessing of MRI data was performed with the fmriprep pipeline[7]. The data analysis was performed using FSL 5.0.9[8]. Results: The accuracy on the verbal irony task correlated with verbal abilities (rho = 0.60**), and mentalization (rho = 0.40*). Latency for verbal irony classification correlated with mentalization reaction time (rho = 0.40**), and inhibitory control (rho = -0.40**). During verbal irony, we observed increased BOLD signal bilaterally in the pSTG, in the RH in the orbital MPFC, SFG, frontal pole, and MFG; and in the LH in the aSTG. Conclusions: The contexts and statements had the desired qualities in that they were well understood by the participants. The psychometric tests showed an association between mentalizing and verbal irony detection, and social abilities. The fMRI study showed that irony recruited areas involved in mentalizing and semantic integration of social contexts. p { margin-bottom: 0.1in; line-height: 115% }

Disclosures: E. Valles-Capetillo: None. **M. Giordano:** None.

Poster

P886. Neural Circuits and Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P886.09

Topic: H.11. Language

Support: Intel Research Award
Burroughs-Wellcome Fund Career Award At the Scientific Interface (CASI)

Title: A unifying computational account of temporal processing in natural speech across cortex

Authors: *V. VO[1], S. JAIN[2], N. M. BECKAGE[1], H.-Y. CHIEN[4], C. OBINWA[3], A. G. HUTH[2]; [1]Emerging Technologies Research, Intel Labs, Intel Corp., Hillsboro, OR; [2]Neuroscience,

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Abstract: To understand natural speech, it has been proposed that human cortex processes temporal information through a hierarchy of representations, from short-timescale regions like auditory cortex (AC) to longer timescales in prefrontal cortex (PFC) and precuneus (PrCu). Existing computational accounts of this phenomenon cannot directly model responses to natural stimuli, and collapse data across subjects and brain regions, limiting their predictive power and resolution. Here we propose an account that predicts responses to ecologically valid stimuli and implicitly captures the hierarchy, while providing all benefits of single-subject, single-voxel predictions. Interpreting the estimated models reveals a diversity in voxel function across cortex, tied to each region's timescale. We first built a multi-timescale recurrent neural network (MT-RNN) trained as a self-supervised language model, where each unit integrates temporal information at a fixed rate. We then extracted stimulus features from each MT-RNN unit to build multi-timescale encoding models (MT-EMs). MT-EMs were trained on fMRI data from 7 subjects listening to 5 hours of narrative English stories using ridge regression. MT-EMs significantly predicted brain responses on held-out data in much of the temporal lobe, PrCu and PFC, and captured known timescale differences across regions. Next we simulated experimental manipulations from prior fMRI studies of temporal context effects [Lerner 2011; Yeshurun 2017; Chien 2020]. These in silico experiments tested timescale properties of both MT-RNN itself and voxels in cortex using MT-EMs. In all tests, MT-RNN units exhibited effects in accordance with their assigned timescale. And despite only learning to predict BOLD responses in a passive listening task, MT-EMs could successfully replicate effects across cortex in nearly every test. While AC was sensitive to word-level manipulations and integrated information quickly, PrCu and PFC integrated information slowly and were sensitive to long-timescale manipulations. Lastly, we probed MT-RNN to infer what linguistic information was encoded in different units. We found that short-timescale units capture token-level information like part-of-speech or entity type, while longer timescales capture discourse-level information like narrative topic. Combined with the implied computational isomorphism between MT-RNN and cortical processing, these results provide new evidence that different timescale regions in the brain may preferentially process different linguistic features. This is the first unified, predictive account of temporal context effects in cortex during speech perception.

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Poster

P950. Computational Modelling of Synaptic Networks: Topology and Structure

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P950.01

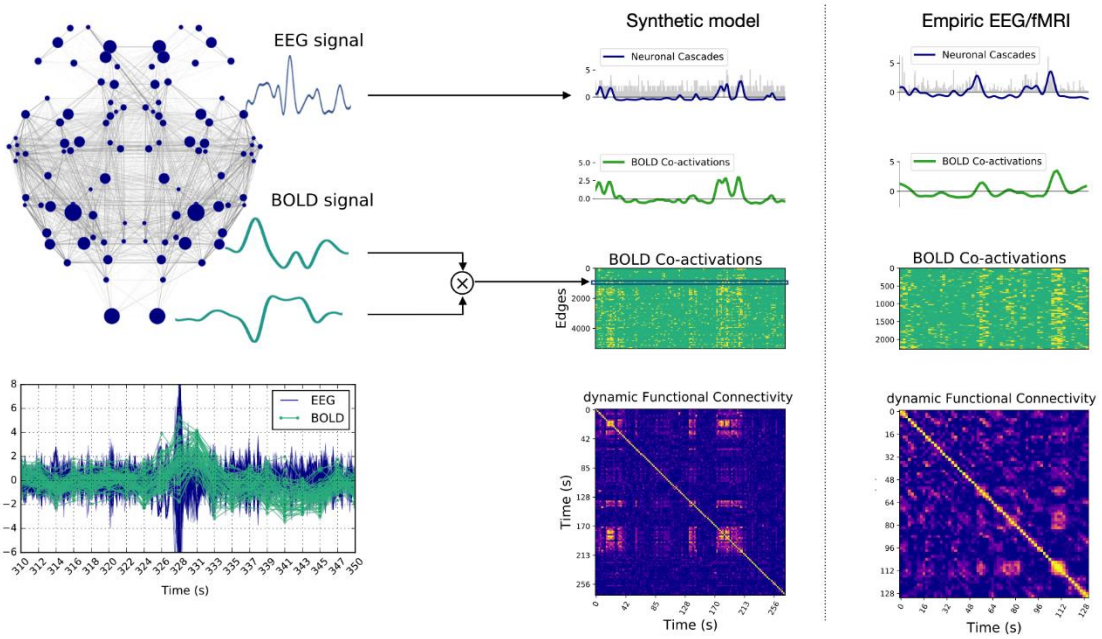
Topic: B.09. Network interactions

Support: Agence Nationale de la Recherche (ANR): ANR-17-CE37-0001 - CONNECTOME
EC | Horizon 2020 (EU Framework Programme for Research and Innovation): 945539 (SGA3)
EC | Horizon 2020 (EU Framework Programme for Research and Innovation): 826421

Title: Neuronal cascades shape whole-brain functional dynamics at rest

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Abstract: At rest, mammalian brains display remarkable spatiotemporal complexity, switching between different brain states at an infra-slow timescale (~tens of seconds). While the phenomenology of the resting state dynamics is valuable in distinguishing healthy and pathological brains [1], little is known about its underlying mechanisms. Using full-brain network modeling [2], we show that neuronal cascades spontaneously emerge at an infra-slow timescale when neuronal populations are coupled via a detailed structural connectome. Neuronal cascades occur under conditions of near-criticality and are characterized by epochs of increased deviations from baseline firing rate activity. The ignition and subsequent propagation of cascades depend upon the brain state and connectivity of each region. The largest cascades produce bursts of Blood-Oxygen-Level-Dependent (BOLD) co-fluctuations at pairs of regions across the brain, which explain most of the functional connectivity [3] and shape resting state dynamics. We experimentally confirm these theoretical predictions. We demonstrate the existence and stability of intermittent epochs of functional connectivity comprising BOLD co-activation bursts in mice. We then provide evidence for the leading role of the neuronal cascades in humans with simultaneous EEG/fMRI recordings. These results show that neuronal cascades are a major determinant of spontaneous fluctuations in brain dynamics at rest.
Bibliography:
[1] Maria Giulia Preti, Thomas AW Bolton, and Dimitri Van De Ville. The dynamic functional connectome: State-of-the-art and perspectives. Neuroimage, 160:41-54, 2017. [2] Paula Sanz-Leon, Stuart A Knock, Andreas Spiegler, and Viktor K Jirsa. Mathematical framework for large-scale brain network modeling in the virtual brain. Neuroimage, 111:385- 430, 2015 [3] Farnaz Zamani Esfahlani, Youngheun Jo, Joshua Faskowitz, Lisa Byrge, Daniel P. Kennedy, Olaf Sporns, and Richard F. Betzel. High-amplitude cofluctuations in cortical activity drive functional connectivity. Proceedings of the National Academy of Sciences, 2020



Disclosures: **G. Rabuffo:** A. Employment/Salary (full or part-time):; Institut de Neurosciences des Systèmes - Aix-Marseille Université. **J. Fousek:** A. Employment/Salary (full or part-time):; Aix-Marseille University. **C. Bernard:** A. Employment/Salary (full or part-time):; Institut de Neurosciences des Systèmes - Aix-Marseille Université. **V.K. Jirsa:** A. Employment/Salary (full or part-time):; Aix-Marseille University.

Poster

P950. Computational Modelling of Synaptic Networks: Topology and Structure

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P950.02

Topic: B.09. Network interactions

Title: Combined spatial and connective efficiency organizes the topology of mouse brain regions

Authors: *N. R. SMITH[1], A. BORZOU[4], S. N. MILLER[2], J. KASPER[3], S. AMEEN[4], J. M. SCHWARZ[4], J. D. HOMMEL[3];
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Abstract: The mammalian brain is a complex set of highly interconnected, yet anatomically and functionally distinct, regions. Together, these brain regions are organized into a functional neural network, but it remains unclear what factors drive this organization. One possible factor driving the spatial orientation of discrete brain regions is the structural connectivity of the network which promotes efficient function, where a network is efficient if regions that are well-connected are placed closer together. The Allen Mouse Brain Connectivity Atlas provides a high-resolution structural connectivity map of mouse brain regions derived from a series of viral tracing experiments. With these data, we explored our hypothesis that regions which are highly interconnected are located proximal to each other resulting in an efficient spatial organization that is related to the network connectivity. To test this hypothesis, we composed a model of the mouse brain by first simplifying complex brain region geometry to spheres while maintaining original structure volume. Then, using accompanying axonal projection strength data from the Atlas, we defined the spatial-connective efficiency for the placement of brain regions in 3D space as the strength of axonal projections between each region divided by the distance between regions. With this parameter defined, we formed a weighted sphere packing problem using Atlas-derived connectivity and brain region volume data. We utilized the CVXPY convex optimization library for Python to model a solution for the maximal spatial-connective efficiency, and therefore the optimal theoretical placement, of the 26 wild-type mouse brain regions which compose the hypothalamus. We validated these results by implementing a Monte Carlo simulation to this weighted sphere packing problem and compared both solutions to the true mouse brain. We found that the mouse hypothalamus' true configuration correlates significantly with the theoretical configuration that optimizes for spatial-connective efficiency via both convex optimization and Monte Carlo simulation. This correlation intensified when we slightly adjusted the hypothalamus' true configuration to account for our non-overlapping, spherical model. These results suggest that the brain's spatial organization is partly driven by the efficiency of the network connectivity, which may have significant impact in understanding the processes governing early brain development.

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Poster

P950. Computational Modelling of Synaptic Networks: Topology and Structure

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P950.03

Topic: B.09. Network interactions

Support: KVPY Fellowship
IISER Pune

Title: Network topology determines neuronal phase-of-firing codes

Authors: *M. R. PRATYUSH, C. ASSISI;
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Abstract: Neuronal networks encode information as spatiotemporal patterns of activity. In a spatiotemporal coding scheme, often referred to as a phase-of-firing code, inputs to a group of neurons are encoded by the phase at which they spike with respect to a common oscillation. In earlier studies , the phase code was implemented using a disconnected group of neurons, each unrestrained by the activity of the other, and capable of firing at any phase of the common oscillation. In contrast, in neuronal networks, the topology of the network must necessarily limit the space of possible responses. It is known that several regions of the brain encode sensory information using a phase-of-firing code, with recurrent network connections and a common oscillation. We ask, how does the structure of the network influence this phase code?
To address this question we simulated a neuronal network with balanced excitatory and inhibitory inputs to each neuron. When we stimulated this network with a uniform input, different groups of neurons were transiently synchronized. We show that the topology of the inhibitory sub-network determines the identity of neurons that form transiently synchronized groups. The order in which these transiently synchronous groups fired was stable to perturbations and periodic. Further, we found, for a given network, a profusion of periodic solutions are possible. In fact, using a small network with 81 neurons, nearly 10^9 periodic solutions were possible. Different initial conditions led the network to one of several such possibilities. How does one reliably access one out of these 10^9 possibilities? We show that heterogeneous inputs to subsets of neurons, coupled with an oscillatory drive, can lead the system to a particular stable periodic orbit. The phase at which these groups fired depended on the amplitude of inputs each received. Further, the dynamics of the network could be switched between different periodic orbits using transient heterogeneous perturbations.
Our study highlights general principles governing the dynamics of balanced networks driven by a common oscillation. Several regions of the brain show characteristic global oscillations, such as the hippocampal theta and gamma rhythms, and oscillations in the mammalian and insect olfactory networks. In addition, certain behaviours like sniffing in rodents or antennal flicking in insects induce a periodic time-dependent stimulus, resulting in imposed ‘extrinsic’ oscillations that drive the neuronal networks involved in sensory processing. The general principles elucidated in this study can potentially help in understanding the role of oscillations in the functioning of these systems.

Disclosures: M.R. Pratyush: None. **C. Assisi:** None.

Poster

P950. Computational Modelling of Synaptic Networks: Topology and Structure

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Program #/Poster #: P950.04

Topic: B.09. Network interactions

Support: NSERC 210977
NSERC 210989
NSERC 05830
FRQNT Doctoral Scholarship

Title: Paradoxical disinhibition of pyramidal neurons following parvalbumin optogenetic activation

Authors: *E. GIRAUD[1], M. LYNN[2], J.-C. BEIQUE[2], J.-P. THIVIERGE[1];
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Abstract: The ability of brain regions to regulate information processing depends intimately on how distinct cell types are functionally organized into networks. In the prefrontal cortex (PFC), an elaborate repertoire of activity arises from the interaction of single excitatory (E) and inhibitory (I) neurons through their network connectivity. It remains unclear, however, how each neuronal subtype contributes to the population activity of the surrounding network. To examine the contribution of different neuronal subtypes to cortical dynamics, we employed in vitro recordings of PFC circuits and computational modeling with a diversity of cell types. We recorded and stimulated spiking activity in acute slices of PFC by combining electrophysiological and optogenetic methods on high-density multi-electrode arrays containing 4,096 closely spaced electrodes. To parse out the contribution of distinct cell types, we developed a spike sorting technique to distinguish putative regular-spiking excitatory neurons from fast-spiking inhibitory interneurons. Our sorting algorithm was validated using a targeted combination of viral and optogenetic strategies to activate parvalbumin (PV) interneurons. When probing different subsamples of cortical ensembles, we found that optogenetically activating PV neurons led to a counter-intuitive increase in excitatory pyramidal (PYR) activity and a decrease in overall inhibition. To gain a mechanistic understanding of these findings, we developed a theoretical framework based on linear response theory that encompassed somatostatin (SOM), parvalbumin and pyramidal cell populations. With this framework, we derived analytically the steady-state response of neuronal subtypes under targeted perturbations. We found that global inhibition stemming from PV and SOM neurons decreased when PV neurons were externally driven. This resulted in a disinhibition of pyramidal neurons, thus leading to an increase in their activity. We confirmed these analytical findings in a leaky integrate-and-fire model with PV, SOM, and PYR subtypes connected in a realistic circuit. In this model, pyramidal neuron disinhibition allowed for gain modulation of excitatory activity. Together, these results provide insights on how E and I cells can affect overall network activity through complex connectivity motifs. Moreover, they reveal that a complex mapping between the various cell-type ensembles within cortex underlie network responses, and that such responses could generate counter-intuitive network interactions.

Disclosures: E. Giraud: None. **M. Lynn:** None. **J. Beique:** None. **J. Thivierge:** None.

Poster

P950. Computational Modelling of Synaptic Networks: Topology and Structure

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Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P950.05

Topic: B.09. Network interactions

Support: Harvard University
HHMI

Title: Multi-stable attractor dynamics in cortical layer 1: theory and in vivo voltage imaging experiments

Authors: *Y. QI[1], H. TIAN[1], V. J. PAROT[2], D. J. WONG-CAMPOS[1], X. LI[1], P. PARK[1], S. BEGUM[1], A. PREECHA[1], A. E. COHEN[1];
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Abstract: Cortical layer 1 (L1) comprises a sparse population of purely inhibitory interneurons. Recent work has suggested that L1 interneurons integrate sensory and neuromodulatory inputs and exert a dis-inhibitory effect on underlying cortical pyramidal cells by inhibiting deeper-layer interneurons. These results have led to the hypothesis that L1 might play a role in attentional gating of sensory processing. However, the circuit mechanisms by which L1 integrates sensory and modulatory information remain unclear. Many L1 interneurons have axons that extend laterally and synapse reciprocally onto each other, forming a network of lateral mutual inhibition. We used numerical simulations to investigate the spatiotemporal excitability of the L1 network. We used an adaptive exponential integrate-and-fire neuron model, which captured realistic firing properties but was simple enough to enable large-scale network simulations. We positioned the cells at realistic densities and used distance-dependent synaptic strengths calibrated from patch clamp measurements in brain slice. We then simulated the effect of different spatial and temporal patterns of conductance-based stimuli. A simple two-cell network exhibited winner-take-all dynamics under tonic stimulation. This circuit motif appeared functionally equivalent to a flip-flop circuit in digital logic, a resettable short-term memory. In larger networks, gradually increasing strength of tonic network-wide excitation led initially to gradually increasing firing rate, followed by an abrupt transition to a sparse winner-takes all pattern in which a small percentage of the cells fired tonically while the rest were silent. The identities of the winner neurons could be biased by small excitatory inputs timed to match the onset of the global excitatory stimulus. We propose that this circuit mechanism might be an underlying mechanism of attentional fixation triggered by strong arousal. Experiments using all-optical electrophysiology (optogenetics combined with calcium or voltage imaging) are underway to compare these simulation results to circuit dynamics *in vivo*.

Disclosures: Y. Qi: None. **H. Tian:** None. **V.J. Parot:** None. **D.J. Wong-Campos:** None. **X. Li:** None. **P. Park:** None. **S. Begum:** None. **A. Preecha:** None. **A.E. Cohen:** None.

Poster

P950. Computational Modelling of Synaptic Networks: Topology and Structure

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Program #/Poster #: P950.06

Topic: B.09. Network interactions

Support: NIH Grant K01EB027184
NIH Grant DP2NS122605

Title: Stimulation-mediated reverse engineering of silent neural networks

Authors: *X. REN, I. BOK, S. BHATT, E. MASTERSON, J. PHILLIPS, A. D. VAREBERG, A. HAI;
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Abstract: Reconstructing connectivity of neuronal networks from single cell activity is essential to understanding brain function, but the challenge of deciphering connections from populations of silent neurons has been largely unmet. Most studies rely primarily on active units to derive connectivity, and largely exclude non-firing or minimally active neurons. However, numerous groups present evidence that more than half of the neurons in the brain are “silent”, firing at very low frequencies and having highly specialized receptive fields. In this work we demonstrate a supervised learning method to analyze the use of stimulation for determining connectivity between postsynaptic neurons and heterogenous populations of both silent and active presynaptic neurons in a leaky integrate and fire (LIF) neuron model. We generated spike trains for populations of neurons (size ranging between 200 and 2000 cells) recorded during stimulus response. 66% of the spike trains corresponded to “silent” neurons (firing at 1 spike/min, or 0.017 Hz) and the rest corresponded to responsive neurons (firing at 20 Hz). We show that repetitive stimulation epochs can evoke activity that is sufficient for determining the synaptic weights of the entire neuronal population. For the realistic neuronal population that includes silent neurons, the algorithm fails to predict the weights without stimulation (RMSE = 3.59 ± 0.42 for inferred vs. real synaptic weights, Pearson correlation $r = 0.44$, $p < 0.01$). When the population was sufficiently stimulated, the algorithm was able to derive weights with high accuracy (RMSE is 0.35 ± 0.06, Pearson correlation $r = 0.97$, $p \sim 0$, 15 stimulations, 100 iterations). We use these findings to further characterize the performance of our algorithm for determining weights from excitatory, inhibitory and unconnected neurons within the population, and compare the ability of the method to predict spike trains for increasingly larger population sizes. The testable weight predictions about the number and protocol of the required stimulations

is expected to enhance future efforts for deriving neuronal connectivity and drive new experiments to better understand brain function.

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Poster

P950. Computational Modelling of Synaptic Networks: Topology and Structure

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Program #/Poster #: P950.07

Topic: B.09. Network interactions

Support: NIH Grant UF1NS115821-01
The University of Chicago Dean's Scholarship
Neuroscience Research Metcalf Fellowship

Title: Spiking neural networks show input-dependent sloppiness in synaptic architectures

Authors: *T. JABRI, J. N. MACLEAN;
Dept. of Neurobio., The Univ. of Chicago, Chicago, IL

Abstract: Local circuitry in the mammalian neocortex is characterized by statistical regularities of synaptic connectivity. However, the dynamical consequences of these regularities remain unclear. All complex systems can be defined by 'sloppy' dimensions, meaning that their behavior is unmodified by large changes to specific parameter combinations, and 'stiff' dimensions which are the opposite. In the case of the neocortex, sloppiness in synaptic architectures would be crucial to allow for naturalistic spiking dynamics to be maintained in response to a diversity of input patterns, changes in connections through learning and neuromodulation. Using simulations on spiking neural networks operating in a naturalistic spiking regime, we determine the stiff and sloppy parameters of synaptic architectures and the impact of three classes of input (brief, continuous, and cyclical) on model sloppiness. Large-scale algorithmically-generated connectivity parameter values revealed that specific combinations of excitatory and inhibitory connectivity are stiff and that all other architectural details are sloppy. Moreover, while the extent of viable synaptic architectures capable of producing sustained naturalistic spiking diminished with the brevity of input, the stiff dimensions are comparable across input classes. We also find that experimentally estimated connectivity probabilities are similarly stiff and sloppy when compared to the architectures that we identified algorithmically. This suggests that simple statistical descriptions of spiking dynamics are a sufficient and parsimonious description of network behavior when examining structure-function relationships at the mesoscopic scale. This study provides evidence highlighting the importance of the interrelationship of excitatory and inhibitory connectivity to establishing stable spiking dynamics in the neocortex.

Disclosures: T. Jabri: None. **J.N. MacLean:** None.

Poster

P950. Computational Modelling of Synaptic Networks: Topology and Structure

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Program #/Poster #: P950.08

Topic: B.09. Network interactions

Support: NIH grant R01MH112746
NIH grant R01MH108590
SFARI Pilot Award

Title: Personalized cortical circuit modelling implicates synaptic dynamics in functional variation across individuals

Authors: *R. COOPER[1], M. DEMIRTAS[5], J. B. BURT[1], A. M. HOWELL[2], J. JI[1], A. ANTICEVIC[3], J. D. MURRAY[4];
[2]Neurosci., [3]Psychiatry, [4]Dept. of Psychiatry, [1]Yale Univ., New Haven, CT; [5]Univ. Pompeu Fabra, Barcelona, Spain

Abstract: Popular for its ability to non-invasively image the macroscopic anatomical and functional connections in the brain, functional magnetic resonance imaging (fMRI) of the human cortex has revealed promising results concerning the reliability and stability of individual-level cortical connectomics. However, the synapse-level mechanisms underlying inter-individual variability are not well-understood, such as the respective roles of long-range white-matter structural connectivity vs. cortical physiological dynamics. One approach to bridging these mechanistic gaps is using biophysically-based neural circuit models of large-scale brain dynamics which can be quantitatively fit to empirical neuroimaging data. In this study we have utilized a circuit model with neurobiologically interpretable parameters to model functional connectivity (FC) at the individual level in healthy subjects, finding that such a model is able to capture differences between subjects.
We selected 879 subjects from the Human Connectome Project, from which we generated parcellated, resting-state FC matrices. We employ a cortical circuit model developed by Demirtas et al. whose free parameters represent synapse-level activity, allowing the macroscopic inter-individual variations apparent in fMRI scans to be understood in terms of the underlying cellular architecture. A key advantage of this model lies in its assumption that local circuit properties are heterogeneous across the cortex, following a large-scale gradient related to cortical hierarchy and indexed by the T1w/T2w structural MRI measure. Using this low-dimensional circuit model, we generate simulated FC matrices which are optimized to maximally fit the respective empirical data for the left hemisphere of the cortex at the level of individual subjects as well as the group average.

Our circuit model, with hierarchical heterogeneity in local circuit properties, is sensitive to subject-level differences in FC. Allowing a hierarchical, heterogeneous distribution of weight parameters across the cortex substantially improves the model's ability to fit empirical FC data by specifically adding to the model the flexibility to capture leading components of inter-individual variation. Further, model parameters related to cortical physiological dynamics explain the majority of variation across subjects, while subject-level structural connectivity failed to significantly capture variation. Thus, our model supports the notion that microcircuit properties related to cortical physiology and dynamics contribute to neural variability across individuals in healthy populations.

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Poster

P950. Computational Modelling of Synaptic Networks: Topology and Structure

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Program #/Poster #: P950.09

Topic: B.09. Network interactions

Support: NIH Grant R01MH112746

Title: Noradrenergic neuromodulation of spatiotemporal dynamics in a large-scale model of mouse cortex

Authors: *Q. GU[1], **D. LI**[1], B. FULCHER[3], V. ZERBI[4], J. D. MURRAY[2];
[2]Dept. of Psychiatry, [1]Yale Univ., New Haven, CT; [3]Univ. of Sydney, Camperdown, Australia; [4]Neural Control of Movement Lab, ETH Zurich, Zurich, Switzerland

Abstract: The locus coeruleus (LC) is a small brainstem nucleus whose projections throughout the brain mediate neuromodulation by norepinephrine (NE). It remains poorly understood how this key neuromodulatory system regulates local brain dynamics and reshapes functional network interactions. A recent experimental study (Zerbi et al., 2019, *Neuron*) showed, through fMRI in mice, that chemogenetic activation of LC increases BOLD functional connectivity across cortex. We analyzed the data from that study and further report significantly increased variance and reduced autocorrelation of cortical BOLD signals after LC activation. To investigate the neural microcircuit mechanisms capable of generating these spatiotemporal dynamic changes, we developed a nonlinear biophysical mean-field dynamical circuit model of the mouse cortex. Interestingly, the reduction in BOLD autocorrelation could not be captured with previously-developed linearized models in which neural activity fluctuates about a single fixed point. This motivated us to examine the bistable regime of excitatory-inhibitory local circuits in which activity stochastically transitions between a bursty up and a quiescent down state. Based on

neurophysiology, we implemented LC-NE modulation via increased excitatory neuronal gain, and found that this reproduces all the key experimental observations, when in a down-state-dominating regime. When the neuronal gain increases further, up states dominate, which produces a U/inverted-U-shaped modulation curve, a well-known phenomenon in LC studies linked to cognitive enhancement. At the large-scale network level, gain modulation reshapes the effective dimensionality of cortex-wide neural activity patterns. Finally, we tested gain modulation of stimulus representation and propagation in the large-scale model, by applying a pulse input to somatosensory areas and testing stimulus decoding accuracy using activities of the non-stimulated areas. We found that increasing gain improves distributed stimulus representation, inline with the improvement in perception due to the LC-NE modulation. Together, we propose a circuit mechanism for the functional role of LC-NE system in reshaping local and large-scale cortical dynamics. The model, which links from neuronal effects of neuromodulation to local and large-scale cortical dynamics, also provides a platform to flexibly investigate various dynamical and cognitive phenomena at both local and long-range scales *in silico*.

Disclosures: Q. Gu: None. **D. Li:** None. **B. Fulcher:** None. **V. Zerbi:** None. **J.D. Murray:** None.

Poster

P959. Experimental Computational Tools: Cellular and Molecular

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Program #/Poster #: P959.01

Topic: I.06. Computation, Modeling, and Simulation

Support: NIH Grant NS011613

Title: Interfacing Computational Neuronal Models Generated with NEURON Simulation Software with Living Cells Using a Real-Time Dynamic Clamp System

Authors: M. W. NOWAK[1], L. KORBEL[1], A. KANE[1], B. K. PANAMA[2], M. HINES[3], *N. CARNEVALE[3], G. C. L. BETT[2], R. L. RASMUSSON[2];
[1]Cytocybernetics, Inc., Buffalo, NY; [2]Jacobs Sch. of Med., Univ. at Buffalo, State Univ. of New York, Buffalo, NY; [3]Yale Univ., New Haven, CT

Abstract: p { margin-bottom: 0.1in; direction: ltr; line-height: 120%; text-align: left; orphans: 2; widows: 2 }a:link { color: #0563c1 } The NEURON simulation software package (neuron.yale.edu) is the leading *in silico* system for building and testing computational models of neurons and neural networks. We combined the previously offline NEURON simulation program with real-time, dynamic clamp system (Cybercyte DC1, Cytocybernetics, NY) allowing comprehensive NEURON cell models to be directly interfaced with living cells during

electrophysiological recording experiments. In Synthetic Cell mode, the action potential (AP) from the NEURON model is used to voltage clamp a cell expressing a cloned ion channel. The experimentally-obtained cloned channel current is scaled appropriately then put into the dynamic clamp NEURON model and summed with the model currents. In this mode, the effects of cloned ion channel currents on NEURON model APs can be studied. We used the Akemann and Knopfel (2006) NEURON model of a Purkinje neuron to voltage clamp HEK cells stably expressing Kv11.1 (hERG). Members of the EAG family of potassium channels (eag, erg, elk) have been shown to modulate neuronal AP firing (Bauer and Schwarz, 2018). Adding the relatively slowly gating hERG current to the Purkinje neuron model decreased the frequency of AP firing and slightly depolarized resting membrane potential. For the Purkinje model, baseline resting membrane potential (RMP) was -60.9±0.4 mV with an AP firing frequency of 10.0±1.6 Hz (n=6). Addition of hERG current (21.0±1.8 pA tail current at +30 mV) reduced AP firing (1.6±1.6 Hz) and slightly hyperpolarized RMP to -62.4±0.3 mV (n=5). Decreasing HERG tail current to 10.7±0.7 pA resulted in an AP firing frequency of 7.4±1.9 Hz and RMP of -61.0±0.6 mV (n=5). Our results show that combining NEURON simulations with real-time dynamic clamp system expands the capabilities of electrophysiology research. Applications include delineating the role of ion channel mutations underlying neurological disorders on neuronal function and in drug discovery to identify drugs targeting expressed ion channels using neuronal activity of the NEURON model as the read-out.

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Poster

P959. Experimental Computational Tools: Cellular and Molecular

Location: Virtual Only

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Program #/Poster #: P959.02

Topic: I.06. Computation, Modeling, and Simulation

Support: NIH NINDS award NS103906

Title: Computational modeling predicts acidic microdomains in the glutamatergic synaptic cleft

Authors: T. FEGHHI[1], *R. X. HERNANDEZ[2,3,4], M. STAWARSKI[5], C. I. THOMAS[7], N. KAMASAWA[7], A. LAU[1], G. T. MACLEOD[6,4,5];
[1]Dept. of Physics, Florida Atlantic Univ., Boca Raton, FL; [2]Intl. Max Planck Res. Sch. (IMPRS) for Brain and Behavior, [3]Integrative Biol. and Neurosci. Grad. Program, [4]Jupiter Life Sci.

Initiative, [5]Wilkes Honors Col., [6]Brain Inst., Florida Atlantic Univ., Jupiter, FL; [7]Electron Microscopy Facility, Max Planck Florida Inst., Jupiter, FL

Abstract: At chemical synapses, synaptic vesicles release their acidic contents into the synaptic cleft leading to the expectation that the cleft should acidify during neurotransmission. However, fluorescent pH probes targeted to the cleft of conventional glutamatergic synapses in both fruit flies and mice reveal cleft alkalinization, rather than acidification. To understand why, we created a reaction-diffusion scheme where we modeled pH dynamics at the *Drosophila* neuromuscular junction as glutamate, adenosine triphosphate (ATP) and protons (H^+) are released into the cleft. With bicarbonate and phosphate buffering systems as well as plasma membrane calcium-ATPase (PMCA) activity incorporated into the model, we predict substantial cleft acidification at release sites following neurotransmitter release for fractions of a milliseconds. Thereafter, the cleft rapidly alkalinizes and remains alkaline for over 100 milliseconds, as the PMCA removes H^+ from the cleft in exchange for calcium ions from adjacent pre and postsynaptic compartments. Thus, recapitulating the empirical data. The magnitude and time course of alkalinization is observed to be influenced by the small volume of the cleft. In addition, we observe that phosphate, but not bicarbonate buffering is effective at ameliorating the magnitude and time course of the acidic spike. Yet, both buffering systems are effective at ameliorating cleft alkalinization.

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Poster

P959. Experimental Computational Tools: Cellular and Molecular

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Program #/Poster #: P959.03

Topic: I.06. Computation, Modeling, and Simulation

Support: NIH NIDCD #1R01DC019278

Title: A systematic, data-driven analysis reveals cortical neuron-type morphologies for dense recordings using simulation-based machine learning

Authors: V. HAYNES, *S. CROOK, Y. ZHOU;
Arizona State Univ., Tempe, AZ

Abstract: Advancements in multi-channel recordings of single-unit activity (SUA) *in vivo* present an opportunity to examine how neurons should be grouped based on spatially varying extracellular action potentials (EAPs). Previous methods for grouping units based on EAPs from sparse recordings inherited the limitations of single-channel recordings due to the restriction to

the time domain. This predominantly resulted in binary classifications of putative excitatory and inhibitory types in the literature. However, recent studies using high-density electrodes suggest that some morphological properties of cortical neurons could be inferred from multi-channel recordings. Jia et al. (2019) used novel measures of EAP spatial variations with unsupervised learning to show that propagation velocity of EAPs across channels can differentiate two types of excitatory cells. Buccino et al. (2018) combined computational models of morphologically realistic neurons that simulate EAPs and deep learning techniques to show that dense electrode arrays can be used to differentiate more neuron-types in principle. Here, we improve on these results using a more systematic, data-driven approach that characterizes spatial and multi-timescale dynamic patterns in dense recordings, determines which morphological properties are observable from spatiotemporal EAPs, and develops a useful classification scheme for plausible neuron-types recorded *in vivo*.

We address the problem of grouping neurons based on spatiotemporal EAPs using automated feature engineering and computational modeling to uncover reliable morphological types given fundamental biophysical constraints for recordings of SUA. Our systematic, data-driven pipeline leverages an unsupervised method for extracting patterns spanning channels and timescales while also differentially representing individual units. We simulate a pool of units from diverse cortical neuron-type models at many spatial locations. This allows discovery of multiple unique spatial templates where combinations of these templates are reflected in unit representations that are predictive of variations in dendritic features of the underlying neuron-types. We characterize plausible latent morphologies based on the unit representations across all neuron-types and the spatial templates. Finally, we demonstrate how our approach addresses inference of neuron-types and their gross morphologies by comparing experimental and simulated units in the same latent morphological space.

References

Jia, X. *et al. Journal of Neurophysiology* **121**, 1831–1847 (2019).
Buccino, A. P. *et al. Journal of Neurophysiology* **120**, 1212–1232 (2018).

Disclosures: V. Haynes: None. **S. Crook:** None. **Y. Zhou:** None.

Poster

P959. Experimental Computational Tools: Cellular and Molecular

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Topic: I.06. Computation, Modeling, and Simulation

Support: ICMR-SRF (Grant No. ISRM/11(61)/2017)
IBRO International Travel Grant

Title: Quantum mechanics/ molecular mechanics analysis, synthesis and biological evaluation of epalrestat based mutual-prodrugs for diabetic neuropathy

Authors: *S. CHOUDHARY, O. SILAKARI;
Pharmaceut. Sci. & Drug Res., Punjabi Univ., PATIALA, India

Abstract: In the current study, a quantum mechanics/molecular mechanics (QM/MM) based biotransformation study was performed on synthetically feasible mutual-prodrugs of epalrestat which have been identified from an in-house database developed by us. These prodrugs were submitted to quantum polarized ligand docking (QPLD) with the CES1 enzyme followed by MM-GBSA calculation. Electronic aspects of transition state of these prodrugs were also considered to study the catalytic process through density functional theory (DFT). ADMET analysis of these prodrugs was then carried out to assess the drug-likeness. On the basis of in-silico results, the best five prodrugs were synthesized and further evaluated for their neuroprotective potential in high-fat diet-streptozotocin (HFD-STZ) induced diabetes in the rat model. Clinically relevant molecular manifestations of diabetic neuropathy including aldose reductase (ALR2) activity and oxidative stress markers such as reduced glutathione (GSH), catalase (CAT), and thiobarbituric acid reactive substances (TBARS) were determined in blood plasma as well as tissues of the brain and kidneys. The histopathological examination of these organs was also carried out to see the improvement in structural deformities caused due to neuropathy. Finally, in-vivo pharmacokinetic study was performed for the best two prodrugs to assess the improvement in biopharmaceutical attributes of parent drugs. Overall, EP-G-MFA and EP-MFA have significantly reduced the hyperglycemia-induced ALR2 activity, levels of oxidative stress markers, and manifested about a two-fold increase in the biological half-life (T1/2) of parent drugs. The overall findings of this study suggest that methyl ferulate conjugated prodrugs of epalrestat may be considered as potential protective agents in diabetic neuropathy.

Disclosures: S. Choudhary: None. **O. Silakari:** None.

Poster

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Program #/Poster #: P959.05

Topic: I.06. Computation, Modeling, and Simulation

Title: Improved spatial registration method for highly multiplexed gigapixel immunohistological image datasets in brain mapping studies

Authors: J. JAHANIPOUR[1], B. ROYSAM[2], A. SEDLOCK[1], *D. MARIC[1];
[1]NINDS/NIH, Bethesda, MD; [2]Electrical & Computer Engin., Univ. of Houston, Houston, TX

Abstract: Mapping brain tissue cytoarchitecture using multiplexed immunohistochemical (MP-IHC) techniques has recently gained increasing interest in the neuroscience imaging field. The major challenge in mining high content MP-IHC information from brain tissue slice preparations is the need to screen myriad of biomarkers of interest using iterative immunostaining and multispectral fluorescence microscopy imaging of same tissue samples processed in multiple sequential rounds to cumulatively acquire highly enriched (>50-plex) biomarker image datasets. The challenge with this type of technique is to spatially register the acquired image channels in each round of staining/imaging to precisely correct for microscope stage alignment, stitching artifacts, and tissue deformations. Transitioning from relatively small megapixel scale image datasets to extremely large gigapixel scale image datasets, such those generated from scans of human brain tissues, poses an extra challenge of image alignment due to the extreme size of the images that can be in the order of multi-gigapixels per channel. Traditional methods, such as feature-based registration, are adequate to estimate a robust affine transformation in small-scale images, but they fail to accurately estimate a robust transformation in large-scale images. To address this challenge on large-scale images, we developed a fully automatic pipeline to align each MP-IHC staining/imaging round to each subsequent round with sub-pixel accuracy using a common reference channel (nuclei or high contrast brightfield) in each imaging round. First, we use phase correlation method in Fourier domain to estimate the global shift between images due to stage alignment imprecision to correct for differences in tissue slide mounting onto the scanning microscope stage prior to each round of imaging. Next, we use an optical flow approach to estimate the pixel-to-pixel movement between images caused by tissue deformation due to tissue processing. The proposed method provides global, parallel, robust, parameter-free registration for large-scale multi-round epifluorescence images irrespective of image size, which is the current limitation of existing traditional image registration methods.

Disclosures: J. Jahanipour: None. **B. Roysam:** None. **A. Sedlock:** None. **D. Maric:** None.

Poster

P959. Experimental Computational Tools: Cellular and Molecular

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P959.06

Topic: I.06. Computation, Modeling, and Simulation

Title: Network simulations with popular programming languages that outperform or match C language

Authors: *C. FIETKIEWICZ, T. MUENZBERG;
Hobart and William Smith Colleges, Geneva, NY

Abstract: MOTIVATION
Many computational neuroscientists write simulation software using common programming

languages. However, performance differences between programming languages are generally not well known.
METHODS
A test model comprised a network of canonical Hodgkin-Huxley neurons (Hodgkin and Huxley 1952) with voltage-gated sodium and potassium conductances and monosynaptic connections. One neuron received an intracellular stimulus in order to generate network activity. Performance was evaluated based on the length of run times for the overall simulation.
Five programming languages were tested: C, Java, Julia, JavaScript, and Python. Python was also evaluated with a performance enhancing compiler known as Numba. C is the de facto performance standard, while the other languages are popular due to various reasons, including ease of use and a large user base.
Each language can be categorized as either compiled or interpreted. Compiled languages included C, Java, Julia, and the Python/Numba combination. Interpreted languages included Javascript and standard Python (without the Numba performance enhancer).
RESULTS
Run times were calculated in two ways: (i) the gross difference between start and stop times, similar to a stopwatch, and (ii) precise measurement of exact CPU usage via profiling software. Java and Julia exhibited the shortest run times. C was at least 15% slower, even with full compiler optimization. This contradicts a common assumption that C is necessarily as fast as any general purpose language. Python with Numba was at least 20% slower, as compared to Java and Julia.
Interpreted languages have poorer performance because instruction interpretations require additional translation at the time of execution. We included them because of their popularity. As expected, the interpreted languages were much slower. Javascript was approximately 200% slower, meaning that the run time for Javascript was 3 times as large as the run time for Java or Julia. However, Python without Numba was practically incomparable with run times on the order of 10,000 times as large as those of Java and Julia.
CONCLUSIONS
The compiled languages performed well, as expected. However, Java and Julia outperformed C. This counters a common assumption that C automatically provides the highest possible performance. Though Python with Numba was the slowest of the compiled languages, desirable features of Python may be an acceptable tradeoff for some. Javascript was expectedly slower, which should be considered in developing web-based simulators. Python without Numba, however, yielded incomparably low performance, and modelers should be aware of this deficiency.

Disclosures: C. Fietkiewicz: None. **T. Muenzberg:** None.

Poster

P959. Experimental Computational Tools: Cellular and Molecular

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P959.07

Topic: I.06. Computation, Modeling, and Simulation

Support: NSF Graduate Research Fellowship

Title: End-to-end neocortical cell type classification from current-clamp electrophysiology recordings using convolutional neural network

Authors: *R. WANG[1], S. HAN[1], M. GAJOWA[2], C. LIU[3];
[1]Dept. of Electrical Engin. and Computer Sci., [2]Dept. of Mol. and Cell Biol., [3]Dept. of Electrical Engin. and Computer Sci. and Helen Wills Neurosci. Inst., Univ. of California, Berkeley, Berkeley, CA

Abstract: Neuronal type classification problem has been present in neuroscience since Ramón y Cajal's presentation of the neuron doctrine that highlighted the ample diversity of neurons. Today, we classify neurons by genetic makeup, anatomy, and physiology, enabling cell type-specific studies on the role of particular neurons. However, subtle differences in both neuronal type definitions and classification pipelines make comparing and reproducing experimental outcomes based on cell types challenging. Leveraging the ground truth data published in the Allen Institute Cell Types Database, which contains anatomical and morphological descriptions, genetic types, and electrophysiology features of neurons in the mouse neocortex, we constructed a robust and efficient convolutional neural network that successfully classifies neurons according to their genetic label or broad type (excitatory or inhibitory) solely using current-clamp electrophysiology recordings. Previous solution to this cell type classification consisted of dimensionality reduction juxtaposed with clustering using calculated action potential features. This technique resulted in a reported accuracy of 84.13±0.81% for classification among the five primary neuron types: excitatory pyramidal (Pyr), parvalbumin (PV), somatostatin (SST), 5HT3a, and vasoactive intestinal peptide (VIP) cells (Ghaderi et al., Front. Neurosci., 2018). In comparison, our single pipeline method is fast and streamlined while simultaneously achieving competitive test set accuracies with a single current-clamp trace as the input. The 95% confidence intervals for the test set accuracies of the presented deep learning architecture are 96.35±0.30% for correct differentiation between excitatory cells and inhibitory interneurons; and 88.52±0.26% for successful classification among Pyr cells, SST, PV, VIP, and neuron-derived neurotrophic factor (Ndnf) interneurons. After training, this end-to-end convolutional neural network-based classification method does not require any specific knowledge or human intervention for quick identification of the cell type with high accuracy.

Disclosures: R. Wang: None. **S. Han:** None. **M. Gajowa:** None. **C. Liu:** None.

Poster

P972. Software Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P972.01

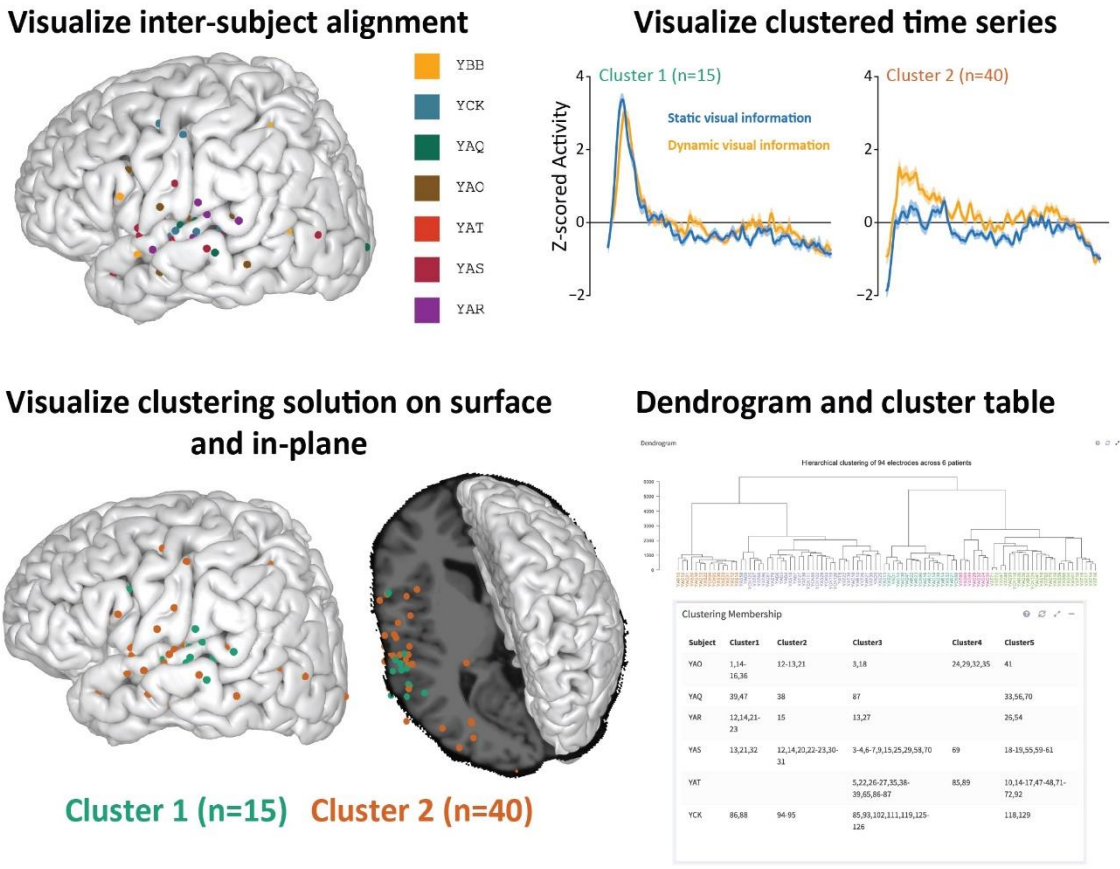
Topic: I.07. Data Analysis and Statistics

Support: 5R24MH117529-02

Title: Gui-driven clustering of group-level intracranial EEG data in RAVE

Authors: *J. MAGNOTTI[1], X. ZHANG[1], Z. WANG[2], Y. ZHANG[1], M. BEAUCHAMP[1]; [1]Univ. of Pennsylvania, Philadelphia, PA; [2]Rice Univ., Houston, TX

Abstract: Intracranial electroencephalography (iEEG) is a popular technique that encompasses the use of surface and depth electrodes to directly record activity of small population of neurons from the human brain. We have developed RAVE ("R" Analysis and Visualization of iEEG), a freely available, GUI-driven software package purpose-built for iEEG data that takes as input raw iEEG data and outputs publication-ready graphs and analyses of both single- and multi-subject data. Because iEEG electrode placement is guided by clinical utility, a critical step in an iEEG analysis is determining which electrodes relate to the phenomenon under study. To address this problem, we developed a RAVE module implementing single-subject and group-level clustering for iEEG data. Critically, this module follows the RAVE software philosophy: keep the users close to the data; rigorous statistical methods; play well with others. Cluster centroids are visualized as time series and cluster membership shown on the subject's cortical surface or brain volume using the built-in 3dViewer. For group data, the module automatically handles inter-subject alignment using surface-based (for surface electrodes) or volume-based (for depth electrodes) alignment tools. We graphically display not only the primary clustering results, but also cluster diagnostics, centroid plots, and distance plots to provide information to the user about the relevant trade-offs of particular clustering solution. We have initially implemented k-means (or medians) clustering alongside hierarchical clustering (with user-selectable distance and linkage methods). The modular, open-source nature of RAVE allows for easy expansion of our existing solution, allowing for outside contributions and extensions to more sophisticated algorithms. The module allows users to input/output data to/from the module using open data standards—the module is fully functional even if the data were not pre-processed using RAVE, and it outputs data and figures that can be edited in other software. For more details, see: https://openwetware.org/wiki/RAVE



Disclosures: J. Magnotti: None. **X. Zhang:** None. **Z. Wang:** None. **Y. Zhang:** None. **M. Beauchamp:** None.

Poster

P972. Software Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P972.02

Topic: I.07. Data Analysis and Statistics

Title: A Novel Graphical User Interface for Large Scale MEA Recordings Provides Insight into the Evolution of Epileptiform Activity and Tracks Seizures over Space and Time

Authors: *R. PARRISH, A. MAHADEVAN;
Xenon Pharmaceuticals, Burnaby, BC, Canada

Abstract: High definition multi-electrode array (MEA) has enabled *in vitro* and *ex vivo* neuronal measurements at high spatial resolution to record local field potentials, action potentials, and

network activity with relative ease. Whilst we have advanced recording systems with over 4000 electrodes, with capabilities to record data at over 20K Hz, it still presents computational challenges to handle, process, extract and view information from these large recordings. It can be quite challenging and frustrating for researchers to extract and view even a single channel that has more than a billion points, let alone process a group of channels. We will present a computational method, and an open source toolkit built on Python, rendered on a web browser using Plotly's Dash for extracting, viewing, and creating interactive visualization. In addition to extracting and viewing small chunks of the high frequency data or entire traces of low frequency (down sampled) data, it provides a framework to collect user input, to analyze channel groups, generate raster plots, view quick summary measures for local field potential activity, detect and isolate noise channels and generate plots and visualization in both time and frequency domain. We also present a seizure detection method using sliding convolution windows, which can be used to detect the onset of seizures in a group of channels, start and end time, rate of seizure speed, and distance of seizure spread across the region of interest. We will demonstrate the utility of this toolkit, using datasets collected from the 3Brain BioCAM duplex system. The recording has 4096 channels, sampled at 20K Hz, over a duration of 1 hour. For the current analysis, we demonstrate the toolkit and methods with a low frequency dataset (0 to 2048 Hz) over a small group (100 to 300) of channels. Using this toolkit, we will present data demonstrating a novel finding relevant to the transition from tonic-clonic seizures to status epilepticus from slices bathed in low Mg^{2+} artificial cerebrospinal fluid. Furthermore, we will present data demonstrating how tracking rates of seizures and the speed of seizure spread can help one gain mechanistic insight into the epileptic phenotype. In summary, with advances in HD MEA recording systems with high spatial and time resolution, limited tools are available for researchers to process these big data sets. We will demonstrate a user-friendly toolkit to analyze HD MEA data and demonstrate the utility of this new graphic user interface with novel biological findings.

Disclosures: R. Parrish: E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Employee of Xenon and may hold stock or stock options in the company. **A. Mahadevan:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Employee of Xenon and may hold stock or stock options in the company.

Poster

P972. Software Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P972.03

Topic: I.07. Data Analysis and Statistics

Support: NINDS R01NS092388
NIGMS 5U54GM104942-03

Title: Vesselvio: a novel and open-source application for vasculature dataset analysis and visualization

Authors: *J. R. BUMGARNER, R. J. NELSON;
Neurosci., West Virginia Univ., Morgantown, WV

Abstract: Vascular networks are essential for the delivery and clearance of nutrients, waste, and trafficking immune cells to and from tissue. These networks are highly organized, and their structure often can indicate and dictate states of health and disease. Quantitative analysis of these networks can improve the characterization of disease states and may inform future therapeutic strategies. Numerous advancements in imaging technologies and open-source segmentation software have led to the generation of large-scale and high resolution cerebrovasculature datasets. However, these advances have not been met with open-source and easily accessed analysis software packages. We have developed VesselVio, an open-source application for the analysis and visualization of vasculature datasets. Novel branch point filtering and radius analysis techniques are implemented and tested against ground truth datasets. The utility and compatibility of VesselVio is demonstrated via the analysis of female and male mouse 3D inferior colliculus segmentations and 2D human retinal fundus images.

Disclosures: J.R. Bumgarner: None. **R.J. Nelson:** None.

Poster

P972. Software Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P972.04

Topic: I.07. Data Analysis and Statistics

Support: SPARC Project, NIH Common Fund, NIH Office of the Director, Award OT2 OD030541

Title: Sparc connectivity knowledge base: discovering autonomic knowledge

Authors: *T. GILLESPIE[1], M. SURLES-ZEIGLER[1], J. MENKE[2], D. P. NICKERSON[3], B. OZYURT[1], B. DE BONO[3], M. E. MARTONE[1];
[1]Neurosciences, [2]Ctr. for Res. in Biol. Systems, UCSD, La Jolla, CA; [3]Auckland Bioengineering Inst., The Univ. of Auckland, Auckland, New Zealand

Abstract: The Stimulating Peripheral Activity to Relieve Conditions (SPARC) program is a NIH-funded consortium whose overarching goal is to improve the understanding of how the

autonomic nervous system (ANS) interacts with end organs and the central nervous system. A major objective of SPARC is to use this knowledge to develop the next generation of neuromodulator devices as effective therapies for diseases of the nervous system. To assist in that effort, the SPARC Data and Resource Center (DRC) is fielding an infrastructure for sharing data and tools for mapping the ANS. A centerpiece of this infrastructure is the SPARC Connectivity Knowledge base (SCKB) and associated visual map of ANS circuitry. The SCKB contains knowledge about CNS-ANS-end organ circuitry derived from SPARC data and scientific literature, in a form that supports reasoning. All connections are annotated with the SPARC vocabularies to ensure anatomical structures are used consistently. The SCKB currently contains two levels of semantic connectivity knowledge: circuit and individual connections. A circuit represents a detailed model of ANS connectivity associated with a particular organ, e.g., bladder or functional circuits, e.g., defensive breathing. These circuits are created through interviews with SPARC investigators, anatomical experts and the scientific literature. They contain detailed representations of neuron populations giving rise to ANS connections, including mappings of the locations of cell bodies, dendrites, axon segments and synaptic endings involved in a particular circuit. Circuits are modeled using ApiNATOMY, a knowledge model and tool suite specifically created to represent biological connectivity. These detailed circuits are supplemented with more general knowledge on connectivity between CNS nuclei, ANS ganglia, nerves, and end organs derived from the scientific literature. To facilitate extracting this knowledge from the literature and maintaining relevance over time, we developed a Natural Language Processing (NLP) pipeline, which extracts connectivity relationships between unique anatomical structures within a sentence from the scientific literature. The SCKB is being used to create a queryable visual atlas of ANS circuitry through the SPARC portal using a familiar street-map interface. Connectivity knowledge obtained from the SCKB is routed via hand-drawn anatomical features to provide highly flexible and explorable “flatmaps” linked to relevant SPARC data and knowledge. The SCKB provides a central location to populate, discover, and query ANS connectivity knowledge over multiple scales for SPARC and the neuroscience community at large.

Disclosures: T. Gillespie: None. **M. Surles-Zeigler:** None. **J. Menke:** None. **D.P. Nickerson:** None. **B. Ozyurt:** None. **B. de Bono:** None. **M.E. Martone:** None.

Poster

P972. Software Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P972.05

Topic: I.07. Data Analysis and Statistics

Support: NIH Grant R01 HL130249
BCM McNair Scholar Program

Title: An Automated Respiratory Data Pipeline for Waveform Characteristic Analysis

Authors: *S. LUSK[1], C. S. WARD[1], A. CHANG[3], A. TWITCHELL-HEYNE[1], G. ALLEN[3], R. RAY[2];
[2]Deparment of Neurosci., [1]Baylor Col. of Med., Houston, TX; [3]Rice Univ., Houston, TX

Abstract: Whole body barometric flow through plethysmography is used to study respiratory output in mouse and rat genetic and disease models. Turn-key commercial and custom systems are both used to gather multiple data streams including respiratory-pressure waveforms, O_2 and CO_2 measurements, and other data points for a reasonable estimation of tidal volume and key metabolic parameters. These experiments typically produce large amounts of data for analysis that are time consuming and difficult to organize. For analysis, the investigator may use cumbersome hand annotation subject to observer bias or several software products may be combined to manage raw data, statistical analysis, and graphing. Commercial software is prohibitively expensive or bundled with turn-key systems that limits options for those using bespoke respiratory measurement systems. To address these deficiencies, we developed a software pipeline for processing raw respiratory recordings and associated meta-data (experimental design, age, weight, temperature, etc.) into operative respiratory outcomes, publication-worthy graphs, and robust statistical analyses. This pipeline consists of three modules. The first module is a Python based graphical user interface (GUI) for uploading all relevant data files and allows the user to define and label both independent and dependent variables relevant to the experimental design, statistical analysis, and graphical outputs. In the second module (Breathcaller) the data are processed with a Python program to segment the respiratory waveform based on user defined thresholds for movement artifacts and unique features. Breathcaller quantifies respiratory cycle components and demarks unique features including sighs, apneas, expiratory reflexes, and user defined entities. The module also allows for browsing the raw signals with markup of annotated breaths to facilitate validation of breath detection, and to permit manual annotations of features if desired. The third module (SG-Runner) is an R-based program that accepts the Breathcaller output along with configuration files from the GUI to graph the desired variables while performing a linear mixed effects statistical model analysis of the data with appropriate post-hoc tests. Following graph generation, statistically significant differences in respiratory variables are automatically highlighted in the graphs. The open-source program offers a facile and highly customizable platform for automated handling and analysis of animal respiratory and metabolic data that sets the stage for high-throughput studies and machine learning approaches on large data sets.

Disclosures: S. Lusk: None. **C.S. Ward:** None. **A. Chang:** None. **A. Twitchell-Heyne:** None. **G. Allen:** None. **R. Ray:** None.

Poster

P972. Software Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P972.06

Topic: I.07. Data Analysis and Statistics

Title: The ΔF/F's are too small! A computational method for removing light contamination in two-photon calcium imaging

Authors: *B. LAU[1], G. ZIPURSKY[3], J. GAUTHIER[2];
[2]Biol., [1]Swarthmore Col., Swarthmore, PA; [3]New York Univ., New York, NY

Abstract: Two-photon (2P) imaging of calcium indicators has enabled major advances in systems neuroscience. Many applications of this technique rely on accurate estimates of ΔF/F, the change in fluorescence ascribed to each neuron. One potential source of error in ΔF/F measurements is from contamination by background light, which increases the denominator (resting fluorescence) without affecting the numerator (deviations from rest). To test whether current cell finding techniques are susceptible to background light contamination, we applied them to a simulation where ground truth was known (Song et al. 2021, J Neurosci Methods). Surprisingly, two popular methods greatly underestimated the true ΔF/F (CaImAn, median 0.02 of true ΔF/F; Suite2P, median 0.27). Moreover, the degree of underestimation varied widely across cells (CaImAn quartile range 0.0082-0.031; Suite2P 0.21-0.32). Underestimation could introduce unexpected errors into spike inference algorithms or analyses that try to explain variance. Inconsistent errors across cells might also obscure activity patterns, such as functional cell classes that could otherwise be identified by the amplitude of their transients.
To obtain more accurate measurements of ΔF/F, we developed a technique to remove background contamination using a post-hoc computational analysis. We constructed an estimator of cell brightness at rest (ΔF/F denominator) using a simple model of 2P fluorescence. When applied to simulated data, our method estimated ΔF/F nearly perfectly on average (median 1.10) and with a tighter distribution of ratios (quartile range 0.90-1.12). When applied to real data, where ground truth is not available, our method found much higher values of ΔF/F than popular methods (median ratio 7.9, quartile range 6.9-8.9), consistent with improved background compensation.
More accurate and consistent estimates of ΔF/F could improve spike inference and other analyses that rely on absolute ΔF/F values. This might allow for detection of new subclasses of neurons, or improve identification of special kinds of calcium events, such as complex spikes in Purkinje cells or CA1 pyramidal cells. More generally, correcting published ΔF/F values has the potential to alter scientific conclusions.

Disclosures: B. Lau: None. **G. Zipursky:** None. **J. Gauthier:** None.

Poster

P972. Software Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P972.07

Topic: I.07. Data Analysis and Statistics

Support: VA Merit Award I0RX002969
PVA Research Foundation
Taylor Foundation for Chronic Diseases

Title: Core principles for the implementation of the Neurodata Without Borders data standard

Authors: *M. L. REIMER[1], L. BANGALORE[2], A. M. TAN[3], S. G. WAXMAN[4];
[1]Neurol., Dept. of Neurol. and Ctr. for Neurosci. and Regeneration Research, Yale Univ. Sch. of Medicine, New Haven, CT 06510, USA; Rehabil. Res. Center, Veterans Affairs Connecticut Healthcare Syst., West Haven, CT; [2]Dept. of Neurol. and Ctr. for Neurosci. and Regeneration Research, Yale Univ. Sch. of Med., West Haven, CT; [3]Neurol., Yale University/VA Connecticut Healthcare Syst., West Haven, CT; [4]Neurol., Yale University, Neurosci. and Regeneration Res. Ctr., West Haven, CT

Abstract: The Neurodata Without Borders data standard (NWB) unifies diverse modalities of neurophysiology data. It defines how to structure and store data and is streamlined for research settings. Integrating NWB with a database unleashes its full potential to promote collaboration, standardize analyses, capitalize on historical data, and ensure data integrity.
To demonstrate the benefits of this computational innovation--which allows the incorporation of big data analytical principles into a standard laboratory setting--we implemented the NWB standard to streamline database operations with DataJoint in Jupyter notebooks, a user-friendly open document format. Specifically, we illustrated this process by converting open-field animal behavioral data into the NWB data standard. This permitted rapid application of data query filtering techniques within the DataJoint pipeline and Jupyter notebooks. Taken together, our previous report provided ample evidence that support the powerful utility of applying data standardization and warehousing principles to laboratory behavioral data.
Here, we present an extension of this prior work demonstrating the advantages of implementing the NWB and DataJoint pipeline within microscopic imaging protocols, e.g., z-stack 3D optical images collected from histological tissue samples. Additionally, we highlight the next generation of Jupyter notebooks, “Jupyter Labs”. As invaluable pedagogical tools, we will continue to use simple programming examples in this presentation.
Databases leveraging the NWB data structure are the foundation of analytical systems used by academic leaders including the Allen Institute and the International Brain Laboratory. The joint use of NWB with DataJoint transforms traditional laboratory datasets and workflows. Our Jupyter examples will showcase the analytical and collaborative advantages of adopting these technologies. Finally, this workflow can be tailored to other modalities by researchers interested in evaluating and implementing an NWB-based data storage and analysis pipeline.

Disclosures: M.L. Reimer: None. **L. Bangalore:** None. **A.M. Tan:** None. **S.G. Waxman:** None.

Poster

P972. Software Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P972.08

Topic: I.07. Data Analysis and Statistics

Support: Lee Kong Chian School of Medicine, Nanyang Technological University Singapore Dean's Postdoctoral Fellowship
Lee Kong Chian School of Medicine, Nanyang Technological University Singapore Nanayang Assistant Professor Start-up Grant.

Title: Coldcuts: an open source r package to draw brain segmentations

Authors: *G. A. D'AGOSTINO, S. R. LANGLEY;
Lee Kong Chiang Sch. of Medicine, Nanyang Technological Univ., Singapore, Singapore

Abstract: The generation of large datasets from the brain, such as imaging or transcriptomics, requires the use of analytical pipelines which run entirely on open source programming languages such as R and its environments. However, there are few options to relate the results of these pipelines back to the original structural context of the brain without going through third party software, and these options are often limited in scope. Moreover, copyright-free precise representations of brain structures are useful for didactic purposes and can be hard to find. We present coldcuts, an R software package to quickly render a previously generated brain segmentation as ordered sets of polygons that can be easily plotted in 2 dimensions using any plotting library. coldcuts accepts a variety of compressed inputs such as NIfTI, NRRD or RAW binary formats, slices the voxel array along the 3 anatomical planes and automatically draws polygons for every slice with high precision. Structure search and maximum projections are supported, as well as the integration with any dataset for which a structural correspondence can be drawn easily, such as large scale brain transcriptomic studies. As an example we present complete drawings for several human, ape, mouse and Drosophila melanogaster brain segmentations, together with functions to plot transcriptomic datasets such as GTEx and Brainspan. Additionally, 3D representations of every segmentations are supported, both in an interactive and static format.

Disclosures: G.A. D'Agostino: A. Employment/Salary (full or part-time):; Lee Kong Chian School of Medicine, Nanyang Technological University Singapore Dean's Postdoctoral Fellowship. **S.R. Langley:** A. Employment/Salary (full or part-time):; Lee Kong Chian School of Medicine, Nanyang Technological University Singapore Nanayang Assistant Professor Start-up Grant..

Poster

P972. Software Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P972.09

Topic: I.07. Data Analysis and Statistics

Title: Eeg and deep learning - which features to feature? using a gated recurrent network to predict future feature performance

Authors: *J. SLATER[1], J. HOFER[2], B. VIERCK[2];
[1]Stratus, Irving, TX; [2]Positronic AI, Chesterfield, MO

Abstract: Deep learning models for EEG frequently utilize features generated from the data instead of, or in addition to, the raw data itself. Which features to use and how many can be a major stumbling block in model design. To solve this problem, we used a Gated Recurrent Unit (GRU) model run for a small number of epochs to screen features for subsequent inclusion in a multi-feature model - either a Long Short-Term Memory Unit (LSTM) or Convolutional Neural Network (CNN) model. Both models are designed to solve the short-term memory issues of standard recurrent neural networks (RNNs) when evaluating lengthy times series. The "gates" within these models can learn which data in a numeric sequence is important to keep or throw away. The key difference between a GRU and LSTM is that a GRU contains 2 gates (*reset* and *update*) while an LSTM contains three (*input*, *output* and *forget*). A GRU model is less complex and computationally faster than an equivalent LSTM. GRUs perform well with smaller data sets but are usually outperformed by LSTMs especially if there are critical relationships in the time series related to long sequences with long-distance relationships. Two publicly available EEG datasets were utilized - the first containing subjects with mild traumatic brain injury (mTBI) and controls, and a second including subjects diagnosed with Parkinson's disease and controls. For each dataset, multiple features were generated from the EEG data, including a power spectral decomposition (PSD), Lyapunov exponent (LYA), Higuchi Fractal Dimension, and Lempel Ziv Complexity for each electrode, as well as interhemispheric coherence (IC) between matching electrode pairs. A GRU model was constructed for each feature, run for a limited number of epochs, then the average classification accuracy was calculated as a measure of model performance. The generated features were then used, singly and in combination as inputs to each of two different model types - CNN and LSTM. 75% of the files were used for training with 25% withheld for model validation. The primary outcome measure was the maximum validation accuracy achieved. PSD + IC using a CNN model achieved an 82.6% accuracy for the Parkinson's feature set, and PSD + IC + LYA using a LSTM model achieved a 75.4% accuracy for the mTBI data set. Features with the highest average accuracy scores from the GRU model consistently added to the performance of the subsequent multi-feature model - both CNN and LSTM. Some of features with lower average GRU accuracy scores were still later determined to add value to the multi-feature models. This study validates the use of a GRU model as an initial screening tool for feature selection.

Disclosures: **J. Slater:** A. Employment/Salary (full or part-time):; Stratus. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Stratus. **J. Hofer:** A. Employment/Salary (full or part-time):; Positronic AI. **B. Vierck:** A. Employment/Salary (full or part-time):; Positronic AI. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Positronic AI.

Poster

P972. Software Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:30 AM - 10:30 AM

Program #/Poster #: P972.10

Topic: I.07. Data Analysis and Statistics

Support: Kavli Foundation
Simons Foundation

Title: Nwb widgets, a python library for interactive visualizations of nwb data

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Abstract: Neurodata Without Borders (NWB) is emerging as a field standard for exchanging cellular neurophysiology data, providing a structure to share their data in a way that maximizes reusability. In order to facilitate exploration of published NWB data, we have developed an interactive data visualization library, NWB Widgets, to provide informative, efficient, and intuitive visualizations of NWB datasets. NWB Widgets parses the NWB file to provide automatic visualizations of any type of data. Data objects in NWB files are assigned “neurodata types” that indicate what type of data they are (e.g. voltage trace, spike times, body position tracking etc.). NWB Widgets defines a library of visualizations, and maps these visualizations to corresponding neurodata types. This allows it to provide a set of common visualizations for each neurodata instance (e.g. voltage traces are shown as traces over time, spike times are shown as a raster plot, and body position is shown as X vs Y). If no visualizations are available for a particular neurodata type, NWB Widgets searches back through the inheritance tree and uses visualizations from the first parent type with visualizations defined. This approach allows us to efficiently cover the entire space of neurodata types, as well as automatically cover any types in neurodata extensions that subclass core types. NWB Widgets can also be easily extended to create a custom visualization for an existing or new neurodata type. NWB Widgets uses ipywidgets for interactivity. These components let you navigate through the data by setting parameters like time windows, channels selections, and unit selections. The plots are then

rendered using Plotly or matplotlib. The data visualizations are organized in an accordion structure and rendered on-the-fly, only when (or if) they come into view. This allows us to define many visualizations without increasing the code runtime. When a visualization is rendered, it only reads the data in the window of interest from the NWB file. This allows us to visualize data from very large files (as long as the view window is not very large). Our goal is for NWB Widgets to facilitate data exploration and reuse across a variety of platforms. Using the "ros3" HDF5 driver, NWB Widgets can stream data directly from any public s3 bucket, such as those in the DANDI Archive. This is particularly fast if run in DANDI Hub, a free jupyter hub that is configured to work well with the DANDI Archive. The code is open source, and other projects may import and use its components. NWB Explorer imports NWB Widgets, and utilizes the defined visualizations to enable data exploration on their platform.

Disclosures: B. Dichter: None. **L. Tauffer:** None. **C. Baker:** None. **S. Sharda:** None. **M. McCormick:** None. **O. Ruebel:** None.

Poster

P004. Cell Lineage

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P004.01

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH 5P20GM103620-08

Title: Breasi-crispr: an efficient genome editing method to interrogate protein localization and protein-protein interactions in the embryonic mouse cortex

Authors: N. K. TIWARI[1], B. L. MEYERINK[1,3], P. KC[1], C. M. KITTOCK[1,3], A. KLEIN[3], C. EVANS[2], **L.-J. PILAZ**[1,4];
[1]Pediatrics and Rare Dis., [2]Histology Core, Sanford Res., Sioux Falls, SD; [3]Basic Biomed. Sci., Univ. of South Dakota Sanford Sch. of Med., Vermillion, SD; [4]Pediatrics, Univ. of South Dakota Sanford Sch. of Med., Sioux Falls, SD

Abstract: Since the introduction of CRISPR/Cas 9-based genome editing, many advancements have been made to improve the technology's efficiency and specificity and broaden its applications. One such method is "Efficient additions with ssDNA inserts-CRISPR" or Easi-CRISPR. This technique is highly efficacious at generating knock-in or point-mutant mouse models using ssDNA as a template for homology directed repair (HDR). Here, we combine Easi-CRISPR with in utero electroporation in embryonic mouse brains. Together, this method is called Breasi-CRISPR (Brain Easi-CRISPR). Breasi-CRISPR enables knock-in of both short and long epitope tag sequences in genes of interest with high efficiency. It allows for tagging of endogenous proteins in a vast number of embryonic brain cells. Using Breasi-CRISPR, we were

able to detect epitope tagged proteins with both low and high expression levels, such as: ACTIN LMNB1, EMERIN, FMRP, NOTCH1, FXR1, ZBTB7A and RPL22 by immunohistochemistry as soon as one day after electroporation at embryonic day 13 (E13). Co-electroporating CRISPR reagents with an EGFP reporter at this timepoint, we observed efficient gene editing in up to 50% of the EGFP+ cells two and five days after electroporation, and in more than 30% of all the cells located in the electroporated region. This high efficiency gene editing enabled the visualization of tagged proteins by Western Blotting in lysates from individual cortices 2 days after electroporation. With pooled brain lysates from individual litters, we had success performing co-immunoprecipitation (co-IP) experiments with endogenously tagged proteins to test for protein-protein interactions. Moreover, we performed co-IP mass-spectrometry analyses of the Breasi-CRISPR tagged autism-related protein FMRP to discover its interactome in the embryonic cortex. This analysis uncovered interactions between FMRP, ribosomes, and the spliceosome as well as proteins involved with complement activation, histones and intermediate filaments. Finally we demonstrate that Breasi-CRISPR enabled the tagging of endogenous proteins with EGFP in 20% of all the electroporated cells expressing a tdTomato reporter, allowing live-imaging of endogenous proteins in organotypic brain slices as soon as 2 days after electroporation. Together, these data show Breasi-CRISPR is a powerful tool with many applications that will help elucidate the role of pertinent proteins at early timepoints in neurodevelopment.

Disclosures: N.K. Tiwari: None. **B.L. Meyerink:** None. **P. Kc:** None. **C.M. Kittock:** None. **A. Klein:** None. **C. Evans:** None. **L. Pilaz:** None.

Poster

P004. Cell Lineage

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P004.02

Topic: A.01. Neurogenesis and Gliogenesis

Support: AHA Scientist Development Grant
NARSAD Young Investigator Grant

Title: Exon junction complex factor, Rbm8a, is requiredfor cortical interneuron development

Authors: *Y. MAO[1], C. MCSWEENEY[2], F. DONG[2], M. CHEN[3], N. CROWLEY[2], J. MOTT[2]; [2]Biol., [1]Penn State Univ., University Park, PA; [3]Penn State Univ., STATE COLLEGE, PA

Abstract: The formation of the nervous system requires a balance between proliferation, differentiation and migration of neural progenitors. Mutations in genes regulating development impede neurogenesis and lead to neuropsychiatric diseases, such as autism spectrum disorders (ASDs) and schizophrenia (SZ). Recently, mutations in nonsense mediated mRNA decay (NMD)

genes have been associated with ASDs, intellectual disability (ID), and SZ. Here, we seek to examine the role of exon junction complex (EJC) gene, *Rbm8a*, in the development of the nervous system. We selectively knocked out RBM8a in neural stem cells, and the resulting mice exhibited microcephaly, early postnatal lethality, and altered distribution of excitatory neurons in the neocortex. Moreover, *Rbm8a* haploinsufficiency in the central nervous system results in a decrease in cell proliferation in the ganglionic eminences (GE). A significant decrease in parvalbumin (PV)+ and neuropeptide Y (NPY)+ interneurons in the cortex, and abnormal distribution of interneurons are observed. Consistently, neurons in the cortex of knockout (KO) mice show a significant decrease in GABA frequency. To further examine the role of Rbm8a in interneuron differentiation, conditional KO (cKO) of *Rbm8a* in Nkx2.1 interneuron progenitor cells affects Nkx2.1 progenitor proliferation and alters interneuron distributions. Taken together, these data indicate the critical role of Rbm8a in interneuron development, and establish that perturbation of this gene leads to profound developmental deficits.

Disclosures: **Y. Mao:** A. Employment/Salary (full or part-time):; Penn State University. **C. McSweeney:** None. **F. Dong:** None. **M. Chen:** None. **N. Crowley:** None.

Poster

P004. Cell Lineage

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P004.03

Topic: A.01. Neurogenesis and Gliogenesis

Support: T32 CA080621

Title: Identification of human astrocyte progenitors and their association to glioblastoma subtypes

Authors: *S. VAN GULDEN, C.-Y. PENG, A. K. LINDEN, J. A. KESSLER; Northwestern Univ., Chicago, IL

Abstract: Understanding human astrocyte lineage cells and their involvement in brain malignancies has been limited due to inaccessibility of human brain tissue. Here we present an approach for generating astrocyte progenitors and astrocytes using human pluripotent stem cells (hPSCs), as a model for studying the developmental diversity of astrocytes and how it correlates with glioblastoma (GBM) subtypes. Using a combination of biochemical, molecular, and single cell transcriptomic techniques, we characterized the hPSCs-derived astrocytes at two developmental time points and demonstrated that cells generated follow normal developmental progression along the astroglial lineage. Due to the heterogeneous nature of our culture at the astrocyte precursor stage, we used a combination of surface markers to isolate and characterize four different progenitor populations that arise during astrocyte differentiation. Transcriptional

analysis revealed a correlation between gene expression profiles in hPSC-derived astrocyte progenitor populations with proneural and mesenchymal GBM subtypes. Additional characterization of these progenitor populations revealed genes associated with Notch signaling and TGF-β signaling pathways. These findings suggest that hPSC-derived astrocyte progenitors and astrocytes share developmental mechanisms replicated by the cell types found in GBM tumors. Therefore, understanding how the generation of glioma subtypes is linked to the developmental processes that regulate neural and glial diversification has important implications for the understanding and treatment of this disease.

Disclosures: S. Van Gulden: None. **C. Peng:** A. Employment/Salary (full or part-time):; Northwestern University. **A.K. Linden:** None. **J.A. Kessler:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Northwestern University.

Poster

P004. Cell Lineage

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P004.04

Topic: A.01. Neurogenesis and Gliogenesis

Support: Spanish MICINN PID2019-105218RB-I00
Fundación Ramón Areces CIVP9A5928

Title: Lineage Commitment Of Daughter Cells Generated From Single Pallial NG2- and GFAP-expressing Progenitors

Authors: *A. OJALVO-SANZ, R. SÁNCHEZ-GONZÁLEZ, L. LÓPEZ-MASCARAQUE; Cajal Inst., Madrid, Spain

Abstract: Neural Progenitor Cells (NPCs) are generated from a pool of neuroepithelial cells that begin to transition into Radial Glial Cells (RGC). These cells, in partially overlapped waves, first producing several self-amplifying symmetric divisions and then RGCs divide asymmetrically generating either neurons, astrocytes, NG2-glia or oligodendrocytes. Few embryonic cortical progenitors appear to be lineage-multipotent, suggesting the existence of progenitors committed to one specific lineage. Other studies reveal a different spatio-temporal pattern related to the progenitor cell potential, reinforcing the idea that **NPCs constitute a heterogeneous population**. In addition, the NG2-glia is another remarkable cell type that serves as a progenitor. Then, it is known how NPCs produce the diverse cell progeny, although many unknowns about lineage relationships and progenitor heterogeneity remain unravelled. To address this, we looked at the derived-cell progeny of single GFAP- or NG2-expressing progenitors targeted with stable

and heritable labelling. To target single NG2- or GFAP-progenitors within the pallial SVZ we used either **UbC-(NG2-PB)-StarTrack** or **UbC-(GFAP-PB)-StarTrack**, respectively. After *in utero* electroporation of those StarTrack strategies, at different mouse embryonic stages (E12, E14, E16) and neonatal (P0), we performed a clonal analysis of their derived-cell progeny at P30. Our findings provide fundamental aspects of the fate and lineage cell potential of GFAP- or NG2-expressing progenitors during development.

Disclosures: A. Ojalvo-Sanz: None. **R. Sánchez-González:** None. **L. López-Mascaraque:** None.

Poster

P004. Cell Lineage

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P004.05

Topic: A.01. Neurogenesis and Gliogenesis

Title: Sibling Cell Relationships of Single Subpallial Progenitor Cells Using the UbCStarTrack

Authors: *R. SÁNCHEZ-GONZÁLEZ, A. OJALVO-SANZ, S. BARRIOLA, L. LÓPEZ-MASCARAQUE;
cajal insitutute, madrid, Spain

Abstract: Over the last decades a growing number of evidences pointed out on the heterogeneity of neural progenitor cells (NPCs). Neural development requires a perfect balance between proliferation and differentiation of NPCs to give rise a functional adult nervous system. In our research, we are focused on the heterogeneity of NPCs, to unravel the diversity of the different neural lineages. NPCs give rise to distinct neural cell types at different embryonic and adult stages increasing the cell diversity in the brain. Using a multi-color genetic lineage tracing system, UbC-StarTrack, we targeted specific progenitor cells to analyze the cell type and location of their adult cell-derived progeny as well as their lineage relationships. To this end, we designed different StarTrack strategies to permanently label the individual progenitor cells and their progeny accordingly with the identity of the NPCs (Sánchez-Gonzalez et al 2020a). At this regard, we performed a new PiggyBac transposase under the expression of Gsh-2 promoter (called UbC-(Gsh-2-hyPB)-StarTrack) to specifically target progenitor cells located in the ventral areas of the ventricular surface of early mouse embryo. The derived-cell progeny of those subpallial progenitors produce different neural types (astrocytes, oligondrocytes, NG2 and even neurons), displaying diverse clonal patterns in relation to both their ontogenic origin and cell identity.Our findings provide fundamental aspects of the progenitor potential, cell fate and the clonal relationships of their derived-cell progeny that might help to gain new insights into their behavior, complexity and functionality.

Disclosures: R. Sánchez-González: None. **A. Ojalvo-sanz:** None. **S. Barriola:** None. **L. López-mascaraque:** None.

Poster

P004. Cell Lineage

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Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P004.06

Topic: A.01. Neurogenesis and Gliogenesis

Support: ACS Grant RSG-128945

Title: Response to codon biased transcripts changes in a defined neural lineage.

Authors: *R. STEWART, S. ALLEN, D. FOX;
Duke Univ., Durham, NC

Abstract: Codon bias occurs when certain codons appear more frequently than their synonymous counterparts. This phenomenon occurs throughout all forms of life and has been shown to be important in determining protein expression at the steps of both transcription and translation. Although the impact of codon bias has not been thoroughly studied during animal development and in different tissues, we are beginning to understand how cellular response to codon biased transcripts intersects with cell identity and proliferative state. We have undertaken one of the first studies of cell and tissue-specific differences in codon bias in the model organism *Drosophila* and have uncovered some of the first evidence that tissues in the same animal respond differently to codon usage. Specifically, in cell types within the developing *Drosophila* brain, we find robust protein expression from reporters enriched in rarely used codons, whereas most other tissues in the fly do not express such reporters. Within the brain itself, neural stem cells are unable to express proteins encoded by rare-codon enriched genes, while differentiated neurons can. Cell identity appears tied to ability to express transcripts enriched in rare codons, as experimentally blocking neuronal differentiation drastically reduces expression of a rare-codon enriched reporter. To uncover molecular regulators that enable neurons to specifically express rare-codon enriched reporters, we have initiated a reverse genetic screen. This screen used RNAi and the UAS-GAL4 system to knock down such potential regulators enriched in neuroblasts or neurons. We assessed the role of each gene in enabling translation of rare-codon enriched transcripts by measuring expression of our rare-codon enriched reporter. Results of the screen will be presented at the poster. Using our *Drosophila* model, we may ultimately identify new regulatory mechanism during neuronal development and function: cellular response to codon bias.

Disclosures: R. Stewart: None. **S. Allen:** None. **D. Fox:** None.

Poster

P004. Cell Lineage

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P004.07

Topic: A.01. Neurogenesis and Gliogenesis

Title: Loss of Ezh2 within the medial ganglionic eminence alters lineage commitment in cortical interneuron progenitors

Authors: *C. RHODES, M. SOHN, D. ASOKUMAR, Y. ZHANG, R. DALE, T. PETROS; NIH, Bethesda, MD

Abstract: GABAergic interneurons are a heterogenous cell population and their dysfunction is associated with numerous brain disorders. Interneurons are born in the ganglionic eminences (MGE, LGE and CGE), with distinct subtypes arising from different GE domains. Despite substantial evidence suggesting that initial interneuron subtypes are specified within these neurogenic niches, only a handful of fate-determining genes have been identified. One possible explanation is that progenitors contain epigenetic signatures directing cells towards a particular interneuron fate that are not yet apparent in the transcriptome. Ample evidence indicates that epigenetic mechanisms induce heritable changes in gene expression and are critical in mediating cell fate and differentiation in a variety of tissues. Yet we lack a comprehensive understanding of how epigenetic states influence interneuron cell fate in normal development and disease models.To explore the relationship between interneuron cell fate and epigenetic mechanisms in neural progenitors, we performed single cell RNA (scRNA-Seq) and single cell assay for transposase-accessible chromatin (scATAC-Seq) sequencing to determine the transcriptome and chromatin accessibility in the embryonic MGE, LGE, CGE and cortex. This allows us to screen for candidate enhancers and other non-coding regions regulating expression of specific interneuron subtypes. We detected distinct genes and corresponding chromatin profiles that are restricted to specific progenitor zones and interneuron lineages. By examining ATAC peaks in lineage-specific genes across cell types, we detected putative enhancers associated with specific GEs and potentially distinct interneuron subtypes. To expand on these initial observations, we generated a conditional knockout of *Enhancer of zeste homolog 2* (*Ezh2*) in MGE progenitors. Ezh2 regulates histone methylation and acts as a transcriptional repressor; thus loss of Ezh2 likely disrupts chromatin architecture and gene expression. These Ezh2 cKO mice displayed a significant decrease in PV+ interneurons in multiple brain regions. We are currently performing scRNAseq and scATACseq on the MGE of Ezh2 cKO to assess how the transcriptome and chromatin landscape is altered in these mice. These insights increase our understanding of how epigenetic modifications regulate interneuron fate and could open new avenues for understanding how disease-associated genes or genomic loci could perturb interneuron fate and maturation.

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Poster

P004. Cell Lineage

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Program #/Poster #: P004.08

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH R01 NS119690
NIH R01 NS096240
NIH R01 MH110555
NIH R21 NS092034-01A1

Title: Dual embryonic origins of the SNc define cohorts with distinctive molecular and anatomical properties

Authors: *M. PEREIRA LUPPI[1], M. AZCORRA[2], G. CARONIA-BROWN[2], J.-F. POULIN[4], Z. GAERTNER[2], S. GATICA[2], O. MORENO RAMOS[2], N. NOURI[2], M. DUBOIS[5], C. RAMAKRISHNAN[6], L. E. FENNO[7], Y. KIM[6], K. DEISSEROTH[8], F. CICCHETTI[5], D. A. DOMBECK[3], R. AWATRAMANI[2];
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Abstract: Dopamine (DA) neurons in the ventral tier of the substantia nigra pars compacta (SNc) degenerate prominently in Parkinson's disease, while those in the dorsal tier are relatively spared. Defining the molecular, functional, and developmental characteristics of each SNc tier is crucial to understand their distinct susceptibility. We demonstrate that *Sox6* expression defines a ventral and a dorsal tier in the SNc. The *Sox6+* ventral tier includes an *Aldh1a1+* population and is enriched in gene pathways that underpin vulnerability. This dorso-ventral division extends beyond vulnerability - *Sox6+* neurons project to the dorsal striatum and show activity correlated with acceleration. *Sox6-* neurons project to the ventral striatum and respond to rewards. Moreover, we show that this adult division is partially encoded early in development. Overall, our work demonstrates a dual origin of the SNc that results in DA neuron cohorts with distinct molecular profiles, projections, and functions.

Disclosures: M. Pereira Luppi: None. **M. Azcorra:** None. **G. Caronia-Brown:** None. **J. Poulin:** None. **Z. Gaertner:** None. **S. Gatica:** None. **O. Moreno Ramos:** None. **N. Nouri:**

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Poster

P004. Cell Lineage

Location: Virtual Only

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Program #/Poster #: P004.09

Topic: A.01. Neurogenesis and Gliogenesis

Support: NSF Grant 1553764
NSF Grant 1338188

Title: Quantifying radial glial clone morphology and dynamics in the developing zebrafish brain

Authors: L. B. DOSTER[1], N. L. BROCKWAY[1], Z. T. COOK[2], *T. A. WEISSMAN[1];
[1]Biol., Lewis & Clark Col., Portland, OR; [2]Neurol., Oregon Hlth. & Sci. Univ., Portland, OR

Abstract: During neurogenesis, individual mitotic radial glial cells produce postmitotic daughter neurons, forming small clones of related cells. These radial glial clones serve as a fundamental unit for neural proliferation during brain development; however, little is known about the functional relationship among clonally related cells. Sister cells can ultimately function in the same neural circuit and be involved in cortical organization (Yu et al., 2009; Li et al, 2012; Ohtsuki et al., 2012). Further, clonally related cells can undergo coordinated cell death during neurogenesis (Brockway et al., 2019). To better understand the functional role of clones, we have developed quantitative criteria to classify and measure radial clones in the developing zebrafish hindbrain. Using Brainbow expression to color-code and track radial clones in vivo, we applied semi-automated techniques to measure four main aspects of radial clones: 1) the number and size of cells within each clone; 2) spatial orientation; 3) the color associations among cells within a clone; and 4) the temporal dynamics of radial clones. Using these measures, we compared clones quantitatively across different conditions. These tools help to track clones and measure clonal dynamics during neural development; ultimately we aim to understand how radial clone organization contributes to brain function.

Disclosures: L.B. Doster: None. **N.L. Brockway:** None. **Z.T. Cook:** None. **T.A. Weissman:** None.

Poster

P014. Stem Cell Approaches in Neurodegenerative / Neuropsychiatric Disorders

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Program #/Poster #: P014.01

Topic: A.03. Stem Cells and Reprogramming

Support: NIH/NINDS grant NS112910
DoD grant W81XWH2010186

Title: Similarities and differences of reprogrammed neurons via direct conversion and human induced pluripotent stem cells in modeling DYT1 dystonia

Authors: *B. DING;
Univ. of Louisiana At Lafayette, Lafayette, LA

Abstract: DYT1 dystonia is a hereditary neurological movement disease. It is caused by a heterozygous deletion in *TOR1A* gene, resulting in the loss of one glutamate residues in the carboxy-terminal region of TOR1A protein (ΔE). Although ΔE has been shown to be a loss-of-function mutation, the mechanisms underlying ΔE leads to changes in the nervous system to cause dystonia remain poorly understood. While rodent models provide insights into disease mechanisms, significant species-dependent differences exist because animals with the identical heterozygous mutation fail to show pathology. Excitingly, reprogramming of human neurons from adult fibroblasts overcomes the limitation of inaccessibility to patient neurons and provides an unprecedented approach in deciphering the molecular pathogenesis underlying neurological diseases. In this study, DYT1 neurons have been generated via two strategies: (1) direct conversion from patient fibroblasts using lentiviral delivery of transcription factors, and (2) induced pluripotent stem cells (iPSCs)-based induction and differentiation. Using different combinations of reprogramming factors, different neuronal subtypes could be generated, including generic neurons (GNs), motor neurons (MNs), and cholinergic interneurons (ChINs). Directly converted MNs (diMNs) and iPSC-derived MNs (iMNs) have been systematically examined and compared in modeling DYT1 dystonia. Diseased neurons generated by both approaches showed similar phenotypes in impaired neurodevelopment, abnormal nuclear morphology, disrupted nucleocytoplasmic transport and dysregulation of nuclear lamin B1. Interestingly, diMNs showed more severe deficits than iMNs. Thus, diMNs are excellent materials for pathogenesis study based on immunocytochemistry. Compare to diMNs, iMNs possess unique advantages, such as much higher purity and yield, and the shorter culture time to reach the mature stage (3 weks vs 6 weeks). Generation of large amount of highly pure neurons via iPSC-based approach makes it possible to biochemically identify dysregulated factors in diseased neurons.

Disclosures: B. Ding: None.

Poster

P014. Stem Cell Approaches in Neurodegenerative / Neuropsychiatric Disorders

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P014.02

Topic: A.03. Stem Cells and Reprogramming

Support: CFI Grant 037755 to J.L.
2016 NARSAD Young Investigator Grant 24990 to J.L.

Title: Bipolar disorder iPSC-derived neural progenitor cells display aberrant store-operated calcium entry and neurodevelopment

Authors: *T. HEWITT[1], B. ALURAL[1], J. WANG[2], M. TILAK[1], N. JONES[1], S. D. SHERIDAN[2], R. H. PERLIS[2], J. LALONDE[1];
[1]Mol. and Cell. Biol., Univ. of Guelph, Guelph, ON, Canada; [2]Ctr. for Quantitative Hlth., Massachusetts Gen. Hosp., Boston, MA

Abstract: Bipolar disorder (BD) is a chronic and disabling condition characterized by alternating episodes of severe depression and mania. The large majority of cellular research conducted about BD focuses on phenotypes present at the mature neuron stage, even though some manifestations of BD may occur earlier in development. This begs the question of whether it can be considered a neurodevelopmental disease. Although deviations in neuron development are suspected to contribute to the pathophysiology of BD, the precise molecular underpinnings remain unknown. In cells, calcium plays a central role in development via signaling cascades. As such, calcium accumulates in the endoplasmic reticulum (ER) from where it can be rapidly released in response to a second messenger. Calcium levels are, therefore, tightly regulated through store-operated calcium entry (SOCE)—a mechanism that promotes calcium influx through ORAI channels when the ER is depleted. Recently, we found striking evidence of aberrant SOCE activity in BD patient-derived induced pluripotent stem cells (iPSCs) differentiated into neural progenitor cells (NPCs) in comparison to iPSC-NPCs derived from healthy control individuals. Additionally, an RNA-sequencing experiment using this model uncovered upregulated neurodevelopmental processes in the BD-derived NPCs. This led us to hypothesize that calcium dynamics and neurodevelopment were aberrant in the BD-derived progenitors and neurons. To begin testing this, the sharp decrease in SOCE between BD patient-derived and control iPSC-NPCs was found to correlate with reduced STIM/Orai puncta formation. Probing the premature differentiation phenotype, we sought to compare the progenitors' capacity to self-renew and differentiate as neurons. To do this, we employed an EdU proliferation assay and a neurosphere formation assay. These led to the discoveries that BD-derived NPCs exhibited slower rates of proliferation, and faster rates of neurite outgrowth and migration. Finally, to model these processes in a 3D context, we generated 50-day old cerebral organoids from healthy- and BD-derived iPSCs. Using immunocytochemistry for radial glia markers PAX6 and pVimentin (dividing radial glia), we found that BD-derived organoids displayed smaller and thinner subventricular zones. As these zones are the sites of neurogenesis in developing and mature brains, this decreased area suggests a reduced progenitor cell pool due to premature differentiation. Together, our findings suggest that both SOCE and development are dysregulated in BD-derived NPCs, which implicates neurodevelopment in the pathophysiology of BD.

Disclosures: T. Hewitt: None. **B. Alural:** None. **J. Wang:** None. **M. Tilak:** None. **N. Jones:** None. **S.D. Sheridan:** None. **R.H. Perlis:** None. **J. Lalonde:** None.

Poster

P014. Stem Cell Approaches in Neurodegenerative / Neuropsychiatric Disorders

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Program #/Poster #: P014.03

Topic: A.03. Stem Cells and Reprogramming

Support: Maryland Stem Cell Research Fund, 2019-Mscrfd-5093

Title: Establishment of a Spatially Separated Microfluidic Culture System to Study the Regeneration of Axons from Human iPSC-Derived Sensory Neurons in the Context of Diabetic Peripheral Neuropathy

Authors: *M. JAMES, A. VENKATESAN, M. POLYDEFKIS, M. FARAH;
Johns Hopkins Univ. Sch. of Med., Baltimore, MD

Abstract: Diabetic peripheral neuropathy (DPN) is the most common complication of diabetes and is estimated to affect over 50% of the almost 35 million people diagnosed with diabetes in the United States. One important aspect of this disease that is not yet well understood is why nerves from people with diabetes mellitus 2 (DM2) regenerate more slowly than control patients, especially within the epidermal layer. We believe the neuronal origin of the axon, and more importantly, the target environment has a large effect on the regenerative capacity of sensory axons. I.e., that sensory axons from diabetic patient neurons will regenerate more slowly than controls, and that no matter the sensory axon origin, axons will regenerate more slowly in the epidermal environment of a diabetic patient than that of a control patient. To study this pathogenesis, we are creating a spatially separated co-culture system of sensory neurons and target tissue (skin) in microfluidic devices. We have established a system for culturing sensory neurons within these devices and for studying the regenerative capacity of those cells. Human induced pluripotent stem cell (iPSC)-derived sensory neurons were cultured in cell body compartments of microfluidic devices and allowed to extend axons into the axonal compartments. We have successfully matured and cultured these neurons in microfluidic devices and observed long axons, ~ 3mm, in un-injured axonal compartments. We have confirmed the generation of mature sensory neurons by staining for Islet 1 (ISL1) and POU domain, class 4, transcription factor 1 (POU4F1) also known as brain-specific homeobox/POU domain protein 3A (BRN3A). To quantify the efficiency of differentiation within the devices, we calculated the percent positive cells for both ISL1 (95%) and Brn3A (87%) within the cell body compartment, which confirmed our ability to reliably differentiate these cells within microfluidic devices at a high efficiency. Once the sensory neurons reached maturity, axotomies were performed in some of the axonal compartments by vigorous vacuum aspiration to severe the axons at the entrance to

the compartment, which allows us to track axonal regrowth and regeneration. Axons reliably regrew into the axonal compartment, and they continued to grow for at least 7 days post-axotomy. With this work, we have successfully been able to create a system for reliably studying distal axon growth and regeneration in human iPSC derived sensory neurons. This system also creates the opportunity for future spatially separated sensory neuron co-culture experiments for the study of DPN and other sensory neuropathies.

Disclosures: M. James: None. **A. Venkatesan:** None. **M. Polydefkis:** None. **M. Farah:** None.

Poster

P014. Stem Cell Approaches in Neurodegenerative / Neuropsychiatric Disorders

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P014.04

Topic: A.03. Stem Cells and Reprogramming

Support: University of Georgia, Start-up Funds

Title: Deciphering stress-related sympathetic neuropathology in familial dysautonomia using induced pluripotent stem cell-based disease modeling

Authors: *H. WU[1], W. YU[1], H. LIU[1], F. LEFCORT[2], N. ZELTNER[1];
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Abstract: Familial dysautonomia (FD) is a neurodevelopmental and neurodegenerative disorder that mainly affects the peripheral nervous system, especially the sympathetic nervous system (SNS). One of the most devastating symptoms in patients with FD is the dysautonomic crisis, a severe dysregulation and dysfunction of the SNS triggered by stress. Interestingly, brain function which is the main stress responding center is minimally impacted in FD. This highlights the importance of the SNS in FD pathology. To date, the role of SNS dysfunction in FD, specifically, the pathological mechanism in sympathetic neurons (symNs) that leads to developmental defects, degeneration and FD crisis, remains unclear. A challenge of that is the lack of a accessible human model system comprised of symNs. Here, we adopt the human pluripotent stem cell (hPSC)-based disease modeling to investigate the defects in FD patient, healthy control and isogenic control-derived symNs. We show in early stages of differentiation, FD iPSCs display inefficient neural crest (NC) specification. When differentiating into sympathetic neuroblasts, FD NCs have a lower survival rate, yet remaining progenitor cells still generate symNs. These results reveal early developmental defects of SNS in FD. Using microelectrode array and calcium imaging, we found that FD symNs are spontaneously hyperactive. Accordingly, FD symNs drive higher cardiomyocyte beating in a co-culture system. We further confirmed symN hyperactivity using primary symN from mice lacking ELP1, the main gene mutation in FD. To understand the molecular cause of hyperactivity, we show that

ELP1 rescue does not rescue hyperactivity, suggesting the complexity of FD mechanisms in human. In addition, we found decreased intracellular norepinephrine (NE) levels in FD symNs, suggesting impaired autoregulatory function. Indeed, FD symNs expressed less norepinephrine transporter (NET), a NE reuptake transporter, and displayed decreased NE reuptake ability. Treating control symNs with NET inhibitor increased their activity, which supports our hypothesis. Furthermore, a clinically tested FD drug dexmedetomidine significantly suppressed FD symN hyperactivity, showing that symN pathology is a key feature in FD. Finally, we performed a mini drug screen and showed that our hPSC-based platform can be used for future FD drug discovery. Together, our studies show that symN hyperactivity caused by ill-controlled neurotransmitter release may contribute to FD pathology in patients. Our *in vitro* system allows the examination of molecular mechanisms underlying symN defects in FD and shows that hPSC-symN is a competent tool for studying SNS diseases.

Disclosures: H. Wu: None. **W. Yu:** None. **H. Liu:** None. **F. Lefcort:** None. **N. Zeltner:** None.

Poster

P014. Stem Cell Approaches in Neurodegenerative / Neuropsychiatric Disorders

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P014.05

Topic: A.03. Stem Cells and Reprogramming

Support: APVV-17-0642
APVV-18-0515
VEGA 2/0110/20
JPsustaiND-681043

Title: Ipsc-astrocytes derived from patients with Alzheimer's disease show altered Ca^{2+} signalling and ability to degrade tau protein.

Authors: V. BREZOVAKOVA, S. JADHAV, D. CIZKOVA, *E. SYKOVA;
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Abstract: Astrocytic tau pathology has been described in many neurodegenerative diseases including Alzheimer's disease (AD). Growing evidence points to major structural and functional differences between human and rodent astrocytes, thus prompting the research on human glial cells. Induced pluripotent stem cells (iPSC) represent a valuable tool to model and study human cells in health and disease. We generated human astrocytes from iPSCs derived from patients with sporadic AD (sAD), familial AD (fAD), and non-demented controls (NDC). Upon *in vitro* differentiation, all clones gained astrocyte-specific morphological and proteomic features without considerable inter-clonal differences. Since reactive astrocytes contribute to the dysfunction of Ca^{2+}signalling in AD, we analyzed the Ca^{2+} activity of individual clones upon

glutamate stimulation. We found that sAD astrocytes exhibited Ca^{2+} overload accompanied by hyperphosphorylation of protein kinase C- alpha/beta II when compared to healthy astrocytes. Moreover, sAD astrocyte secretome altered Ca^{2+} influx in neurons derived from non-demented control iPSCs (iNeurons). Astrocytes actively participate in amyloid-beta metabolism, however, their involvement in tau processing is yet to be elucidated. We investigated the secretome of astrocytes for their potential to metabolize tau protein *in vitro*. We show that tau induces the secretion of astrocytic gelatinases, specifically metalloproteinase 9 and 2 (MMP-9, MMP-2). Both MMP -9 and MMP -2 were responsible for the degradation of monomeric and aggregated tau species *in vitro*. Moreover, we found that the degrading capacity of astrocyte conditioned media (ACM) from NDC and sAD was significantly higher than that from fAD ACM. The degrading capacity of ACM also correlated with the levels of pERK1/2, a kinase involved in MMP secretion. ACM-degraded tau was less toxic when added to iNeurons than "native tau" *in vitro*. Our study unveils a mechanism by which astrocytes can modulate tau pathology. The activity of metalloproteinases secreted by astrocytes contributes to neuroprotection via attenuation of tau toxicity on neurons.

Disclosures: V. Brezovakova: None. **S. Jadhav:** None. **D. Cizkova:** None. **E. Sykova:** None.

Poster

P014. Stem Cell Approaches in Neurodegenerative / Neuropsychiatric Disorders

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P014.06

Topic: A.03. Stem Cells and Reprogramming

Support: PhD studentship grant from the UK Medical Research Council

Title: Protein Kinase A inhibition modulates endogenous levels of dipeptide repeat proteins in C9orf72 ALS/FTD patient iPSC-derived neurons

Authors: *S. SALOMONSSON[1,2], N. V. LICATA[3], K. M. WILSON[1,2], M. PANCHER[4], V. ADAMI[4], A. M. ISAACS[1,2], A. PROVENZANI[3];
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Abstract: Introduction
A well-characterised feature of Amyotrophic Lateral Sclerosis (ALS) and Frontotemporal Dementia (FTD) associated with C9orf72 mutation is non-canonical repeat associated non-AUG (RAN) translation of a G_4C_2 repeat expansion, producing disruptive dipeptide repeat proteins (DPRs) in the patient brain. The mechanisms by which DPRs are produced and escape

degradation are not well understood, particularly in terms of endogenous levels in patient-derived neurons. We investigate novel small molecule modulators of DPR levels through a primary screen and validate the hits using independent patient iPSC-derived motor neuron lines.

Methods

N=3 independent patient iPSC lines were used. Differentiation of mature spinal cord motor neurons was induced through a 2D dual SMAD inhibition protocol. iPSC-derived motor neurons (iPSC-MNs) were treated with small molecules identified in a primary high throughput screen of RAN translation modifiers, which screened a library of compounds using a cell model expressing a (G_4C_2)x58-GFP reporter construct and an AUG-driven RFP control. Endogenous poly-GP and poly-GA levels in iPSC-MNs were quantified through MSD immunoassay.

Results

Four hits from the primary screen were carried forward to testing in patient iPSC-MNs. Two of the small molecules targeted the cyclic AMP pathway, whereof one inhibited Protein Kinase A (PKA) activity and stood out as significantly effective in reducing DPR levels in patient iPSC-MNs, although there were donor-dependent response differences to treatment. Donor-dependent differences in baseline DPR levels were observed, which did not correlate with repeat length.

Conclusion

We found a role for PKA activity in modulation of endogenous DPR levels, which is a novel investigative avenue in the search for specific therapeutic targets for C9orf72-associated ALS/FTD. The patient iPSC-MN lines responded differently to treatment, which may be due to differences in DPR turnover rates or RAN translation machinery, highlighting the importance of utilising patient-derived cell lines from several independent genetic backgrounds when investigating therapeutic targets for genetic diseases.

Disclosures: S. Salomonsson: None. **N.V. Licata:** None. **K.M. Wilson:** None. **M. Pancher:** None. **V. Adami:** None. **A.M. Isaacs:** None. **A. Provenzani:** None.

Poster

P014. Stem Cell Approaches in Neurodegenerative / Neuropsychiatric Disorders

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P014.07

Topic: A.03. Stem Cells and Reprogramming

Support: Lieber Institute for Brain Development
The MGH Collaborative Center for X-Linked Dystonia-Parkinsonism
Maryland Stem Cell Research Fund
NIH Grant T32MH015330

Title: Deciphering the molecular phenotypes underlying the differential degeneration of the adult neostriatum in X-linked Dystonia-Parkinsonism (XDP) through postmortem brain tissues and iPSC-derived striatal organoids

Authors: *L. D'IGNAZIO[1,2], T. SAWADA[1], A. S. FELTRIN[1], K. J. M. BENJAMIN[1,2,3], R. S. JACOMINI[1,2], B. QAMAR[1], R. ARORA[1,6], B. TORRES[1], M. ZABOLOCKI[7], C. BARDY[7], K. E. DIFFENDERFER[8], A. R. PANKONIN[8], T. A. EVANS[1,2], W. T. HENDRIKS[9,10], T. M. HYDE[1,3], J. E. KLEINMAN[1,3], D. R. WEINBERGER[1,2,3,4,5], D. C. BRAGG[9,10], A. C. M. PAQUOLA[1,2], J. A. ERWIN[1,2,3,4];
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Abstract: X-linked Dystonia-Parkinsonism (XDP) is an inherited adult-onset movement disorder affecting predominantly male of Filipino descent. At molecular level, a disease-specific SINE-VNTR-Alu (SVA) retrotransposon insertion occurs within intron 32 of *TAF1*, core member of the TFIID complex mediating the RNA pol II promoter initiation. Histopathological analyses unveiled a progressive differential degeneration in the adult neostriatum of XDP patients. However, whether and how a partial loss of *TAF1* function and/or an SVA-driven pathological gain of function contribute to XDP is yet unknown. While all XDP males are usually affected, females are obligate heterozygous carriers not manifesting the full syndrome. Because of X-chromosome inactivation (XCI), females transcriptionally silence the majority of genes on one X chromosome, including *TAF1*. Understanding how XDP manifests in female carriers can reveal key features about XDP pathophysiology. Here, we analyze female wild-type and carrier-derived induced pluripotent stem cells (iPSCs) as well as postmortem brain tissues to identify intrinsic properties of *TAF1* expression that may underlie differential degeneration in XDP. We characterized iPSCs from female carrier individuals, identifying a set of isogenic lines where one clonal iPSC line expresses the wild-type X and two other clonal iPSC lines express the XDP haplotype. We found that *TAF1 is* decreased in female wild-type iPSCs, XDP carrier iPSCs, and several regions of the female post-mortem brain, compared to males. However, in the striatum, *TAF1* is not sex-differentially expressed, perhaps indicating a regional sensitivity to *TAF1* expression. Also, in the caudate nucleus, *TAF1* expression decreases after 15 years of age, implying cell-type, age and sex specific mechanisms regulating its expression, and highlighting the importance of disease-relevant cellular models. To recapitulate the mature and complex XDP cellular system, we developed a novel protocol to differentiate female carrier and male control, XDP, and SVA-deleted XDP iPSCs into striatal organoids. Through an extensive phenotypical and molecular characterization, we found that striatal organoids are mainly composed of medium spiny neurons (MSNs) and striatal interneurons, exhibiting a functional network activity up to at least 1 year. Our striatal organoid model can serve as a valuable 3D cellular model of XDP,

recapitulating key molecular features of the disease. This will contribute to elucidate mechanisms modulating the transcriptional dysregulation in XDP, shedding light upon novel insights of XDP etiology.

Disclosures: L. D'Ignazio: None. **T. Sawada:** None. **A.S. Feltrin:** None. **K.J.M. Benjamin:** None. **R.S. Jacomini:** None. **B. Qamar:** None. **R. Arora:** None. **B. Torres:** None. **M. Zabolocki:** None. **C. Bardy:** None. **K.E. Diffenderfer:** None. **A.R. Pankonin:** None. **T.A. Evans:** None. **W.T. Hendriks:** None. **T.M. Hyde:** None. **J.E. Kleinman:** None. **D.R. Weinberger:** None. **D.C. Bragg:** None. **A.C.M. Paquola:** None. **J.A. Erwin:** None.

Poster

P048. Autism: Synaptic and Cellular Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P048.01

Topic: A.07. Developmental Disorders

Support: Max Planck Society

Title: Som-specific knockout of the ASD candidate gene PTEN results in elevated fear and anxiety and disrupts local circuitry in the central amygdala

Authors: *T. W. HOLFORD, M. BOLTON;
Max Planck Florida Inst., Jupiter, FL

Abstract: Autism spectrum disorder (ASD) is a complex disorder with large individual variability, where every case has differences in the type and severity of symptoms. Despite the recent increase in diagnoses, scientists have advanced considerably less in their understanding of the mechanisms of ASD because few individual genes that are implicated in ASD are mutated in much more than 1% of patients. One proposed mechanism is that the dysfunction of GABAergic interneurons may play a role in the development and progression of the disorder by interrupting the excitatory and inhibitory balance of neural networks. In our research, we elucidate the role of one class of interneurons in ASD by knocking out a high-risk gene (phosphatase and tensin homologue on chromosome ten, or PTEN) selectively in somatostatin- expressing (SOM+) interneurons. We found that mice with a SOM+ conditional knockout of PTEN had elevated levels of anxiety and fear recall, suggesting a disruption of amygdala function. We then investigated potential dysfunction at the cellular and circuit levels using confocal microscopy, electrophysiology, and 2P local circuit mapping. We found that SOM+ cells lacking PTEN were overgrown morphologically, with larger cell bodies and larger, more complex dendritic arbors. Additionally, SOM+ cells in the central amygdala (CeA) lacking PTEN had elevated levels of excitatory drive from the basolateral amygdala (BLA) as well as a drastic disruption of lateral inhibition within the CeA, seen by decreased connection probability and reduced inhibitory post

synaptic currents. Given what is known about central amygdala circuitry, these deficits in CeA SOM+ neuron activity conceivably underlie the fear and anxiety-related phenotype observed in mice with a conditional SOM+ PTEN knockout.

Disclosures: T.W. Holford: None. **M. Bolton:** None.

Poster

P048. Autism: Synaptic and Cellular Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P048.02

Topic: A.07. Developmental Disorders

Support: Autism Spectrum Program of Excellence (ASPE)

Title: Different neurexin isoforms uniquely contribute to temporal aspects of the behavioral response to food deprivation

Authors: *B. L. BASTIEN, M. P. HART;
Genet., Univ. of Pennsylvania, Philadelphia, PA

Abstract: Structural and functional properties of neural circuits underlie behavior, and perturbations in either can lead to abnormal, dysfunctional, or sometimes debilitating behavioral changes. The conserved synaptic adhesion molecules neurexins impact structure and function of circuits and are associated with autism spectrum and schizophrenia, disorders characterized by distinct behavioral phenotypes. Neurexins impact synaptic function and some behaviors, but significant gaps remain in our understanding of the molecular mechanisms that connect neurexins to behavior. We analyzed the *C. elegans* behavioral response to food deprivation in order to test the role of genes, including neurexin, in a conserved behavior with an experimentally defined neuronal circuit. Using the "WorMotel" platform and unbiased computer vision image analysis, we analyzed activity levels of hundreds of hermaphrodite *C. elegans* while fed or food deprived. While control worms significantly increase locomotor activity when food deprived (sustained >8 hours), worms lacking *nrx-1* (the singular neurexin in *C. elegans*) have a significantly reduced response to food deprivation (on food, there is no difference in locomotor activity levels between control and *nrx-1* worms). Loss of *nrx-1* also disrupted responsiveness to octopamine, a key neuromodulator in the food deprivation response circuit that increases locomotor activity. *nrx-1* has two major isoforms: the long α and short γ (lacking extracellular domains). Using isoform-specific mutation alleles and isoform-specific transgenic expression in neurons, we found initial response to food deprivation (hour 1) and octopamine require the short isoform, while sustained response to food deprivation (beyond hour 1) required only the long isoform, which was not required for response to octopamine. Loss of *nlg-1*, the singular neuroligin in *C. elegans* and the canonical postsynaptic binding partners of *nrx-1*, had

no impact on the food deprivation response. Loss of *nlg-1* in *nrx-1* mutants (*nlg-1*; *nrx-1* double mutants) eliminated the *nrx-1* phenotype in long-isoform dependent sustained responses to food deprivation, but not short isoform (which is unable to bind *nlg-1)* dependent initial responses or octopamine responsiveness. This demonstrates a novel role for *nrx-1* isoforms and a non-synergistic interaction with *nlg-1* in controlling temporal aspects of a single behavior at the molecular and potentially circuit level. Our work uncovers novel molecular complexity for the role of *nrx-1* in circuits and behavior, emphasizing the utility of using behavioral readouts for understanding gene function in neurons, circuits, and behaviors in parallel.

Disclosures: B.L. Bastien: None. **M.P. Hart:** None.

Poster

P048. Autism: Synaptic and Cellular Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P048.03

Topic: A.07. Developmental Disorders

Support: NIH/NIGMS 5P20GM103641-07
SC EPSCOR 18-SR04

Title: Counteracting epigenetic mechanisms in human neuronal morphogenesis and maturation

Authors: *F. RITCHIE[1], J. VACHARASIN[1], A. CULVER[1], S. LIZARRAGA[1], S. CHEON[1], A. BAGNELL[1], M. MCCORD[1], C. PAPENDORP[2], E. CHUKWURAH[1], A. SMITH[1], M. COWEN[1], P. GHATE[1], S. DAVIS[1], J. LIU[2];
[1]Univ. of South Carolina, Columbia, SC; [2]Brown Univ., Providence, RI

Abstract: Autism Spectrum Disorders (ASD) are highly heritable neurodevelopmental disorders characterized by social and verbal impairments as well as repetitive behaviors. Large human genetic studies showed that genes encoding chromatin and transcriptional regulatory proteins are among the most commonly mutated genes in ASD. Absent, Small, or Homeotic 1-like (ASH1L) is a histone methyltransferase that has previously been identified as a major genetic risk factor for ASD. ASH1L methylates Histone H3 on Lysine 36 (H3K36), which mainly coincides with transcriptional activation. ASH1L is proposed to counteract the activity of Polycomb Repressor Complex 2 (PRC2) by preventing the tri-methylation of Histone H3 on Lysine 27 (H3K27me3). We identify an overrepresentation of genes associated with neuronal projection development or morphogenesis that are highly correlated with ASH1L in the developing human cerebral cortex. Hence, this suggests that ASH1L could have a role in regulating neuronal morphogenesis. We find that knockdown of ASH1L in human stem cell-derived neurons alters different parameters of neuronal morphogenesis. Specifically, we report that ASH1L-depleted neurons have decreased neurite outgrowth, but have increased cell soma size and growth cone areas.

Additionally, we show that deficits in ASH1L lead to an increased percentage of DCX-positive neurons compared to control cultures. DCX is a marker of immature cortical neurons suggesting that ASH1L knockdown potentially leads to delayed maturation. We further find that ASH1L phenotypes are rescued by inhibition of the catalytic activity of PRC2. Taken together, these results indicate that the counteracting activities of ASH1L vs. PRC2, are important for the control of neuronal morphogenesis and maturation in human experimental systems.

Disclosures: F. Ritchie: None. **J. Vacharasin:** None. **A. Culver:** None. **S. Lizarraga:** None. **S. Cheon:** None. **A. Bagnell:** None. **M. McCord:** None. **C. Papendorp:** None. **E. Chukwurah:** None. **A. Smith:** None. **M. Cowen:** None. **P. Ghate:** None. **S. Davis:** None. **J. Liu:** None.

Poster

P048. Autism: Synaptic and Cellular Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P048.04

Topic: A.07. Developmental Disorders

Title: Transcriptomic profiling and bioinformatic analysis reveals modulation of mTOR-associated genes following glycine type 1 transport (GlyT1) inhibition in BALB/c mice

Authors: A. C. RUCKER[1], S. E. JONES[1], S. I. DEUTSCH[2], *J. A. BURKET[1];
[1]Mol. Biol. and Chem., Christopher Newport Univ., Newport News, VA; [2]Psychiatry and Behavioral Sci., Eastern Virginia Med. Sch., Norfolk, VA

Abstract: Mechanistic Target of Rapamycin (mTOR) has been implicated in several monogenetic forms of autism spectrum disorder (ASD). The NMDA receptor (NMDAR), a regulator of mTOR complex 1 (mTORC1), has emerged as a viable pharmacotherapeutic target in ASD. VU0410120 (VU120), a highly selective glycine (GlyT1) transporter inhibitor, improved social and cognitive behaviors in BALB/c mice, a model of impaired sociability. Transcriptomic and bioinformatic analyses of archived microarray data were conducted to determine strain differences and prosocial effects of VU120 in BALB/c and Swiss Webster (SW) mice. Frontocortical RNA was subjected to transcriptome profiling using the Affymetrix microarray. Briefly, global prosocial responses of individual mice were ranked and assigned to prosocial response categories. In vehicle- and VU120-treated groups of BALB/c mice, samples designated with the highest and lowest total ratings of social behaviors were used for microarray analysis; SW mice were chosen for comparison (n=4/group). Gene expression was studied using Affymetrix Transcriptome Analysis Console (TAC), Cytoscape, DAVID, and KEGG software. Principal component analysis identified clustering of vehicle-treated BALB/c mice irrespective of high or low sociability; BALB/c mice non-responsive to VU120 treatment clustered with vehicle-treated BALB/c mice. Importantly, a separate cluster for BALB/c mice responsive to treatment was observed independent of vehicle-treated groups and in closer proximity to the

vehicle-treated SW mice. Subsequent targeted analyses of 103 protein coding genes involved in regulation of mTOR signaling pathway were explored. Between strain comparisons of differentially expressed genes showed significant fold changes of mTOR-associated genes (PPKAA2, DEPTOR, APC, CAD, PPP2R2A) in BALB/c mice relative to SW (-1.20 < or > 1.20, $p \leq 0.05$; FDR $p \leq 0.05$). Comparisons were made between BALB/c mice that displayed increases in sociability following VU120 treatment (responders) to BALB/c mice that did not respond to treatment (non-responders). BALB/c responders showed significant differences in expression of regulators of mTOR (IRS1, PIK3CG, PIK3C2B, RAC1, PPP2R2A, RPS6KB2, EPM2A, DDIT4). Gene ontology analysis revealed significant fold enrichment for biological processes with upregulation in BALB/c non-responders compared to responders related to negative regulation of TOR signaling ($p = 0.013$), semaphorin signaling ($p = 0.006$), neuronal migration ($p = 0.004$) and neuronal development ($p = 0.021$). Overall, these data encourage exploration of a therapeutic role for NMDAR activation in regulation of the mTOR signaling pathway.

Disclosures: A.C. Rucker: None. **S.E. Jones:** None. **S.I. Deutsch:** None. **J.A. Burket:** None.

Poster

P048. Autism: Synaptic and Cellular Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P048.05

Topic: A.07. Developmental Disorders

Support: PACS1 Syndrome Research Foundation

Title: Neurons from PACS1 syndrome have altered expression of transcripts involved in axon outgrowth and impaired neural network activity

Authors: *L. RYLAARSDAM[1], A. D. GUEMEZ-GAMBOA[2];
[2]Physiol., [1]Northwestern Univ., Chicago, IL

Abstract: PACS1 syndrome is a neurodevelopmental disorder characterized by intellectual disability, autism spectrum disorder (ASD), epilepsy, distinct craniofacial abnormalities, and global developmental delays. PACS1 syndrome results from a recurrent *de novo* missense p.R203W variant in phosphofurin acidic cluster sorting protein 1 (PACS1). Almost nothing is known about how this variant in PACS1 affects the developing nervous system and very few therapeutic options are available for patients. In order to begin to address the impact of the R203W variant on different cell types in the developing brain, I performed single-cell RNA sequencing on cerebral organoids generated from patient and control-derived stem cells. I found a total of 199 unique differentially expressed genes (DEGs) between $PACS1^{+/R203W}$ and control organoids. DAVID Bioinformatics Resources analysis of DEGs and Weighted Gene Co-

expression Network Analysis (WGCNA) of the neuronal cells suggest that PACS1$^{+/R203W}$ neurons have increased expression of genes involved in axon outgrowth and guidance. These processes are critical for proper neural network formation in the developing brain and alterations could have serious consequences for the individual. To test if the electrical properties of neural networks are impaired in PACS1 syndrome, I cultured iPSC-derived neurons on multielectrode arrays. While PACS1$^{+/R203W}$ neurons had the same burst duration for individual electrode bursts, network burst duration was strikingly longer. PACS1$^{+/R203W}$ neurons also exhibited decreased synchrony with a lower percentage of spikes happening inside bursts. Future studies will be focused on determining the mechanism behind these electrophysiological impairments. This work is some of the first to look at the impact of the PACS1 syndrome variant on the developing brain and shows promising avenues of study for determining disease mechanisms in order to develop therapies.

Disclosures: L. Rylaarsdam: None. **A.D. Guemez-Gamboa:** None.

Poster

P048. Autism: Synaptic and Cellular Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P048.06

Topic: A.07. Developmental Disorders

Support: NIH Grant 1K22NS105922-01

Title: Elevated temperature alters the excitation to inhibition (E-I) balance to alleviate cognitive deficits in the Scn2a autism mouse model.

Authors: *Y. AWASTHI[1], Y. SHEN[2], *M. W. ANTOINE[2];
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Abstract: In most mammals (humans included) core body temperature is regulated to a setpoint value between 36.1 and 37.8°C, with an average fluctuation from the target of 0.5°C during a 24-hour day. Despite mechanistic safeguards, major temperature deviations (1-3 °C) from the setpoint occur in the body and in turn the brain. In children, these increases in brain temperature are generally well-tolerated. However, for unknown reasons, large fluctuations in brain temperature in some cases result in deleterious outcomes such as seizures, whereas for select autistic children beneficial effects including improved cognition can occur. Here, we describe a neural mechanism for circuit-level adaptive regulation of cortical activity during large fluctuations in brain temperature. Our study reveals temperature-induced alterations in the excitation to inhibition (E-I) balance that stabilize activity in the cortex. We show that in the Sodium Voltage-Gated Channel Alpha Subunit 2 (*Scn2a*) mouse model for autism with

intellectual disability, these temperature-induced circuit-level alterations can be beneficial by normalizing circuit function and improving higher-level cognition.

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Poster

P048. Autism: Synaptic and Cellular Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P048.07

Topic: A.07. Developmental Disorders

Support: R01DC016385
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Title: Characterizing human iPSC-derived neural progenitor cells in idiopathic autism

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Abstract: The vast majority (80-90%) of autism spectrum disorder (ASD) patients suffer from the idiopathic form of this neurodevelopmental disorder. Tractable models that can be directly correlated to patient phenotypes are essential for moving toward a precision medicine approach to treating ASD, a highly heterogeneous disorder. To address this, our research group has longitudinally phenotyped a cohort of developmentally delayed toddlers who go on to develop ASD as well as typically developing controls. These data include social communication and language assessments, neuroimaging data, eye tracking, and leukocyte transcriptomics. From a subset of these subjects, we generated induced pluripotent stem cell-derived neural progenitor cells (NPCs) and performed single cell RNA sequencing to better understand the processes associated with idiopathic ASD in early prenatal development.
Preliminary results showed a shift in neural progenitor subtype distribution between ASD and typicals. ASD NPCs showed a reduction of SB100+ and Neurod1+ NPCs, with an increase in VCAM1+, LGASL1+ and RBFOX3+ NPCs. ASD-specific transcriptional signatures were associated with IGF1 and retinoic acid signaling. Here we characterize unique properties of NPCs in ASD and their correlations with neuroimaging and behavior data in subjects from a deeply phenotyped cohort, including pilot data exploring sex differences in ASD.

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Poster

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Topic: A.07. Developmental Disorders

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Title: Loss of Tsc1 from striatal direct pathway neurons impairs endocannabinoid-LTD and enhances motor routine learning

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Abstract: Autism spectrum disorder (ASD) is characterized by social and communication deficits, as well as the presence of restricted and repetitive behaviors (RRBs). Over one hundred genes have been associated with ASD risk, including genes that cause syndromic disorders, which are associated with a constellation of neurological, psychiatric and medical conditions. One such disorder is Tuberous Sclerosis Complex (TSC), caused by mutations in either *TSC1* or *TSC2*, in which roughly half of patients meet the clinical criteria for an ASD diagnosis and most have epilepsy. While epilepsy in TSC likely arises from altered excitability in forebrain circuits, the brain regions and cell types important for ASD-related behaviors are less well understood. We hypothesized that alterations in striatal circuits, which mediate motor learning, action selection, and habit formation, might contribute to the RRBs observed in TSC patients with ASD. Striatal function depends on the coordinated activity of two subpopulations of GABAergic cells: direct pathway spiny projection neurons (dSPNs) and indirect pathway spiny projection neurons (iSPNs). dSPNs and iSPNs are distinct in their projection targets, and at the simplest level, bulk activation of dSPNs increases locomotor behavior and action selection while stimulation of iSPNs inhibits movement. To determine how developmental loss of Tsc1 affects striatal synapses and whether this is sufficient to alter striatal-dependent motor behaviors, we generated mice in which *Tsc1* was selectively deleted from either dSPNs or iSPNs. We find that motor routine learning in the accelerating rotarod task is enhanced in mice with *Tsc1* deletion restricted to dorsal striatal dSPNs, but not iSPNs, which also occurs in mice with global haploinsufficiency of *Tsc2*. Further, using whole-cell patch clamp electrophysiology we find that loss of Tsc1 causes a non-cell autonomous enhancement of cortical inputs onto dSPNs but not iSPNs, which is associated with loss of endocannabinoid-mediated synaptic long-term depression (eCB-LTD). Utilizing translating ribosome affinity purification (TRAP), we find

reduced ribosome engagement of multiple mRNAs encoding proteins required for eCB-LTD in Tsc1 KO dSPNs, which provides a potential molecular basis for the eCB-LTD deficit observed in these cells. Together, these findings demonstrate that increased cortical drive of dSPNs resulting from Tsc1 loss is sufficient to enhance the learning of a fixed motor routine, which may have implications for the emergence of restricted, repetitive patterns of behavior in TSC.

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Poster

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Title: Aberrant DJ-1 expression underlies L-type calcium channel hypoactivity in tuberous sclerosis complex and Alzheimer’s disease

Authors: *F. NIERE[1], L. P. CACHEAUX[1], A. UNERI[1], S. NAMJOSHI[1], C. REYNOLDSON[1], J. PENARANDA[1], C. J. MCARDLE[1], X. WANG[1], W. C. TAYLOR[1], S. CRAFT[2], T. MA[2], C. D. KEENE[3], K. F. RAAB-GRAHAM[1];
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Abstract: L-type voltage-dependent Ca2+ channels (L-VDCC) integrate synaptic signals to facilitate a plethora of cellular mechanisms. L-VDCC dysfunction is implicated in several neurological and psychiatric diseases. Despite their importance, signals upstream of L-VDCC activity that regulate their channel density, however, are poorly defined. In disease models with overactive mammalian target of rapamycin complex 1 (mTORC1) signaling (or mTORopathies), including tuberous sclerosis (TS) and Alzheimer’s disease (AD), we report a novel mechanism downstream of mTORC1 signaling that results in a deficit in dendritic L-VDCC activity. Deficits in L-VDCC activity are associated with increased expression of the mTORC1-regulated RNA-binding protein DJ-1. DJ-1 binds the mRNA coding the auxiliary Ca2+ channel subunit α2δ2

responsible for shuttling L-VDCC to the membrane and represses its expression. Moreover, this novel DJ-1/α2δ2/L-VDCC pathway is disrupted in human AD and preclinical models of AD and TS. Our discovery that DJ-1 directs L-VDCC activity and L-VDCC-associated protein α2δ2 at the synapse suggests that DJ-1/α2δ2/L-VDCC is a common, fundamental pathway disrupted in TS and AD that can be targeted in clinical mTORopathies.

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Poster

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Title: Procedural learning abnormalities in individuals with ASD and their biological parents

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Abstract: Sensorimotor impairments and deficits in procedural learning have been found in individuals with autism spectrum disorder (ASD) and their unaffected first-degree relatives (Mosconi et al., 2010). This suggests that procedural learning deficits may be familial in ASD, though no known studies have directly assessed the extent to which procedural learning impairments are associated between individuals with ASD (probands) and their parents (ASD parents) or may vary in relation to separate familial traits associated with ASD. Forty-four probands, 105 of their unaffected biological parents (ASD parents), 27 typically developing (TD) controls, and 47 TD parent controls matched on age, sex, and nonverbal IQ completed an oculomotor test of procedural learning. During testing, peripheral targets alternated between two fixed locations (+/- 6 degrees from center) at set intervals (1500 ms). Saccade latency, change in

latency over trials (rate of learning), and variability of latencies across trials were examined. Parents completed the Broad Autism Phenotype Questionnaire (BAP-Q) to assess subclinical traits associated with ASD. Parents were separated into BAP+ or BAP- groups using previously published cutoff scores (Sasson et al., 2013). No differences in saccade latencies, rate of learning, or variability of saccade latencies were seen for proband or ASD parents compared to control groups. Subgroup analyses indicated that probands whose parents did not show any BAP features (BAP-) showed increased variability of rightward saccade latencies relative to probands with at least one parent showing BAP features (BAP+) and TD controls (Group x direction interaction: F=3.92; p=.03). BAP+ parents showed faster leftward saccade latencies compared to BAP- parents and parent controls (β=-.02; p=.01). Rate of learning was inter-correlated among probands and their parents (h^2= 0.735, p=0.003). Together this suggests that procedural learning abnormalities in ASD may represent an important intermediate phenotype associated with familial risk. Our finding that probands with BAP- parents show increased rightward saccade latency variability during procedural learning is consistent with prior data implicating atypical functioning of left hemisphere striatal chronometric systems (D'Cruz et al., 2009), and further suggests these alterations represent familial trait liabilities independent of subclinical core traits. Findings that rate of learning was correlated among proband subgroups and their parents suggest that procedural learning impairments in ASD are familial and may serve as useful intermediate phenotypes for tracking familial mechanisms of ASD.

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Poster

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Topic: A.07. Developmental Disorders

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Title: Structure of corticostriatal tracts in middle-aged and older adults with autism spectrum disorder

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Abstract: Individuals with autism spectrum disorder (ASD) show atypical white matter (WM) structure through childhood and young adulthood, but little is known about WM differences in middle-aged and older populations with ASD. In typical aging, fractional anisotropy (FA) decreases while mean diffusivity (MD) increases. WM changes in the striatum, a critical relay center, have been implicated in the cognitive decline observed during typical aging. Behavioral activation, inhibition, attention, and reward sensitivity depend on the striatum, and younger persons with ASD show altered FA and MD. Therefore, understanding differences in corticostriatal WM among middle-aged and older ASD/typical control (TC) populations is important. We hypothesized that 1) aging adults with ASD would show decreased FA and increased MD compared to TC and 2) greater age would be associated with decreased FA and increased MD for both groups. Participants aged 40-70 years were recruited with and without ASD. Diagnosis was confirmed by an expert clinician and TC participants had no family/personal history of ASD. Diffusion weighted MRI data were collected on a 3T scanner with one of two protocols (b = 1500 or 1000 s/mm^2) and processed using FSL and TractSeg. Diffusivity measures were extracted for 14 striatal (ST) tracts (7 left [L], 7 right [R]) and harmonized (for protocol) using ComBat. Groups were matched on gender, age, non-verbal IQ, handedness, brain volume, and head motion. Regressions were used to test the effects of age and diagnostic group on ST-tracts while controlling for motion and protocol. 58 participants with and without ASD (ASD n = 27, TC, n = 31) were included in analyses. Significant group differences in MD (ASD>TC) were observed after FDR correction in the R ST-postcentral, L and R ST-parietal, and L and R ST-occipital tracts. Greater age was significantly associated with greater MD in the R ST-postcentral, and the L and R ST-occipital tracts. No significant differences were observed for FA. Diagnosis and age effects were observed for MD across several ST-tracts that connect to cortical regions associated with attention, somatosensory, and visual networks; all of which send critical inputs for cognitive function to the striatum and show differences in aging and ASD. Results are consistent with a recent study indicating that MD is more sensitive to aging than FA. Although much attention has been paid to frontal-cortex and ST-frontal WM in typical aging and younger persons with ASD, our results highlight posterior ST-tract differences in middle-aged and older ASD. Future work will explore the relationship between ST-tracts, associated cortical regions, and neurocognitive measures.

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Poster

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Title: Neural responses to audiovisual speech in infants at increased likelihood for autism and links with language: An ERP pilot study

Authors: *K. DUNHAM[1,2], B. KECELI-KAYSILI[3], A. J. GOLDEN[3], P. SANTAPURAM[3], J. MARKFELD[2], J. I. FELDMAN[2,4], T. WOYNAROSKI[4,5,6,1];
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Abstract: Speech is inherently a multisensory process, wherein synchronized visual cues from the moving mouth complement the dynamic acoustic stream. Past research has shown that typically developing (TD) infants tune in to multisensory speech, looking to the mouth of their communication partners during the first year of life, presumably to access audiovisual cues that facilitate speech processing and language learning. This study aims to determine: (a) if visual cues increase efficiency of speech processing as indexed by event-related potentials (ERPs) in infants at general population-level likelihood for autism (infant siblings of TD children; Sibs-TD), (b) if infants at increased familial likelihood for autism (infant siblings of children with autism; Sibs-AUT) display a lesser boost in speech processing efficiency with access to multisensory versus unisensory cues compared to Sibs-TD, and (c) if individual differences in speech processing efficiency covary with language ability. Forty 12-18 month old infants (20 Sibs-AUT; 20 Sibs-TD matched on sex and chronological age) are being recruited. Participants view videos of a female speaking syllables in audiovisual (auditory speech + synchronous visual mouth movements) and auditory only (auditory speech + still image of the face) conditions. Data are collected using NetStation and a 128-channel Geodesic sensor net (Net Amps 400 amplifier, Hydrocel GSN 128 EEG cap, EGI Systems Inc.). The raw EEG signal is sampled at 1000 Hz and referenced to vertex (Cz). The amplitude of the P2 component (defined a priori as occurring between 160 ms and 340 ms) as measured at Cz is extracted from the average ERP of each participant. Parents also complete the MacArthur-Bates Communicative Development Inventories to report on their child's emerging vocabulary. Preliminary results for data processed to date (5 Sibs-TD; 5 Sibs-AUT) suggest that, on average, Sibs-TD display a *decreased* P2 amplitude in response to audiovisual versus auditory only speech, while Sibs-AUT display reduced differentiation of audiovisual versus auditory only speech as indexed by the P2 amplitude compared to Sibs-TD. Preliminary results also indicate moderate associations between the degree of P2 amplitude suppression and receptive and expressive language across groups. These findings suggest that differential processing of audiovisual speech may emerge early in life and that infants at increased likelihood for autism appear to show reduced differentiation of multisensory versus unisensory speech that may be clinically useful for explaining individual differences in language ability, as well as broader language delays or disorders.

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Poster

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Program #/Poster #: P051.04

Topic: A.07. Developmental Disorders

Support: Dr. Stanley Ho Medical Development Foundation

Title: Disruptions of functional connectivity patterns in Chinese children with ASD - an EEG coherence study

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Abstract: Autism spectrum disorder (ASD) pathophysiology involves disruptions in the brain's oscillatory rhythms that can be captured by electroencephalogram (EEG). Recent EEG studies have shown that coherence patterns of EEG signal can be used to classify subjects into ASD and non-ASD groups. This study aimed to validate whether functional connectivity at rest measured by EEG phase coherence can distinguish children with ASD from typical developing controls in Chinese population. A group of 44 children with ASD and 30 age and gender controlled children (age range: 5 - 12 years) were recruited and recorded in the study. The ASD diagnosis was obtained from a qualified clinician and ASD subjects also met Autism Diagnostic Observation Schedule (ADOS-2) classification criteria. EEG was recorded continuously with a 64-channel Neuroscan system (Compumedics, Australia) at 1000 Hz sampling rate while the subject was at rest with eyes opened. Two bipolar channels registered horizontal and vertical eye-movements. The EEG pre-processing steps included re-referencing to the average mastoid, band-pass filtering in 1-90 Hz range, regressing out the eye-movement artifact, Laplacian transform to minimize the volume conduction effects, epoching into 3-second intervals with 50% overlap, rejection of the high-amplitude epochs and pooling the electrodes into 10 clusters. Phase coherence between the clusters was calculated from the complex FFT spectra of individual epochs in four frequency bands: delta (0.5-4 Hz), theta (4-8 Hz), alpha (8-12 Hz), beta (12-30 Hz). Coherence was compared between the control and ASD groups with a mass-univariate t-test with Bonferroni correction for the multiple comparisons across the cluster pairs and frequency bands. The phase

coherence was significantly reduced in ASD patients in comparison to the controls in 21, 15, 22 and 3 cluster pairs in delta, theta, alpha and beta bands correspondingly. The most affected connections were posterior frontal-occipital pathway for the delta band, right parietal-occipital connection for the theta band, anterior frontal-left parietal for the alpha band and left temporal-left parietal for the beta band. In sum, ASD is associated with reduced connectivity measured by EEG in four frequency bands. The alpha and delta band connectivity was affected stronger than theta and beta bands. The most affected areas were located anterior in the alpha and delta bands and posterior in the theta and beta bands. Our findings extend previously reported disruptions in the resting-state EEG connectivity in children with ASD to Chinese population.

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Poster

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Erasmus MC Fellowship 106958

Title: Personalized Cerebellar Connectomic Signatures in Autism Spectrum Disorder

Authors: *F. D'OLEIRE UQUILLAS[1], E. BUEICHEKÚ[2], J. SEIDLITZ[3], K. STEELE[1], B. LI[1], R. GESUE[1], M. LATIF[1], T. FASULO[1], M. TROTTER[1], J. YAO[1], J. VERPEUT[4], H.-J. BOELE[1], J. SEPULCRE[2], S. S.-H. WANG[1], J. GOMEZ[1];
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Abstract: Cerebellar injury at birth is the largest non-genetic risk factor for Autism Spectrum Disorder (ASD). Although brain regions develop in synchrony over childhood, the detailed connectomic intersections between cerebellar-subcortical-neocortical circuits and ASD remain poorly understood. Using within-subject measures of regional morphological covariation, we

estimated ASD-related alterations along the cerebellar-thalamic-neocortical pathway across childhood.
Methods: T1w MRI data from 69 children from the Autism Brain Imaging Data Exchange (94% males, 40% controls, median age 8±2 years, mean ± SD; range 5 to 17 years old) were used to compute individualized structural covariance networks. Local/intra-region (cerebellar-cerebellar, thalamic-thalamic connections) and distributed/inter-region (cerebellar-thalamic, thalamic-neocortical connections) graph-metrics of topological structural integrity (nodal degree, participation coefficient, edge-betweenness, global efficiency, small-worldness) were used as variables in general linear models to test for age x group interactions, corrected for multiple comparisons (false discovery rate $q<0.05$).
Results: At the local level, we found age by group effects in the nodal degree of cerebellar dentate nuclei ($p=0.015$), and thalamic medial ventral reuniens (mvR) nuclei ($p=0.02$). At the distributed level, we found effects in the participation coefficient of cerebellar dentate nuclei ($p=0.014$), medial geniculate (MGN) ($p=0.001$) and mvR ($p=0.001$) thalamic nuclei. Alterations in edge-betweenness were found from thalamic mediodorsal- lateral nuclei to inferior prefrontal gyri ($p=0.018$), ventral anterior nuclei to orbitofrontal cortex ($p=0.010$), central medial nuclei to parieto-occipital cortex ($p<0.0001$), and ventral anterior magnocellular nuclei to lateral occipital cortex ($p=0.043$).
Conclusions: The coordinated structural development of the cerebellar-thalamic-neocortical pathway is atypical in ASD children. A compromised nexus between cerebellum and neocortex is characterized by topological alterations in cerebellar dentate nuclei, MGN and mvR thalamic nuclei at both local and distributed levels. This investigation builds groundwork for studies that can evaluate the cerebellum as an important bellwether and site for intervention in ASD.

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Poster

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Title: Effects of visual and proprioceptive inputs on postural stability in individuals with autism spectrum disorder (ASD)

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Abstract: Sensorimotor impairments are prevalent in autism spectrum disorder (ASD), though the degrees to which disruptions of individual sensory modalities affect motor control and execution in patients are not well understood. Previous studies have suggested that individuals with ASD show increased postural sway when visual information is occluded or proprioceptive illusion is introduced. The current study aimed to evaluate the interactive effect of both visual and proprioceptive inputs on postural stability and complexity in individuals with ASD. Forty-one individuals with ASD (ages 10-35 years) and 18 typically developing (TD) controls completed static stances on a force platform. Lightweight tendon vibrators (TVs) were attached to each of the Achilles tendons of participants' ankle joints. They vibrated at a frequency of 80 Hz when turned on, creating a transient proprioceptive illusion of lengthening of tendons and leaning forward. Participants completed four testing conditions: TVs turned on or off (TVon vs. TVoff) and with or without lights (LTon vs. LToff) in the room. Postural sway variability was quantified using the center of pressure (COP) trajectory length and COP standard deviation in the anterior-posterior (COP_{AP}) and mediolateral (COP_{ML}) directions. Postural sway complexity was quantified using the mutual information of COP_{AP} and COP_{ML} and detrended fluctuation analysis (DFA) of each COP time series. Individuals with ASD showed increased COP length and COP standard deviation in both directions relative to TD controls. Female individuals with ASD showed COP length and COP_{ML} standard deviation reductions relative to male patients, but this sex difference was not identified in TDs. COP length and COP_{AP} standard deviation and DFA in both directions increased significantly in TDs during TVon conditions. DFA in the ML direction also increased in TDs during LTon trials relative to LToff stances. Neither sensory manipulation (TVoff, LToff) showed an impact on COP or DFA measures in individuals with ASD. Our results suggest that while postural control in TDs is affected by degradation of proprioceptive or visual feedback, alterations of these sensory inputs did not disrupt postural stability in ASD indicating these sensory feedback mechanisms show minimal contributions to postural control in patients. Our study also is the first to identify sex differences during gross motor tasks involving multiple sensory interferences in ASD suggesting studies investigating ASD-specific sex differences in sensorimotor functioning are needed to clarify key neurodevelopmental processes that may vary across male and female patients.

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Poster

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Topic: A.07. Developmental Disorders

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Title: Abnormal alpha power spectra, modulation, and connectivity in low-functioning individuals with autism with intact exogenous attention

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Abstract: Some studies have shown that individuals with autism have attenuated resting-state EEG alpha (8 - 13 Hz) power and less modulation due to sensory stimulation. However, such findings were inconsistent across studies and limited to high-functioning autism (HFA), which may not represent the entire autism population. In this study, I recorded 256-channel EEG of adults with low-functioning autism (LFA; n=6) performing an exogenous cueing task after training, along with adults with HFA (n=6) and those typically developed (TD; n=6). In the experiment, participants used a computer mouse to identify a target that spatially coincides within a rectangular cue (valid cue; 80% of trials) or not (invalid cue; 20%). I adapted independent component and wavelet analyses to examine the time course of the alpha power change along the sequence of stimulus windows: blank baseline (500ms), fixation (500ms), cue (600ms), and target (500ms). The response time (RT) and visually evoked potential (VEP) were also examined. The results showed that all participants responded faster for the valid cue than the invalid cue condition, although the overall RTs were slower in the LFA than the HFA and TD groups. The permutation test of VEPs of the two cue conditions also showed a significant difference in all groups, demonstrating intact exogenous attention in these measures. However, participant groups showed quite different patterns of alpha oscillation over Oz and Fz. In TD, the alpha power peaked in the blank baseline and gradually decreased along the course of the stimulus presentation. The HFA group showed a similar pattern of alpha power change as TD but had a reduced alpha power peak in the baseline. The LFA group showed much more attenuated alpha power than the HFA group and showed no alpha change across stimulus windows. The topographic map of all channels also confirmed these findings. The functional connectivity analysis revealed a strong frontal-occipital alpha (10 Hz) coherence in the TD and HFA groups but no such connectivity in the LFA group. Instead, the LFA group had robust local connectivity in the frontal and posterior areas. The theta (5 Hz) band activity also showed a similar pattern of connectivity. While exploratory, the results replicated the primary finding of our previous study

that alpha oscillation is critically impaired in LFA, demonstrating the importance of the inclusion of individuals with LFA in studies. Importantly, if the alpha oscillation reflects a control mechanism of cortical activities, as some have claimed, these results indicate that the cortical alpha activity could serve as an electrophysiological marker for severe cases of autism.

Disclosures: K. Sung: None.

Poster

P051. Autism: Physiology and Systems II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P051.08

Topic: A.07. Developmental Disorders

Support: NICHD Autism Center of Excellence P50 HD093079
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Robert Shoes Fund
Scott & Jennifer Fearon

Title: Distinct Auditory ERPs in Autism with and without Disproportionate Megalencephaly

Authors: *P. DWYER[1], S. VUKUSIC[2], A. SILLAS[2], C. D. SARON[2], S. M. RIVERA[1];
[1]Psychology, [2]Ctr. for Mind and Brain, UC Davis, Davis, CA

Abstract: Introduction
Prior research suggests that autism spectrum development (ASD) with disproportionate megalencephaly (ASD-DM) is a meaningful neurophenotype differing behaviourally from ASD with normal brain size (ASD-N), with implications for development and cognitive abilities. This study explored the functional correlates of DM in early development by examining auditory ERPs in ASD-DM, ASD-N, and typical development (TD) with normal brain size (TD-N).
Methods
Included participants were 47 TD-N (40 male, M_{age} = 36.72mos, M_{DQ} = 104.71), 78 ASD-N (67 male, M_{age} = 38.35mos, M_{DQ} = 56.60), and 14 ASD-DM (12 male, M_{age} = 37.64mos, M_{DQ} = 56.83). Groups were selected not to differ in age and (ASD only) cognitive ability. DM was defined by total cerebral volume (structural MRI) to height ratios 1.5 SDs above TD mean. While watching a quiet video, participants heard, via headphones, brief complex 50-80 dB tones (~250 trials/intensity; ISI 1-2s). 61-channel EEG was sampled at 1 kHz, average-referenced, and processed using SOBI ICA. P1 values were mean amplitudes over fronto-central channels ±50 ms around the grand-average peak/intensity: 73-173 ms (50 dB), 59-159 ms (60 dB), 46-146 ms

(70 dB), and 45-145 ms (80 dB). N2 values were quantified as mean voltages over fronto-central channels between 201-350 ms. Amplitudes were entered into mixed ANOVA (group x hemisphere x intensity).

Results

There was a main effect of group on P1 amplitudes, $F = 3.10$, $p < .05$, driven by attenuated responses in ASD-DM relative to TD-N, $F = 9.83$, $p = .003$, as well as trending smaller responses in ASD-DM than ASD-N, $F = 3.46$, $p = .07$. ASD-N and TD-N did not differ, $F = 0.79$, $p = .37$. There were no P1 main effects or interactions involving hemisphere or intensity. There was also a trend towards an effect of group on N2 amplitudes, $F = 2.66$, $p = .07$. This was driven by attenuation of the N2 in ASD-DM, $F = 3.71$, $p = .06$, and ASD-N, $F = 3.74$, $p = .06$, relative to TD-N. N2 amplitudes in ASD-DM and ASD-N did not differ, $F = 0.60$, $p = .44$. An omnibus main effect of intensity, $F = 9.77$, $p < .0001$, was driven by larger N2 responses to 70 dB than 50 dB tones and to 80 dB than 50 or 60 dB tones.

Discussion

Across intensities, the P1 response appears selectively attenuated in ASD-DM, while ASD-N and TD-N show similar ERPs in the P1 time window. In contrast, in the later N2 window, ASD-DM and ASD-N both exhibit similar ERPs that trend towards attenuation compared to TD-N. Overall, these results suggest DM status in ASD is associated with appearance of atypicality in cortical auditory ERPs more quickly after stimulus onset. However, the neurophysiological basis of this observation is as yet unclear.

Disclosures: P. Dwyer: None. **S. Vukusic:** None. **A. Sillas:** None. **C.D. Saron:** None. **S.M. Rivera:** None.

Poster

P051. Autism: Physiology and Systems II

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Program #/Poster #: P051.09

Topic: A.07. Developmental Disorders

Support: 1R21AG065621-01A1
1R01NS121120-01

Title: Atypical cortical and subcortical brain activation associated with precision visuomotor control in autistic adults

Authors: *Z. WANG[1], S. COOMBES[1], D. VAILLANCOURT[1], D. SHIRLEY[1], G. VALCANTE[1], A.-M. ORLANDO[1], R. ROMERO[1], B. KARMAKAR[1], A. WAGLE SHUKLA[1], M. MOSCONI[2];
[1]Univ. of Florida, Gainesville, FL; [2]Univ. of Kansas, Lawrence, KS

Abstract: Although conceptualized as a childhood disability, autism spectrum disorder (ASD) has increasingly been recognized as a lifelong neurological condition that could significantly impact adult functioning and quality of life. A recent clinical study has identified a more than 25-fold increased risk of Parkinsonism from two independent cohorts of autistic adults, suggesting that neurological conditions may evolve and even become more prominent as affected individuals age. However, studies focused on neuromotor control in autistic adults are scant, and we do not yet know to what degrees autistic adults might be at risk of developing neurological symptoms and neurophysiological systems show abnormal levels of degeneration. Sixteen autistic adults ages 30-73 years and 19 age-, sex-, and IQ-matched non-autistic controls completed a visually guided precision grip task during fMRI. Participants pressed an MRI-compatible load cell with their thumb and forefinger while viewing two horizontal bars from a visual display inside the scanner. Participants pressed to move the lower force bar upwards to the height of the static target bar and maintained a constant grip force for 2 sec followed by a one sec rest. A total of 4 blocks (10 trials/ block) were administered and alternated with 10 sec rest. The target bar was set at 15% of individuals' maximum voluntary contraction (MVC). Grip force variability and peak force were examined. Whole brain, voxelwise analyses were conducted on the fMRI imaging data. Our results showed no group differences in MVC. Autistic adults showed both mean and peak force reductions relative to non-autistic controls. Increased BOLD signal was observed in well-established regions of the visuomotor network including motor and premotor cortex, inferior parietal lobule, extrastriate visual cortex, thalamus, putamen, and cerebellum. Autistic adults showed greater BOLD activation than controls in the left precentral (M1) and postcentral (S1) gyrus, bilateral inferior frontal gyrus (IFG), and right cerebellum lobule VIII (CBL.VIII). Increased age was associated with increased task-related activation in bilateral IFG and right CBL.VIII in ASD. In contrast, increased age was associated with activation reductions of the left M1, RIFG, and right CBL.VIII in non-autistic controls. In autistic adults, decreased activation of RIFG was associated with more severe clinical ratings of repetitive behaviors. Overall, these results provide new evidence that neuromotor alterations in autistic adults involve aberrant cortical and subcortical organization that may contribute to critical aging-related clinical issues in ASD.

Disclosures: Z. Wang: None. **S. Coombes:** None. **D. Vaillancourt:** None. **D. Shirley:** None. **G. Valcante:** None. **A. Orlando:** None. **R. Romero:** None. **B. Karmakar:** None. **A. Wagle Shukla:** None. **M. Mosconi:** None.

Poster

P051. Autism: Physiology and Systems II

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Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P051.10

Topic: A.07. Developmental Disorders

Title: The transgenerational inheritance of circadian disturbances in a valproic-acid (VPA) model of Autism Spectrum Disorder

Authors: *S. FERRARO, N. DE ZAVALIA, N. QUTEISHAT, S. AMIR;
Concordia Univ., Montreal, QC, Canada

Abstract: Autism Spectrum Disorder (ASD) is a heterogenous group of disorders with similar and overlapping symptomology but differential clinical progression and severity. Disturbances within the circadian system are a prevalent pathological accompaniment of ASD but remain unaddressed in ASD individuals, presenting significant lifelong challenges. Over the past two decades the incidence of ASD diagnoses has continued to rise, leading to the suggestion that epigenetic mechanisms may underlie both the emergence of maladaptive behaviours seen in the disorder and the increased prevalence of ASD within the population. Previous research has documented that both the ASD-like behaviours and the neurobiological alterations resulting from *in utero* exposure to valproic acid (VPA), persist into the F3 generation. In this study we explored whether comorbid circadian disturbances previously described in the VPA-exposed F1 generation were also present in the F2 and F3 generations. We produced a patrilineal breeding scheme wherein 5-month-old Wistar males exposed to either saline (n = 6) or VPA (n = 6) *in utero* were mated with naïve females (n = 12) to produce the F2 generation. Males from the F2 generation were similarly mated to produce the F3 line and all animals underwent circadian locomotor analysis. We demonstrate that the emergence of disorganized circadian behaviour arising from altered photic-entrainment capacity is present in F2 and F3 VPA-exposed lineages. Moreover, we show a sexually dimorphic persistence of these behaviours wherein males, but not females, demonstrate a longer and more stable presentation of circadian rhythm disruption across all three generations. These findings demonstrate that deleterious comorbid disturbances in the circadian timing-system can also be transmitted through generations and emphasize the urgent need for novel therapies addressing ASD comorbidities.

Disclosures: S. Ferraro: None. **N. De Zavalia:** None. **N. Quteishat:** None. **S. Amir:** None.

Poster

P095. Potassium Channels: Mechanisms of Action

Location: Virtual Only

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Program #/Poster #: P095.01

Topic: B.04. Ion Channels

Support: NIH Grant NIHR21EY031813-01A1

Title: Peptide Lv augments intermediate conductance calcium-dependent potassium (IK_{Ca}) channels

Authors: *D. L. PHAM[1], G. Y.-P. KO[2];
[1]Vet. Integrative Biosci., [2]Vet Integrative Biosci., Texas A&M Univ., College Station, TX

Abstract: Peptide Lv is an endogenous secretory peptide (~40 amino acids) expressed in various organs and tissues including the neural retina, brain, and vascular endothelium. Peptide Lv is named for its ability to increase the mRNA and protein expression of L-type voltage-gated calcium channels (LTCCs) in cultured neurons and cardiomyocytes. Peptide Lv is upregulated in the neural retina and retinal microvascular endothelium in diabetic animals and patients with proliferative diabetic retinopathy. Although it is able to promote endothelial proliferation and pathological angiogenesis, the detailed mechanism of its angiogenic action is unclear. Endothelial potassium (K) channels are important for endothelial hyperpolarization and function since inhibition of endothelial K channels dampens the endothelial cell (EC) proliferation, an essential property for angiogenesis. The hypothesis is that the angiogenic action of peptide Lv is through endothelial K channels. We found the treatment with peptide Lv in cultured ECs increased the mRNA and protein levels of intermediate conductance calcium-dependent potassium (IK_{Ca}) channels. Interestingly, peptide Lv had no effect on other endothelial K channels such as ATP-sensitive potassium (K_{ATP}) and small conductance K_{Ca} channels. Whole-cell patch-clamp recordings on cultured ECs were performed to further examine the effects of peptide Lv on endothelial hyperpolarization and endothelial IK_{Ca}. Cultured ECs treated with peptide Lv was significantly hyperpolarized, and peptide Lv significantly enhanced IK_{Ca} current density. Blocking IK_{Ca} dampened peptide Lv-elicited EC proliferation. Thus, the angiogenic action of peptide Lv is in part through the augmentation of endothelial IK_{Ca}. This work is funded by NIHR21EY031813-01A1 to GK.

Disclosures: D.L. Pham: None. **G.Y. Ko:** None.

Poster

P095. Potassium Channels: Mechanisms of Action

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P095.02

Topic: B.04. Ion Channels

Support: NIH Grant NS116327

Title: Sumoylation of the kv4.2 ternary complex increases current amplitude and surface expression by blocking internalization

Authors: *M. WELCH[1], L.-A. R. JANSEN[2], D. J. BARO[3];
[1]Georgia State Univ., Hampton, GA; [2]Georgia State Univ., Atlanta, GA; [3]Dept Biol, Georgia State Univ. Dept. of Biol., Atlanta, GA

Abstract: A family of Kv4 -subunits mediate a transient A-type potassium current, IA. Kv4 channels exist as ternary complexes with potassium channel interacting protein (KChIP1-4) and dipeptidyl peptidase like protein (DPP6/10). Multiple ancillary proteins associate with Kv4 throughout its lifetime to regulate function and expression. How interactions within the Kv4 macromolecular complex are modulated is poorly understood. Small ubiquitin-like modifier (SUMO) is an 11kD peptide post-translationally added to lysine (K) residues to control protein-protein interactions. To test if Kv4 SUMOylation modulates ternary complex interactions, human embryonic kidney (HEK) cells were co-transfected with 3 plasmids encoding a Kv4.2 GFP fusion protein (Kv4.2g), KChIP2a and DPP10. SUMOylation was or was not increased by co-expressing plasmids encoding SUMO and the SUMO conjugating enzyme, Ubc9. Whole cell patch clamp recordings showed increasing SUMOylation in HEK cells expressing Kv4.2g ternary complexes produced a significant 35% increase in IA maximal conductance (Gmax) (t test $p<0.05$), and cell surface biotinylation experiments revealed a significant 30% increase in Kv4.2g surface expression (t test $p<0.05$). A biotin internalization assay showed increasing Kv4.2g ternary complex SUMOylation significantly reduced channel internalization by 65% (t test $p<0.05$). Experiments were repeated using a mutant Kv4.2g construct where a previously identified SUMOylation site, K579, was mutated to arginine (R). Overexpressing SUMO and Ubc9 with Kv4.2g K579R +KChIP2a +DPP10 no longer produced the significant increase in IA Gmax (t test $p=0.98$) or surface expression (t test $p=0.53$), or the significant decrease in internalization (t test $p=0.34$). Ternary complex internalization depends upon the levels of endocytosis and subsequent recycling. Blocking endocytosis with Pitstop2 significantly increased IA Gmax by 57% (One Way ANOVA, Tukey's post hoc $F(3,38)=7.7$, $p<0.05$). Including SUMO peptide in the patch pipette significantly increased IA Gmax by 56% (Tukey's $p<0.05$). No further significant increase was observed when SUMO was co-applied with Pitstop2 (Tukey's $p=0.83$). Pitstop2 mimicked and occluded the effect of SUMO. Surprisingly, the association of Kv4.2g with the major adaptor for clathrin-dependent endocytosis, AP2, was not affected by overexpressing SUMO and Ubc9 (t test $p=0.66$). These data suggest that the effect of SUMOylation is downstream of endocytosis. Future experiments will test if SUMO affects Kv4.2g recycling. In sum, we show SUMOylation of Kv4.2g K579 increases IA Gmax mediated by the ternary complex by reducing internalization.

Disclosures: M. Welch: None. **L.R. Jansen:** None. **D.J. Baro:** None.

Poster

P095. Potassium Channels: Mechanisms of Action

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Program #/Poster #: P095.03

Topic: B.04. Ion Channels

Support: F32NS108519

R21NS101648
R01NS114210
R01HL144071

Title: SPHKAP anchors type-I PKA at neuronal somatic ER-PM junctions formed by Kv2.1 channels

Authors: *N. C. VIERRA[1], D. VAN DER LIST[1], M. KIRMIZ[2], J. S. TRIMMER[1];
[1]Physiol. & Membrane Biol., Univ. of California, Davis, Davis, CA; [2]Mol. and Cell. Physiol., Stanford Univ. Sch. of Med., Stanford, CA

Abstract: The voltage-gated K^+ (Kv) channel Kv2.1 is present in dense clusters on the plasma membrane (PM) of the somata, proximal dendrites, and axon initial segments of most brain neurons. In addition to its significant role in controlling electrical excitability, Kv2.1 also serves a non-canonical structural function in tethering the PM to the endoplasmic reticulum (ER) to form ER-PM junctions. To identify specific neuronal functions linked to Kv2.1-associated ER-PM junctions, we have started to define their molecular composition by immunopurifying (IPing) Kv2.1 from mouse brain and analyzing the IPed complexes by protein mass spectrometry. This approach allowed us to show that Kv2.1-associated ER-PM junctions enable compartmentalized Ca^{2+} signaling in the neuronal soma by promoting the clustering of the voltage-gated L-type Ca^{2+} channel (LTCC) Cav1.2 at ER-PM junctions (Vierra et al., eLife e49953, 2019). We have now identified cAMP-dependent protein kinase A (PKA)-related signaling proteins that also associate with Kv2.1 at ER-PM junctions. PKA regulates critical neuronal functions, including transcription, synaptic plasticity, and electrical activity. Spatiotemporal specificity of PKA substrate phosphorylation is determined by its binding to A-kinase anchoring proteins (AKAPs) that target PKA to spatially restricted signaling microdomains. Prominent in our Kv2.1 IP experiments was an AKAP called SPHKAP. Immunolabeling of brain sections from mice and rats of both sexes and cultured rat hippocampal neurons revealed co-clustering of SPHKAP with Kv2.1 and PKA-RIα, suggesting a unique role for PKA-RI at somatic ER-PM junctions. We also determined that SPHKAP clustering was sensitive to Kv2 channel-mediated ER-PM junction formation, as mice lacking Kv2 channels showed reduced somatic SPHKAP cluster size. In neurons, activation of certain PKA-associated signaling pathways is initiated by Ca^{2+} and PKA crosstalk enabled by AKAPs that place PKA near LTCCs. To test whether SPHKAP enables Ca^{2+}/PKA crosstalk at somatic ER-PM junctions, we performed shRNA-mediated knockdown of SPHKAP in cultured rat hippocampal neurons. We found that SPHKAP knockdown significantly reduced the amount of PKA molecules in close proximity to somatic Cav1.2 channels, impaired depolarization-evoked Ca^{2+} influx, and decreased LTCC-dependent activation of CREB and c-Fos, two transcription factors involved in learning and memory. Together, these data reveal that SPHKAP is targeted to somatic ER-PM junctions formed by Kv2.1 channels and promotes PKA-dependent regulation of somatic Ca^{2+} signals that are coupled to depolarization-dependent gene expression.

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Poster

P095. Potassium Channels: Mechanisms of Action

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Program #/Poster #: P095.04

Topic: B.04. Ion Channels

Support: NIH Grant F31NS110192
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Title: Modulating Hippocampal Glutamate Release with Kv2-Mediated Endoplasmic Reticulum/Plasma Membrane Junctions

Authors: *L. C. PANZERA[1], I. CHO[1], A. N. LEEK[2], M. M. TAMKUN[2], M. B. HOPPA[1]; [1]Dartmouth Col., Hanover, NH; [2]Colorado State Univ., Fort Collins, CO

Abstract: Endoplasmic reticulum/plasma membrane (ER/PM) junctions act as subcellular hubs important for mediating calcium homeostasis, protein localization and lipid exchange. Although a number of ER proteins can interact with lipids on the PM, specific proteinaceous PM anchors that form stable ER junctions have remained elusive. The voltage-gated potassium channel Kv2.1 is the first PM protein found to engineer ER/PM junctions through interaction and recruitment of the transmembrane ER protein VAMP-associated protein (VAP). While Kv2.1 has a canonical conducting role in shaping electrical activity in the somatodendritic compartment, a non-conducting population of Kv2.1 channels form ER/PM junctions occupying ~10% of the neuronal soma. These sites are involved in trafficking, vesicle fusion, and the delivery of membrane proteins to the PM in addition to being sites of neuron-glia contact. Here, we present data in cultured hippocampal neurons that loss of Kv2.1 decreases the ability of the somatic ER to fill with calcium during electrical stimulation. More importantly, and in contrast to Kv2.1 dogma, we go on to demonstrate that Kv2.1 is enriched in synaptic terminals where it exclusively engages in a non-conducting role, significantly altering both ER calcium handling and synaptic function. Using noninvasive optical indicators, we reveal that genetic knockdown of Kv2.1 impairs stimulation-evoked calcium uptake into the ER and decreases synaptic vesicle exocytosis. Further, localization of Kv2.1 as well as VAP recruitment to terminals were found to be important for maintaining the unique role of these ER/PM junctions with regards to several aspects of synaptic physiology.

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Poster

P095. Potassium Channels: Mechanisms of Action

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Program #/Poster #: P095.05

Topic: B.04. Ion Channels

Support: NIH FI2 GM120004
NIH Z01-HD008755
NIH ZIC DC000081

Title: Cav2.3 is required for A-type K^+ current regulation of spontaneous synaptic input in hippocampal dendrites

Authors: *J. G. MURPHY[1], J. J. GUTZMANN[1], L. LIN[1], J. HU[1], R. S. PETRALIA[2], Y.-X. WANG[2], D. A. HOFFMAN[1];
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Abstract: The transient voltage-gated K^+ current (I_A) mediated by Kv4.2 in CA1 hippocampal pyramidal neurons regulates dendritic excitability, synaptic plasticity, and learning. We report that the voltage-gated Ca^{2+} channel Cav2.3 forms an ion channel complex with Kv4.2 and promotes I_A in CA1 pyramidal neurons. We found that Ca^{2+} binding auxiliary subunits of the Kv4.x family, known as K^+ channel interacting proteins (KChIPs), mediate Cav2.3 Ca^{2+} influx-mediated stabilization of Kv4.2 plasma membrane localization. We characterized the Cav2.3-Kv4.2 complex using immunofluorescence colocalization, coimmunoprecipitation, electron microscopy, FRAP, and FRET. Functionally, Cav2.3 Ca^{2+}-entry was found to promote whole-cell I_A due to an increase in Kv4.2 surface expression in heterologous cells and hippocampal neurons. Pharmacological block of Cav2.3 in neurons reduced whole-cell I_A which was less effective in Cav2.3 knockout (KO) mouse neurons. We also found reduced whole-cell I_A and an impaired proximal-distal dendritic I_A gradient in Cav2.3 KO neurons. Furthermore, reduced dendritic I_A in Cav2.3 KOs was found to occlude I_A-mediated regulation of excitatory synaptic currents and spine Ca^{2+} transients. These results reveal an intermolecular Cav2.3-Kv4.2 complex impacting basal synaptic function in CA1 hippocampal neurons.

Disclosures: J.G. Murphy: None. **J.J. Gutzmann:** None. **L. Lin:** None. **J. Hu:** None. **R.S. Petralia:** None. **Y. Wang:** None. **D.A. Hoffman:** None.

Poster

P095. Potassium Channels: Mechanisms of Action

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Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P095.06

Topic: B.04. Ion Channels

Support: MRC UKRI Grant MR/N0137941/1

Title: Transient ultrasound stimulation has lasting effects on neuronal excitability and whole-cell potassium currents.

Authors: *B. CLENNELL, T. G. J. STEWARD, B. W. DRINKWATER, D. J. WHITCOMB; Univ. of Bristol, Bristol, United Kingdom

Abstract: Background: Transcranial ultrasound stimulation can acutely modulate brain activity, but the lasting effects on neurons are unknown. We have previously shown that brief ultrasound stimulation resulted in the enhancement of intrinsic excitability of cultured cortical rat neurons, and this effect lasted for at least 8 hours (Clennell et al. 2021. Brain Stimul. 14(2):217-225). We subsequently investigated the effect on whole-cell sodium and potassium currents.
Objective: To assess the effect of transient ultrasound stimulation on neuronal excitability and whole-cell ion currents.
Methods: Primary rat cortical neurons were stimulated with a 40 second, 200kHz pulsed ultrasound stimulation or sham-stimulation. Intrinsic excitability was investigated through whole-cell patch-clamp recording by evoking action potentials (APs) in response to somatic current injection. Whole-cell sodium and potassium currents were recorded in response to sequential voltage steps in the presence of relevant ion channel blockers. Recordings were taken at set timepoints following ultrasound stimulation: 0-2 hours, 6-8 hours, 12-14 hours and 24-26 hours.
Results: In the 0-2 hour window, neurons stimulated with ultrasound displayed an increase in the mean frequency of evoked APs of 32% above control cell levels ($p = 0.023$). After 6-8 hours this increase was measured as 44% ($p = 0.0043$). By 12-14 hours this effect was eliminated and remained absent 24-26 hours post-stimulation. These changes to AP firing rates occurred in conjunction with statistically significant differences in AP kinetics that were similarly eliminated by 24-hours. We did not find any significant differences in the magnitude or kinetics of whole-cell sodium currents at any timepoint. Conversely, we did observe statistically significant increases in whole-cell potassium currents at 0-2hr ($\Delta I_{peak} = 24\%$, $p = 0.004$), 6-8hr ($\Delta I_{peak} = 20\%$, $p = 0.001$) and 12-14hr ($\Delta I_{peak} = 25\%$, $p < 0.001$) hours post-stimulation, which may explain the observed enhancement of AP repolarisation rates. Similar to the observed effects on excitability and AP kinetics, these changes were absent by 24-hours.
Conclusion: Stimulating neurons with 40 seconds of ultrasound enhances their excitability for up to 8 hours in conjunction with modifications to action potential kinetics. In addition, ultrasound induces sustained increases in whole-cell potassium currents suggesting changes to voltage-gated potassium channel function that may underpin the observed modifications to AP firing**.** These results can inform the application of transcranial ultrasound in experimental and therapeutic settings.

Disclosures: B. Clennell: None. **T.G.J. Steward:** None. **B.W. Drinkwater:** None. **D.J. Whitcomb:** None.

Poster

P095. Potassium Channels: Mechanisms of Action

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Topic: B.04. Ion Channels

Support: NWO FOM 16NEPH02
NWO VICI 865.17.003

Title: Optical voltage recordings reveal multiple gradients in action potential repolarization in the proximal axon

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Abstract: The axon initial segment (AIS) is a unique region of the proximal axon where the action potential (AP) is initiated to propagate forward into the axon and backward into the soma and dendrites. Within the AIS, the speed of AP repolarization is critical for the voltage-gated sodium channels to recover from inactivation, thereby determining the temporal dynamics for the next AP. Axons contain a large variety of potassium channels shaping repolarization, of which voltage-gated potassium channels (K_V, specifically K_V1) and large-conducting calcium- and voltage-dependent potassium (BK_{Ca}) channels directly mediate AP repolarization in the AIS. To better understand the detailed temporal and spatial properties of AP repolarization and the contribution of these channels, we used voltage-sensitive dye imaging to optically record the AP from the first 90 µm of rat L5 pyramidal axons in acute slices, which allowed us to compare the AP shape from regions within the same axon, including the myelinated internode. By optimizing holographic illumination, we restricted excitation light to the region of interest, which minimized light scattering from other neurites and maximized signal accuracy, enabling voltage-calibrated optical recordings at 50 µs and ~3 µm, temporal and spatial resolution, respectively. The results showed that the AP characteristics highly differed between axonal domains, with AP half-width being progressively narrower towards the distal AIS and within the internode in comparison to the proximal AIS (~350 µs vs ~720 µs, respectively, $n > 30$, $p < 0.001$). Next, we pharmacologically investigated which potassium channels contribute to setting the AP width in each axonal domain. Blocking K_V channels with 4-AP revealed an increase in the AP width in all regions of the axon. The effect of 4-AP became more pronounced with distance from the soma, with the width being increased to ~140% in the proximal AIS and to ~250% in the myelinated internode, consistent with the known expression of K_V channels. In contrast, application of Iberiotoxin, a blocker of BK channels, specifically increased the AP half-width in the proximal AIS to ~125% but not in other axonal domains. We are complementing these experimental approaches with computational simulations in COMSOL, studying the concerted action of

different ion channels at the nanoscale level. To summarize, using holographic voltage imaging, we show that distinct mechanisms control the AP repolarization in the forward and backward direction.

Disclosures: N.A.K. Hanemaaijer: None. **J.D. Elberse:** None. **M.H.P. Kole:** None.

Poster

P095. Potassium Channels: Mechanisms of Action

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P095.08

Topic: B.04. Ion Channels

Support: NSF Grant 1456302
NSF Grant 1645199
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NIH GRANT DC012938

Title: Kv3.1 enables sub-millisecond spikes in upper vocal-motor neurons

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Abstract: The speed and accuracy of muscle control and coordination for execution of fine motor skills depend on the spiking activity of upper motor neurons. In mammals, there is robust evidence that specific subclasses of these neurons contain unique excitable features that may enable the temporal precision required for fast, complex behaviors. Zebra finches provide an excellent model system for testing how a unique population of upper motor neurons provide the temporal precision required for a rapid, precise behavior. Their songs consist of motifs containing syllables with complex acoustic structure on a sub-second time scale. This behavior is controlled by pallial nuclei that form microcircuits considered analogous to those in the layered mammalian neocortex. Specifically, the vocal-motor projection neurons of the robust nucleus of the arcopallium (RAPNs) provide direct input to brainstem vocal and respiratory neurons, and are spatially separated from the upper-motor neurons of the dorsal intermediate arcopallium (AIdPNs), a region implicated in the control of somatic motor functions such as wing flapping and hopping. RA and AId have similar molecular profiles, suggesting that RA may have evolved as a specialization of AId, which is consistent with the motor theory of vocal learning origin. Here we provide strong evidence linking the expression of the Kv3.1 channel to the narrow spike

waveform produced by RAPNs. In whole cell current clamp recordings performed in brain slices from adult male finches, RAPNs exhibit fast spiking action potentials (APs) with narrower, sub-millisecond half-widths and larger maximum repolarization rates than in AIdPNs. We also show that the AP waveform in RAPNs is more sensitive to pharmacological compounds targeting Kv3.x channels than in AIdPNs. These electrophysiological findings are complemented by *in situ* hybridization data indicating that Kv3.1 is the primary transcript encoding TEA-hypersensitive channels in finch RA. We suggest that Kv3.1 regulates the intrinsic excitability of RAPNs on a sub-millisecond time scale and propose that this property allows a subclass of upper motor neurons to operate as highly specialized control units for the production of rapid and precise movements in the zebra finch. This specialization of RAPNs closely resemble those of Betz cells identified in mammals that have wide-caliber myelinated axons, fast action potential (AP) conduction velocities and ultra-narrow AP half-widths (<500μs). Thus, our findings in the finch may be of general interest in understanding the regulation of excitability in specialized cortical motor circuits in vertebrates.

Disclosures: B. Zemel: None. **A. Nevue:** None. **P.V. Lovell:** None. **C.V. Mello:** None. **H.P. von Gersdorff:** None.

Poster

P095. Potassium Channels: Mechanisms of Action

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Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P095.09

Topic: B.04. Ion Channels

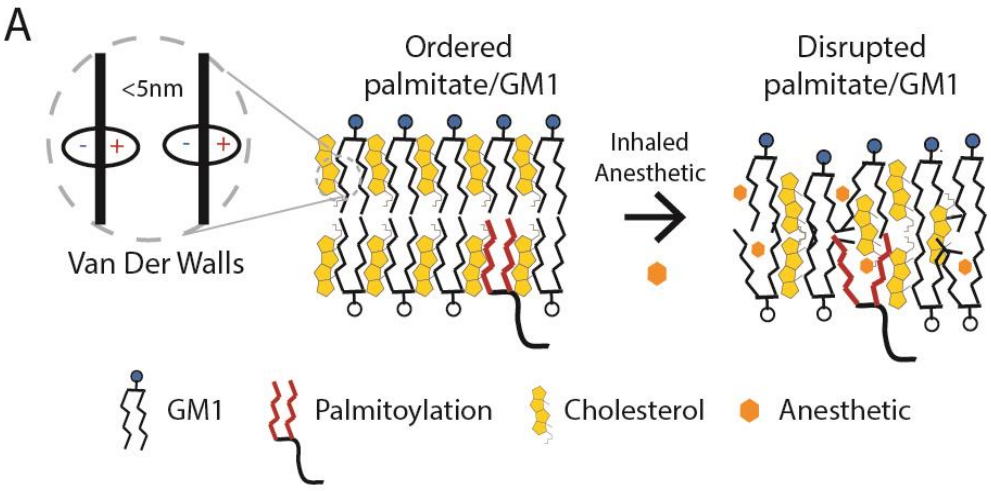
Support: DP2NS087943
R01NS112534

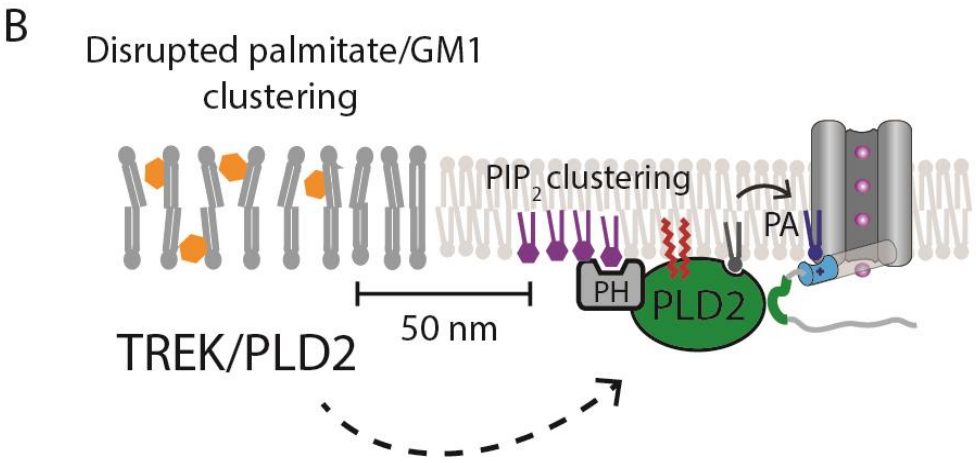
Title: A molecular basis for membrane-mediated inhaled anesthesia.

Authors: *S. HANSEN[1], M. A. PAVEL[2], R. LERNER[3];
[1]The Scripps Res. Inst., Jupiter, FL; [2]Scripps Res. Inst., Jupiter, FL; [3]Scripps Res. Inst., La Jolla, CA

Abstract: For 150 years anesthetics were thought to act on the plasma membrane, but a plausible site within the membrane had never been found. Here we show that inhaled anesthetics (chloroform and isoflurane) activate Twik-related potassium channel subtype 1 (TREK-1) through a two-step membrane-mediated mechanism. The membrane mediated site is located within clusters of ganglioside (GM1) containing lipids (see figure A). The clusters are ordered, cholesterol dependent, and bind palmitoylated proteins (i.e., proteins with a saturated 16 carbon lipid covalently attached). The anesthetics dislodge palmitoylated proteins including phospholipase D2 (PLD2). Once free from GM1 lipids, PLD2 translocates to PIP_2 containing

lipids where it encounters its substrate PC and activates TREK-1 (see figure B). General anesthetics chloroform, isoflurane, diethyl ether, xenon, and propofol all activate PLD2 in a live cell assay. The distance PLD2 travels during activation is less than 50 nm and imaged by two color super resolution microscopy of fluorescently labeled lipids and proteins. The short distance traveled allows for very fast activation of PLD2 and the channel. A catalytically dead PLD2 robustly blocks anesthetic TREK-1 currents in cell patch-clamp. Localization of PLD2 renders a TRAAK channel anesthetic sensitive, a channel that is otherwise insensitive. In the whole brain of flies, anesthesia perturbs GM1 clusters and PLD^{null} flies resist anesthesia. Our results establish a membrane-mediated target of inhaled anesthesia and suggest PA helps set thresholds of anesthetic sensitivity *in vivo*. Anesthetic induced movement of proteins between GM1 and PIP_2 clusters establishes a potent and dynamic layer of nanoscopic lipid signaling. And the disruption of lipid order by high concentration of anesthetic suggests a chaotropic effect contributes to the membrane-mediated mechanism of inhaled anesthesia.





Disclosures: S. Hansen: None. **M.A. Pavel:** None. **R. Lerner:** None.

Poster

P095. Potassium Channels: Mechanisms of Action

Location: Virtual Only

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Program #/Poster #: P095.10

Topic: B.04. Ion Channels

Support: CAMH Discovery Fund

Title: A data-driven approach for identifying gene expression-electrophysiological property relationships

Authors: *A. NIGAM, S. L. HILL, S. TRIPATHY;
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Abstract: As the influence of genes on gene expression is extensively studied with next generation sequencing technologies, the effects of gene expression on neuronal electrophysiological properties remains poorly understood. With the help of in silico approaches, we can study a large sample size of cells at a given time, infer what is happening at the cellular and molecular level, and predict future directions for experimental work. Here, to study the relationship between gene expression patterns and electrophysiological properties in silico, we developed an analysis approach to assess these relationships using large patch-seq datasets.
We looked at all the genes and ten electrophysiological properties and filtered for gene-ephys pairs that are expressed in at least 5 cells or more in our inhibitory cell datasets (Gouwens *et al*., 2020; Scala *et al*., 2020). Next, we applied linear mixed models where we looked at the influence of gene expression on an electrophysiological property at different cell type resolutions in the patch-seq dataset from mouse visual cortex and mouse motor cortex. These different cell type resolution models (Class, Subclass, and Cluster) correspond to the taxonomic classification of these neurons. The linear mixed models are then applied on each gene and electrophysiological property combination. Further, we looked into the significant gene-ephys combinations to observe which resolution is the best to study gene expression and electrophysiological property. We concluded that the gene expression - electrophysiology property relationships at the class level are the best to study. Specifically, we found multiple significant gene expression - electrophysiology property pairs from the mouse visual cortex dataset that belonged to either voltage gated ion channel genes or genes already known to be associated with epilepsy. Moreover, we have been focusing on exploring the associations between the 40 potassium voltage gated ion channel genes (Ranjan *et al*., 2019) and electrophysiological properties at the subclass and cluster levels. Such gene expression - electrophysiology property associations will further increase our understanding of the mechanisms of the neurological diseases they are implicated in.
Eventually, our goal is to better understand how these genes correlated with ephys differences might intersect with genes previously linked with epilepsy in humans, with the goal of better understanding and interpreting genetic variants associated with epilepsy. Our next goal is to check whether the gene expression - electrophysiology property associations can be replicated in mouse and human excitatory cells datasets.

Disclosures: A. Nigam: None. **S.L. Hill:** None. **S. Tripathy:** None.

Poster

P113. Spike-Timing Dependent Plasticity

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P113.01

Topic: B.07. Synaptic Plasticity

Title: A dendritic mechanism for retrieval-dependent memory consolidation

Authors: *E. GERON, W. GAN;
NYU, New York, NY

Abstract: Retrieval-dependent processing of a new memory improves its long-term retention. Although the positive effect of retrieval on memory strength was recognized at the beginning of the 20th century, its underlying neuronal mechanisms remain unclear. To study this, we trained mice with a modified fear conditioning paradigm, where a presentation of a reminder tone, shortly after tone-shock pairing, improved long-term memory retention. We next constructed a platform with which mice were conditioned while their brains were imaged via two-photon microscopy. We found that reminders induced a substantial increase in Ca^{2+} spike generation in apical dendrites of layer 5 pyramidal neurons (L5 PNs) of the primary motor cortex. The potency of reminders to evoke Ca^{2+} spikes was transient, occurring within less than 30 minutes after tone-shock pairing. Notably, different dendritic branches of individual L5 PNs exhibited Ca^{2+} spikes with different amplitudes. On branches with high Ca^{2+} spike amplitudes, spines were more likely to be activated before the spikes, and to undergo long-lasting weakening (depotentiation). In contrast, spines on branches with lower Ca^{2+} spike amplitudes tended to be activated after Ca^{2+} spike generation, and only spines with high activity during spiking underwent weakening. In addition, optogenetic inhibition of CaMKII, shortly after reminder-induced spikes, disrupted spine weakening, spike transience, and conditioned freezing. These results reveal that early reminders trigger the generation of dendritic spikes to cause branch-specific synaptic plasticity to support retrieval-dependent improvement of memory retention.

Disclosures: E. Geron: None. **W. Gan:** None.

Poster

P113. Spike-Timing Dependent Plasticity

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P113.02

Topic: B.07. Synaptic Plasticity

Support: The Academy of Finland (decision Nos. 318879, 326494, 326495, 345280)
European Union's Horizon 2020 Framework Programme for Research and Innovation under the Specific Grant Agreement 945539 (Human Brain Project SGA3)

Title: Integrating neuron-glia knowledge and computational modeling to understand cortical synaptic plasticity

Authors: T. MANNINEN[1], A. SAUDARGIENE[2], ***M.-L. LINNE**[1];
[1]Fac. of Med. and Hlth. Technol., Tampere Univ., Tampere, Finland; [2]Neurosci. Inst., Lithuanian Univ. of Hlth. Sci., Kaunas, Lithuania

Abstract: Synaptic plasticity is expressed in the brain in diverse forms across multiple timescales. In the neuron-centric view, changes in neuronal activity and synaptic plasticity depend solely on the intrinsic properties of neurons and the information transmitted. However, non-neuronal glial cells, particularly astrocytes, have recently been shown to dynamically modulate synaptic transmission and plasticity. Several biochemical and biophysical mechanisms of astrocytes have been proposed to be involved in synaptic transmission and plasticity and the mechanisms involved seem to depend on the developmental stage of an animal, brain area, and neural circuitry. We are interested in studying different forms of synaptic plasticity in cortical synapses during postnatal development. We use our previously developed computational model for spike-timing-dependent long-term depression (t-LTD) at layer 4 to layer 2/3 synapse in somatosensory cortex as the base of our work (Manninen et al., 2020). The study involved using a multitude of data from molecular, biochemical, and electrophysiological experiments in rodent somatosensory cortex during postnatal development. The model links presynaptic, postsynaptic, and astrocytic mechanisms to the time window of t-LTD induction. In our model, postsynaptically released endocannabinoids increase astrocytic calcium signaling which in turn induces exocytosis of glutamate. Glutamate exocytosis from an astrocyte can further activate presynaptic N-methyl-D-aspartate receptors coupled with calcineurin signaling and influence synaptic properties dynamically. In the present study, we performed computational sensitivity analysis for the key model variables with an aim to extend the model to express different plasticity forms of cortical synapses in addition to t-LTD. Our results contribute to better understanding of synaptic plasticity mechanisms during postnatal development in health and disease. The novelty of our work is that we incorporate not only the neuronal mechanisms but also several astrocytic mechanisms responsible for synaptic plasticity.

Disclosures: T. Manninen: None. **A. Saudargiene:** None. **M. Linne:** None.

Poster

P113. Spike-Timing Dependent Plasticity

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P113.03

Topic: B.07. Synaptic Plasticity

Support: Life Sciences Postdoctoral Fellowship
Vallee Foundation
National Institutes of Health

Title: Thalamic inputs to cortical layer 5 neurons strengthen within a subcellular critical period

Authors: *C. E. YAEGER, D. VARDALAKI, N. J. BROWN, M. T. HARNETT;
McGovern Inst. for Brain Res., MIT, Cambridge, MA

Abstract: On the cellular level, neural plasticity is a function of pre and post-synaptic properties, such as the pattern of presynaptic innervation, postsynaptic receptor content and integration, and their convergent activity. During postnatal development, plasticity occurs in functional cascades, where specific synapses are plastic during defined windows of time. To achieve precise synaptic changes, do single neurons maintain distinct plasticity rules for functionally-specific inputs? Here we show that the apical oblique dendrites of layer 5b neurons in primary visual cortex have a developmentally restricted window of plasticity for feedforward thalamic input. We find that within layer 5b neurons, the oblique dendritic compartment is the sole recipient of input from the dorsolateral geniculate nucleus of the thalamus. In adult cortex, synapses on oblique dendrites integrate inputs linearly with minimal calcium influx and lack the capacity for NMDA-dependent potentiation, unlike neighboring basal dendrites. However, oblique dendrites can sustain NMDA-dependent nonlinearities, calcium spikes, and plasticity during early postnatal development (P14-21). The transition to adult properties is apparent by postnatal day 28 and requires vision. Plasticity during this developmental time window results in the robust recruitment of AMPA receptors, and by adulthood, oblique dendrites have a higher AMPA/NMDA ratio compared to basal dendrites. Early visual experience therefore changes the postsynaptic landscape in oblique dendrites, resulting in a decreased propensity for synaptic potentiation, while basal dendrites remain plastic into adulthood. The presence of strong, linear thalamic input to the apical obliques early in development may contribute to the stability of feedforward representations while experience-dependent plasticity is ongoing in other dendritic compartments.

Disclosures: C.E. Yaeger: None. **D. Vardalaki:** None. **N.J. Brown:** None. **M.T. Harnett:** None.

Poster

P113. Spike-Timing Dependent Plasticity

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P113.04

Topic: B.07. Synaptic Plasticity

Title: The contribution of different postsynaptic Ca^{2+} sources and Ca^{2+}-permeable AMPA receptors to timing-dependent LTP elicited by two distinct low repeat STDP paradigms at SC-CA1 synapses

Authors: *B. KHODAIE[1,2], E. CEPEDA-PRADO[1], E. EDELMANN[1,2,3], V. LESSMANN[1,2,3]; [1]Inst. of Physiology, Otto-von-Guericke Univ., Magdeburg, Germany; [2]OVGU ESF-funded Intl. Grad. Sch. ABINEP, Magdeburg, Germany; [3]Ctr. for Behavioral Brain Sci. (CBBS), Magdeburg, Germany

Abstract: STDP is a cellular model for learning that employs low frequency repeats of coincident action potential (AP) firing in pre- and postsynaptic neurons to induce timing-dependent LTP (t-LTP). Recent evidence showed that t-LTP at SC-CA1 synapses is equally robust as high-frequency stimulation induced conventional LTP. While conventional SC-CA1 LTP depends on postsynaptic Ca^{2+} rise through NMDA receptors and L-type VGCCs, the contribution of intracellular and extracellular Ca^{2+} sources to t-LTP is not well understood. We set out to decipher the origin of postsynaptic Ca^{2+} that triggers induction of SC-CA1 t-LTP elicited by physiologically relevant low repeat STDP stimulation. We performed whole cell patch clamp recordings in acute hippocampal slices from 4 weeks old male C57Bl6 mice and used either a canonical (1EPSP/1AP) or a burst (1EPSP/4APs) STDP protocol, which was repeated just 6 times at 0.5 Hz, to induce t-LTP in CA1 pyramidal cells. The importance of postsynaptic Ca^{2+} signaling for t-LTP was initially tested with application of the Ca^{2+} chelator BAPTA (10 mM) via the patch pipette, which blocked t-LTP induced by both protocols. Bath application of APV (50 µM) and Nifedipine (25 µM) were used to test the role of NMDAR and L-type VGCCs. Contribution of mGluR signaling was tested by co-application of the mGluR1 antagonist YM298198 (1 µM), and the mGluR5 antagonist MPEP (10 µM). Contribution of GluA2-lacking calcium-permeable AMPARs (cp-AMPARs) was tested by the bath applied inhibitors NASPM (100 µM) or IEM-1460 (100 µM). To test the role of ER Ca^{2+}stores, we used bath application of 2-APB (100 µM) to inhibit IP3 receptors, and intracellular ryanodine (100 µM) to block ryanodine receptors (RyRs). Our results show that t-LTP induction by the 2 protocols (6x 1:1 and 6x 1:4) recruited different induction mechanisms. Interestingly, both protocols depended on activity of cp-AMPARs, whereas only the canonical 6x 1:1 protocol depended on Ca^{2+} entry via NMDARs and L-type VGCCs. While group I mGluRs (mGluR1 and mGluR5) contributed to t-LTP induction with the burst protocol, the canonical 6x 1:1 protocol induced t-LTP did not involve group I mGluRs. However, activation of IP3 and RyR receptor sensitive internal Ca^{2+} stores was necessary to allow t-LTP with either of the 2 protocols. Thus, while our study reveals distinct mechanisms for the initial postsynaptic Ca^{2+} rise required for the 2 low repeat t-LTP protocols, both paradigms critically depend on sustained Ca^{2+} release from the ER and Ca^{2+} influx through cp-AMPARs during test stimulation after the induction paradigm. Funding: OVGU ESF-founded International graduate school ABINEP.

Disclosures: B. Khodaie: None. **E. Cepeda-Prado:** None. **E. Edelmann:** None. **V. Lessmann:** None.

Poster

P113. Spike-Timing Dependent Plasticity

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P113.05

Topic: B.07. Synaptic Plasticity

Support: NIH 5R00MH118423

Title: Local excitatory control of CA1 interneurons shows diverse plasticity rules in the awake, behaving mouse

Authors: *P. PAUDEL, K. BAUGH, S. MCKENZIE;
Neurosciences, Univ. of New Mexico, Albuquerque, NM

Abstract: Memories are believed to be stored in the pattern of synaptic connectivity between neurons. Coincident neural activity between connected cells is known to change synaptic strength, but the rules for how activity affects connectivity *in vivo* are largely unknown. Area CA1 of the hippocampus has been the focus of most *in vivo* electrophysiological investigations of hippocampal function, but excitatory neurons in this region primarily influence each other indirectly through inhibitory interneurons. Therefore, we tested the spike-timing dependent plasticity rules governing the excitatory synaptic strength onto local inhibitory cells. The red-shifted opsin ChrimsonR was expressed under the parvalbumin (PV) promoter to target fast-spiking interneurons, and Channelrhodopsin 2 (ChR2) was expressed under the CaMKIIα promoter to target primarily pyramidal cells. To induce plasticity, red (635 nm) and blue (473 nm) light were synchronously pulsed at 8 Hz to drive rhythmic coincident activity in pyramidal and PV+ interneurons. The effects of plasticity were assessed by measuring dose response curves to red and blue light delivered before and after pairing. Bidirectional changes in excitability were observed in both pyramidal cells and interneurons, in response to blue and red light. Even in sessions where plasticity was observed, simultaneously recorded units could show stable responses. In a second experiment, units were detected online and cross correlations between spike trains were calculated in real time to identify putatively connected pairs. After a baseline recording, firing of the presynaptic pyramidal cell was used to trigger 10 ms of red light to activate ChrimsonR in PV+ interneurons. In a subset of putatively connected pairs, this protocol strongly increased the correlation in firing between the pre- and post-synaptic cell pairs. The increase in coupling strength correlated with the efficacy of the closed-loop protocol in driving short-time scale spike-spike correlations, as expected from a Hebbian learning rule. Together, these findings show the potential for heterogeneous plasticity rules at the excitatory to inhibitory

synapse in area CA1 of the hippocampus. Future experiments will explore the consequences of this plasticity on neural coding and memory.

Disclosures: P. Paudel: None. **K. Baugh:** None. **S. McKenzie:** None.

Poster

P113. Spike-Timing Dependent Plasticity

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P113.06

Topic: B.07. Synaptic Plasticity

Support: K.J has received funding from the Belgian National Fund for Scientific Research - FNRS (ID application: 34959817, https://www.frs-fnrs.be/en/)

Title: Effect of switches in brain states on calcium-based plasticity rules: a computational study for sleep-dependent memory consolidation.

Authors: *K. JACQUERIE, C. MINNE, G. DRION;
Electrical Engin. and Computer Sci., Univ. of Liege, Liege, Belgium

Abstract: Sleep contributes to memory through a mechanism called sleep-dependent memory consolidation. Little is known about its underlying physiological and functional mechanisms, but it involves global and long-lasting switches in neuronal population activity, called the wake-sleep pattern. At the network level, neuronal populations show transitions from active states to oscillatory states. Zooming in at the cellular level, neurons switch from *tonic firing to bursting*. In parallel, memory and learning rely on functional changes in neuronal connectivity through a process called *synaptic plasticity*. The main goal of this project is to investigate how these transient switches in brain rhythmic activity, or brain states, affect synaptic plasticity and learning. To this end, we exploit recent advances in the mathematical modeling of cellular-induced fast and localized switches in brain states [Drion, 2018; Jacquerie, 2021]. These models make the *generation of brain state switches independent from synaptic weights* between neurons and thus compatible with synaptic plasticity. We investigate the interaction between switches in brain states and learning using an associative memory task, which quantifies the ability of neurons coding for concomitant events to strongly connect together. To do so, we embed a neuron conductance-based model with a *calcium-dependent plasticity rule* in which internal calcium concentration governs the direction and the amplitude of the synaptic changes [Shouval, 2002; Graupner and Brunel, 2016], where internal calcium dynamics are regulated by the activation of NMDA receptors and voltage-gated calcium channels. This rule is parametrized to fit experimental data guaranteeing the correct behavior in wake state [Sjostrom, 2001]. The synaptic plasticity rule is shown to fail to reproduce sleep-dependent memory consolidation theories such as synaptic homeostasis hypothesis [Tononi, 2020] or active systems consolidation

[Born, 2012] for any parameter values. Indeed, regardless of the connection strengths established during the day, they all converge to a fixe point during the night, which resets the circuit connectivity during sleep without showing any down-selection or reinforcement. Adding the effect of neuromodulators on calcium-dependent plasticity rule solves this problem by modifying calcium signal transduction pathways, hence the synaptic rule properties. The proposed model makes it therefore possible to track the evolution of synaptic wiring during sleep-wake switches, as well as to study sleep related mechanisms that can affect plasticity rules.

Disclosures: K. Jacquerie: None. **C. Minne:** None. **G. Drion:** None.

Poster

P113. Spike-Timing Dependent Plasticity

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P113.07

Topic: B.07. Synaptic Plasticity

Title: Are synaptic plasticity rules compatible with sleep-dependent memory consolidation?

Authors: *C. MINNE[1], K. JACQUERIE[2], G. DRION[3];
[1]Univ. of Liège, Fraiture, Belgium; [3]Electrical Engin. and Computer Sci., [2]Univ. of Liege, Liege, Belgium

Abstract: Among all essential functions of sleep, research has highlighted its preponderant role in memory formation and consolidation. At the cellular level, *synaptic plasticity*, the ability of neurons to modify their connections, is the cellular mechanism responsible for learning and memory. Therefore, sleep and synaptic plasticity are two linked concepts in the context of memory consolidation.
A large amount of experimental and modeling work has extracted mechanisms of synaptic plasticity, such as e.g. *spike-timing-dependent plasticity*, that occur during wakefulness [Bi and Poo, 1998]. But are they compatible with memory consolidation during sleep [Tononi, 2020; Born, 2016]?
Here, we tackle this question by focusing on the behavior of *phenomenological* models of plasticity, *i.e.* mathematical models based on spike-timing, during sleep. To this end, we build a 3-cell circuit composed of conductance-based models capable of reproducing faithfully the neuronal activity switches observed during the sleep-wake cycle [Drion, 2018; Jacquerie, 2021]. Plasticity is taking place between a pre- and a postsynaptic cell and implemented using well-known plasticity rules from the literature: the pair-based [Abbott, 2000] and triplet [Pfister, 2006] models. We show that those rules, which are successful in reproducing experimental observation in waking state, are no longer compatible with memory consolidation after a sleeping period. Interestingly, the outcome is not determined by the synaptic plasticity rule itself but rather by the choice of saturation bounds applied to the synaptic connection. We show that

modeling the effect of neuromodulators on the learning rule during the sleep-wake cycle, for instance by switching to synaptic depression only during sleep [Gonzalez-Rueda, 2018], permits to reconcile classical synaptic plasticity rules with sleep-dependent memory consolidation.

Disclosures: C. Minne: None. **K. Jacquerie:** None. **G. Drion:** None.

Poster

P119. Structural Plasticity: Mechanisms

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Program #/Poster #: P119.01

Topic: B.07. Synaptic Plasticity

Support: NIMH (R01MH077303)
Sealy Center for Structural Biology and Molecular Biophysics (SCSB) at UTMB

Title: Interplay between hevin, SPARC, and MDGAs: modulators of neurexin-neuroligin transsynaptic bridges

Authors: *S. FAN, M. MACHIUS, G. RUDENKO;
Dept. of Pharmacol. and Toxicology, The Univ. of Texas Med. Br., Galveston, TX

Abstract: Hevin (also known as SPARCL1) promotes synapse formation and stabilizes neurexin-neuroligin trans-synaptic bridges *in vivo*. Hevin is secreted predominantly by astrocytes, and its synaptogenic effects are antagonized by related protein, SPARC, which shares ~65% sequence identity in the two major domains (the FS-EC tandem). A third protein, the membrane-tethered MDGA, blocks neurexin-neuroligin trans-synaptic bridges. What's more, heparan sulfate (HS), a glycan modification on neurexins promotes neurexin binding to neuroligin, adding an additional dimension to the diversity and regulation of neurexin-neuroligin interactions. The exact molecular underpinnings of this interaction network that regulates neurexin-neuroligin trans-synaptic bridges is largely unknown. Here, we show that hevin and SPARC both bind neurexin and neuroligin, and they can compete with each other. Our 2.3 Å hevin FS-EC crystal structure reveals both profound similarities and profound differences with the SPARC counterpart. We show that hevin and SPARC FS domains are structurally conserved, and they both house nanomolar affinity binding sites for neurexin and neuroligin. In contrast, the hevin and SPARC EC domains are structurally very divergent, and they both essentially don't interact with neuroligin. SPARC's antagonist action compared to hevin likely is rooted in the absence of a long N-terminal region that is found in hevin but not SPARC. Intriguingly, we also show that the hevin FS domain competes with MDGA for an overlapping binding site on neuroligin; this shared site may be in the vicinity of a putative HS binding site on neuroligin. Furthermore, the hevin EC domain binds the extracellular matrix protein, collagen (like SPARC), which may provide an avenue to tether neurexin-neuroligin trans-synaptic bridges to

the extracellular matrix surrounding synapses. Collectively, our data provide mechanistic insight into a portfolio of molecules, both secreted and membrane-tethered, that cooperate together to regulate neurexin-neuroligin trans-synaptic bridges, impacting synapse formation and ultimately shaping neural circuits.

Disclosures: S. Fan: None. **M. Machius:** None. **G. Rudenko:** None.

Poster

P119. Structural Plasticity: Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P119.02

Topic: B.07. Synaptic Plasticity

Support: NIH/NINDS NS111976
NIH/NINDS NS110385

Title: Extracellular kinases: The role of Vertebrate Lonesome Kinase (VLK) in Eph-ecting the synaptic recruitment of NMDA receptors.

Authors: *P. CHANDER[1], H. R. WASHBURN[1], S. D. KOLLURU[1], T. J. PRICE[2], M. B. DALVA[1];
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Abstract: While protein function modification by intracellular phosphorylation is one of the more widely appreciated mechanisms mediating protein-protein interactions, the function of extracellular phosphorylation and their cognate protein kinases have remained obscure. One important event that requires extracellular phosphorylation is the direct interaction between the receptor tyrosine kinase EphB2 and the GluN1 subunit of the NMDAR. The EphB family of receptor tyrosine kinases are essential for spine synapse formation and synaptic localization of the NMDAR. However, little is known about the kinases that mediate phosphorylation of extracellular domains. Recent studies from the Dixon and Whitman labs have identified two protein kinases families the mediate most extracellular phosphorylation. Of these, the Vertebrate Lonesome kinase (VLK; PKDCC) is a putative tyrosine kinase with six family members. We have begun to examine the function of these kinases focusing on the tyrosine kinase VLK, an essential gene, with knockouts dying at birth from respiratory failure. Other notable phenotypes are cleft palate, polydactyly and defects in axon guidance, phenotypes that overlap with some of those found in knockouts of EphBs and their ligands the ephrin-Bs. To further characterize VLK function, we examined the subcellular localization and regulation of VLK in neurons. Since VLK is secreted in platelets, we asked if ephrin-B2 activation of EphB2 might result in VLK release into the media from neurons. Ephrin-B induces VLK secretion from cortical neurons.

Moreover, ephrin-B activation results in increased transcriptional activation of VLK. To begin to examine the mechanism for VLK release, we fractionated the brain. Remarkably, in both vesicle and synaptosome fractions, VLK co-fractioned with synaptic vesicle markers. To determine if VLK is localized in the cellular secretory pathway, we expressed a VLK-mCherry fusion construct in cortical neurons. VLK was distributed as discrete puncta through the axons. Live cell imaging revealed that VLK puncta underwent active bidirectional transport in axons of neurons and colocalized with pre-synaptic protein Synaptophysin 1 (SYP1). We are currently examining whether stable VLK puncta are associated with postsynaptic markers. Overall, data suggest that VLK is regulated by EphB signaling and may be associated with presynaptic sites.

Disclosures: P. Chander: None. **H.R. Washburn:** None. **S.D. Kolluru:** None. **T.J. Price:** None. **M.B. Dalva:** None.

Poster

P119. Structural Plasticity: Mechanisms

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Program #/Poster #: P119.03

Topic: B.07. Synaptic Plasticity

Support: NIH/NINDS NS111976
NIH/NINDS NS110385

Title: Molecular Mechanisms of the EphB-NMDA Receptor Extracellular Interaction

Authors: H. R. WASHBURN[1], P. CHANDER[2], S. D. KOLLURU[2], T. J. PRICE[3], ***M. B. DALVA**[2];
[2]Neurosci., [1]Thomas Jefferson Univ., Philadelphia, PA; [3]Sch. of Behavioral and Brain Sci., Univ. of Texas at Dallas, Dallas, TX

Abstract: Synapse formation and function is regulated by several families of transsynaptic adhesion proteins that span the synaptic cleft, guide synaptic development, and regulate mature synaptic function. Of these families, the EphB family of receptor tyrosine kinases are required for spine synapse formation and the synaptic localization of the N-methyl-D-aspartate receptor (NMDAR). Normal brain function requires that NMDARs are properly localized to synaptic sites, and synaptic NMDARs are required for the induction of structural and synaptic plasticity. At the mature synapse, EphB2 binding the membrane bound ligands, the ephrin-Bs, to the ligand binding domain (LBD) of EphB2 drives a direct extracellular interaction between the EphB2 receptor and NMDAR. The direct extracellular EphB-NMDAR interaction requires post-translational modification of the extracellular domain of EphB2. Unbiased mass spectrometry data showed that a specific tyrosine reside, Y504, in the fibronectin type III (FN3) repeat domain of the extracellular region of EphB2 undergoes ephrin-B2 ligand-dependent phosphorylation.

Furthermore, functional analysis of EphB2 phosphorylation mutants indicated that Y504 is necessary and sufficient for the EphB-NMDAR interaction. Protein phosphorylation is a nearly universal mechanism used by cells to regulate intracellular and extracellular processes. The majority of phosphoproteins are intracellular; however, mass-spec and other approaches have shown recently that numerous extracellular proteins are phosphorylated. Here, we explore the possible mechanisms of EphB2 Y504 phosphorylation. We identify a secreted extracellular tyrosine kinase, vertebrate lonesome kinase (VLK), that is capable of phosphorylating EphB2 at Y504 and induces EphB2 interaction with the NMDA receptor. Additionally, loss of VLK results in a loss of the Eph-NMDA receptor interaction. Our findings also show that VLK fractionates with synaptic vesicles and colocalizes with synaptic vesicle markers, suggesting a mechanism of pre-synaptic secretion into the synaptic cleft. Together, these studies impact our understanding of the role and importance of extracellular protein-protein interactions in synaptic development and function.

Disclosures: H.R. Washburn: None. **P. Chander:** None. **S.D. Kolluru:** None. **T.J. Price:** None. **M.B. Dalva:** None.

Poster

P119. Structural Plasticity: Mechanisms

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Program #/Poster #: P119.04

Topic: B.07. Synaptic Plasticity

Support: NIH Grant NS111976-01A1
NIH Grant NS115441-01

Title: Vertebrate Lonesome Kinase phosphorylates EphB2 receptors on an extracellular residue to drive NMDA-dependent pain

Authors: *H. ELAHI[1], S. N. HASSLER[2], M. KUME[1], S. SHIERS[1], M. B. DALVA[3], T. J. PRICE[1];
[1]Univ. of Texas at Dallas, Richardson, TX; [2]Univ. of Houston, Houston, TX; [3]Neurosci., Thomas Jefferson Univ., Philadelphia, PA

Abstract: EphB/ephrinB signaling plays a key role in synaptic function and is linked to several neuropathological conditions, including neuropathic pain. The EphB2 receptor tyrosine kinase is known to interact with *N*-Methyl-D-aspartate glutamate receptors (NMDAR), resulting in increased surface retention of NMDARs at the synapse and an enhancement of postsynaptic currents. As changes in NMDAR signaling have been heavily implicated in the development of both acute and chronic pain, targeting this receptor interaction may produce clinically relevant therapeutics. We have recently demonstrated that the phosphorylation of a tyrosine residue

(Y504) on the extracellular domain of the EphB2 receptor is necessary and sufficient to induce its interaction with NMDARs and cause mechanical pain hypersensitivity. The kinase responsible for this phosphorylation is unknown. We hypothesized that Vertebrate Lonesome Kinase (VLK) may be the key effector. Since the phosphorylation site of interest is located on the extracellular face of the EphB2 receptor, a secreted tyrosine kinase like VLK is an ideal candidate. Our results provide strong support for our hypothesis that VLK drives pain via extracellular phosphorylation of EphBs. We show that intrathecal injection of VLK resulted in long-lasting mechanical hypersensitivity in mice. Additionally, the effects observed for VLK were dependent on kinase activity and entirely dependent upon NMDARs because they were blocked by APV. We also show evidence of VLK protein and mRNA expression in dorsal root ganglion and spinal cord neurons of both mice and humans. Our results provide evidence for the involvement of VLK in pain behaviors and spinal cord NMDAR signaling, and identify a completely unique extracellular kinase target for the alleviation of pain.

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Poster

P119. Structural Plasticity: Mechanisms

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Program #/Poster #: P119.05

Topic: B.07. Synaptic Plasticity

Support: UW ICTR

Title: Biochemical Regulation of RhoA Activity and Signaling in Cortical Neurons via DISC1

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[1]Comparative Biosci., The Univ. of Wisconsin-Madison, Madison, WI; [2]Dept. of Psychiatry, Johns Hopkins Univ., Baltimore, MD

Abstract: Structural plasticity of dendritic spines, the sites of most excitatory connections in the brain, is critical for many forms of synaptic plasticity, and ultimately, learning and memory. Long term potentiation (LTP) and long-term depression (LTD) are associated with spine head enlargement and shrinkage, respectively. Structural plasticity is largely mediated by intracellular signaling pathways that cause the reorganization of the actin cytoskeleton in dendritic spines. Small GTPases are potent regulators of synaptic structural plasticity via their direct and indirect influences on actin. Small GTPases are active when bound to GTP and inactive when GDP bound. The direct activation of small GTPases is accomplished by guanine nucleotide exchange factors (GEFs). RhoA activation is known to have a destabilizing effect on the actin cytoskeleton with associated alterations in dendritic spine stability. However, the specific biochemical

mechanisms that control RhoA activity in neurons remain largely uncharacterized. PDZ-RhoGEF is a RhoA-specific guanine nucleotide exchange factor whose expression is restricted to the central nervous system, with levels highest in the neocortex. Further, among RhoA-specific GEFs, PDZ-RhoGEF shows an unusually potent ability to activate RhoA. Nevertheless, owing to a scarcity of studies, minimal is known about the regulation and function of PDZ-RhoGEF in the brain. Here we show that PDZ-RhoGEF destabilizes dendritic spines in cortical neurons and causes cortical-dependent cognitive impairment. We found that PDZ-RhoGEF activity and cellular localization in vitro is controlled by its interaction with the DISC1 scaffolding-like protein, such that DISC1 inhibits PDZ-RhoGEF-mediated RhoA activity. Ongoing studies are examining the functional consequences of the interaction between PDZ-RhoGEF and DISC1 in dendritic spines and in cortical neurons in vivo. Taken together, these findings identify PDZ-RhoGEF as a potent regulator of synaptic stability and cognition, and highlight a previously unrecognized involvement for DISC1 in controlling RhoA activity via PDZ-RhoGEF.

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Poster

P119. Structural Plasticity: Mechanisms

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Program #/Poster #: P119.06

Topic: B.07. Synaptic Plasticity

Support: Air Force Office of Scientific Research FA9550-18-1-0051

Title: Biophysical modeling of actin-mediated structural plasticity

Authors: *M. BONILLA-QUINTANA, P. RANGAMANI;
UCSD, La Jolla, CA

Abstract: Memories are thought to be encoded in the connections between neurons (synapses) which are plastic, increasing or decreasing their strength depending on their activity. The postsynaptic part of most excitatory synapses are located in small protrusions from the dendrites, called dendritic spines, which undergo structural plasticity where a long-lasting synaptic strength increase (long term potentiation, LTP) or decrease (long term depression, LTD) correlates with an increase or decrease in the size of these spines, respectively. Changes in spine size are possible due to a reconfiguration of its cytoskeleton, composed primarily of actin filaments, triggered by a signaling cascade upon LTP/LTD induction. Although many studies have investigated parts of this process, it is challenging to have a full picture due to the small size of the spines, its high concentration of actin, and the diversity of proteins. Therefore, we propose a biophysical 3D model to investigate the dynamics of actin upon LTP/LTD and the resulting changes in the spine size and shape. Similar to previous models describing cell motility, our

model aims to determine the mechanisms underlying the spine reconfiguration. We use partial differential equations to describe the evolution actin, cofilin, and Arp2/3 concentrations, confined to a time-evolving domain representing the spine membrane with Helfrich potential energy. We focus on cofilin and Arp2/3 from all the possible actin-binding proteins (ABPs) because of their principal role in severing and branching actin filaments, respectively. The model allows the study of heterogeneous protein distribution inside the spine, which results in asymmetric and more realistic spine shapes. We show that the timing and location of the interaction between these proteins are crucial to generate long-lasting changes in the spine shape and size. Our model can reproduce the changes upon LTP and LTD by only altering the protein concentrations. Moreover, it can inform about spine abnormalities due to alterations in ABPs characteristic in diseases such as Alzheimer's Disease.

Disclosures: M. Bonilla-Quintana: None. **P. Rangamani:** None.

Poster

P119. Structural Plasticity: Mechanisms

Location: Virtual Only

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Program #/Poster #: P119.07

Topic: B.07. Synaptic Plasticity

Title: Computational benefits of nonlinear synaptic activations

Authors: *Y. SONG[1], M. K. BENNA[2];
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Abstract: We investigate supervised learning in a perceptron-like neural network architecture in which synaptic transmission is governed by nonlinear functions relating the post- synaptic currents to pre-synaptic neural activities. These nonlinearities are shaped by learning rules for binary classification, and can implement curved decision boundaries that are more flexible than hyperplanes. We show that such systems can achieve a larger capacity than the standard perceptron, even if all synaptic nonlinearities are constrained to be monotonically increasing. We discuss both offline optimization schemes and more biologically plausible online learning rules for finding useful decision boundaries, as well as the convergence properties of the proposed learning algorithms.

Disclosures: Y. Song: None. **M.K. Benna:** None.

Poster

P230. Therapeutic Strategies: Preclinical Animal Models: Small Molecule Therapeutics II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P230.01

Topic: C.03. Parkinson's Disease

Support: Sun Pharma Advanced Research Company, Ltd

Title: C-abl inhibitor, k0706/vodobatinib, protects dopaminergic neurons against α-synuclein preformed fibril induced neurodegeneration

Authors: S. S. KARUPPAGOUNDER[1], H. WANG[1], F. AKKENTLI[1], Y. YAMASHITA[2], S. BISEN[2], N. SLOAN[2], B. DANG[2], A. SIGMON[2], H. WOO LEE[2], S. LEE[2], L. WATKINS[3], R. NGUYEN[1], E. KIM[4], S. BRAHMACHARI[1], M. KUMAR[1], S. MANDHANE[5], V. RAMANATHAN[5], N. DAMLE[5], V. L. DAWSON[1], T. M. DAWSON[1];
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Abstract: In Parkinson's disease (PD), oxidative stress contributes to the underlying pathology. Oxidative stress activates c-Abl, a normally auto-inhibited nonreceptor tyrosine kinase, which causes the death of dopamine (DA) neurons in the substantia nigra pars compacta. Activated c-Abl kinase phosphorylates and inhibits parkin ubiquitin function, which leads to accumulation of toxic parkin substrates that contribute to α-synuclein (α-syn) induced neurodegeneration. α-Synuclein misfolding and aggregation play a significant role in progressive neurodegeneration in PD. Genetic inhibition of c-Abl kinase attenuates neurodegeneration in PD models. Thus, modulating the c-Abl-Parkin pathway could be a potential diseasing modifying strategy to prevent or halt the progression of PD. To date, several symptomatic drugs for PD have been developed, but none of them are disease-modifying. K0706, also known as vodobatinib, is a potent orally bioavailable, brain-penetrating small molecule inhibitor of c-Abl. The purpose of this study was to investigate the neuroprotective efficacy of K0706 in the α-syn preformed fibril (PFF) model of PD. K0706 treatment prevents the reduction in dopamine neurons, dopamine levels and the increase in phospho-S129 α-syn and phospho-Y39 α-syn as well as the accompanying behavioral deficits. We also observed a dose-dependent neuroprotective effect of K0706 in this α-syn PFF model of PD. These results suggest that the c-Abl inhibitor, K0706 may be beneficial as a disease modifying therapy in PD. K0706/vodobatinib is currently being evaluated in phase 2 clinical trial, ProSeek, in patients with early stage Parkinson's Disease.

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Poster

P230. Therapeutic Strategies: Preclinical Animal Models: Small Molecule Therapeutics II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P230.02

Topic: C.03. Parkinson's Disease

Support: Michael J Fox Foundation
Inhibikase Therapeutics

Title: Ikt-148009 a c-abl inhibitor: target engagement in parkinson's disease models

Authors: *S. BRAHMACHARI[1], S. S. KARUPPAGOUNDER[2], H. WANG[3], R. NGUYEN[4], S. BISEN[4], Y. YAMSHITA[4], N. SLOAN[4], B. DANG[4], A. SIGMON[4], H. WOO LEE[4], S. MARINO LEE[4], L. WATKINS[5], E. KIM[6], M. KUMAR[3], T. M. DAWSON[7], V. L. DAWSON[7]; [2]Neurol., [1]Johns Hopkins Univ. Sch. of Med., Baltimore, MD; [3]Johns Hopkins Univ., Baltimore, MD; [4]Krieger Sch. of Arts and Sci., Baltimore, MD; [5]Washington & Jefferson Col., Washington, PA; [6]Georgetown Univ., Washington DC, DC; [7]Inst. Cell Engin., Johns Hopkins Univ. Sch. Med., Baltimore, MD

Abstract: c- Abl activation contributes to the pathology of Parkinson's disease (PD). Mitochondrial dysfunction results in oxidative stress, which leads to c-Abl activation, a molecular sensor of oxidative stress. c-Abl phosphorylates α-synuclein (α-syn) at Y39 leads to misfolding of α-syn. Previously, we have shown that Parkin inactivation by c-Abl causes accumulation of pathogenic parkin substrates, PARIS and AIMP2, and subsequently leads to parthanatos-mediated neuronal death. Current therapies focus on reducing the symptoms of the disease, but none of the existing drugs are disease-modifying therapies. Target engagement therapeutic approaches will be beneficial in halting or preventing the progression of PD. Current limitations with small molecule inhibitors are due to poor brain penetration, lack of efficacy, and other adverse effects. In the present study, we evaluated, IkT-148009, a highly potent c-Abl inhibitor, a small-molecule drug designed and engineered to target c-Abl kinase specifically. The primary aim of this current study was to evaluate target engagement of IKT-148009 in the α-syn preformed fibril (PFF) model and the conditional TetP-hA53T α-syn transgenic PD mouse model. The results demonstrate significant inhibition of activated c-Abl in the α-syn PFF and the TetP-hA53T α-syn transgenic PD mouse models, which prevents c-Abl-Parkin mediated demise of dopaminergic neurons. This study provides evidence for the clinical use of, IkT-148009, in the treatment of PD.

Disclosures: S. Brahmachari: None. **S.S. Karuppagounder:** None. **H. Wang:** None. **R. Nguyen:** None. **S. Bisen:** None. **Y. Yamshita:** None. **N. Sloan:** None. **B. Dang:** None. **A. Sigmon:** None. **H. Woo Lee:** None. **S. Marino Lee:** None. **L. Watkins:** None. **E. Kim:** None. **M. Kumar:** None. **T.M. Dawson:** None. **V.L. Dawson:** None.

Poster

P230. Therapeutic Strategies: Preclinical Animal Models: Small Molecule Therapeutics II

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Program #/Poster #: P230.03

Topic: C.03. Parkinson's Disease

Support: NIH NINDS R01 NS109510
R01 NS083845

Title: A brain-penetrant stearoyl-CoA desaturase inhibitor reverses α-synuclein toxicity in synucleinopathy models in vitro and in Parkinson's disease-like mice

Authors: *C. CHUNG[1], S. NUBER[2], D. TARDIFF[1], P. BECHADE[2], T. MCCAFFERY[2], J. CHOI[1], B. CHANG[1,3], W. RAJA[1], E. NEVE[1], C. BURKE[1], X. JIANG[1], P. XU[1], V. KHURANA[1], U. DETTMER[2], K. RHODES[1,4], D. SELKOE[2], R. SCANNEVIN[1,5];
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Abstract: Increasing evidence has shown that Parkinson's disease (PD) impairs midbrain dopaminergic, cortical and other neuronal subtypes in large part due to the build-up of lipid- and vesicle-rich α-synuclein (αSyn) cytotoxic inclusions. We previously identified stearoyl-CoA desaturase (SCD) as a potential therapeutic target for synucleinopathies. A brain-penetrant SCD inhibitor, YTX-7739, was developed and has entered Phase I clinical trials. Here, we report the efficacy of YTX-7739 in reversing pathological αSyn phenotypes in various *in vitro* and *in vivo* model systems. In cell-based assays, YTX-7739 decreased αSyn-mediated neuronal death, reversed the abnormal membrane interaction of amplified E46K (3K) αSyn, and reversed pathological phenotypes in A53T and αSyn triplication patient-derived neurospheres, including dysregulated fatty acid profiles and pS129 αSyn accumulation. In 3K PD-like mice, YTX-7739 readily crossed the blood-brain barrier, decreased unsaturated fatty acids, and prevented the motor behavior deficits and their progressive decline. YTX-7739 administration and decreasing SCD activity through deletion of one copy of the SCD1 gene restored the physiological αSyn tetramer to monomer ratio, dopaminergic integrity, and neuronal survival in 3K αSyn mice. YTX-7739 efficiently reduced pS129+ and PK-resistant in both human wild-type αSyn and mutant 3K mice. Together, these data provide further validation of SCD as a therapeutic target, and YTX-7739 as a clinical candidate, for treating human α-synucleinopathies.

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Yumanity. **P. Bechade:** None. **T. McCaffery:** None. **J. Choi:** A. Employment/Salary (full or part-time):; Yumanity. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Yumanity. **B. Chang:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Yumanity. **W. Raja:** A. Employment/Salary (full or part-time):; Yumanity. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Yumanity. **E. Neve:** A. Employment/Salary (full or part-time):; Yumanity. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Yumanity. **C. Burke:** A. Employment/Salary (full or part-time):; Yumanity. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Yumanity. **X. Jiang:** A. Employment/Salary (full or part-time):; Yumanity. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Yumanity. **P. Xu:** A. Employment/Salary (full or part-time):; Yumanity. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Yumanity. **V. Khurana:** A. Employment/Salary (full or part-time):; Yumanity. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Yumanity. **U. Dettmer:** None. **K. Rhodes:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Yumanity. **D. Selkoe:** F. Consulting Fees (e.g., advisory boards); Prothena. **R. Scannevin:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Yumanity.

Poster

P230. Therapeutic Strategies: Preclinical Animal Models: Small Molecule Therapeutics II

Location: Virtual Only

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Program #/Poster #: P230.04

Topic: C.03. Parkinson's Disease

Support: La Caixa Foundation (LCF/BQ/DI18/1166063)
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JPND-ISCIII (AC20/00121)
MINECO (SAF2016-77541-R)

Title: Gene therapy with VMAT2 reduces age-dependent neuromelanin accumulation and prevents Parkinson's Disease-like motor symptoms in neuromelanin-producing rats

Authors: J. COMPTE, M. GONZÁLEZ-SEPULVEDA, A. NICOLAU, T. CUADROS, J. ROMERO-GIMÉNEZ, A. PARENT, A. LAGUNA, *M. VILA;
Vall d'Hebron Res. Inst., Barcelona, Spain

Abstract: Background: In Parkinson's disease (PD), there is a preferential degeneration of neurons that contain the pigment neuromelanin, especially dopaminergic neurons of the substantia nigra (SN), the loss of which leads to classical motor PD symptoms. We recently developed the first rodent model of human-like neuromelanin production based on the viral vector-mediated nigral expression of melanin-producing enzyme tyrosinase (AAV-hTyr). This has revealed that neuromelanin can trigger PD pathology when accumulated above a specific pathogenic threshold. Because neuromelanin derives from the oxidation of free cytosolic dopamine, we hypothesized that enhancing dopamine vesicular encapsulation with vesicular monoamine transporter 2 (VMAT2) will decrease cytosolic dopamine that can convert to neuromelanin. This approach should slow down the intracellular buildup of neuromelanin that occurs with age and thus prevent, delay or attenuate PD pathology. **Material and methods**: Adult male Sprague-Dawley rats received single unilateral stereotaxic co-injections of AAV-hTyr and AAV-VMAT2 immediately above the SN. Control animals received equivalent amounts of either vehicle, AAV-hTyr or AAV-VMAT2, separately. At selected times post-injection animals were assessed for motor asymmetry and their brains processed for histological analyses. **Results:** VMAT2 overexpression in hTyr-expressing rats reduced intracellular neuromelanin to levels below its pathogenic threshold by lowering the production of dopamine-oxidized neuromelanin precursors. In these animals, reduction of neuromelanin levels was associated with a marked attenuation of Lewy body-like inclusions formation, nigrostriatal degeneration, extracellular neuromelanin accumulation, microglia/macrophage activation, astrocytic presence and PD-like motor deficits. **Conclusions:** Our results demonstrate the feasibility and therapeutic potential of modulating intracellular neuromelanin levels in vivo, thereby opening an unexplored window of potential strategies for PD treatment.

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Poster

P230. Therapeutic Strategies: Preclinical Animal Models: Small Molecule Therapeutics II

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Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P230.05

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Title: CCR3 inhibition reduces recruitment of peripheral immune cells to the eye and brain in inflammatory disease models

Authors: *Y. LOPEZ, *Y. LOPEZ, S. CARYOTAKIS, S. REGE, J. MASUMI, L. AKRAPONGPISAK, A. TEICHERT, S. S. MINAMI, O. DHANDE;
Alkahest, San Carlos, CA

Abstract: Inflammation is a common thread through multiple age-related CNS diseases, appropriately termed inflammaging. One such example is age-related macular degeneration (AMD), the most common cause of irreversible blindness in people over 60 years old. AKST4290, a CCR3 inhibitor, is a promising oral drug for AMD treatment. This is evident in recently completed Phase 2a clinical studies, in which the majority of both treatment-naïve and refractory neovascular AMD (nAMD) subjects demonstrated stable or improved best-corrected visual acuity after 6 weeks of AKST4290 treatment. Preclinical studies using the sodium iodate ($NaIO_3$) induced oxidative stress model were subsequently conducted to examine whether the efficacy observed in nAMD patients was in part due to blocking CCR3-mediated retinal homing of damage-promoting immune cells from the periphery. This in turn could dampen local inflammation and may subsequently modify disease progression. This hypothesis was assessed in 2-month-old male C57BL/6 mice administered $NaIO_3$ and AKST4290 concurrently for 3 days. Flow cytometric analysis of RPE/choroid tissue, the primary site of AMD pathology, illustrates reduced $NaIO_3$-induced infiltration of myeloid, monocyte, neutrophil, and T cell populations with AKST4290. To further elucidate effects of AKST4290 on peripheral T cells, we assessed CCR3 inhibition in another inflammatory model, experimental autoimmune encephalomyelitis (EAE), which is known to have robust T cell infiltration in both the brain and eye. AKST4290 was dosed for 9 days after EAE induction along with a control group that received vehicle only. EAE elicited a significant increase in CD3+ cells in the cerebellum in comparison to control mice. In comparison, AKST4290 significantly reduced this T cell infiltration in the cerebellum. AKST4290 treatment trends towards reduced CD3+ T cells in the retina as well. Together, these data illustrate that CCR3 inhibition modulates immune cell infiltration in both the eye and brain. Furthermore, these data suggest a common mechanistic action of AKST4290 on T cells in inflammatory ophthalmic and neurological disease models.

Disclosures: Y. Lopez: A. Employment/Salary (full or part-time):; Grifols. **S. Caryotakis:** A. Employment/Salary (full or part-time):; Grifols. **S. Rege:** A. Employment/Salary (full or part-time):; Grifols. **J. Masumi:** A. Employment/Salary (full or part-time):; Grifols. **L. Akrapongpisak:** A. Employment/Salary (full or part-time):; Grifols. **A. Teichert:** A. Employment/Salary (full or part-time):; Grifols. **S.S. Minami:** A. Employment/Salary (full or part-time):; Grifols. **O. Dhande:** A. Employment/Salary (full or part-time):; Grifols.

Poster

P230. Therapeutic Strategies: Preclinical Animal Models: Small Molecule Therapeutics II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P230.06

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: PHC Procope 2018
Bayerisch-Französisches Hochschulzentrum/Centre de Coopération Universitaire Franco-Bavarois (BayFrance)

Title: Anti-amnesic and neuroprotective effects of the selective butyrylcholinesterase inhibitor UW-MD-95 in a pharmacological mouse model of Alzheimer's disease

Authors: *T. MAURICE[1], A. CARLES[1], M. HOFFMANN[2], M. SCHEINER[2], L. CROUZIER[1], C. BERTRAND-GADAY[3], A. CHATONNET[3], M. DECKER[2];
[1]MMDN, Univ. Montpellier, EPHE, INSERM, Montpellier, France; [2]Pharmazeutische und Medizinische Chemie, Inst. für Pharmazie und Lebensmittelchemie, Julius-Maximilians-Universität, Würzburg, Germany; [3]DMeM, Univ. Montpellier, INRAE, Montpellier, France

Abstract: Alzheimer's disease (AD) is a devastating dementia characterized by extracellular amyloid-β (Aβ) protein aggregates and intracellular tau protein deposition. Clinically available drugs target acetylcholinesterase (AChE) and indirectly sustain cholinergic neuronal tonus. Butyrylcholinesterase (BChE) also controls ACh turnover and is involved in the formation of Aβ aggregates and maturation of senile plaques. We previously described new carbamate-based drugs, acting as highly potent pseudo-irreversible BChE inhibitors with high selectivity for BChE *vs* AChE and showing promising protective potentials in a pharmacological mouse model of AD [*Hoffman et al., J Med Chem 2019*]. We here characterized the neuroprotective activity of the most promising compound in the series, UW-MD-95, in mice treated intracerebroventricularly with oligomerized $A\beta_{25-35}$ peptide using behavioral, biochemical and immunohistochemical approaches. When injected once, in the 0.3-3 mg/kg dose-range, 30 min before the behavioral tests (spontaneous alternation in the Y-maze, object recognition or passive avoidance), UW-MD-95 showed symptomatic anti-amnesic effects in $A\beta_{25-35}$-treated mice. When injected once-a-day in the same dose-range, the drug prevented the appearance of $A\beta_{25-35}$-induced memory deficits. This effect was lost in BChE knockout mice but observed in their littermate controls. Moreover, the drug repeated treatment prevented $A\beta_{25-35}$-induced oxidative stress (assessed by increased lipid peroxidation or cytochrome c release), neuroinflammation (increased in IL-6 and TNFα levels or GFAP and Iba1 immunoreactivity) in the hippocampus and cortex, and apoptosis (increased Bax level). The novel dual-acting BChE inhibitor UW-MD-95 therefore appeared as a potent neuroprotective drug in the $A\beta_{25-35}$ model of AD, with potentially an impact on Aβ aggregation that could suggest a novel mechanism of neuroprotection.

Disclosures: T. Maurice: None. **A. Carles:** None. **M. Hoffmann:** None. **M. Scheiner:** None. **L. Crouzier:** None. **C. Bertrand-Gaday:** None. **A. Chatonnet:** None. **M. Decker:** None.

Poster

P230. Therapeutic Strategies: Preclinical Animal Models: Small Molecule Therapeutics II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P230.07

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: PIP 2016–2022 n. 0779 (CONICET)
UBACyT 2017–2022 (UBA)
PICD 0031 2016-2022 (FONCyT)

Title: Palmitoylethanolamide prevents neurodevelopmental delay and early hippocampal damage after perinatal asphyxia.

Authors: *M. HERRERA[1], L. UDOVIN[3], T. KOBIEC[2], M. MARTÍNEZ[2], M.-.-M. CAMILA[2], N. TORO-URREGO[4], C. KUSNIER[4], R. KÖLLIKER-FRERS[4], M. OTERO-LOSADA[4], J. LUACES[4], F. CAPANI[4];
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Abstract: Palmitoylethanolamide prevents neurodevelopmental delay and early hippocampal damage after perinatal asphyxia.
Perinatal asphyxia (PA) is a clinical condition brought by a birth temporary oxygen deprivation. This health problem continues to be a morbidity factor in neurodevelopment. Palmitoylethanolamide (PEA) is a neuromodulator well known for its protective effects in neurodevelopmental disorders, such as autism, albeit not deeply studied regarding its particular effects following PA. Recent evidence from our laboratory revealed perinatal treatment with PEA (10 mg/kg) could attenuate cytoskeletal alterations in CA1 hippocampal neurons and improve behavioral outcomes 1 month after PA induction (Herrera et al., 2018). In order to extend these findings, we intended to elucidate whether PEA might exert protective effects earlier on neurodevelopment. Pups (N=75) were tested daily for several reflexes and signs that indicate the maturation of the nervous system. Vulnerability of CA1 neurons was analyzed with Electron Microscopy. Microtubule associated protein-2 (MAP-2) was assessed by Immunohistochemistry and Western Blot at P21. During the first 3 weeks of life, asphyctic rats displayed a significant delay in the appearance of air righting, auditory startle, sensory eyelid, placing and grasp reflexes. A late onset of gait, negative geotaxis and eye opening was also observed. In addition, a slower performance of negative geotaxis was encountered in PA rats from postnatal day 11 (P11) to postnatal day 14 (P14). At P21, ultrastructural signs of neurodegeneration and MAP-2 deficit were found on CA1 hippocampal region. PEA treatment could ameliorate hippocampal damage and reverse the delay in the appearance of gait and motor reflexes. Therefore, PEA appears as a promising neuroprotective agent for the developing injured

brain.
*Herrera MI and Udovin LD have contributed equally to this work.
References: Herrera MI et al. Palmitoylethanolamide Ameliorates Hippocampal Damage and Behavioral Dysfunction After Perinatal Asphyxia in the Immature Rat Brain. Front Neurosci. 2018 Mar 28;12:145.

Disclosures: M. Herrera: None. **L. Udovin:** None. **T. Kobiec:** None. **M. Martínez:** None. **M. Camila:** None. **N. Toro-Urrego:** None. **C. Kusnier:** None. **R. Kölliker-Frers:** None. **M. Otero-Losada:** None. **J. Luaces:** None. **F. Capani:** None.

Poster

P230. Therapeutic Strategies: Preclinical Animal Models: Small Molecule Therapeutics II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P230.08

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Title: The small molecule myeloid cell modulator TQS-168 normalizes the inflammatory phenotype of immune cells from ALS patients and extends survival in the SOD1 G93A ALS mouse model

Authors: *E. CHAVEZ JUAN, D. NESS, B. AJAMI, N. SCHWARTZ, J. HANNESTAD, S. KAKKAR, H. LANCERO;
Tranquis Therapeut., San Mateo, CA

Abstract: Background: Amyotrophic Lateral Sclerosis (ALS) is a rapidly progressing neurodegenerative disease that almost invariably leads to death. While the pathogenesis of ALS is not well understood, myeloid cell dysfunction and inflammation appear to contribute to its progression. To investigate this possibility, we developed TQS-168, a small molecule myeloid-cell modulator, and tested its effects on peripheral blood mononuclear cells from ALS patients, and in the SOD1 G93A model of ALS (a transgenic mouse that overexpresses a mutant form of human *SOD1*).Methods: Blood samples were obtained from ALS patients, PBMCs were isolated, and the ability of TQS-168 to regulate monocyte phenotype and cytokine release were determined ex vivo. SOD1 G93A mice and wildtype littermates were dosed with TQS-168 by oral gavage from the age of 70 days until death. Weight loss and survival were monitored by an experimenter blinded to the treatment condition. Cytokine levels were measured in plasma samples, and muscle fiber number and size were quantified in muscle tissue.Results: TQS-168 modulated monocyte phenotypes and cytokine production in PBMCs from patients with ALS. In the SOD1 G93A model, TQS-168 slowed weight loss and prolonged survival after symptom onset (defined as >3% decrease in body weight and the onset of motor symptoms). Plasma levels of multiple cytokines and monocyte subsets were regulated by TQS-168 in SOD1 G93A mice, and the correlation between changes in motor function and muscle morphometry was

determined.Conclusions: The small molecule TQS-168 regulates the inflammatory phenotype of myeloid cells in patients with ALS and slows disease progression in a mouse model of ALS. Modulation of cytokines by TQS-168 correlated with survival in SOD1 G93A mice. TQS-168 will enter clinical development to evaluate its potential as a treatment for ALS patients.

Disclosures: E. Chavez Juan: None. **D. Ness:** None. **B. Ajami:** None. **N. Schwartz:** None. **J. Hannestad:** None. **S. Kakkar:** None. **H. Lancero:** None.

Poster

P230. Therapeutic Strategies: Preclinical Animal Models: Small Molecule Therapeutics II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P230.09

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: Work was supported by Janssen Pharmaceutical Companies of Johnson & Johnson.

Title: Glial antioxidant response mediated by Keap1-Nrf2 interaction inhibitors is protective in rodent models of Parkinson's disease and neuroinflammation

Authors: *G. B. DAWE[1], Y. HE[2], N. TAYLOR[2], L. FOURGEAUD[2], M. LE[2], S. ALLHANDS[2], K. J. SPILLER[1], R. ARAS[1], E. A. RAMSBURG[1], H. VENKATESAN[2], A. BHATTACHARYA[2];
[1]Janssen Pharmaceut. Companies of Johnson & Johnson, Spring House, PA; [2]Janssen Pharmaceut. Companies of Johnson & Johnson, San Diego, CA

Abstract: Parkinson's disease (PD) is an incurable neurodegenerative disorder characterized by the progressive loss of dopaminergic neurons in the substantia nigra (SN). Impaired mitochondrial function and elevated levels of oxidized biomolecules in the midbrain of PD patients suggest an inability to mitigate oxidative stress may also be critical in disease pathogenesis. To explore this hypothesis further, we activated the Nuclear factor erythroid 2-related factor 2 (Nrf2) antioxidant response pathway in rodent glial cells and disease models, using two small-molecule inhibitors of its interaction with Kelch-like ECH-associated protein 1 (Keap1). In BV-2 cells and primary mouse microglia, pre-treatment with compound A or B suppressed inflammatory cytokine release following lipopolysaccharide (LPS) stimulation. Additionally, primary rat astrocytes exhibited upregulation of Nrf2-regulated antioxidant response element (ARE) genes and increased glutathione production in response to treatment with either compound. Intraperitoneal (IP) injection of compound B (50 mg/kg) in mice yielded upregulation of several ARE genes in brain tissue at 6 and 18 h time points, suggesting sufficient bioavailability for efficacy studies. Subsequent investigation of plasma and brain tissue 4 h after LPS injection (0.8 mg/kg) revealed modest suppression of inflammatory cytokine levels when

animals were injected with compound B prior to the LPS challenge. Moreover, the loss of SN dopaminergic neurons associated with 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) injection was significantly attenuated in mice that were treated with compound B. The treatment group also displayed diminished microglial activation throughout SN tissue in response to MPTP. In conclusion, our data strengthen the notion that enhancement of the Nrf2 signaling pathway may be beneficial in PD and other inflammatory disorders of the nervous system.

Disclosures: G.B. Dawe: A. Employment/Salary (full or part-time):; Janssen. **Y. He:** A. Employment/Salary (full or part-time):; Janssen. **N. Taylor:** A. Employment/Salary (full or part-time):; Janssen. **L. Fourgeaud:** A. Employment/Salary (full or part-time):; Janssen. **M. Le:** A. Employment/Salary (full or part-time):; Janssen. **S. Allhands:** A. Employment/Salary (full or part-time):; Janssen. **K.J. Spiller:** A. Employment/Salary (full or part-time):; Janssen. **R. Aras:** A. Employment/Salary (full or part-time):; Janssen. **E.A. Ramsburg:** A. Employment/Salary (full or part-time):; Spark Therapeutics. **H. Venkatesan:** A. Employment/Salary (full or part-time):; Janssen. **A. Bhattacharya:** A. Employment/Salary (full or part-time):; Janssen.

Poster

P230. Therapeutic Strategies: Preclinical Animal Models: Small Molecule Therapeutics II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P230.10

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Title: Embelin Abrogates Amyloid beta Induced Neurotoxicity With Emphasis on Neuroinflammation, Neurochemistry and p38-MAPK

Authors: *R. ARORA[1], R. DESHMUKH[2];
[1]ISF Col. of Pharm. Moga Punjab India, Moga, India; [2]Pharmaceut. Sci., MRSPTU, Bathinda, India

Abstract: Embelin Abrogates Amyloid-β-Induced Neurotoxicity with Emphasis on Neuroinflammation, Neurochemistry and p38-MAPK Rimpi Arora*, and Rahul Deshmukh*1.Research Scholar, IKGPTU, Jalandhar, Punjab, India2.Neuropharmacology Division, Department of Pharmacology, I. S. F. College of Pharmacy, Moga- 142001, Punjab, India3.Department of Pharmaceutical Sciences & Technology, Maharaja Ranjit Singh Punjab Technical University, Bathinda-151001, Punjab, India
Abstract Background: Stress activated protein kinases such as p38 MAPK have been implicated in the pathogenesis of AD. Extracellular deposits of amyloid-β (Aβ) in neuronal synapse via p38MAPK have been considered as a major hallmark of Alzheimer's disease (AD). **Purpose:** Here in we have investigated the neuroprotective potential of embelin in Aβ-42 induced neurotoxicity in rats. **Material and Methods:** $A\beta_{1-42}$ oligomer was infused (3nmol/3µl) intracerebroventrically twice on day-1to induced Alzheimer's type dementia in rats. Spatial and

non-spatial memory was assessed at different time intervals and terminally biochemical, neurochemical and neuroinflammatory parameters were determined in rat hippocampal brain tissue. One week following $A\beta_{1-42}$ infused rats were treated with different doses of embelin (EMB) (2.5, 5 and 10mg/kg/p.o.) till 21st day. **Results:** $A\beta_{1-42}$ infusion upregulated p38 MAPK consequently significant deterioration in learning and memory while hippocampal tissue showed elevation in AChE activity, oxidative stress, and pro-inflammatory cytokine levels (TNF-α, IL-β etc.) and disturbed pattern of catecholamines and GABA/glutamate levels. On the other side, EMB significantly attenuated $A\beta_{1-42}$ induced cognitive deficits & other biochemical changes in rats. EMB treated rats showed improved learning and memory as well as able to reduce the burden of hippocampal oxidative stress and pro-inflammatory cytokines and restored catecholamines and GABA/glutamate balance in rats. **Conclusion and Implication:** The pro-cognitive effect of EMB may be due to its antioxidant and anti-inflammatory actions through p38MAPK downregulation. The observed results indicate the therapeutic potential of EMB in cognitive disorders. **Key words:** Alzheimer's disease; Embelin; Amyloid-β; Neuroprotection; Dementia; Cognitive deficit.

Disclosures: R. Arora: None. **R. Deshmukh:** None.

Poster

P241. Frontotemporal, Vascular, and Other Non-Alzheimer Dementias I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P241.01

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R01 NS084817
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Title: Methamphetamine self-administration causes neuronal dysfunction in rat medial prefrontal cortex in a sex-specific and withdrawal time-dependent manner

Authors: *L. CHEN, S. L. CASSODAY, A. DONNER, N. CHOUDHURY, L. AL-HARTHI, X.-T. HU;
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Abstract: Methamphetamine (Meth) is a highly addictive and widely abused psychostimulant. There is no FDA-approved medicine for treating people with Meth use disorders (MUD). Although female and male Meth users are reportedly taking similar quantity of Meth, woman

users appear to suffer greater consequential dependence, and experience more frequent relapse during the abstinence. People with MUD also show sex difference in the severity of Meth-caused cognitive deficits/structural changes in the brain. The mechanism underlying such sex difference is unknown; and has rarely been studied. The medial prefrontal cortex (mPFC), one of the key regulators of cognition and addiction, is profoundly altered by Meth; but the mechanism by which Meth causes dysfunction of mPFC pyramidal neurons is not fully understood. To fill these knowledge gaps, we assessed drug-taking/drug-seeking behaviors and associated mPFC neuron activity in male and female adult rats after Meth self-administration (SA) followed by different withdrawal periods. Male and female F344 rats at age of 4-5 months old were trained to self-administer (SA) Meth (0.01mg/kg/infusion) for 1 week, and then 0.05mg/kg/infusion for another 2 weeks. A subset of rats was tested for drug-seeking behaviors on withdrawal day 2, 3, 9, 16, 23 and 30, respectively. Age-matched saline (SAL)-yoked rats were used as control. Brain slices containing the mPFC were prepared for whole-cell patch-clamp recording after 1-5 days (d, short-term) or 31-38d (long-term) withdrawal from the last Meth-SA. We found that both males and females showed similar drug-taking, total Meth intake and persisting drug-seeking behaviors during various withdrawal periods. Meanwhile, mPFC neuron firing was significantly decreased in Meth-SA male rats after a short-term withdrawal, but increased after a long-term withdrawal, compared to SAL-yoked controls. We also found that similar to the altered firing, evoked Ca^{2+} plateau potentials (indicating Ca^{2+} influx via voltage-gated Ca^{2+} channels) were also significantly decreased in these neurons from Meth-SA male rats after a short-term withdrawal, but increased in them after a long-term withdrawal, compared to SAL-yoked controls. In contrast, there was no significant change in firing or Ca^{2+} spikes of mPFC neurons from Meth-SA female rats, regardless of the withdrawal times. Collectively, our novel findings demonstrate that chronic Meth-induced mPFC neuron dysfunction is sex-specific and withdrawal time-dependent. They also suggest that a delayed response to Meth-induced mPFC neuronal dysfunction could drive females for longer period of Meth-SA that could cause more severe consequences.

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Poster

P241. Frontotemporal, Vascular, and Other Non-Alzheimer Dementias I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P241.02

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant 3R01NS091299-05S1

Title: Tdp-43 expression in dementia-relevant circuits causes axonal degeneration and behavioral deficits in Drosophila

Authors: *R. KEATING GODFREY[1], R. T. BJORK[2], C. WILLIAMS[1], G. HALA'UFIA[3], H. B. COWELL[1], E. M. LEHMKUHL[1], E. ALSOP[4], K. VAN KEUREN-JENSEN[4], D. C. ZARNESCU[1];
[1]Mol. and Cell. Biol., [2]Grad. Interdisciplinary Program in Neurosci., [3]Sch. of Mind, Brain, and Behavior, Univ. of Arizona, Tucson, AZ; [4]TGen, Phoenix, AZ

Abstract: Trans active response (TAR) DNA-Binding protein 43 (TDP-43), an evolutionarily conserved RNA/DNA binding protein that regulates RNA processing, is known to form pathological, cytoplasmic inclusions in numerous neurodegenerative diseases. First identified in ubiquitin-positive inclusions in amyotrophic lateral sclerosis (ALS) and frontotemporal lobar degeneration (FTLD) and more recently in Alzheimer's disease (AD), TDP-43 appears to be a common molecular link among central nervous system neurodegenerative diseases. Given our success modeling TDP-43 proteinopathy in a fly model of ALS, we have developed a novel fly model of dementia by overexpressing human TDP-43 in the mushroom bodies (MBs) of the *Drosophila* brain. Insect MBs form an elaborate, associative network that overlaps in function and gene expression with neurons in the human hippocampus and frontal cortex, making it an appropriate brain structure to model human cognitive disease. We evaluate the effects of TDP-43 proteinopathy in MBs by measuring changes in TDP-43 localization in MB neurons, neurodegeneration in the MB axons, and behavior deficits in working memory and sleep. We find that TDP-43 is mislocalized from the nucleus to the cytoplasm and forms axonal inclusions. This results in both behavioral deficits and age-dependent axonal degeneration in MBs. Importantly, cognitive deficits in working memory and sleep appear prior to our ability to detect axonal degeneration, paralleling what is seen in human disease. Additionally, we sought to identify the molecular targets of TDP-43 in the MBs using RNA immunoprecipitations and RNAseq. These experiments identified several candidate targets of TDP-43 in mushroom bodies, including Dally-like protein (Dlp/GPC6), a translational target that we recently reported in *Drosophila* sALS models and patients. These findings suggest that TDP-43 overexpression in MBs causes FTD-relevant phenotypes that may help uncover novel mechanisms of disease. Future experiments are aimed at identifying additional FTD-relevant targets of TDP-43, including novel therapeutic strategies.

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Poster

P241. Frontotemporal, Vascular, and Other Non-Alzheimer Dementias I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P241.03

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH R01 AG063945
NIH R21 AG059223
AARG-17-533363

Title: Functional role of PLD1 signalosome in Frontotemporal dementia

Authors: *C. NATARAJAN[1], B. THOMPSON[2], B. KRISHNAN[1];
[1]Neurol., [2]UTMB, Galveston, TX

Abstract: Background: Phospholipase D (PLD), a lipolytic enzyme for membrane phospholipids, exists in two isoforms of mammalian phosphatidyl choline (PC) specific PLD; inducible PC-PLD1 and constitutive PC-PLD2. Elevated PLD activity is well documented in clinical studies involving Alzheimer's disease (AD). Our lab was the first to show that elevated PLD1 facilitates synaptic dysfunction and underlying memory deficits driven by amyloidogenic insults by Aß/tau. We observed preservation of memory in response to PLD1 specific small molecule inhibitor in transgenic mouse models and documented preservation of dendritic spines as an underlying basis. In this study, we explored a role for PLD1 in Frontotemporal Dementia in an attempt to explore its pathological role in tauopathy-driven dementia. **Methods:** We studied altered expression/activity of key F-actin signaling elements common to PLD1 signalosome in synaptosomal fractions of human post-mortem FTD brain regions. Synaptic deficits associated with elevated PLD1 expression in human FTD samples was studied by Fluorescence Analysis of Single Synapse Long Term Potentiation (FASS-LTP). Using immunofluorescence, we studied the association of PLD1 with tau, presynaptic, and post-synaptic receptors. **Results:** Elevated PLD1 expression in synaptosomal fractions of post-mortem FTD brain tissues was corroborated by synaptic deficits and association of PLD1 with postsynaptic density protein (PSD95). FASS-LTP assay indicated relatively lower long-term potentiation in synaptosomal fractions of FTD brains as compared to the control brains implying synaptic deficits in FTD subjects. **Conclusion**: In the light of these observations, we conclude elevated PLD1 expression is associated with toxicity of tau causing memory deficits in FTD patients. This PLD1-tau interaction alters PLD signaling partners such as mTOR (mammalian target of rapamycin), cofilin and phosphocofilin. This would implicate the actin-dependent remodeling of synapse to be a key step in the role of PLD1 signalosome in FTD. Delineating the effects of PLD1 signalosome in tauopathies such as FTD will hugely be beneficial in tau-targeting therapeutics for a multipronged approach in the treatment of neurodegenerative disorders.

Disclosures: C. Natarajan: None. **B. Thompson:** None. **B. Krishnan:** None.

Poster

P241. Frontotemporal, Vascular, and Other Non-Alzheimer Dementias I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P241.04

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant AR-067667

Title: 1,3-butanediol ameliorates high-salt diet induced vascular dementia via mirna-histone acetylation

Authors: J. BEHERA, M. SINGH, ***N. TYAGI**;
Univ. of Louisville, Louisville, KY

Abstract: Progressive decline in cognitive function, and memory is on the rise and in fact parallels to that of the hypertension increase in aging humans. The mechanistic basis of high-salt diet (HSD) induced hypertension-associated memory impairment, and dementia pathology is not yet fully understood. Furthermore, it is also currently unknown whether ketone bodies are secreted by brain cells or whether it is responsible for the memory as well. Here, we examined the impact of ketone bodies or the precursor molecule (1,3-butanediol) supplementation on cognitive function through the histone acetylation in the hypertensive mice. To test the hypothesis, we employed adult female wild-type (WT) mice. The mice were fed with HSD (8%), and HSD mice were supplemented with 1,3-Butanediol (2%), and the HSD mice were supplemented with β-hydroxybutyrate (BHB, 0.2%) in the drinking water for 12 weeks. CODA noninvasive BP system (tail-cuff) testing revealed that 1,3-Butanediol or BHB reduced the systolic, and diastolic blood pressure significantly in the hypertensive mice. The results show that 1,3-Butanediol or BHB treatment of hypertensive mice reduced systemic level of the circulating miRNA-128-3p in comparison to the hypertensive mice. Further, the data analysis revealed higher behavioral proficiency, and more robust long-term, and short-term memory in HSD+BHB or HSD+ 1,3-Butanediol mice as compared to the hypertensive mice. This mechanistic study shows that HSD+ 1,3-Butanediol mice display an increased mitochondrial TFAM dependent upregulation of brain derived neurotropic factor (BDNF), and nuclear receptor subfamily 4A (Nr4a) in the mouse hippocampus most likely via an epigenetic activation of the H3K27ac acetylation at the promoters. Hippocampal overexpression of BDNF/Nr4a by Lentiviral BDNF/Nr4a administration rescued the hypertensive-memory impairment. In addition, the anti-miR-128-3p treatment restored the neuroprotective function and reduced the dementia pathology in the hypertensive mice. Taken together, our findings suggest that 1,3-Butanediol or BHB treatment increases the ketone bodies production, and that subsequently improves the cognitive function in the hypertensive mice. Acknowledgment: Financial support from National Institute of Health grant AR-067667.

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Poster

P241. Frontotemporal, Vascular, and Other Non-Alzheimer Dementias I

Location: Virtual Only

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Program #/Poster #: P241.05

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: R01DA0462582 SLChang

Title: Meta-analysis on the molecular mechanisms underlying nicotine modulation of HIV-associated dementia

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Abstract: HIV-Associated Dementia (HAD) is a significant comorbidity that many HIV patients face. Our study utilized the Ingenuity Pathway Analysis (IPA) license from QIAGEN to identify and analyze molecular profiles and pathways underlying nicotine's impact on HAD pathology. The Qiagen Knowledge Base (QKB) defines HAD as "Dementia associated with acquired immunodeficiency syndrome (disorder)." Although much remains unknown about HAD pathology, the curated research findings from the QKB shows 5 upregulated molecules that are associated with HAD+: CCL2 (Chemokine (C-C motif) ligand 2), L-glutamic acid, GLS (Glutaminase), POLG (DNA polymerase subunit gamma), and POLB (DNA polymerase subunit beta). The current study focused on these 5 HAD pathology molecules as the phenotype of interest. The Pathway Explorer tool of IPA was used to connect nicotine-associated molecules with the 5 HAD-associated molecules (HAD pathology molecules) by connecting 29 overlapping molecules (including transcription regulators, cytokines, kinases, and other enzymes/proteins). The Molecule-Activity-Predictor (MAP) tool predicted nicotine-induced activation of the HAD pathology molecules indicating the exacerbation of HAD. However, alternative pathways with more holistic representations of molecular relationships revealed the potential of nicotine as a neuroprotective treatment. It was found that concurrent with nicotine treatment the individual inactivation of several of the intermediary molecules in the holistic pathways caused the downregulation of the HAD pathology molecules. These findings predict that nicotine may have therapeutic properties for HAD when given alongside specific inhibitory drugs for one or more of the identified intermediary molecules.

Disclosures: V. Krishnan: None. **S.L. Chang:** None. **M. Vigorito:** None. **N.K. Kota:** None.

Poster

P241. Frontotemporal, Vascular, and Other Non-Alzheimer Dementias I

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Program #/Poster #: P241.06

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: National Institute on Aging Grant R01AG041762
National Institute on Aging Grant P01AG019724
National Institute on Aging Grant F31AG059378

Title: I Think, Therefore You Feel: Neuropsychological Correlates of Emotion Perception In Neurodegenerative Diseases

Authors: *S. YOKOYAMA[1], Y. CHEN[2], N. Y. XU[2], C. L. BROWN[2,1], A. Y. HUA[2,1], J. H. KRAMER[1], R. W. LEVENSON[2];
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Abstract: When we accurately perceive others' emotions, we engage a variety of cognitive skills. Neurodegenerative diseases lead to deficits in cognitive functioning that could disrupt the ability to perceive others' emotions accurately. In the current study, we explored how cognitive skills (i.e., executive functions and word knowledge) relate to the ability to label and track others' emotions. We recruited 87 participants (59% male; aged 34 to 80), including 63 individuals with frontotemporal dementia and 24 healthy controls, who underwent neuropsychological testing. Three aspects of executive functioning were assessed: (a) working memory, (b) task-switching, and (c) inhibition, using digits backwards, trail making, and the Stroop Task, respectively. Word knowledge was assessed using the Peabody Picture Vocabulary Test. Additionally, participants completed two laboratory tasks assessing different facets of emotion perception: (a) an emotion labeling task that measures the ability to identify specific emotions (e.g., happy, sad) experienced by characters in films, and (b) an emotion tracking task that measures the ability to continuously track the emotional valence of a character in a film using a rating dial. Regression analyses of the three aspects of executive functioning revealed: (a) better working memory (digits backwards) was associated with better performance on both emotion perception tasks, (b) task switching (trails) was not associated with performance on either emotion perception task, and (c) better inhibition (Stroop) was associated with better emotion tracking, but not emotion labeling. In contrast, greater word knowledge was associated with better emotion labeling, but not emotion tracking. All results remained consistent after controlling for participants' age and sex. Findings suggest that the ability to inhibit focus from non-target cues (as measured by the Stroop Task) is important for accurately perceiving others' emotional valence. Conversely, greater word knowledge (as measured by the Peabody Picture Vocabulary Test) appears to be related to the ability to label others' emotions, perhaps reflecting the important role that semantic knowledge plays in emotion labeling.

Disclosures: S. Yokoyama: None. **Y. Chen:** None. **N.Y. Xu:** None. **C.L. Brown:** None. **A.Y. Hua:** None. **J.H. Kramer:** None. **R.W. Levenson:** None.

Poster

P241. Frontotemporal, Vascular, and Other Non-Alzheimer Dementias I

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Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R01NS105971
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The Bluefield Project to Cure Frontotemporal Dementia
BrightFocus Foundation

Title: The granulin linker domain is necessary for proper folding, trafficking, and rescue of lysosome dysfunction: Implications for treatment of Frontotemporal Dementia and Alzheimer's disease.

Authors: *P. MERINO, J. ROOT, G. TAYLOR, L. TROIANO ARAU, T. KUKAR;
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Abstract: Decreased levels of progranulin (PGRN) cause frontotemporal dementia (FTD), neuronal ceroid lipofuscinosis (NCL) and are a risk factor for Alzheimer's disease and Parkinson's disease. PGRN is a glycoprotein composed of 7.5 tandem domains called granulins (GRNs). PGRN can be cleaved by proteases to release individual GRNs. Unfortunately, the precise function of PGRN and GRNs is unclear. Complete deficiency of PGRN/GRNs causes NCL, lysosomal dysfunction, lipofuscin accumulation, neuroinflammation, and ultimately neurodegeneration. Thus, we hypothesized that the primary function of PGRN is in the lysosome. In support of this idea, we and others find that PGRN is predominantly cleaved into GRNs by cathepsins in the lysosome. However, the lysosomal function of PGRN/GRNs is still unresolved. Moreover, there are conflicting reports whether GRNs are benign, beneficial, or detrimental to cellular health and function. To resolve these questions, we focused on understanding the folding and function of GRNs in the lysosome. In this project, we dissected the contribution of the intervening peptide linkers (L1-8) between each granulin domain (GRN 1-7) in human PGRN. We find that the peptide linkers play an important role in folding and structural stability of GRNs. Briefly, transient transfection of GRN2 followed by linker 3 (GRN2+L3) leads to the secretion of a well-folded protein. In contrast, GRN2 lacking linker 3 (GRN2-ΔL3) is expressed but secreted less efficiently and forms higher-order oligomers. Further, GRN2+L3 is internalized and trafficked to the lysosome when administered to cells in the media. In contrast, GRN2-ΔL3 does not traffic to the lysosome and is unable to rescue lysosomal dysfunction in PGRN-deficient cells. We investigated this phenomenon in more detail by making a series of GRN2 constructs with mutated linker sequences. Using this strategy, we aimed to determine if any additional peptide sequence can substitute for linker 3, or if specific amino acids are necessary for proper folding, trafficking, lysosome function, and cellular toxicity. Our data reveal that the presence of the linker is necessary for the folding of GRN2, and the specific sequence of linker 3 is important for the proper folding of the protein**.** Taken together, our results provide evidence that GRN linker sequences are necessary for protein folding and function. Most importantly, our data suggest GRNs themselves are the bioactive

component of PGRN and are sufficient to maintain lysosome function and homeostasis and are potential therapeutics to treat FTD caused by *GRN* mutations.

Disclosures: P. Merino: None. **J. Root:** None. **G. Taylor:** None. **L. Troiano Arau:** None. **T. Kukar:** None.

Poster

P241. Frontotemporal, Vascular, and Other Non-Alzheimer Dementias I

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Program #/Poster #: P241.08

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NINDS R01-NS121125-01

Title: Evaluating the role of PABPC4 as a potential novel modifier of TDP-43 pathology in FTLD-TDP

Authors: *U. SHETH[1,2], N. FINCH[1], M. BAKER[1], N. R. GRAFF-RADFORD[3], B. F. BOEVE[4], R. PETERSEN[4], D. S. KNOPMAN[4], K. A. JOSEPHS[4], B. E. OSARKSSON[3], R. RADEMAKERS[1], D. W. DICKSON[1,5], Y. ZHANG[1], L. PETRUCELLI[1,2], M. M. VAN BLITTERSWIJK[1,2], T. F. GENDRON[1,2];
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Abstract: Frontotemporal dementia (FTD), the second most common form of dementia affecting people under the age of 65, is characterized by atrophy of the frontal and temporal lobes. Frontotemporal lobar degeneration (FTLD) is used to denote its pathological diagnosis. Approximately 50% of all FTLD and 95% of all Amyotrophic Lateral Sclerosis (ALS) cases are characterized by the abnormal cleavage, phosphorylation and aggregation of TDP-43, a DNA and RNA binding protein, highlighting pathological overlap between these diseases. Thus, reducing accumulation of toxic TDP-43 presents a promising therapeutic approach. Our exciting preliminary data show that polyadenylate-binding protein 4 (PABPC4) reduces accumulation of pathological TDP-43. Through transcriptomic studies, we discovered that higher levels of frontal cortex *PABPC4* mRNA correlate with greater survival in patients with FTLD. Further, we found a significant inverse correlation between frontal cortex *PABPC4* and phosphorylated TDP-43 (pTDP-43). It is thus noteworthy that PABPC4, a predominantly cytoplasmic protein involved in regulating mRNA stability, was reported to co-aggregate with TDP-43. To examine the effect of PABPC4 on pathological TDP-43, we knocked down or overexpressed PABPC4 in cultured cells overexpressing various TDP-43 species. We noted increased pTDP-43 when PABPC4 was knocked down and, conversely, decreased pTDP-43 when PABPC4 was overexpressed. These findings are congruent with the inverse relationship between *PABPC4* and pTDP-43

accumulation in the frontal cortex of FTLD cases. These early studies uncover PABPC4 as a potential suppressor of toxic TDP-43 accumulation. Studies are underway to define the mechanisms through which this occurs, uncovering the significance of this relationship in post-mortem human tissue, and the involvement of other proteins in the PABP family.

Disclosures: U. Sheth: None. **N. Finch:** None. **M. Baker:** None. **N.R. Graff-Radford:** None. **B.F. Boeve:** None. **R. Petersen:** None. **D.S. Knopman:** None. **K.A. Josephs:** None. **B.E. Osarksson:** None. **R. Rademakers:** None. **D.W. Dickson:** None. **Y. Zhang:** None. **L. Petrucelli:** None. **M.M. Van Blitterswijk:** None. **T.F. Gendron:** None.

Poster

P241. Frontotemporal, Vascular, and Other Non-Alzheimer Dementias I

Location: Virtual Only

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Program #/Poster #: P241.09

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NMRC/OFIRG/0036/2017
MOE2017-T3-1-002

Title: Intermittent fasting: A potential modifier of DNA methylation landscape in a chronic cerebral hypoperfusion mouse model of Vascular Dementia

Authors: *S. SELVARAJI[1,2,3], M. EFTHYMIOS[4], S. ROGER[5,6], K. LIM[7], K. MITCHELL[3], C. CHEN[3,8], T. ARUMUGAM[10,11,9];
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Abstract: Vascular dementia (VaD), the second most common cause of dementia worldwide, is a progressive neurodegenerative disorder that leads to cognitive decline. It is characterised by reduced cerebral blood flow to the brain (cerebral hypoperfusion), white matter hyperintensities and increased blood-brain barrier permeability. With age being a high-risk factor for VaD, coupled with a rapidly aging population, the number of VaD cases are on the rise. With a rapid step-wise decline in disease progression, the associated mortality is high, thus emphasising the

need to tackle the issue at the soonest. Further, current treatments for VaD patients are purely symptomatic. The unknown pathophysiology of VaD underpins the difficulty of targeting the root cause of the problem. While clinicopathological considerations have been made in the research of VaD, molecular mechanisms of the disease manifestation remain largely unexplored. Given the lifestyle-associated risk factors for VaD such as diabetes and hypertension, the environment seems to play a key role in modulation of gene expression. The gene-environment interaction, in the form of epigenetic alterations, could thus perhaps explain the underlying mechanistic changes observed in the pathophysiology of VaD. This study thus aims to investigate one such epigenetic alteration in a chronic cerebral hypoperfusion (CCH) mouse model - DNA methylation. A representative VaD mouse model was established using the bilateral common carotid artery stenosis (BCAS) method that involves surgically inserting micro-coils in C57BL/6 male mice. The coils were left in the mice for different timepoints of 7, 15 and 30 days respectively to study the temporal differences in the DNA methylation landscape. Additionally, as a nexus between the gene-environment interaction, intermittent fasting (IF), which involves time restricted feeding, was introduced as a prophylactic intervention. Hence, a parallel group of mice with the same timepoints were fasted for 16 hours every day for a period of 4 months before BCAS surgery was performed. At the end of the respective timepoints, the mice were sacrificed, and the DNA extracted from the cerebral cortex were sent for DNA methylation sequencing. This study, for the first time, shows that BCAS-induced CCH altered the DNA methylation landscape compared to the controls. Interestingly, with IF, the DNA methylation landscape was re-modelled and showed signs of reversal of the methylation status. IF ameliorated cognitive impairments induced under CCH and attenuated the neuropathological alterations as well. Therefore, IF shows promise in offering potential beneficial effects under CCH.

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Poster

P249. Parkinson's Disease: Cellular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P249.01

Topic: C.03. Parkinson’s Disease

Support: NIH grant R01 NS105783

Title: Non-canonical role of PINK1 in modulating PKA function and dendrite outgrowth.

Authors: *S. K. SOMAN, R. K. DAGDA;
Pharmacol., Univ. of Nevada Reno, Reno, NV

Abstract: Background: Over more than 40 loss of function mutations in PTEN-induced Kinase 1 (PINK1) causes early-onset Parkinson's disease (PD) in humans. In the mitochondrion, full-length PINK1 is a serine-threonine kinase with a predominant role in enhancing mitophagy in oxidatively stressed neurons. However, emerging evidence suggests an extra-mitochondrial role of post-translationally processed, cleaved PINK1 (c-PINK1) in promoting neuronal development and plasticity. Like c-PINK1, Protein Kinase A (PKA) activity is vital for neuronal physiology, and *in vitro* and *in vivo* models of PD have shown that a deficiency in PKA function contributes to PD pathology. Our previous studies have shown that c-PINK1 and PKA interact to modulate neuronal plasticity by stabilizing dendritic networks through the extracellular release of Brain-Derived Neurotrophic Factor (BDNF). **Problem:** *The molecular mechanism(s) and spatio-temporal dynamics by which PINK1 promotes PKA-mediated dendrite outgrowth remains to be determined*. **Methods:** Primary Cortical Neurons (PCNs), prepared from 14 to 15-day C57BL/6 and PINK1-KO mouse embryos, were used as an experimental model. Kinetin (50µM) was used to activate endogenous PINK1 pharmacologically. Western blotting and ELISA were used to measure PKA subunit expression and phosphorylation patterns. Statistical analysis was performed with GraphPad Prism 5.0. Results are expressed as mean ±SEM from three independent experiments. The data analysis was performed by the experimentalist blind to experimental groups. One-way ANOVA with Bonferroni's post-hoc correction or Student's t-test were used as appropriate. **Results:** A significant reduction in the endogenous level and activation status of various subunits of the PKA holoenzyme was observed in PINK1-KO primary cortical neurons (PCN's) compared to WT-PCN's, suggesting that a loss of PINK1 function leads to reduced global neuroprotective PKA activity. In addition, in *vitro* PKA activity assays suggest that PINK1-mediated activation of PKA occurs within 8 hrs. and plateaus by 12 hours following treatment of PCNs with kinetin. Mechanistically, c-PINK1 enhances the autocatalytic-mediated activation of PKA and reduces the activation threshold by phosphorylating Type II regulatory subunits of PKA. To further elucidate the correlation between PINK1 activity and PKA function in dendrites, FRET-based PKA sensors were employed to analyze the spatio-temporal dynamics of PKA activation by PINK1 in dendrites of PCNs. **Conclusion:** Overall, our study demonstrates that PINK1 modulates PKA function and highlights the extra-mitochondrial and neuroprotective roles of PINK1 in neurons.

Disclosures: S. K. Soman: None. **R. K. Dagda:** None.

Poster

P249. Parkinson's Disease: Cellular Mechanisms I

Location: Virtual Only

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Program #/Poster #: P249.02

Topic: C.03. Parkinson's Disease

Support: IBRO Travel Grant

CAPES
FAPERJ
CNPq

Title: Layer-specific enteric glial cell response and gut immune events during the neurodegenerative process in the mouse model of Parkinson's disease induced by 6-hydroxydopamine

Authors: *B. THOMASI[1], L. VALDETARO[1], L. S. HAYASHIDE[1], A. M. N. FERNANDES[1], A. MUSSAUER[1], M. L. SILVA[1], M. RICCIARDI[1], N. ROCHA[1], A. C. FARIA-MELIBEU[1], M. L. RIBEIRO[1], P. CAMPELLO-COSTA[1], V. MOURA-NETO[2], A. TAVARES-GOMES[1]; [1]Univ. Federal Fluminense, Niterói, Brazil; [2]Univ. Federal do Rio de Janeiro, Rio de Janeiro, Brazil

Abstract: Enteric glial cells (EGCs) constitute the majority of the neural population of the enteric nervous system and are found in all layers of the gastrointestinal tract (GIT). They are active in enteric functions such as immunomodulation, as evidenced by the regulation of the intestinal epithelial barrier (IEB). Both EGCs and IEB have been described as altered in Parkinson's disease (PD). Our previous results working with the mouse model of PD induced by 6-hydroxydopamine (6-OHDA) showed a colonic inflammatory response marked by increased IBA1 and GFAP immunoreactivity in the colonic mucosa. Our goal was to explore colonic histology, EGCs, and immune components in the GIT layers during the neurodegenerative process induced by 6-OHDA. C57Bl6 male mice were unilaterally injected with 6-OHDA in the striatum and another group of animals operated was used as controls. Animals were submitted to an indirect motility assay through feces water analysis. Colon tissue was extracted for different analyses at 48h, one week (1w), and two weeks (2w) post-lesion. Through H&E staining and immunofluorescence, we evaluated the myenteric ganglia. For biochemical analyses, the neuromuscular and mucosal layers were microdissected to Western blotting. 6-OHDA injected animals present less water percentage in their feces only at 1w and 2w post-lesion ($n=7$, $p\leq0.05$), pointing to dysmotility. At 48h post-lesion, GFAP and p-p38MAPK expression increased in the neuromuscular layer ($n=5$ and $n=3$, respectively, $p\leq0.05$) meanwhile no modifications in TNF-α, GDNF, and occludin expression in the mucosal layer. 1w post-induction, myenteric ganglia area ($n=3$, $p\leq0.05$) and neuromuscular GFAP content increased ($n=5$, $p\leq0.05$) as well as mucosal TNF-α ($n= 4$, $p\leq0.001$), GDNF ($n=6$, $p\leq0.05$), and occludin expression ($n=4$, $p\leq0.05$). Also, the intraganglionic glia presented an altered morphology with an ameboid profile and less S100β immunolabeling. 2w after model induction, ganglia area were still increased ($n=3$, $p\leq0.05$) with less S100β staining and occludin expression in the 6-OHDA group ($n=3$, $p\leq0.05$). At 48h time after neurodegeneration induction, we detected an increase in GFAP in the neuromuscular layer, suggesting that it may be a primary event for the upregulation of GDNF, TNF-α, and occludin in the intestinal mucosa. In addition, EG seems to elicit a plastic molecular response through time. EGCs are a complex and plastic cell type that regulates immune interactions in the gut, contributing to inflammatory responses. So, EG modulation may be an early enteric sign induced by parkinsonian neurodegeneration followed by inflammatory, dysmotility signs, and IEB structural plasticity.

Disclosures: B. Thomasi: None. **L. Valdetaro:** None. **L.S. Hayashide:** None. **A.M.N. Fernandes:** None. **A. Mussauer:** None. **M.L. Silva:** None. **M. Ricciardi:** None. **N. Rocha:** None. **A.C. Faria-Melibeu:** None. **M.L. Ribeiro:** None. **P. Campello-Costa:** None. **V. Moura-Neto:** None. **A. Tavares-Gomes:** None.

Poster

P249. Parkinson's Disease: Cellular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P249.03

Topic: C.03. Parkinson's Disease

Title: Death-associated protein kinase (DAPK1) and its role in a 6-OHDA mouse model of Parkinson's disease.

Authors: *M. SANTORO, A. T. ADAM, R. K. LAM, C. BACHMEIER, M. SHAMLOO; Neurosurg., Stanford Univ., Palo Alto, CA

Abstract: Death-associated protein kinase (DAPK1) belongs to a large family of Ca^{2+}/calmoduline regulated serine/threonine kinases. Its cellular functions entail regulation of the cell cycle, autophagy, phagocytosis, and apoptosis. Mounting evidence suggests the involvement of DAPK1 in neurodegenerative diseases due to its dysregulated activity linked to Ca^{2+}/glutamate neurotoxicity and the initiation of pro-apoptotic cell pathways. In addition, a strong link between variants of DAPK1 and the incidence risk of Alzheimer's disease has been reported in several studies. To date, the role of DAPK1 in Parkinson's disease (PD) is poorly understood. In light of the pivotal roles of DAPK1 on cell survival and cell death, we hypothesize that dysregulation of DAPK1 activity in the aged brain contributes to the apoptotic cell demise of nigrostriatal dopaminergic neurons observed in PD patients. To answer our questions, we identified a lead molecule with selective inhibitory activity against DAPK1. Three analog compounds were synthesized and selected based on their IC_{50} for *in-vitro* screening. *In-vivo* experiments relied on male C57BL/6J mice aged 4 and 19 months. In addition, male mice C57BL/6J mice 12 weeks old were stereotaxically infused in the striatum with 2 µl of a 0.05 % solution of ascorbic acid containing 10 µgr of 6-OHDA per µl. We observed DAPK1 protein levels significantly down-regulated in the hippocampus of aged mice (55.6 % ± 24.9). Oppositely, protein levels of the inactive form of DAPK1 (phosphorylated on the serine 308) are increased against total DAPK1 (n=10 per group, $p < 0.05$). A similar trend was observed in the 6-OHDA treated mice with total protein levels of DAPK1 decreased. *In-vitro* assessment of toxicology and neuroprotective effects of our three novel DAPK1 inhibitors showed high tolerability of our analogs in human microglial cells and rat cortical neurons with a survival rate above 95 %. On the other hand, pretreatment of rat cortical neurons did not protect them against NMDA neurotoxicity. Furthermore, incubation of human microglia with our novel DAPK1

inhibitors does not impair phagocytosis. Our findings provide robust evidence for DAPK1 involvement in PD. Elucidation of autophagy and phagocytosis pathways using knocked-out DAPK1 mice will be conducted using an α-synuclein mouse model to elucidate the role of DAPK1 in amyloidosis and dopaminergic neuron vulnerability.

Disclosures: M. Santoro: None. **A.T. Adam:** None. **R.K. Lam:** None. **C. Bachmeier:** None. **M. Shamloo:** None.

Poster

P249. Parkinson's Disease: Cellular Mechanisms I

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Program #/Poster #: P249.04

Topic: C.03. Parkinson’s Disease

Title: Dopaminergic Neurons generated from iPSCs carrying mutations in either SNCA, GBA or LRRK2 recapitulate salient Parkinson’s disease phenotypes

Authors: *A. FATHI, S. DICKERSON, A. MUSKINSKY, M. MILLER, W. POON, J. LIU; Fujifilm Cell. Dynamics Inc., Madison, WI

Abstract: Human iPSC-derived dopaminergic neurons offer a developmentally and physiologically relevant *in-vitro* model of human midbrain dopaminergic neurons that innervate different regions in the CNS including the forebrain and striatum. Loss of dopaminergic neurons cause decreased dopamine levels in the CNS and result in neurodegenerative conditions including Parkinson’s disease (PD). Dopaminergic neurons decline with age and are selectively vulnerable to oxidative stress generated by dopamine oxidation that increases with age. The present study outlines the generation and characterization of iPSC derived dopaminergic neurons from two PD patients that inherited either the GBA (N370S) or LRRK2 (G2019S) mutation and genetically engineered SNCA A53T iPSCs. End stage dopaminergic neurons derived from healthy as well PD donors were assessed for GBA activity, neuronal MEA activity, and alpha-synuclein-mediated protein aggregation. These results recapitulated the many features of PD in a dish. Thus, this panel of human iPSC-derived dopaminergic neurons carrying disease-specific mutations can be used for various in vitro applications to uncover mechanistic insights of dopaminergic neuronal degeneration and identification of novel therapeutic targets.

Disclosures: A. Fathi: A. Employment/Salary (full or part-time):; FUJIFILM Cellular Dynamics, Inc., Madison, WI USA. **S. Dickerson:** A. Employment/Salary (full or part-time):; FUJIFILM Cellular Dynamics, Inc., Madison, WI USA. **A. Muskinsky:** A. Employment/Salary (full or part-time):; FUJIFILM Cellular Dynamics, Inc., Madison, WI USA. **M. Miller:** A. Employment/Salary (full or part-time):; FUJIFILM Cellular Dynamics, Inc., Madison, WI USA. **W. Poon:** A. Employment/Salary (full or part-time):; FUJIFILM Cellular Dynamics, Inc.,

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Poster

P249. Parkinson's Disease: Cellular Mechanisms I

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Topic: C.03. Parkinson’s Disease

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JPB Foundation

Title: Neuronal NLRP3 is a parkin substrate that drives neurodegeneration in Parkinson's Disease

Authors: *N. PANICKER[1], T.-I. KAM[1], H. WANG[1], M. KUMAR[3], S. BRAHMACHARI[1], A. JHALDIYAL[1], J. HINKLE[4], F. AKKENTLI[4], X. MAO[5], E. XU[6], S. S. KARUPPAGOUNDER[1], E. T. HSU[2], S. KANG[2], O. PLETNIKOVA[7], J. C. TRONCOSO[8], V. L. DAWSON[9], T. M. DAWSON[9];
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Abstract: Intraneuronal accumulation of α-synuclein aggregates and the subsequent death of substantia nigra *pars compacta* (SNpc) dopamine (DA) neurons are hallmarks of Parkinson’s disease (PD). Microglial activation of the Nod-Like Receptor Protein-3 (NLRP3) inflammasome has been well-documented in various neurodegenerative diseases, including PD. We show here that loss of activity of the PD-associated E-3 ubiquitin ligase parkin in mouse and human DA neurons results in spontaneous neuronal NLRP3 inflammasome assembly, leading to DA neuron death. Parkin normally inhibits inflammasome priming by ubiquitinating and targeting NLRP3 for proteasomal degradation. Loss of parkin activity also contributes to assembly of active NLRP3 inflammasome complex via mitochondrial-derived reactive oxygen species (mitoROS) generation through the accumulation of another parkin ubiquitination substrate, ZNF746/PARIS.

Inhibition of neuronal NLRP3 inflammasome assembly prevents degeneration of DA neurons in familial and sporadic PD models. Strategies aimed at limiting neuronal NLRP3 inflammasome activation hold promise as a disease modifying therapy for PD.

Disclosures: N. Panicker: None. **T. Kam:** None. **H. Wang:** None. **M. Kumar:** None. **S. Brahmachari:** None. **A. Jhaldiyal:** None. **J. Hinkle:** None. **F. Akkentli:** None. **X. Mao:** None. **E. Xu:** None. **S.S. Karuppagounder:** None. **E.T. Hsu:** None. **S. Kang:** None. **O. Pletnikova:** None. **J.C. Troncoso:** None. **V.L. Dawson:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); V. Dawson is a founder of Valted, LLC and holds an ownership equity interest in the company. **T.M. Dawson:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Dr. T. Dawson is a founder of Valted, LLC and holds an ownership equity interest in the company..

Poster

P249. Parkinson's Disease: Cellular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P249.06

Topic: C.03. Parkinson’s Disease

Support: Michael J Fox foundation Grant 18366
Tech Launch Arizona Grant MDH21

Title: Phenotyping Parkinson's disease in patient-derived fibroblasts

Authors: *A. MCROBBIE-JOHNSON, E. CARRUTH, M. J. CORENBLUM, L. MADHAVAN;
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Abstract: Parkinson’s disease (PD) is a common age-related neurodegenerative disorder characterized by debilitating motor deficits and neurocognitive decline. It affects more than 10 million people worldwide, however, has no cure. A major roadblock impeding the development of effective therapeutics has been the inadequate understanding of PD etiopathology, coupled with its relatively late diagnosis at an advanced stage. Our previous work reported specific cellular and molecular alterations in skin fibroblasts of late-onset sporadic PD subjects, that distinguished them from fibroblasts of healthy controls (Teves et al. 2018). Based on these findings, the current work (1) further validates fibroblast features highly predictive of PD, and (2) assesses whether particular changes in the fibroblasts can track PD severity and progression. More specifically, we examined several fibroblast lines obtained from sporadic PD and healthy control individuals at <5 years from diagnosis (early-stage disease) and at post-mortem (end-stage disease). These fibroblast lines were subjected to a range of assays examining cell growth,

morphology, viability, redox status, mitochondrial and autophagic function. It was found that fibroblasts from early-stage PD patients were significantly smaller and rounder, and showed higher rates of proliferation, compared to age- and sex-matched control lines. This was associated with higher reactive oxygen species (ROS) levels (DCFDA assay), mitochondrial dysfunction (Seahorse Mito Stress test) and lower viability rates (MTT assay) in the PD fibroblasts. Additionally, PD cells displayed increased numbers of autophagic vesicles (electron microscopy), and greater autophagic flux as indicated by changes in the standard autophagy markers, LC3II and P62 (western blotting). Correlatively, a significantly higher expression of the PD-relevant protein alpha-synuclein (α-syn), a target of autophagy, was noted in the PD cells. Interestingly, preliminary data from the fibroblasts of the post-mortem cases indicated slower growth rates but further elevated levels of ROS, as well as highly reduced viability in the PD fibroblasts. In essence, these data suggest that patient-derived skin fibroblasts can not only serve as a useful system for diagnosing sporadic PD, but potentially also monitoring PD progression and severity.

Disclosures: A. McRobbie-Johnson: None. **E. Carruth:** None. **M.J. Corenblum:** None. **L. Madhavan:** None.

Poster

P249. Parkinson's Disease: Cellular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P249.07

Topic: C.03. Parkinson’s Disease

Support: Euskampus Foundation
France Parkinson's Foundation

Title: Cell-type specific alteration of excitability in the substantia nigra pars reticulata of parkinsonian mice

Authors: *L. DELGADO ZABALZA[1], M. GARRET[2], J. BAUFRETON[1], C. MIGUELEZ[3]; [1]Inst. of Neurodegenerative disorders, Univ. of Bordeaux, CNRS-UMR 5293, Bordeaux, France; [2]Inst. for Cognitive & Integrative Neurosci. Aquitaine, Univ. of Bordeaux, CNRS-UMR 5287, Bordeaux, France; [3]Pharmacol., Univ. of the Basque Country (UPV/EHU), Leioa, Vizcaya, Spain

Abstract: The *substantia nigra pars reticulata* (SNr) is the main output nucleus of the basal ganglia (BG), a subcortical network involved in motor, cognitive and associative functions. Recently, parvalbumin-expressing SNr subpopulation has been associated with motor functions; however the impact of dopamine depletion on the excitability of motor-related SNr-PV neurons is unknown. By combining immunohistochemical and electrophysiological *in vitro* approaches in

PVCre::Ai9T mice, we demonstrated that SNr-PV neurons exhibit different anatomical, molecular and electrophysiological profiles than non PV-expressing SNr neurons. Moreover, we revealed that only SNr-PV neurons lose their excitability in the unilateral 6-OHDA mouse model of PD, due to a reduction in background sodium currents in these neurons. Currently, we are using chemogenetic approaches to improve motor symptoms in parkinsonian mice and to demonstrate a causative relationship between SNr-PV excitability and motor function.

Disclosures: L. Delgado Zabalza: None. **M. Garret:** None. **J. Baufreton:** None. **C. Miguelez:** None.

Poster

P249. Parkinson's Disease: Cellular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P249.08

Topic: C.03. Parkinson’s Disease

Support: NIH grant R01NS115809
American Parkinson Disease Association (APDA) grant

Title: S100b alters intrinsic firing and ca^{2+}activity of dopaminergic neurons: implications for parkinson's disease.

Authors: *E. A. BANCROFT[1], G. PANDEY[1,2], S. M. ZARATE[1], R. SRINIVASAN[1,2]; [1]Neurosci. & Exptl. Therapeut., Texas A&M Univ. Col. of Med., Bryan, TX; [2]Texas A&M Inst. for Neurosci., College Station, TX

Abstract: Parkinson’s disease (PD) is associated with an abnormal increase in S100B within the midbrain and cerebrospinal fluid. In addition, overexpression of S100B in mice accelerates the loss of substantia nigra pars compacta (SNc) dopaminergic (DA) neurons, suggesting a role for this protein in PD pathogenesis. We found that in the mouse SNc, S100B-labeled astrocytic processes completely envelop the somata of tyrosine hydroxylase (TH) positive DA neurons. Based on this finding, we rationalized that abnormal increases in extracellularly secreted S100B by astrocytic processes in the SNc could alter DA neuron activity, thereby causing dysregulated midbrain function. To test this hypothesis, we measured the effect of bath perfused S100B peptide on the frequency and amplitude of spontaneous Ca^{2+} fluxes in identified TH+ and TH- midbrain neurons from 3-week old mouse primary midbrain cultures. Acute exposure to 50 pM S100B caused a 2-fold increase in Ca^{2+} flux frequency only in TH+ DA neurons. The L-type voltage-gated calcium channel (VGCC) inhibitor, diltiazem eliminated S100B-mediated increases in DA neuron calcium flux frequency, while the T-type specific VGCC blocker mibefradil failed to inhibit the stimulatory effect of S100B. We next asked if changes in intrinsic firing of DA neurons are upstream of the observed S100B-mediated changes in Ca^{2+} activity. We

performed whole cell electrophysiology in identified TH+ and TH- midbrain neurons to determine the effect of acute S100B application on recorded APs in aCSF containing AP5, NBQX, and Bicuculline. Interestingly, acute S100B application increased AP frequency in TH+ DA neurons alone. Because AP firing frequency is known to be regulated by activity of voltage-gated potassium channels (VGKCs), we also determined the effect of acute S100B application on Kv currents in midbrain neurons. Surprisingly, only TH+ DA neurons displayed an inhibition of Kv currents in the presence of 50 pM S100B, suggesting that S100B may inhibit activity of voltage-gated potassium channels (VGKCs) specifically expressed on DA neurons. Together, our results suggest that exposure to S100B pathologically alters intrinsic firing in midbrain neurons, thereby altering spontaneous Ca^{2+} activity. We propose that these changes are initiated via an extracellular interaction between S100B and VGKCs expressed in DA neurons. Our findings are relevant to understanding the complex mechanisms underlying DA neuron loss during PD.

Disclosures: E.A. Bancroft: None. **G. Pandey:** None. **S.M. Zarate:** None. **R. Srinivasan:** None.

Poster

P249. Parkinson's Disease: Cellular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P249.09

Topic: C.03. Parkinson’s Disease

Support: NIH grant P50-NS098685
NIH/ORIP Yerkes National Primate Center base grant (P51-OD011132)

Title: Ultrastructural plasticity of pallidothalamic terminals in the motor thalamus of <MPTP-treated parkinsonian monkeys: A 3D> quantitative analysis.

Authors: *G. J. MASILAMONI[1], H. KELLY[2], A. J. SWAIN[3], Y. SMITH[4];
[1]Yerkes Natl. Primate Res. Center, UDALL Cen, Atlanta, GA; [2]Yerkes Natl. Primate Ctr., Atlanta, GA; [3]Yerkes Natl. Primate Res. Ctr., Atlanta, GA; [4]Yerkes Res. Ctr., Udall Ctr. Excel. For Parkinson's Dis. and Dept. of Neurol., Atlanta, GA

Abstract: In the primate thalamus, the parvocellular ventral anterior nucleus (VApc) and the centromedian nucleus (CM) are under the control of tonic inhibitory inputs from large GABAergic terminals from the internal globus pallidus (GPi). In Parkinson’s disease and animal models of parkinsonism, the loss of nigrostriatal dopamine results in an increased inhibitory pallidothalamic outflow and complex changes in firing rates, firing patterns, and oscillatory firing properties of thalamocortical neurons (Kammermeier et al., 2016, J Neurophysiol 116:2869). Giving that the maintenance of tonic GABAergic synaptic transmission at high presynaptic firing rates is associated with specific ultrastructural features in terminals volume,

number and size of synapses and volume/number of mitochondria in the thalamus and other brain regions (Wanaverbecq et al., 2008, J Neurosci 28:11848; Barberis, 2020, Neuropharmacology 169:107643), we used a Single Block Facing/Scanning Electron Microscopy (SBF/SEM) high resolution 3D electron microscopic approach to compare the morphometry of single GPi terminals in the motor thalamus of 2 control and 2 MPTP-treated parkinsonian monkeys. Our findings demonstrate that pallidothalamic terminals in VApc and CM undergo major ultrastructural changes in parkinsonian monkeys: (1) increased volume terminal volume in both nuclei, (2) increased surface area of synapses in both nuclei, (3) increased number of synapses/GPi terminals in the CM, but not VApc, (4) increased total volume of mitochondria/terminals in both nuclei and increased number of mitochondria in CM. Our findings also show striking morphological differences in terminal volume, number/area of synapses and volume/number of mitochondria between GPi terminals in VApc and CM in control monkeys. In conclusion, results of this study demonstrate that GABAergic pallidothalamic terminals are endowed with a high level of structural plasticity that may contribute to the development and maintenance of the abnormal increase in pallidal outflow to the thalamus in the parkinsonian state. Furthermore, the evidence for ultrastructural differences between GPi terminals in VApc and CM suggests that morphologically distinct pallidothalamic terminals underlie specific physiological properties of pallidal inputs to VApc and CM in normal and diseased states.

Disclosures: G.J. Masilamoni: None. **H. Kelly:** None. **A.J. Swain:** None. **Y. Smith:** None.

Poster

P249. Parkinson's Disease: Cellular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P249.10

Topic: C.03. Parkinson’s Disease

Support: William N. and Bernice E. Bumpus Foundation
Helis Foundation

Title: Pathogenic α-Synuclein inactivates translation repressor eIF4E-BP1 in Parkinson’s diseas

Authors: *M. KHAN[1], T. M. DAWSON[2], V. L. DAWSON[3];
[1]Johns Hopkins Med. Institutions, Baltimore, MD; [2]Inst. for Cell Engin., Johns Hopkins Univ. Inst. for Cell Engin., Baltimore, MD; [3]Inst. Cell Engin., Johns Hopkins Univ. Sch. Med., Baltimore, MD

Abstract: Inhibition of cap-dependent translation by active eIF4E-BP1 (4E-BP1) serves as evolutionarily conserved survival mechanism under various stress conditions which include oxidative stress, immune stress and unfolded protein response. 4E-BP1 is a 5’-Cap dependent

translation inhibitor of mRNA transcripts. In recent years, neuroprotective roles of active 4E-BP1 have emerged in sporadic, autosomal recessive and dominant Parkinsonism. However, posttranslational status of 4E-BP1 is unknown in Parkinson's disease (PD) models and human PD brain. Here we report that 4E-BP1 is post-translationally phosphorylated and inactivated in both PD models and human PD brain. We applied novel proteomics approaches to explore the underlying molecular basis of 4E-BP1 inactivation and translation upregulation in α-Synuclein PFF (Pre-Formed Fibril) mice and cellular models. Our proteomics and Single Molecule Pulldown (SiMPull) data strongly suggest that α-Syn PFF directly interacts with TSC2 and destabilizes TSC1-TSC2 complex, which results in persistent mTORC1 activation and 4E-BP1 inactivation. We generated transgenic models to overexpress active 4E-BP1 in α-Syn toxic background. Overexpression of 4E-BP1 in α-Syn PFF treated neuron shows neuroprotection and block α-Syn pathology. Double transgenic α-Syn:4E-BP1 flies show dopamine neuron protection, rescue motor deficits and mitochondrial defects. Taken together, we provide novel molecular evidences of 4E-BP1 inactivation in Parkinson's disease and restoration of 4E-BP1 activity serves as a conserved protective mechanism against PD pathogenesis.

Disclosures: M. Khan: None. **T.M. Dawson:** None. **V.L. Dawson:** None.

Poster

P249. Parkinson's Disease: Cellular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P249.11

Topic: C.03. Parkinson's Disease

Support: MOH-OFLCG18May-0002

Title: Investigating the role of Parkin in neuronal homeostasis and Parkinson's disease pathogenesis

Authors: *W. TANG[1], J. L. THUNDYIL[2], S. YEOW[3], A. NAIR[4], G. LIM[5], C. CHAI[6], T.-P. YAO[8], K. LIM[7];
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Abstract: Brain lipid dyshomeostasis has been proposed as an early pathological event that triggers the neurodegeneration of energy-demanding dopaminergic neurons in Parkinson's disease (PD). In line with this notion, several studies have reported the presence of lipid-enriched vesicles in the Lewy Bodies found in PD brain tissues and various experimental models of PD.

Moreover, several genetic risk factors for PD (for example, *PLA2G6*, *GBA*, and *SREBF1*) have known molecular functions in lipid homeostasis, suggesting a potential involvement of lipid dysregulation in PD pathogenesis. Intriguingly, the PD-linked protein Parkin has been reported to functionally interact with the fatty acid translocase CD36 and the major regulators of lipid metabolism, namely Sterol Regulatory Element Binding Proteins (SREBPs). However, Parkin modulation on brain lipid metabolism and its potential relevance in dopaminergic neuronal death remain elusive. In this study, we demonstrate by means of multiple experimental models that Parkin regulates SREBP2-Lipoprotein Lipase (LPL) pathway in neurons. We show that Parkin expression level positively correlates with neuronal LPL protein level and activity in human neuroblastoma cell line and a Parkin knockout (KO) mouse model. We identified SREBP2, an important transcription factor involved in lipid metabolism and cholesterol biosynthesis, as a potential mediator between Parkin and LPL. Supporting this, CRISPR-mediated KO of SREBP2 prevented Parkin effect on LPL expression. We further demonstrate that Parkin-SREBP2-LPL pathway regulates the intracellular deposition of lipid droplets (LDs), cellular organelles that are associated with neurodegeneration. Interestingly, this pathway is upregulated upon cellular exposure to PD-linked oxidative stress, which is known to induce accumulation of intracellular LDs. Finally, our data demonstrate that inhibition of either LPL or SREBP2 exacerbates cell deaths triggered by excessive oxidative stress. Taken together, our findings reveal a novel pathway implicating Parkin, SREBP2, and LPL in neuronal lipid homeostasis and LD formation in a PD-relevant context.

Disclosures: W. Tang: None. **J.L. Thundyil:** None. **S. Yeow:** None. **A. Nair:** None. **G. Lim:** None. **C. Chai:** None. **T. Yao:** None. **K. Lim:** None.

Poster

P288. Motor Neuron Disease: Human Genetics and Cellular Mechanisms II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P288.01

Topic: C.06. Neuromuscular Diseases

Support: NIH Grant - R01 NS105473

Title: Altered mitochondrial dynamics disrupts endoplasmic reticulum-mitochondria contacts in Huntington’s disease

Authors: *N. SAWANT, S. KUMAR, P. REDDY;
Texas Tech. Univ. Hlth. Sci. Ctr., Lubbock, TX

Abstract: Huntington's disease (HD) is a fatal, progressive neurodegenerative disease, clinically characterized by chorea, seizures, cognitive decline, and emotional disturbances. Recent research suggests that abnormal interactions between mutant huntingtin (mHTT) and mitochondrial

fission protein - dynamin-related protein 1 (DRP1), induces free radical production and causes excessive mitochondrial fragmentation. Further, the excess fragmentation may also disrupt mitochondrial association with other cell organelles such as the endoplasmic reticulum (ER), as previously reported in other neurodegenerative diseases. These specialized contact sites with ER, known as mitochondria associated membranes (MAMs) are critical for maintaining calcium homeostasis, as mitochondria are major Ca2+ storage sites, therefore disrupted MAMs could be key in HD pathogenesis. **Experiment design:** Mutant HD STHdh$^{Q111/Q111}$ and wild type STHdh$^{Q7/Q7}$ cells were used for studying proteins such as IP3R3, GRP75, VDAC1 and Mfn2 involved in the ER-mitochondria interaction. The cells were treated with mitochondrial division inhibitor 1 (Mdivi1), an inhibitor of Drp1, to see if reduced mitochondrial fragmentation improves MAM contacts. **Methods:** Immunoblotting, immunofluorescence, immunoprecipitation, and co-localization (using pDsRed2-Mito and GFP-Sec61-β constructs for labelling mitochondria and ER respectively) techniques were used to study MAMs and associated proteins, along with mHTT. **Results:** The MAM contacts which were diminished in STHdh$^{Q111/Q111}$ cells, were significantly increased upon Mdivi1 treatment, along with increased IP3R3 and GRP75 interactions. Further, Mdivi1 increased IP3R3 and Mfn2 levels in both wild-type and mutant groups and mHTT localization was less nuclear after treatment. **Conclusion:** Taken together these findings suggest that Mdivi1 reduces excessive mitochondrial fragmentation, reduces mHTT toxicity in HD neurons, and restores the ER-mitochondrial contacts.

Disclosures: N. Sawant: None. **S. Kumar:** None. **P. Reddy:** None.

Poster

P288. Motor Neuron Disease: Human Genetics and Cellular Mechanisms II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P288.02

Topic: C.06. Neuromuscular Diseases

Support: 1R01 NS108191

Title: Investigating androgen receptor dimerization as a novel mediator of toxicity in SBMA

Authors: *A. LISBERG[1], D. E. MERRY[2];
[1]Dept. Biochem. and Mol. Biol., [2]Dept Biochem & Molec Biol, Thomas Jefferson Univ., Philadelphia, PA

Abstract: Spinobulbar muscular atrophy (SBMA) is a progressive neuromuscular degenerative disease caused by a polyglutamine (polyQ) expansion in the androgen receptor (AR). This mutation causes misfolding and aggregation of the protein as well as cell dysfunction and death. While advances in the understanding of the molecular pathogenesis of SBMA have been made,

there is still no effective treatment for this disease. One pathogenic mechanism involves an AR interdomain interaction, deemed the N/C-interaction, that has been shown to be required for polyQ-expanded AR toxicity. This project seeks to understand the mechanism that underlies the role of the N/C interaction in SBMA to develop better therapeutics for this disease. Our initial experiments revealed that the AR N/C interaction is decreased by polyglutamine expansion, consistent with the known impact of the polyglutamine tract on adjacent AR protein structure. This result led us to hypothesize that different forms of the N/C interaction, which occur in either monomeric or dimeric AR, played different roles in SBMA pathogenesis. Using a biochemical assay to evaluate AR dimerization, we found that AR dimerization is increased upon polyglutamine expansion and is decreased upon N/C blockade, supporting the hypothesis that aberrant dimerization may play a role in the toxicity of polyQ-expanded AR. We next tested the hypothesis that AR dimerization, mediated by distinct amino acids within the DNA binding domain, is required for mutant AR aggregation and toxicity, and observed that blocking dimerization decreased nuclear inclusion formation. Investigation into the effect of blocking dimerization on polyQ-expanded AR toxicity in PC12 cells, mouse motor neurons, and iPSC lines is ongoing. Of note, we observed an increased cytoplasmic localization and decreased stabilization of dimerization-blocked AR that is not due to changes in AR nuclear export. Paired with quantitative differential interactome studies that have identified proteasomal components as interactors of interest, this finding suggests that enhanced AR degradation may play a role in the protection seen upon blocking AR dimerization. Further investigation into the role of proteasomal degradation is ongoing. Overall, this work seeks to provide further insight into the molecular mechanisms of SBMA pathogenesis and has identified AR dimerization as a novel mediator of toxicity in this disease.

Disclosures: A. Lisberg: None. **D.E. Merry:** None.

Poster

P288. Motor Neuron Disease: Human Genetics and Cellular Mechanisms II

Location: Virtual Only

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Program #/Poster #: P288.03

Topic: C.06. Neuromuscular Diseases

Support: FWO SB PhD Fellowship 1S46219N
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KU Leuven C1-internal fund C14-17-107
FWO Odysseus grant G0F8516N
ALS Liga Belgium

Title: Morpholino-based TUBA4A knockdown in zebrafish recapitulates ALS-related abnormalities

Authors: *E. VAN SCHOOR[1,2], V. BERCIER[1,2], E. BRAEMS[1,2], P. VAN DAMME[1,3,2], D. R. THAL[2,4], L. VAN DEN BOSCH[1,2];
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Abstract: Aim
Disease-associated variants of *TUBA4A* (alpha-tubulin 4A) have recently been identified in familial ALS patients. It was also shown that this mutated TUBA4A ineffectively forms tubulin dimers *in vitro* [1]. Interestingly, a downregulation of TUBA4A protein expression was suggested in familial as well as sporadic ALS patients [2]. To investigate whether a decreased TUBA4A protein expression could be a driving factor in ALS pathogenesis, we assessed if knockdown of TUBA4A expression in zebrafish could recapitulate an ALS-like phenotype.
Methods
We injected one- to two-cell stage zebrafish embryos from the AB strain with ATG morpholino's against the zebrafish TUBA4A (Tuba8l2). Western blotting was used to confirm the TUBA4A knockdown. To analyze axonal length, fish were fixed with 4% PFA and stained using an antibody against synaptic vesicle 2 (SV2). Motor behavior was assessed using the touch-evoked escape response (TEER). Finally, TUBA4A protein expression levels were assessed in post-mortem brain tissue of ALS patients.
Results
TUBA4A protein downregulation was indeed observed in post-mortem motor cortex of ALS patients. When we performed an ATG morpholino-based knockdown of zebrafish TUBA4A, this induced axonal branching defects at 48 hpf (hours post fertilization) as observed by SV2 immunohistochemistry. In addition, a decrease in maximal and average velocity as well as time and distance travelled was observed using the TEER assay. Both phenotypes were morpholino dose-dependent and could be rescued by the addition of wild-type *TUBA4A* mRNA, but not the ALS-related mutant A383T *TUBA4A* mRNA.
Conclusions
Knockdown of zebrafish TUBA4A using an ATG morpholino induces an axonal branching phenotype as well as motor behavior defects, reflecting a motor neuron disease-related phenotype. Addition of wild-type *TUBA4A* mRNA rescues this phenotype, further strengthening the causal relation with TUBA4A knockdown. Importantly, TUBA4A downregulation was observed in ALS post-mortem motor cortex tissue. Our data point towards an involvement of disturbances in the expression of TUBA4A in ALS disease pathogenesis.
References[1] J.A. Clark, *et al*. "A case for microtubule vulnerability in amyotrophic lateral sclerosis: altered dynamics during disease", *Front. Cell. Neurosci*., vol. 10, pp. 1-16, 2016.[2] A.M. Helferich *et al*., "Dysregulation of a novel miR-1825/TBCB/TUBA4A pathway in sporadic and familial ALS", *Cell. Mol. Life Sci*., vol. 75, no. 23, pp. 4301-4319, 2018.

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Poster

P288. Motor Neuron Disease: Human Genetics and Cellular Mechanisms II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P288.04

Topic: C.06. Neuromuscular Diseases

Support: DOD-TSCRP W81XWH-17-1-0151

Title: Molecular mapping of genetic modifiers in Tuberous Sclerosis (TSC) disease severity throughgenomic approaches

Authors: *A. HOWELL, D. ENRIQUEZ, C. LEGENDRE, P. SWAMINATHAN, H. FRY, S. KANESHAMOORTHY, W. LIANG, V. NARAYANAN, S. RANGASAMY;
Tgen, Phoenix, AZ

Abstract: Tuberous sclerosis complex (TSC) is an autosomal dominant neurogenetic disorder, in which affected individuals are heterozygous for a mutation in either the *TSC1* or *TSC2* gene, thereby causing constitutive activation of the mTOR signaling pathway. Phenotypic variability (PV) in TSC is well recognized, but its molecular basis is not understood. Variability in phenotype can be seen even within a single family (intrafamilial phenotypic variability [IPV]), where all affected individuals have the same TSC gene mutation. IPV clearly indicates that *TSC1* or *TSC2* mutation alone does not account for the observed phenotype and suggests a potential role for modifying factors. Mechanisms currently proposed for PV in monogenic disorders includes modifying effects of unlinked genes (**genetic modifiers**), epigenetic factors, mosaicism, allelic skewing of gene expression, and environmental or other stochastic factors. At the Center for Rare Childhood Disorders (C4RCD), we have assembled a cohort of 16 TSC-afflicted families that exhibit phenotypic variability between parent and child. Using whole exome and RNA sequencing, we aim to (1) identify aberrant gene expression, aberrant splicing, and mono-allelic expression in severe vs mildly afflicted patients (2) identify genetic modifiers in WES data that may be correlated with expression differences in severe vs mildly afflicted patients. Preliminary results using the R tool DESeq2 demonstrates that the differentially expressed gene set between severe and mild forms of TSC. Gene set enrichment analysis (GSEA) indicate that genes are associated with protein processing in endoplasmic reticulum and cell adhesion molecules. Previous research has shown that in human TSC-associated renal angiomyolipoma cells endoplasmic reticulum (ER) stress can develop due to mTORC1-driven protein translation, supporting our functional enrichment analysis. Additional linear regression models using Lasso will incorporate mutation type, mTOR pathway variants, and patient symptoms to predict differential gene expression, aberrant splicing, and mono-allelic expression (MAE) identified

through the detection of RNA outliers' pipeline (DROP). By understanding the molecular basis of PV, we aim to define a better genetic predicators of disease severity and new treatment for TSC.

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Poster

P288. Motor Neuron Disease: Human Genetics and Cellular Mechanisms II

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Program #/Poster #: P288.05

Topic: C.06. Neuromuscular Diseases

Support: NMRC/OFIRG/0042/2017
NIH R01 NS095793

Title: Tdp-43 is essential for oligodendrocyte myelination by regulating srebf2-mediated cholesterol metabolism

Authors: *S.-C. LING[1], W. HO[1], J.-C. CHANG[1], K. LIM[1], G. TUCKER-KELLOGG[1], A. CAZENAVE-GASSIOT[1], M. WENK[1], E. B. LEE[2], A. NGUYEN[2], S.-Y. NG[1], S. ONG[1], A. VIERA-ORTIZ[2];
[1]Natl. Univ. of Singapore, Singapore, Singapore; [2]Dept. of Pathology and Lab. Med., Univ. of Pennsylvania, Philadelphia, PA

Abstract: Cholesterol metabolism operates autonomously within the central nervous system (CNS), where the majority of cholesterol resides in myelin. We demonstrate that TDP-43, the pathological signature protein for amyotrophic lateral sclerosis (ALS) and frontotemporal dementia (FTD), regulates cholesterol metabolism in oligodendrocytes. TDP-43 binds directly to mRNA of SREBF2, the master transcription regulator for cholesterol metabolism, and multiple mRNAs encoding proteins responsible for cholesterol biosynthesis and uptake, including *HMGCR*, *HMGCS1*, and *LDLR*. TDP-43 depletion leads to reduced SREBF2 and LDLR expression, and cholesterol levels *in vitro* and *in vivo*. TDP-43-mediated changes in cholesterol levels can be restored by reintroducing SREBF2 or LDLR. Additionally, cholesterol supplementation rescues demyelination caused by TDP-43 deletion. Furthermore, oligodendrocytes harboring TDP-43 pathology from FTD patients show reduced HMGCR and HMGCS1, and co-aggregation of LDLR and TDP-43. Collectively, our results indicate that TDP-43 plays a role in cholesterol homeostasis, and cholesterol dysmetabolism may be implicated in TDP-43 proteinopathies-related diseases.

Disclosures: S. Ling: None. **W. Ho:** None. **J. Chang:** None. **K. Lim:** None. **G. Tucker-Kellogg:** None. **A. Cazenave-Gassiot:** None. **M. Wenk:** None. **E.B. Lee:** None. **A. Nguyen:** None. **S. Ng:** None. **S. Ong:** None. **A. Viera-Ortiz:** None.

Poster

P288. Motor Neuron Disease: Human Genetics and Cellular Mechanisms II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P288.06

Topic: C.06. Neuromuscular Diseases

Title: Effects of SOD1 aggregates on the electrophysiological properties of hESC-derived motor neurons

Authors: E. MOON[1], *G. AARON[2], A. O'NEIL[3];
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Abstract: Amyotrophic lateral sclerosis (ALS) is a progressive neurodegenerative disease characterized by the dysfunction and death of motor neurons in the spinal cord, brain stem, and motor cortex. Approximately 10% of all ALS cases are familial (fALS) and have been linked to a variety of genetic mutations. About 20% of fALS cases are linked to a mutation in the *superoxide dismutase 1 (SOD1)* gene. Wild type SOD1 is an antioxidant protein responsible for capturing superoxide radicals and preventing oxidative damage. In individuals with SOD1-linked fALS, SOD1 mutations confer a toxic gain of function, resulting in abnormal protein aggregation. Interestingly, SOD1 aggregates are specifically toxic to motor neurons, causing previously healthy cells to die. Other types of neurons also incorporate SOD1 aggregates but show no signs of toxicity. Furthermore, research in both mouse and human pluripotent stem cell models of fALS have shown that motor neurons with SOD1 mutations have different electrical properties and firing patterns compared to wild type controls. Some findings suggest that motor neurons with SOD1 mutations are hyperexcitable while others suggest that cells are hypoexcitable. This apparent contradiction has led to the hypothesis that hyperexcitability may be an early, presymptomatic neuroprotective mechanism and that motor neurons transition into a phase of late hypoexcitability following disease onset and progression. The reasons for these changes in excitability are not fully understood, but work in both mice and humans has shown alterations in ion channel expression and properties as possible causes. Here, we use motor neurons differentiated from human embryonic stem cells with a GFP-Islet1 reporter and matured in culture for 21 days. Following maturation, we add H46R SOD1 aggregates to the culture medium for 72-120 hours. Using whole-cell patch clamp analysis, we show that hESC-derived motor neurons treated with SOD1 aggregates have a significantly depolarized resting membrane

potential and action potential threshold compared to healthy controls, potentially linking SOD1 aggregates to electrophysiological changes.

Disclosures: E. Moon: None. **G. Aaron:** None. **A. O'Neil:** None.

Poster

P288. Motor Neuron Disease: Human Genetics and Cellular Mechanisms II

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Topic: C.06. Neuromuscular Diseases

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Central ALF VLL-841161
Lokal ALF RV-941634
Ulla-Carin Lindquist Stiftelse

Title: Superoxide dismutase-1 aggregate strain type in human and transgenic mice amyotrophic lateral sclerosis

Authors: *T. BRANNSTROM[1], I. SIGFRIDSSON[1], E. STENVALL[1], K. FORSBERG[2], P. M. ANDERSEN[2], S. L. MARKLUND[1];
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Abstract: Mutations in SOD1 are a common known cause of ALS. Both ALS patients and transgenic mice develop aggregates of hSOD1 in motor neurons. By Binary Epitope Mapping it has been shown that in symptomatic mice two different types of aggregates (denoted A and B) can arise (Bergh et al. *Proc Natl Acad Sci U S A*, 2015, 112:4489-94). Inoculation of minute amounts of these into spinal cords of asymptomatic hSOD1 transgenic mice initiates spreading, exponentially growing templated hSOD1 aggregation concomitantly with premature fatal motor neuron disease (Ekhtiari Bindhendi et al. *J. Clin. Invest.* 2016:126:2249-53).Only for the hSOD1G127Gfs*7 truncation mutation (first described by Andersen et al, *Brain,* 1997, 120:1723-37) has it been possible to ascertain the strain type (A) in human tissue (Ekhtiari Bindhendi et al. *Acta Neuropathol.* 2018:136:939-953).Here we explored whether prion-competent mutant hSOD1 aggregates also exist in human ALS using a proximity detection kit and our set of anti-SOD1 antibodies. Using this setup we can show that the Strain type (A,B) is possible to specifically detect also in histologically processed tissues. Using the proximity detection kit it is possible to ascertain the cell type in which the specific strain type is occurring in. For the D90A human transgenic mice there is a difference in cellular spread between type A

and type B aggregates.Our results suggest that templated spread of hSOD1 aggregation might be cell type specific not only in hSOD1 transgenic models, but possibly also in human ALS.

Disclosures: T. Brannstrom: None. **I. Sigfridsson:** None. **E. Stenvall:** None. **K. Forsberg:** None. **P.M. Andersen:** F. Consulting Fees (e.g., advisory boards); Biogen. **S.L. Marklund:** None.

Poster

P288. Motor Neuron Disease: Human Genetics and Cellular Mechanisms II

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Program #/Poster #: P288.08

Topic: C.06. Neuromuscular Diseases

Support: TARGET ALS, the Muscular Dystrophy Association
NIH (R21NS109847)
NIH R01GM46779
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NIH R01CA207893
NIH R01CA248397
NIH R37NS057553

Title: Novel Regulation of C9ORF72 Dipeptide Repeat Protein Synthesis by an Upstream Open Reading Frame, Initiation Factor DAP5, and Ribosome Stalling

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[1]Univ. of Massachusetts Med. Sch., Worcester, MA; [2]Dept. of Microbiology, New York Univ. Sch. of Med., New York, NY

Abstract: Expansion of the G_4C_2(n) repeat in the *C9ORF72* intron is the most common genetic cause of amyotrophic lateral sclerosis (ALS) and frontotemporal dementia (FTD). Each year 5,000 Americans are diagnosed with ALS and FTD is the most common form of dementia in people under 60 years old, and currently there are no cures. The expansion of the G_4C_2(n) repeat leads to loss of the C9ORF72 protein, the toxicity of accumulated repeat RNA, and toxicity of dipeptide repeats (DPRs) translated from the expansion. Using dynamic ribosome profiling on iPSCs from ALS patients, we identified three initiation sites in the intron upstream of G_4C_2. The initiation sites regulate production of DPRs in different reading frames, to produce poly-GA and poly-GP. We identified ribosomes on unspliced G_4C_2-containing RNA, indicating this is the substrate for translation of the repeat. G_4C_2 RNA translation is 5' cap-independent but inhibited by the initiation factor DAP5. DAP5 is an eIF4G homolog that lacks the eIF4E binding site and

it regulates translation of the G_4C_2(n) repeat. These results define novel translational mechanisms of expanded G_4C_2-containing RNA in ALS and FTD.

Disclosures: H.M. van 't Spijker: None. **E. Stackpole:** None. **S. Almeida:** None. **O. Katsara:** None. **B. Liu:** None. **K. Shen:** None. **R.J. Schneider:** None. **F. Gao:** None. **J.D. Richter:** None.

Poster

P293. Motor Neuron Disease: Therapeutics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P293.01

Topic: C.06. Neuromuscular Diseases

Support: Genuv Inc.

Title: Trametinib protects motor neurons from degeneration by enhancing autophagy in the SOD1 G93A amyotrophic lateral sclerosis model mice

Authors: *S.-Y. LEE, Y. CHUN, H. KIM, S. HAN;
Genuv Inc., Seoul, Korea, Republic of

Abstract: Amyotrophic lateral sclerosis (ALS) is a neurological disease characterized by selective degeneration of motor neurons, that causes muscle paralysis and eventual death. To date, there is no clear therapeutics for improving the symptoms of ALS patients. Previously, we screened a MEK inhibitor, trametinib (SNR1611) from a FDA-approved drug library by its neurogenerative and neuroprotective effect. Here, we confirmed that oral administration of trametinib to SOD1-G93A transgenic mice increased motor performance and grip strength, and improved survival rate. Trametinib protected motor neuronal loss in the spinal cord. Trametinib also increased the level of autophagic markers (LC3, LAMP1 and cathepsin D), and significantly reduced the abnormal aggregation of p62 in spinal motor neurons. These clearly indicate that treatment of trametinib significantly improved the autophagy flux. A higher level of SOD1 protein was observed in cells expressing SOD1-G93A than in cells expressing SOD1-WT. Trametinib not only reduced the level of intracellular SOD1 but also prevented the formation of SOD1-postive aggregates which found only in cells expressing mutant SOD1. The combined data provides clear evidence that MEK inhibition might be a novel therapeutical approach for ALS by enhancing autophagy and eliminate disease-causing mutant proteins. Currently, trametinib is a repurposed drug candidate in the clinical trial (Phase I/IIa) for the indication of ALS in Korea (ClinicalTrials.gov identifier: NCT04326283).

Disclosures: S. Lee: None. **Y. Chun:** None. **H. Kim:** None. **S. Han:** None.

Poster

P293. Motor Neuron Disease: Therapeutics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P293.02

Topic: C.06. Neuromuscular Diseases

Title: Development and efficacy of iPSC-derived neural progenitor cells genetically engineered to produce GDNF as a therapy for ALS

Authors: *A. WU[1], A. LAPERLE[2], V. A. MOSER[3], G. LAWLESS[4], P. AVALOS[5], C. N. SVENDSEN[6];
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Abstract: Amyotrophic Lateral Sclerosis (ALS) is a fatal neurodegenerative disease that affects approximately 30,000 individuals within the US at a given time. ALS is characterized by progressive spinal and cortical motor neuron loss, with paralysis and death typically within 4 years of diagnosis. There is no cure, and current FDA-approved drugs only minimally slow progression. Transplanting supportive glial cells engineered to release glial cell line-derived neurotrophic factor (GDNF) is a promising treatment. Our group has generated and extensively characterized human fetal-derived neural progenitor cells (NPCs) lentivirally transfected to produce GDNF. These cells engraft in the spinal cord and slow the loss of choline acetyltransferase (ChAT)-positive motor neurons in the $SOD1^{G93A}$ rat model of ALS. We have recently completed a Phase 1/2a trial that showed the safety of delivering these cells to the spinal cords of ALS patients as a first in-human cell and gene therapy approach. While promising, use of fetal-derived NPCs is constrained by (i) the ethical concerns of human fetal tissue, (ii) scalability of the starting material and (iii) regulation of the GDNF release. To address these challenges, we have assessed human induced pluripotent stem cells (iPSC) as an alternative, renewable cell source that can differentiate to NPCs. In initial studies we generated iPSC-derived NPC lines lentivirally transduced to express GDNF (iNPC-GDNF). Single cell RNA-seq showed that while there was strong similarity and some overlap between iPSC-derived and fetal-derived NPCs, only a minority of the iNPCs clustered with the fetal-derived cells. To establish whether these cellular differences would be relevant for the functional effect of the cells iNPC-GDNF cells were transplanted unilaterally in the lumbar spinal cord of $SOD1^{G93A}$ rats that were then sacrificed at disease onset. While disease onset was not altered with cell transplants, hindlimb motor function was significantly improved in the treated limb compared to the non-treated side and to non-injected controls. Immunohistochemistry (IHC) for human specific cytoplasm (SC121) showed engraftment of iNPC-GDNF cells. Preliminary studies (n=3) to quantify ChAT+ motor neuron number and size indicate a protective effect on the treated side relative to

the non-treated side. Additionally, we have evaluated iNPC lines with a tetracycline-inducible GDNF construct. Inducible expression systems could tailor the GDNF dose to individual patients and provide the safety feature of turning off GDNF expression if required. These new iPSC-based NPC lines present a scalable and promising new regulatable cell and gene therapy for ALS.

Disclosures: A. Wu: None. **A. Laperle:** None. **V.A. Moser:** None. **G. Lawless:** None. **P. Avalos:** None. **C.N. Svendsen:** None.

Poster

P293. Motor Neuron Disease: Therapeutics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P293.03

Topic: C.06. Neuromuscular Diseases

Support: Dementia Consortium: Alzheimer's Research UK, Eisai, Eli Lilly, JPNV and Takeda

Title: High-throughput screening (HTS) for identification of a tool compound targeting the interaction of SRSF1 with C9orf72 to prevent nuclear export in ALS and FTD

Authors: H. TRAN[1], D. J. BORROWS[3], H. WAN-PING[3], D. JONES[2], M. CHAMBERS[2], M. DA SILVA[1], G. HAUTBERGUE[3], T. RAMESH[3], **R. VAN DE BOSPOORT**[1], J. CONNELL[4]; [1]Charles River Labs., Leiden, Netherlands; [2]Charles River Labs., Saffron Walden, United Kingdom; [3]Sheffield Inst. for Translational Neurosci. (SiTRaN), Sheffield, United Kingdom; [4]Alzheimer's Res. UK, Cambridge, United Kingdom

Abstract: Hexanucleotide repeat expansions (HREs) in intron-1 of the *C9ORF72* gene are the most common known genetic cause of amyotrophic lateral sclerosis (ALS) and frontotemporal dementia (FTD). Thesemicrosatellite repeat sequences (GGGGCC, G_4C_2) contribute to neuronal injury via three non-mutually exclusive mechanisms. A growing body of evidence implicates toxic RNA and protein gain-of-functions due to abnormal nuclear export and repeat-associated non-AUG (RAN) translation of intron-retaining repeat transcripts into neurotoxic dipeptide repeat proteins (DPRs) as one of the main drivers of pathogenicity. Repeat RNA sequestration of SRSF1 triggers the nuclear export of repeat transcripts into the cytoplasm while its depletion inhibits the nuclear export of pathological *C9ORF72* transcripts and production of DPRs, thereby preventing neurotoxicity in reporter cells, patient-derived neurons and *Drosophila* models of C9ORF72-related disease (Hautbergue *et al*, 2017). Modulation of SRSF1 by morpholino mediated knockdown in a pure RAN translation zebrafish model further reduced DPR levels (unpublished Ramesh et. al.).
To identify tool compounds targeting the interaction of SRSF1 with C9orf72, an HTRF assay

was established in 1536-well format to detect proximity of tagged SRSF1 to tagged G_4C_2 RNA. This robust assay (S/B 10.1±0.1 and Z' 0.91±0.01) allowed for high throughput screening (HTS) of 50,000 compounds selected from our compound library collection for their significant structural diversity and CNS-differentiating physicochemical properties, lead-likeness and overall solubility profile. Following frequent hitter analysis (>4 pBSF) and structural alerts review (PAINS, BMS-Lilly), 503 compounds were selected based on their ability to inhibit TR-FRET signal. Preliminary clustering analysis showed good SAR.
Our data demonstrates the robustness of our HTRF assay for hit finding and shows how this in collaboration with academia accelerates identification of potential tool compounds. Our current efforts focus on further selection to <100 compounds with selectivity against counter assays, prior to future validation of their potency in inhibiting the SRSF1-dependent nuclear export of C9orf72-repeat transcripts and the production of DPRs in *in vitro* (reporter human cells, patient-derived neurons) and *in vivo* (zebrafish, mouse) models of C9ORF72-ALS/FTD at SITraN. The ADME profiles of promising compounds will also be conducted in the zebrafish and mouse models of disease.

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Poster

P293. Motor Neuron Disease: Therapeutics

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Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P293.04

Topic: C.06. Neuromuscular Diseases

Support: Target ALS
R01NS101986
R37NS057553

Title: Poly(gr) and poly(ga) in cerebrospinal fluid of c9orf72-als/ftd patients as diagnostic and antisense oligonucleotide target engagement biomarkers

Authors: G. KRISHNAN[1], D. RAITCHEVA[3], D. BARTLETT[3], M. PRUDENCIO[4], D. MCKENNA-YASEK[1], S. LADHA[5], O. KING[1], S. J. BARMADA[6], T. M. MILLER[7], R. BOWSER[5], J. K. WATTS[2], L. PETRUCELLI[4], R. H. BROWN[1], M. KANKEL[3], F.-B. GAO[1]; [1]Neurol., [2]RNA Therapeut. Inst. and Dept. of Biochem. and Mol. Pharmacol., Univ. of Massachusetts Med. Sch., Worcester, MA; [3]Neuromuscular & Movement Disorders, Biogen, Cambridge, MA; [4]Neurosci., Mayo Clin., Jacksonville, FL; [5]Neurobio., St. Joseph's Hosp. and Med. Ctr. and Barrow Neurolog. Inst., Phoenix, AZ; [6]Neurol., Univ. of Michigan, Ann Arbor, MI; [7]Neurol., Washington Univ., Saint Louis, MO

Abstract: GGGGCC repeat expansion in *C9ORF72* is the most common genetic cause of both ALS and FTD. Toxic dipeptide repeat (DPR) proteins can be translated from both sense and antisense repeat RNAs, but it is unknown whether they can serve as biomarkers of target engagement in clinical trials testing therapies directed against the repeat RNA. We developed sensitive assays that reliably detect poly(GA) and poly(GR) in the cerebrospinal fluid (CSF) of *C9ORF72* patients, thus potentially serving as valid fluid-based diagnostic biomarkers. CSF poly(GA) and poly(GR) levels do not correlate with age at disease onset, disease duration, or score on the ALS Functional Rating Scale, and the average levels of these DPR proteins are similar in symptomatic and pre-symptomatic *C9ORF72* patients. However, in a human patient treated with antisense oligonucleotide (ASO) targeting *C9ORF72*, CSF poly(GA) and poly(GR) levels decreased significantly faster than poly(GP) levels. Thus, CSF poly(GA) and poly(GR) may serve as sensitive biomarkers for target engagement in *C9ORF72* patients, addressing a crucial need to determine the effectiveness of ASOs and other *C9ORF72*-directed therapies in planned clinical trials.

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Poster

P293. Motor Neuron Disease: Therapeutics

Location: Virtual Only

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Program #/Poster #: P293.05

Topic: C.06. Neuromuscular Diseases

Support: R01AG068130
R01NS093416
R25GM109441

Title: Inhibiting elevated GCPII activity in aging mouse muscle reduces frailty

Authors: *T. R. JOE[1], C. TALLON[2], R. WISEMAN[3], A. G. THOMAS[4], B. S. SLUSHER[5]; [1]Cell Biol., [2]Neurol., [3]Pharmacol. and Mol. Sci., Johns Hopkins Univ., Baltimore, MD; [4]Johns Hopkins Drug Discovery, Johns Hopkins Univ. Sch. of Med., Baltimore, MD; [5]Johns Hopkins Drug Discovery, Baltimore, MD

Abstract: Glutamate carboxypeptidase II (GCPII) is a neuropeptidase that catalyzes the conversion of N-acetyl-aspartyl-glutamate (NAAG) into N-acetyl-aspartate (NAA) and Glu. In the PNS, GCPII is expressed in Schwann cells and activated macrophages and is involved in regulating the synaptic pruning of neuromuscular junctions (NMJs) during normal development. Recently, we observed increased GCPII expression in the muscle of the $SOD1^{G93A}$ mouse model of amyotrophic lateral sclerosis (ALS) that was selectively associated with infiltrating activated macrophages. When using 2-(Phosphonomethyl)-pentanedioic acid (2-PMPA), a potent and selective inhibitor of GCPII, we observed a significant delay in muscle function loss and denervation in the ALS mouse. ALS and aging share similarities including the loss of motor neurons and degeneration of the skeletal muscle resulting in reduced synaptic inputs and fine motor skills, muscle weakness or wasting, and neuro-inflammation. Due to these similarities, we determined whether GCPII levels were altered in muscle of aged mice similarly to ALS mice. We collected gastrocnemius and soleus muscle tissue from 4-, 12-, and 20-month-old C57BL/6 mice and examined GCPII protein expression and enzymatic activity levels. We observed low expression of GCPII in 4- and 12-month-old mice gastrocnemius muscle and significantly increased expression in 20-month-old mice. Like in ALS, we observed co-localization of GCPII staining on infiltrating activated macrophages in aged gastrocnemius muscle. We then began treating 15-months-old mice, with daily 2-PMPA, for 5 months. Monthly frailty scoring was performed using an index that tracks a spectrum of aging-related characteristics. After 5 months of treatment, 2-PMPA treated mice had significantly reduced frailty index scores compared with vehicle-treated mice. Additionally, there was a significant delay in body weight loss and body temperature decline. These studies demonstrate that blockade of GCPII activity has potential therapeutic benefits for slowing aging-related frailty.

Disclosures: T.R. Joe: None. **C. Tallon:** None. **R. Wiseman:** None. **A.G. Thomas:** None. **B.S. Slusher:** None.

Poster

P293. Motor Neuron Disease: Therapeutics

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Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P293.06

Topic: C.06. Neuromuscular Diseases

Support: A Long Swim

Title: Motor cortex with TDP-43 pathology display dysregulated metabolite and energy homeostasis

Authors: *B. HELMOLD[1], M. GAUTAM[1], A. GUNAY[1], N. CHANDEL[2,3], H. OZDINLER[1,4,5,6];
[1]Davee Dept. of Neurol. and Clin. Neurolog. Sci., [2]Dept. of Urology, Feinberg Sch. of Med., [3]Simpson Querrey Inst. for BioNanotechnology, [4]Robert H. Lurie Comprehensive Cancer Ctr., [5]Les Turner ALS Center, Feinberg Sch. of Med., [6]Chem. of Life Processes Inst., Northwestern Univ., Chicago, IL

Abstract: TDP-43 is a DNA/RNA binding protein that shuttles between nucleus and cytoplasm and is involved in key cellular functions. TDP-43 pathology is the most commonly detected proteinopathies observed in a broad spectrum of neurodegenerative diseases including amyotrophic lateral sclerosis (ALS). Mitochondria is at the center of energy production and generation of other metabolites required for cellular function. TDP-43 pathology impairs mitochondrial health and function, which results in the observation of mitochondrial defects in the upper motor neurons. To understand metabolite dysregulation in ALS motor cortex due to TDP-43 pathology, we performed metabolomic profiling of motor cortex isolated from prpTDP-43^{A315T} mouse model of ALS. Motor cortex of postnatal day 90 prpTDP-43^{A315T} and respective WT mice were dissected out and the metabolites were isolated followed by mass spectrometry profiling of metabolites. Careful analysis of metabolome revealed that several key metabolites such as NAD+, ATP, 2-hydroxyglutarate, glutathione disulfide, and phophoenol pyruvate were differentially regulated due to TDP-43 pathology. This analysis also revealed purine, arginine, glutamate, aspartate, methionine metabolism were among the key metabolite pathways differentially regulated. Proper balance of NAD+/NADH is critically important for ATP generation and metabolite alterations. Recently, a compound that can improve NAD+/NADH balance, namely nicotinamide mononucleotide (NMN), was shown to increase levels of NAD+, improve the health and function of mitochondria, and lead to increased survival in mouse models of mitochondrial disease. We treated corticospinal motor neurons with NMN *in vitro* and our results suggest that NMN improves health of diseased CSMN by improving NAD+/NADH balance. Thus, our study shows that in ALS motor cortex there is significant dysregulation of key metabolites and energy deficiency and maintaining NAD+/NADH ratio may help restore metabolite and energy homeostasis. Improving the health and function of mitochondria and energy production could potential prove to be a therapeutic strategy for ALS.

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Poster

P293. Motor Neuron Disease: Therapeutics

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Program #/Poster #: P293.07

Topic: C.06. Neuromuscular Diseases

Title: Inhibiting C1q improves compound muscle action potential and reduces neurodegenerative damage in the $SOD1^{G93A}$ mouse model

Authors: *V. MATHUR, **Y. ANDREWS-ZWILLING**, L. KUHN, S. SANKARANARAYANAN, E. CAHIR-MCFARLAND, T. YEDNOCK;
Annexon Biosci., South San Francisco, CA

Abstract: Inhibiting C1q improves compound muscle action potential and reduces neurodegenerative damage in the $SOD1^{G93A}$ mouse model
Authors: Vidhu Mathur*, Yaisa Andrews-Zwilling, Logan Kuhn, Sethu Sankaranarayanan, Ellen Cahir-McFarland, Ted Yednock
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Amyotrophic lateral sclerosis (ALS) is a sporadic or genetic disease associated with peripheral loss of synaptic connectivity at neuromuscular junctions and central loss of motor neurons. The $SOD1^{G93A}$ mouse model has been widely used to study a familial form of ALS. C1q, the initiating molecule of the classical complement cascade, marks synapses in the central nervous system for glial elimination during normal development, while triggering aberrant synapse loss in neurodegenerative disorders. In ALS, C1q additionally tags the neuromuscular junction within the peripheral nervous system prior to its removal by macrophages. Despite data showing that embryonic knock out of C1q was not protective in a mutant SOD1 model, we hypothesized that excessive synaptic pruning initiated by C1q contributes to motor deficits in ALS and that pharmacologically inhibiting this process would be beneficial. We treated adult $SOD1^{G93A}$ mice with an anti-C1q blocking antibody from 7 to 16 weeks of age. Treatment resulted in reduction of C1q in plasma, spinal cord and muscle tissue, along with inhibition of downstream classical complement activation after 9 weeks of treatment. Treated mice showed significant improvement in the amplitude and reduced latency of compound muscle action potential, demonstrating increased synaptic connectivity at the neuromuscular junction upon C1q inhibition. Furthermore, anti-C1q treatment reduced Nf-L levels in CSF and plasma of $SOD1^{G93A}$ mice, marking the reduction in neuronal damage in this model. These data indicate that anti-C1q therapy preserves synapses of the neuromuscular junction and reduces neuronal damage in an ALS model. A Phase 2 study of ANX005 anti-C1q therapy in ALS patients is ongoing (clinical trials.gov NCT04569435).

Disclosures: **V. Mathur:** A. Employment/Salary (full or part-time):; Annexon Biosciences. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Annexon Biosciences. **Y. Andrews-Zwilling:** A. Employment/Salary (full or part-time):; Annexon Biosciences. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Annexon Biosciences. **L. Kuhn:** A. Employment/Salary (full or part-time):; Annexon Biosciences. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Annexon Biosciences. **S. Sankaranarayanan:** A. Employment/Salary (full or part-time):; Annexon Biosciences. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Annexon Biosciences. **E. Cahir-McFarland:** A. Employment/Salary (full or part-time):; Annexon Biosciences. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Annexon Biosciences. **T. Yednock:** A. Employment/Salary (full or part-time):; Annexon Biosciences. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Annexon Biosciences.

Poster

P293. Motor Neuron Disease: Therapeutics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P293.08

Topic: C.06. Neuromuscular Diseases

Support: CIRM CLIN2-09284

Title: Long-term safety and survival of GDNF-secreting neural progenitors transplanted into the spinal cord of patients with ALS

Authors: P. AVALOS[1], R. H. BALOH[2], P. JOHNSON[3], R. LEWIS[2], K. ROXAS[1], G. LAWLESS[1], P. ALLRED[2], W. TOURTELLOTTE[4], E. MACKLIN[5], *C. N. SVENDSEN[1]; [1]Regenerative Med. Inst., [2]Neurol., Cedars-Sinai Med. Ctr., Los Angeles, CA; [3]Cedars-Sinai Med. Ctr., Spine Center, CA; [4]Pathology and Lab. Med., Cedars-Sinai Med. Ctr., Los Angeles, CA; [5]Biostatistics, Massachusetts Gen. Hosp., Boston, MA

Abstract: Amyotrophic Lateral Sclerosis (ALS) is a progressive neurodegenerative disease involving motor neuron loss that leads to paralysis and death within 3-5 years of diagnosis, with no effective treatments or cures. Dysfunctional astrocytes may contribute to motor neuron loss, while the growth factor glial cell line-derived neurotrophic factor (GDNF) provides motor neuron protection. Human fetal cortical-derived neural progenitor cells were expanded under cGMP and transduced with lentivirus encoding GDNF (CNS10-NPC-GDNF). Following transplantation, these cells differentiated into astrocytes, released GDNF and protected spinal

cord motor neurons in preclinical animal studies. CNS10-NPC-GDNF were then unilaterally transplanted into the lumbar spinal cord of 18 ALS subjects in a Phase 1/2a dose escalation study using a new minimally invasive surgical device with the primary outcome being safety at one year with secondary outcomes including muscle strength preservation in the treated versus non-treated leg. Muscle strength was protected in the treated leg over time, which reached significance in the low dose cohort at the 12-month time point. Asymptomatic benign neuromas at the dorsal root entry zone of some transplant sites were incidental post-mortem findings in some cases. 9 of 14 subjects who have died due to disease progression have been analyzed and have unilateral transplants releasing GDNF in the spinal cord. In a few of the transplant sites, cells were present in the ventral horn but in most cases cells were located in the dorsal regions of the cord. There was no positive or negative effect on the survival of motor neurons. This study reached the primary endpoint of safety and is the first combined gene and cell therapy approach for ALS to show both cell survival and growth factor release for at least 24 months.

Disclosures: P. Avalos: None. **R.H. Baloh:** None. **P. Johnson:** None. **R. Lewis:** None. **K. roxas:** None. **G. Lawless:** None. **P. Allred:** None. **W. Tourtellotte:** None. **E. Macklin:** None. **C.N. Svendsen:** None.

Poster

P293. Motor Neuron Disease: Therapeutics

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Program #/Poster #: P293.09

Topic: C.06. Neuromuscular Diseases

Support: R21NS081182
R01NS097903
R37NS033123
U01NS103883

Title: Normalization of C9ORF72 pathology in vivo by targeting Staufen 1

Authors: *D. R. SCOLES, S. PAUL, W. DANSITHONG, M. GANDELMAN, C. T. NELSON, K. P. FIGUEROA, S. M. PULST;
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Abstract: Expansion mutations in the *C9ORF72* G4C2 repeat are causative of both amyotrophic lateral sclerosis (ALS) and frontotemporal dementia (FTD), two devastating diseases affecting a significant segment of the population. We have previously shown elevated mTOR and other autophagy-related proteins in fibroblast cells derived from patients with ALS/FTD and other neurodegenerative diseases, associated with overabundance of the RNA binding protein Staufen1 (STAU1). The objectives of this study are to determine if STAU1 functions mechanistically in

regulating mTOR expression and if targeting STAU1 could restore C9ORF72-related phenotypes *in vivo*. Previous studies had demonstrated that STAU1 interactions with 5'-UTRs of mRNAs enhance translation. We performed blot overlay assays demonstrating that STAU1 directly interacted with the 5'-UTR of mTOR. Following this, we established a reporter assay with the mTOR 5'-UTR upstream of luciferase, and consistent with increased translation, STAU1 overexpression increased the luciferase signal while the mRNA abundance remained constant. Next, to determine whether reducing STAU1 abundance could modify C9ORF72 phenotypes *in vivo*, we crossed C9ORF72-BAC mice with *Stau1* knockout mice (*C9ORF72/Stau1+/-)*. We found that in 16-week-old *C9ORF72/Stau1+/-* mice spinal cord neurodegeneration was attenuated, as indicated by restored levels of the neurotransmitter Chat, the pan-neuronal marker NeuN and the astrocyte reactivity marker Gfap. In addition, we observed that *Stau1* haploinsufficiency restored markers of defective autophagy, including mTOR, p-mTOR, p62 and LC3-II. Histological analysis of spinal cords revealed polyGA, a product of repeat associated non-AUG translation (RAN translation) that accompanies C4C2 repeat expansion in C9ORF72, showed the typical aggregates and increased immunoreactivity in both neurons and neuropil, features that were absent in mice haploinsufficient for *Stau1*. We conclude that Stau1 overabundance leads to abnormal autophagy and related toxic pathways associated with DPR proteinopathy that can be restored by targeting Stau1 expression. These data support STAU1 as a therapeutic target for C9ORF72-related FTD or ALS as well as other neurodegenerative diseases in which STAU1 is overabundant.

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Poster

P293. Motor Neuron Disease: Therapeutics

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Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P293.10

Topic: C.06. Neuromuscular Diseases

Support: R01AG068130
R01NS093416
R25GM109441

Title: GCPII inhibition prolongs survival and delays muscle mass and function loss in aged C57BL6/J mice

Authors: *C. TALLON[1], T. R. JOE[2], R. WISEMAN[3], A. G. THOMAS[4], B. S. SLUSHER[5]; [1]Neurol., [2]Cell Biol., [3]Pharmacol. and Mol. Sci., Johns Hopkins Univ., Baltimore, MD; [4]Johns Hopkins Drug Discovery, Johns Hopkins Univ. Sch. of Med., Baltimore, MD; [5]Johns Hopkins Drug Discovery, Baltimore, MD

Abstract: Sarcopenia is the progressive loss of muscle mass and function and is a natural part of the aging process. Sarcopenia can contribute to frailty and increase the likelihood of falls and fractures in older adults. With our aging population, sarcopenia is predicted to have significant negative economic ramifications, and thus being able to slow the process would be beneficial. Although the precise mechanisms leading to sarcopenia is currently an area of active research, increased inflammation has been reported in both aging human and mouse muscle tissues and is thought to contribute towards sarcopenia. Our lab has observed elevated expression of the enzyme glutamate carboxypeptidase II (GCPII) in aged mouse muscle that is specifically associated with infiltrating activated macrophages. GCPII cleaves N-acetylaspartylglutamate (NAAG) into NAA and glutamate and its activity has been observed to regulate glutamatergic transmission and drive synaptic pruning at the neuromuscular junction (NMJ) during development. Our lab recently observed a significant delay of muscle function loss and NMJ denervation in an ALS mouse model using the potent GCPII inhibitor 2-(Phosphonomethyl)-pentanedioic acid (2-PMPA). To determine whether 2-PMPA treatment could delay muscle function loss in aged animals, we treated 15-month old C57BL6/J mice with 100mg/kg of 2-PMPA IP daily for 5 months and monitored monthly body weight, grip strength, compound muscle action potential (CMAP), open field behavior, end stage muscle weights and survival. Compared to vehicle treated mice, we observed a significant delay in body weight loss and grip strength after 1 and 4 months on 2-PMPA therapy. 2-PMPA treatment also delayed CMAP amplitude decline, with treated animals having 20% greater values after 4 months of treatment, but the effect did not reach significance. 2-PMPA-treated animals also exhibited elevated ambulation in the open field test after 4 months of treatment. After 5 months of treatment, gastrocnemius and bicep muscle weights were significantly higher in 2-PMPA treated animals than vehicle treated mice. Most noteworthy, 2-PMPA treatment significantly prolonged survival, with survival of 8% versus 58% of vehicle and 2PMPA-treated mice, respectively, after 5 months of treatment. These data support GCPII inhibition as a potential therapeutic strategy to delay muscle function loss related to aging and improve survival.

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Poster

P300. Mechanisms of Neurotoxicity V

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P300.01

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH, NINDS 5R01NS115126
Mayo Clinic Center for Biomedical Discovery
Mayo Clinic Center for MS and Autoimmune Neurology

Title: Identification of neuronal autoantigens in patients with progressive multiple sclerosis

Authors: P. KABIRAJ, B. CLARKSON, R. JOHNSON, *C. HOWE;
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Abstract: Multiple sclerosis (MS) is one of the leading causes of neurological deficits in the US. While current therapeutic options for controlling the relapsing-remitting phase of the disease are effective and widely employed, most patients with MS still eventually enter a progressive phase that is marked by permanent, irreversible loss of neurologic function. At present, there are no drugs or therapeutic strategies available to halt this insidious process. It has long been known that CD8+ T cells predominate in active MS lesions and are associated with axon injury and disease progression. However, the pathogenic role of these cells is still poorly understood. Based on extensive animal model data we have proposed that anti-neuronal CD8+ T cells that specifically target demyelinated axons perpetrate the damage and loss of neural circuitry that underlies disease progression in MS. In order to identify the HLA class I antigens targeted by these CD8+ T cells we hypothesized that B cell responses facilitated by T cell help will result in the presence of serum autoantibodies directed against the same protein antigens recognized by the effector T cells. Using liquid-chromatography mass spectrometry (LCMS) and antibody-mediated identification of autoantigens (AMIDA), we have started to identify the repertoire of neuronal antigens recognized by the immune system in patients with progressive MS. We have generated induced pluripotent stem cell clones from 6 MS patients and 4 healthy controls and we have generated neural stem cells and induced neurons from this material. These patient-specific neurons were stimulated with IFNg for 72 hr to induce upregulation of MHC class I molecules, as we have previously described. Following lysis, MHC class I molecules were immunoprecipitated with the W6/32 clone to capture HLA-A, B, and C molecules. Bound peptides were eluted and analyzed via LCMS. In parallel, lysates from iPSC-derived neurons and from resected human brain tissue were adsorbed against human IVIg and then incubated with biotinylated immunoglobulin isolated from a pool of serum from 3 patients with MS or from control subjects. Following immunoprecipitation on Protein G HP SpinTrap columns, the bound proteins were eluted and analyzed by mass spectrometry. To date we have identified 5 unique peptides and we are now testing CD8+ T cell responses against these peptides presented by autologous protein antigen-fed dendritic cells.

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Poster

P300. Mechanisms of Neurotoxicity V

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Program #/Poster #: P300.02

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH MH109382

Title: Role of PERK-B in INSTI-induced adverse effects in HAND

Authors: *S. GHURA, S. BOND, C. AKAY-ESPINOZA, K. L. JORDAN-SCIUTTO;
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Abstract: Combined antiretroviral therapies (ART) have increased the life expectancy of people living with HIV (PLWH), however about 50% of PLWH present with a spectrum of neurocognitive impairments termed HIV-associated neurocognitive disorder (HAND). Although the exact mechanisms remain unknown, clinical data indicate loss of gray and white matter in specific brain regions critical for learning and memory. These alterations are induced by several factors including HIV itself and ART. We have previously demonstrated that ART therapeutics, specifically integrase inhibitors (INSTIs), that are widely used as first-line therapeutics to control HIV infection, are neurotoxic and inhibit differentiation of oligodendrocytes. At the cellular level, HAND is often characterized by increased endoplasmic reticulum (ER) stress which leads to activation of the unfolded protein response (UPR). Acute UPR activation, a cytoprotective response, aims to alleviate ER stress, whereas chronic UPR activation can be maladaptive. Importantly, we have demonstrated that ART-induced neurotoxicity and detrimental effects on oligodendrocytes are mediated by UPR. A downstream event in UPR is the activation of ER stress sensor PKR-like ER kinase (PERK), which has been implicated in several neurodegenerative disorders, including HAND. Recent studies reported three non-synonymous SNPs of PERK occurs in 31% of the general population. These SNPs are genetically linked forming haplotype B (PERK-B) distinct from the more common haplotype (61%), PERK-A. PERK-B has a putative higher kinase activity, potentially predisposing individuals to neuronal dysfunction. Notably, PERK-B has been associated with an increased risk for progressive supranuclear palsy, a neurodegenerative disease. Data from our lab demonstrates that HAND patients who express PERK-B are associated with worse cognitive global deficit scores. However, our understanding of the role of PERK and effects of PERK haplotypes in mechanisms underpinning cellular dysfunction in HAND remains elusive. In the current study, we hypothesize that PERK plays a critical role in ART-mediated UPR in the context of HAND. More specifically, PERK-B augments INSTI-induced UPR activation in neurons and oligodendrocytes, contributing to HAND pathology. We have developed a novel mouse model that expresses PERK-B. *In vitro* and *in vivo* data from this model supports the role of PERK-B in HAND pathology. The novel mouse model will examine how the different haplotypes modulate cell-specific mechanisms underlying HAND pathology which could validate if PERK-B is a risk factor for worse cognitive impairment in patients with HAND.

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Poster

P300. Mechanisms of Neurotoxicity V

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Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P300.03

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: Families for SCN2A foundation
SLC6A1 connect

Title: In vitro characterization of mitochondrial and metabolic activities in patients presenting with SLC6A1 mutations associated epileptic encephalopathy

Authors: *F. S. ROUSSEL, C. N. DENNYS, A. HARTLAUB, S. SINHA RAY, J. A. SIERRA-DELGADO, S. B. LIKHITE, A. BRADBURY, K. C. MEYER;
Ctr. For Gene Therapy, Res. Inst. Nationwide Children's Hosp., Columbus, OH

Abstract: Mutations in the SLC6A1 gene cause an epileptic encephalopathy, characterized by absence, atonic and myoclonic seizures. Patients also present with developmental delay, EEG abnormalities, language impairment and intellectual disability. Solute carrier family 6 member 1 (SLC6A1) encodes for the GABA transporter 1 (GAT-1) and is mainly expressed in the brain. The protein is responsible for the reuptake of GABA into presynaptic neurons and glia, clearing the extracellular space and modulating neurotransmission. Since little is known about the disease mechanisms and the cell types involved, the development of in vitro models to study the disease in the context of patient cell lines and for testing of new therapeutics is urgently needed. We have previously developed a rapid reprogramming method to convert patient skin fibroblasts into induced Neuronal Progenitors Cells (iNPC) that can further be differentiated into induced Astrocytes (iAs). Here we utilized this methodology in addition to a rapid chemical conversion method to produce neurons (iNs) from patient fibroblasts to investigate the impact of SLC6A1 mutations in both iNs and iAs. These cells were analyzed for GAT-1 levels, markers of neurological stress (elevated BIP, and dysfunctional mitochondria) as well as GABA production and secretion. Interestingly, we found that iNPCs, iAs, and iNs had unique metabolic requirements to survive in vitro, suggesting sensitivity to select nutrients. Morphological assessment of iNs showed normal neuronal morphology, however iAs had a distinct morphology indicative of activation. Co-culturing patient iAs with healthy mouse GFP+ neurons lead to reduced neuron survival. Next, we assessed the mitochondrial and metabolic profiles in patient iAs to determine if aberrant metabolism contributed to iAs disease mechanisms. Seahorse analysis indicated that cellular glycolysis and mitochondria activity do not differ significantly from healthy controls. Combined, this data suggests SLC6A1 iAs contribute to disease pathology in an unidentified mechanism. Further investigations to understand underlying disease cause and the role of iAs and iNs in the disease physiopathology will assist in the development for a novel treatment for patients with SLC6A1 mutations.

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Poster

P300. Mechanisms of Neurotoxicity V

Location: Virtual Only

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Program #/Poster #: P300.04

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: R01CA205166
R01CA231267

Title: Neurotoxicity associated with a novel anticancer drug, beta-lapachone (ARQ-501).

Authors: *H. XIA[1], A. BEHROUZI[1], H. RODRIGUES DE OLIVEIRA[2], J. C. FEHRENBACHER[1];
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Abstract: Chemotherapy-induced peripheral neuropathy (CIPN) is a major dose-limiting side effect of many efficacious anticancer drugs and there currently are limited therapeutics to alleviate the symptoms of CIPN. Despite this knowledge, many investigational drug studies still do not characterize the incidence of CIPN in preclinical or clinical studies. Our previous work identified that oxidative DNA damage within sensory neurons contributes to CIPN, therefore drugs that enhance the generation of reactive oxygen species (ROS) within sensory neurons may induce neurotoxicity. Recent studies have identified NAD(P)H:quinone oxidoreductase 1 (NQO1) as a potential drug target for the treatment of various cancer types. A class of investigational quinones, including beta-lapachone, are bioactivated by NQO1 and can thereby cause futile redox cycling between the quinone and hydroquinone species, generating high amounts of ROS. There are currently multiple clinical trials investigating the use of beta-lapachone as a therapeutic agent; however, the potential for neurotoxicity has not been assessed in this class of drugs. In this study, we aim to determine whether beta-lapachone induces neurotoxicity in sensory neurons.
Primary cultures derived from dorsal root ganglion (DRG) of male Sprague Dawley rats (150g) were used to assess NQO1 expression in sensory neurons and to determine whether beta-lapachone modulates sensory neuron function. Cultures (7d post-harvest) were treated with 0.3, 3, 10, or 30 µM beta-lapachone for 2 hours, after which normal media was replenished. Calcitonin gene-related peptide (CGRP) release was stimulated by capsaicin (30 nM) 24h after the initial beta-lapachone exposure. Sister wells were lysed for protein and immunoblotted for pH2AX, or PFA-fixed for immunofluorescence imaging. Neurite outgrowth analysis was performed on fixed cultures using the neurite outgrowth algorithm by ImageXpress (Molecular

Devices).
NQO1 is expressed in cultured small-diameter sensory neurons and in small- and large-diameter neuronal soma of DRG from male adult mice. Treatment of neuronal cultures with beta-lapachone diminished neuronal sensitivity and the total content of CGRP in a concentration-dependent manner. Treatment with beta-lapachone also altered the morphology of sensory neurons in culture, decreasing neurite length in a concentration-dependent manner.
Our data demonstrate that NQO1 is expressed in sensory neurons of the DRG and drugs that target NQO1 are likely to induce CIPN in patients.

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Poster

P300. Mechanisms of Neurotoxicity V

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Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P300.05

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Title: N-acetyl-l-cysteine improves cognitive performance via kynurenine pathway in mice lead poisoning during lactation

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Abstract: The immature brain is singularly sensitive to lead (Pb) neurotoxicity, compromising the cognitive and behavioral attributes which persist even later in adulthood. Several mechanisms have been implicated in the Pb cognitive impairment including calcium homeostasis, oxidative stress generation, changes in neurotransmitters levels and recently, we have demonstrated the kynurenine pathway (KP) participation, specifically, high levels of kynurenic acid (KYNA) were accompanied by cognitive impairment. KYNA is considered as a neuromodulator that has an important role in cognitive processes due to is an endogenous antagonist of NMDA and α7-nicotinic acetylcholine receptors, representing a good target to be modulated. In this line, it has been shown that N-acetylcysteine (NAC) affects the KP, diminishing kynurenine aminotransferase activity (KAT) and decreasing KYNA levels accompanied by an improvement in cognitive parameters. The aim of this work was to evaluate the effect of NAC on Pb poisoning during lactation in KYNA levels, KAT activity and cognitive parameters. Mice pups were exposed via their dams' drinking water (lead acetate 500ppm) from PND0 to PND23. NAC group received 300 mg/day through food. Pb-NAC group received Pb trough drinking water and were fed with NAC at the same time. Control group received normal

water. The long-term memory was evaluated through buried food probe at PND 60. KAT activity and KYNA levels were evaluated in brain by HPLC. As results, Pb group showed a significant increase in time looking for food, compared to controls. This increase was abolished when mice were treated with NAC at the same time as with Pb. KYNA levels increased in the presence of Pb, but co-treatment with NAC decreased them. KAT activity decreased in the NAC and Pb-NAC groups. These results suggest that NAC can be a good treatment to decrease KYNA levels in Pb poisoning where altered cognitive parameters can be improved.

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Poster

P300. Mechanisms of Neurotoxicity V

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Program #/Poster #: P300.06

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH NINDS R25NS114309
NSF DBI2051440

Title: The effect of methotrexate treatment on neuroinflammation gene expression in pediatric cancer patients

Authors: G. SHEETS[1,2], R. MANDAL[3,2], B. MOORE[2], A. UMRIGAR[2], E. BEATON[1], M. STARK[4], M. NORMAN[2], P. PRASAD[4], J. ZABALETA[2], L. LI[2], C. HICKS[2], *F. TSIEN[2]; [1]Univ. of New Orleans, New Orleans, LA; [2]Louisiana State Univ. Hlth. Sci. Ctr., New Orleans, LA; [3]Tulane Univ., New Orleans, LA; [4]Children's Hosp. New Orleans, New Orleans, LA

Abstract: Methotrexate, a structural analogue of folic acid, is one of the most effective and widely used drugs for treating various types of cancer, particularly leukemias and lymphomas. Methotrexate treatment in pediatric patients has been associated with the long-lasting development of detrimental neurological and psychosocial sequela following cancer survival. These deficits, termed late effects, persist after the methotrexate exposure and may include abnormal behavior such as unusual aggression, problems with executive functioning and processing speed as well as mental disorders like ADHD, depression, and anxiety. Our study includes a retrospective review of neurological and psychosocial evaluations from pediatric cancer survivors enrolled in the Late Effects Clinic at Children’s Hospital New Orleans, Louisiana (CHNOLA). We also use targeted RNA microarray and bioinformatic analysis of neuroinflammation genes in brain specimens obtained from deceased patients from the Pathology Department at CHNOLA. Results from the present study will provide information

regarding gene-environment interactions and thus reveal candidate risk genes and pathways contributing to neurocognitive and psychiatric late effects.

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Poster

P300. Mechanisms of Neurotoxicity V

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Program #/Poster #: P300.07

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Title: A novel multimodal compound, AR1001 attenuates multimodal and cognitive dysfunction in a 3xTg mouse model of Alzheimer's disease

Authors: *S. KIM[1], H. KIM[2], B. KANG[3], S. KANG[4], J. JUNG[5];
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Abstract: Alzheimer's disease is a neurodegenerative disease characterized by the production of β-amyloid proteins and hyperphosphorylation of tau protein. Inflammation and apoptotic severity were highly correlated with earlier age at onset of Alzheimer's disease and were also associated with cognitive decline. This study aims to examine whether the AR1001 could modulate CREB, autophagy and Wnt signal, cognitive deficit and take neuroprotective effects in 3x Tg mouse model of Alzheimer's disease. AR1001 protected their cognitive impairment and improved their memory function by daily oral administration for 60 days consecutively. In addition, AR1001 alleviated the acute inflammation induced by oAβ. Using a QDoc-labeled aptamer and molecular beacon detected to whole brain, the endogenous long noncoding miRNA, the constitutive U6 miRNA, and miR-106b and miR-7 could be detected in the cytoplasm of a normal mouse brain, cultured hippocampal neurons, and an Alzheimer's disease model mouse brain, respectively. The fluorescence recovered by aptamer and molecular beacon correlated highly with the amount of miR-7 and miR-106b, as measured by confocal microscope. We propose novel CLARITY as a simple, fast, inexpensive, and sensitive method for miRNA detection, with a very low background, which could be applied to a variety of research or diagnostic purposes. By simultaneously achieving ∼50-fold signal amplification and offers a robust and versatile approach for detecting miRNAs in situ, including in thick tissues where high background undermines the performance of unamplified FISH. Furthermore, AR1001 played a role in the down-regulation of caspase-3/9 proteins, MMP2/9 and reduced the early apoptosis and oAβ deposits that normally followed APP processing in 3xTg AD mice. The results of this study

showed that the treatment of AR1001 had neuroprotective effects in oAβ-deposited 3x Tg AD mouse model.

Disclosures: **S. Kim:** A. Employment/Salary (full or part-time):; FULL. **H. Kim:** None. **B. Kang:** A. Employment/Salary (full or part-time):; full. **S. Kang:** A. Employment/Salary (full or part-time):; salary (full). **J. Jung:** A. Employment/Salary (full or part-time):; employment.

Poster

P300. Mechanisms of Neurotoxicity V

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P300.08

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: Canadian Institute of Health Research (CIHR)
Natural Sciences and Engineering Research Council (NSERC)

Title: Ketamine-induced neurotoxicity mediated through endoplasmic reticulum stress

Authors: *N. RIGG[1], F. A. ABU-HIJLEH[1], V. PATEL[2], R. K. MISHRA[3];
[1]Psychiatry and Behavioural Neurosci., McMaster Univ. Neurosci. Grad. Program - Minds, Hamilton, ON, Canada; [2]Psychiatry and Behavioural Neurosci., McMaster Univ., Hamilton, ON, Canada; [3]Psychiatry and Behavioural Neurosci., McMaster Univ. Hsc-4N 78, Hamilton, ON, Canada

Abstract: Ketamine has traditionally been used as a dissociative anesthetic. However, there is growing concern over the increased use of ketamine in recreational and therapeutic settings due to potential toxic effects. Recent studies have demonstrated that ketamine is cytotoxic in several cell lines, such as fibroblasts, hepatocytes, uroepithelial cells, and hippocampal cells. Ketamine has been shown to cause disturbances in calcium signalling, increased reactive oxygen species (ROS) production, and mitochondrial dysfunction leading to apoptosis. However, it is not yet clear whether endoplasmic reticulum (ER) stress plays a role in ketamine neurotoxicity in striatal neurons. Disturbances to ER homeostasis, such as calcium dysregulation and ROS, can initiate ER-mediated cell death, which is characteristic of many neurodegenerative diseases. Thus, the purpose of this study was to determine whether ketamine is neurotoxic through an ER stress-dependent pathway and to elucidate the underlying mechanism. Mouse striatal cells were treated with ketamine (0.01mM, 0.1mM, 1mM) or DMEM for 12 to 72 hrs. Following treatment, intracellular calcium was measured, and changes in the expression of ER stress markers were evaluated using RT-qPCR. MTT results revealed that ketamine (1mM) was toxic to striatal cells, as demonstrated by a time-dependent decrease in cell viability after 24 hrs compared to controls. Ketamine reduced basal intracellular calcium levels in a dose-dependent manner at all times and induced a decreased response to KCl depolarization compared with control cells after 24 hrs.

Ketamine upregulated pro-apoptotic ER stress markers', which quickly favoured pro-apoptotic signalling over cell survival. Initial upregulation of glucose-regulated protein-78 (GRP78), the master regulator of ER stress, was observed, while prolonged exposure showed decreased expression. Increased expression of pro-apoptotic C/EBP homologous protein (CHOP) observed throughout the experiment demonstrates ER-stress-mediated apoptosis. Further, the expression of ER-resident neurotrophic factors (MANF and CDNF), which aid in cell survival during stress, decreased after 48 hrs of treatment. Taken together, our results propose that chronic ketamine exposure impairs calcium signalling, inducing severe ER stress and triggering a dynamic neuronal response resulting in ER stress-dependent apoptosis, which may be mediated through PERK- elF2a-ATF4 signalling. These results further support the need for caution when prescribing this drug.

Disclosures: N. Rigg: None. **F.A. Abu-Hijleh:** None. **V. Patel:** None. **R.K. Mishra:** None.

Poster

P300. Mechanisms of Neurotoxicity V

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P300.09

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Title: A mouse model of adrenomyeloneuropathy develops progressive CNS pathology

Authors: *C. KAYATEKIN, K. SINGH, K. KEEFE, R. CHIANG, M. HANUS, J. GANS, W. HUANG, D. KUMAR, J. ESCOBEDO, E. TEEPLE, Z. ZHAO, D. WANG, A. KLOSS, J. PROTO, T. GLADYSHEVA;
Sanofi, Massachusetts, MA

Abstract: Adrenoleukodystrophy, or ALD, is a rare neurometabolic disease that is caused by mutations in the *ABCD1* gene, which encodes for a transporter responsible for shuttling very long chain fatty acids (VLCFAs) into the peroxisome for degradation. It is a debilitating disorder, which has a spectrum of clinical presentations. The more severe form is a rapidly progressing demyelinating disease (cerebral ALD or CALD with a life expectancy of 2-4 years after onset), which often strikes in childhood and can be potentially halted by a successful hematopoietic stem cell transplant from donor without an *ABCD1* mutation. Another form, adrenomyeloneuropathy or AMN, is a slower progressing degeneration of the spinal cord. While an *ABCD1* knockout mouse model exists, it does not develop the pathological hallmarks of ALD. To identify disease phenotypes in this mouse model that can enable therapeutics development, we performed an extensive behavioral, biochemical, and single nuclei transcriptional analysis of the *ABCD1* knockout mouse. We found evidence of progressive accumulation of VLCFAs in the spinal cord, subtle changes in neuronal populations, and white matter pathology that advances with age. These findings support the hypothesis that the buildup of VLCFA in the CNS

contributes to dysfunction and disease, while providing endpoints for disease modifying investigational therapies to treat ALD.

Disclosures: C. Kayatekin: A. Employment/Salary (full or part-time):; Sanofi. **K. Singh:** A. Employment/Salary (full or part-time):; Sanofi. **K. Keefe:** A. Employment/Salary (full or part-time):; Sanofi. **R. Chiang:** A. Employment/Salary (full or part-time):; Sanofi. **M. Hanus:** A. Employment/Salary (full or part-time):; Sanofi. **J. Gans:** A. Employment/Salary (full or part-time):; Sanofi. **W. Huang:** A. Employment/Salary (full or part-time):; Sanofi. **D. Kumar:** A. Employment/Salary (full or part-time):; Sanofi. **J. Escobedo:** A. Employment/Salary (full or part-time):; Sanofi. **E. Teeple:** A. Employment/Salary (full or part-time):; Sanofi. **Z. Zhao:** A. Employment/Salary (full or part-time):; Sanofi. **D. Wang:** A. Employment/Salary (full or part-time):; Sanofi. **A. Kloss:** A. Employment/Salary (full or part-time):; Sanofi. **J. Proto:** A. Employment/Salary (full or part-time):; Sanofi. **T. Gladysheva:** A. Employment/Salary (full or part-time):; Sanofi.

Poster

P300. Mechanisms of Neurotoxicity V

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P300.10

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH Grant R01CA242158
Mayo Clinic Breast SPORE-R50CA116201
Regenerative Medicine Minnesota -RMM 091718 DS 005
Association for Cancer Research-Bosarge Family Foundation-Waun Ki Hong Scholar Award for Regenerative Cancer Medicine -19-40-60-OLIV

Title: Nicotinamide Mononucleotide Protects Against Cisplatin-Induced Mitochondrial Dysfunction and Increases Survival of Cortical Neurons Derived From hiPSC

Authors: *M. RASHID, K.-H. YOO, A. OLIVEROS A, M.-H. JANG;
Mayo Clin., Rochester, MN

Abstract: Mitochondrial dysfunction is thought to play a critical role in a variety of neurological disorders. Chemotherapy-induced cognitive impairment (CICI) is often reported as a neurotoxic side effect of chemotherapy and no effective treatments currently available. Recently, we demonstrated that the NAD+ precursor nicotinamide mononucleotide (NMN) prevents cisplatin, a frontline platinum-based chemotherapy, from inducing impairments in human cortical neurons derived from iPSCs *in vitro*, as well as cognitive deficits in a mouse model of CICI *in vivo*. Despite this, NMN's potential to protect against cisplatin-induced mitochondrial dysfunction remains to be studied. Here we demonstrate that NMN prevents against cisplatin-induced

mitochondrial dysfunction in human cortical neurons derived from hiPSCs. Treatment with cisplatin significantly increased oxidative DNA damage and mitochondrial reactive oxygen species generation (mROS) in our cortical cultures. Furthermore, we found that cisplatin increased the colocalization of the mitochondrial marker TOMM20 with the lysosome marker LAMP1. Interestingly, we detected LAMP1 colocalization with the early and late endosome markers RAB4B and RAB7A, respectively. Additionally, there was colocalization of TOMM20 with LC3B (autophagosome marker). In parallel, cisplatin decreased intracellular ATP content and diminished cell viability. Additionally, cisplatin treatment impaired neuronal morphogenesis, and reduced the expression of the pre- and post- synaptic markers PSD95 and SYP in our hiPSCs derived cortical neurons. These changes were consistent with loss of mitochondrial membrane potential, and loss of cristae membrane and matrix swelling. Importantly, pretreatment with NMN prevented these adverse effects. Taken together, our results suggest that mitochondrial dysfunction and increased oxidative stress are involved in cisplatin chemotherapy-induced cognitive impairment. In support of our recent *in vivo* studies, NMN increased survival in human cortical neurons derived from hiPSCs, while concomitantly preventing cisplatin-induced mitochondrial dysfunction as evidenced by decreasing oxidative stress and preserving mitochondrial structure. Therapeutic agents, such as NMN, may be effective in mitigating cisplatin-induced deleterious effects on mitochondria.

Disclosures: M. Rashid: None. **K. Yoo:** None. **A. Oliveros A:** None. **M. Jang:** None.

Poster

P327. The Blood Brain Barrier in Ischemia and Hemorrhage

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P327.01

Topic: C.08. Ischemia

Support: James and Esther King Biomedical Research Grant 9JK08

Title: Tumor necrosis factor-α inhibition limits hematoma expansion following intracerebral hemorrhage in chronic nicotine-exposed rats.

Authors: *A. K. REHNI, S. CHO, A. P. RAVAL, M. A. PEREZ-PINZON, K. R. DAVE; Neurol., Univ. of Miami Miller Sch. of Med., Miami, FL

Abstract: Spontaneous intracerebral hemorrhage (sICH) is a serious form of hemorrhagic stroke that causes enormous mortality and morbidity worldwide. Tobacco use is an established risk factor of sICH and worsens sICH-outcome. We have shown that chronic nicotine-exposed rats of both sexes display an increased hematoma growth post-sICH. Chronic nicotine exposure increases the levels of tumor necrosis factor-α (TNF-α) in brain microvessels and increased levels of TNF-α receptors[1] are associated with increased risk of sICH. Therefore, we

hypothesized that chronic nicotine exposure-induced increase in TNF-α causes increased hematoma expansion. To test this hypothesis, we evaluated the effect of pharmacological inhibition of TNF-α on hematoma expansion following sICH in nicotine-exposed rats. Rats were treated with nicotine (4.5 mg / kg / day b.w.) using osmotic pumps for 2-3 weeks. Young rats of both sexes were randomized between different treatment groups. Saline (vehicle) or etanercept (TNF-α inhibitor) (6.24 mg/ kg b.w., i.p.) was administered 5 and 2 days prior to the onset of sICH. sICH was induced using intra striatal administration of collagenase in rats. sICH in females was induced during the diestrus stage of estrous cycle. After 24 hours, neurological scores were assessed. Isolated brains were sectioned to measure hematoma volume by a blinded investigator. The hematoma volume of male rats belonging to the TNF-α inhibitor treatment group (90±8 mm^3, n=8) was significantly lower ($p<0.05$) than the control group (135±16 mm^3, n=8). Moreover, the neurological score of male rats in TNF-α inhibitor treatment group (8.5 ± 0.6) was also significantly lower ($p<0.005$) than the control group (10.6 ± 0.3). The hematoma volume observed in the TNF-α inhibitor treated females (82±11 mm^3, n=8) was significantly lower ($p<0.05$) than the controls (127±11 mm^3, n=8). Further, neurological score noted in the TNF-α inhibitor treatment female group (7.9±0.5) was significantly lower ($p<0.001$) than the control group (10.4±0.3). Our results show that TNF-α inhibition limits hematoma growth in previously nicotine-exposed rats suffering from sICH. Therefore, we conclude that TNF-α activation may be involved in mediating chronic nicotine exposure-induced increase in hematoma expansion. This information will help us understand the pathophysiology of increased hematoma growth in sICH patients who are tobacco users and thus may contribute towards identification of future therapeutic strategies. Support: James and Esther King Biomedical Research Grant 9JK08. References: 1) Stroke. 2017;48(10):2710-2715.

Disclosures: A.K. Rehni: A. Employment/Salary (full or part-time):; University of Miami Miller School of Medicine. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Study staff on James and Esther King Biomedical Research Grant 9JK08. **S. Cho:** A. Employment/Salary (full or part-time):; University of Miami Miller School of Medicine. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Study staff on James and Esther King Biomedical Research Grant 9JK08. **A.P. Raval:** A. Employment/Salary (full or part-time):; University of Miami Miller School of Medicine. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Collaborator on James and Esther King Biomedical Research Grant 9JK08. **M.A. Perez-Pinzon:** A. Employment/Salary (full or part-time):; University of Miami Miller School of Medicine. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Collaborator on James and Esther King Biomedical Research Grant 9JK08. **K.R. Dave:** A. Employment/Salary (full or part-time):; University of Miami Miller School of Medicine. B. Contracted Research/Research Grant (principal

investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Principal Investigator on James and Esther King Biomedical Research Grant 9JK08.

Poster

P327. The Blood Brain Barrier in Ischemia and Hemorrhage

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P327.02

Topic: C.08. Ischemia

Support: James and Esther King Biomedical Research Grant 9JK08

Title: Chronic nicotine increases both blood-brain barrier permeability and inflammation in rats of both sexes

Authors: *S. CHO, A. K. REHNI, A. P. RAVAL, M. A. PEREZ-PINZON, K. R. DAVE; Neurol., Univ. of Miami Miller Sch. of Med., Miami, FL

Abstract: As one of the leading causes of preventable death, tobacco use has been found in epidemiological studies to not only increase the risk of but also worsen outcomes following intracerebral hemorrhage (ICH) in both men and women. Previously, we showed that nicotine treatment increased hematoma volumes and neurological deficits after collagenase-induced ICH in both male and female rats. We hypothesize that chronic nicotine exposure causes greater hematoma growth in ICH by increasing blood-brain barrier (BBB) permeability through increased inflammation and decreased tight junction proteins. To test this hypothesis, young Sprague-Dawley rats of both sexes were subcutaneously implanted with an osmotic pump containing either nicotine (4.5 mg / kg / day) or saline (vehicle group). After two to three weeks, animals were perfused with saline, and microvessels were isolated from brain tissue. Brain tissue from female animals was collected during the diestrus stage to avoid hormonal fluctuation. Tight junction proteins were measured using Western blot and normalized to GAPDH. The rate of hydrogen peroxide production was measured using the Amplex Red hydrogen peroxide assay. We previously showed decreased levels of Claudin 3, Claudin 5, and occludin and increased BBB permeability in nicotine-treated rats compared to saline-treated rats in both males and females. In male animals, PECAM-1 levels were 79% ($p<0.05$, n=7 for both groups) lower in nicotine-treated rats than saline-treated rats. In female rats, ZO-1 and PECAM-1 levels were 51% ($p<0.05$, n=6 for saline, n=5 for nicotine) and 43% ($p<0.05$, n=7 for both groups) lower in nicotine-treated animals than saline-treated animals. In addition, TNFα levels were 102% ($p<0.05$) and 64% ($p<0.05$) higher in nicotine-treated rats than saline-treated rats in males (n=9 for saline, n=8 for nicotine) and females (n=6 for both groups), respectively. In male animals, the rate of H_2O_2 production was 51% ($p<0.05$) higher in nicotine-treated rats (n=8) than saline-treated rats (n=8). In female rats, the rate of H_2O_2 production was not statistically different in

nicotine-treated animals than saline-treated animals (n=10 for saline, n=9 for nicotine). Overall, our results show that chronic nicotine exposure increases BBB permeability by decreasing tight junction proteins and increasing inflammation in animals of both sexes. This observed increased BBB permeability and inflammation may be responsible for the increased ICH damage in nicotine-treated rats.

Disclosures: S. Cho: A. Employment/Salary (full or part-time):; University of Miami Miller School of Medicine. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; James and Esther King Biomedical Research Grant - Study Staff. **A.K. Rehni:** A. Employment/Salary (full or part-time):; University of Miami Miller School of Medicine. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; James and Esther King Biomedical Research Grant - Study Staff. **A.P. Raval:** A. Employment/Salary (full or part-time):; University of Miami Miller Sch. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; James and Esther King Biomedical Research Grant - Collaborator. **M.A. Perez-Pinzon:** A. Employment/Salary (full or part-time):; University of Miami Miller Sch. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; James and Esther King Biomedical Research Grant - Collaborator. **K.R. Dave:** A. Employment/Salary (full or part-time):; University of Miami Miller Sch. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; James and Esther King Biomedical Research Grant - PI.

Poster

P327. The Blood Brain Barrier in Ischemia and Hemorrhage

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P327.03

Topic: C.08. Ischemia

Support: James and Esther King Biomedical Research Grant 9JK08
Lois Pope Neuroscience Summer Fellowship
Testing material was provided by RxMP Therapeutics.

Title: Identification of the mechanism by which red cell microparticles limit hematoma growth following intracerebral hemorrhage

Authors: *S. SAMA[1], S. CHO[1], A. K. REHNI[1], W. JY[2], K. R. DAVE[1];
[1]Peritz Scheinberg Cerebral Vascular Dis. Res. Labs., Univ. of Miami Sch. of Medicine, 1420 NW 9th Ave, NRB/203E, Miami, FL; [2]The Wallace H Coulter Platelet Laboratory, Div. of Hematology/Oncology, Univ. of Miami Sch. of Medicine, 1600 NW 10th Ave, RMSB 7109, R36-A, Miami, FL

Abstract: There is no proven therapy for spontaneous intracerebral hemorrhage (sICH), the deadliest stroke sub-type. As hematoma volume correlates with mortality in survivors, limiting hematoma expansion appears to be an effective therapeutic strategy. However, hemostatic agents used today have thromboembolic and neurological complications that outweigh their benefits. In contrast, red cell-derived microparticles (RMPs) are novel universal hemostatic agents with an excellent safety profile[1] as based on earlier studies. We previously showed that RMPs possess potent hemostatic activity[2] and RMP treatment limits hematoma growth in a rat model of sICH. However, the mechanism of action of RMPs is not well understood. In the present study, we investigated how RMPs influence the intrinsic and extrinsic pathways of secondary hemostasis. To evaluate this, we examined clot formation in human plasma deficient in coagulation factors VII, VIII, IX, XI, and XII in the presence and absence of RMPs (2.8×10^8 particles/mL). Pooled human plasma from 53 donors and factor-deficient plasma were purchased from George King Bio-Medical and mixed to generate plasma that is 85%, 92.5% and 96.25% deficient in each of the five factors, and R-times (clotting times) were determined with thromboelastography. RMP concentration was based on earlier studies, and all experiments were performed in triplicate. We found that RMP treatment decreased R-times in 96.25% (21.2 min to 13.4 min) and 92.5% (17.5 min to 13.3 min) deficiency but not in 85% deficiency of factor VII. For factor VIII-deficient plasma, RMP reduced R-times in 92.5% (22.6 min to 17.7 min) but not in 96.25% or 85% deficiency. For factor IX-deficient plasma, RMP treatment reduced R-times in all three degrees of deficiency: in 96.25% (44.3 min to 27 min), 92.5% (27.8 min to 18.7 min), and 85% (25.3 min to 17 min). We observed a similar trend for factor XI-deficient plasma; RMP treatment reduced R-times in 96.25% (22.8 min to 17.9 min), 92.5% (18.1 min to 15.6 min) and 85% (16.9 min to 13.7 min) deficiency. We found that RMP treatment did not decrease R-times for factor XII in all three tested degrees of deficiency. Overall, we found that RMPs significantly ($p<0.05$, $n=3$ each) reduced R-times for plasma deficient in factors VII, VIII, IX, XI but not XII. We are in the process of evaluating the mechanism by which RMP affects primary hemostasis. Our results indicate that RMPs limit hematoma expansion in sICH by enhancing secondary hemostasis through both intrinsic and extrinsic pathways.
References:1) Journal of Cardiovascular Pharmacology and Therapeutics. 2019;24(5):474-83.
2) Thrombosis and Haemostasis. 2013;110(4):751-60.

Disclosures: S. Sama: B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; University of Miami Lois Pope Neuroscience Summer Fellowship. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); RxMP Therapeutics provided the testing material

for the study., James and Esther King Biomedical Research Grant. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); University of Miami has partial ownership of RxMP Therapeutics. The University of Miami is a co-inventor of 2 US patents related to red-cell microparticles. **S. Cho:** A. Employment/Salary (full or part-time):; University of Miami Miller School of Medicine. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; James and Esther King Biomedical Research Grant - Study Staff. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); RxMP Therapeutics provided the testing material for the study.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); University of Miami has partial ownership of RxMP Therapeutics. The University of Miami is a co-inventor of 2 US patents related to red-cell microparticles. **A.K. Rehni:** A. Employment/Salary (full or part-time):; University of Miami Miller School of Medicine. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; James and Esther King Biomedical Research Grant - Study Staff. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); RxMP Therapeutics provided the testing material for the study.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); University of Miami has partial ownership of RxMP Therapeutics. The University of Miami is a co-inventor of 2 US patents related to red-cell microparticles. **W. Jy:** A. Employment/Salary (full or part-time):; University of Miami Miller School of Medicine. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Dr. Jy received grant support from RxMP Therapeutics.. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); RxMP Therapeutics provided the testing material for the study.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Dr. Jy and the University of Miami has partial ownership of RxMP Therapeutics. Dr. Jy is a co-inventor of 2 US patents related to red-cell microparticles. **K.R. Dave:** A. Employment/Salary (full or part-time):; University of Miami Miller School of Medicine. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; James and Esther King Biomedical Research Grant - PI. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); RxMP Therapeutics provided the testing material for the study.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); The University of Miami has partial ownership of RxMP Therapeutics. The University of Miami is a co-inventor of 2 US patents related to red-cell microparticles..

Poster

P327. The Blood Brain Barrier in Ischemia and Hemorrhage

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P327.04

Topic: C.09.Stroke

Support: NIH Grant HL146574
NIH Grant AG064422
AHA Fellowship 20PRE35210605

Title: Fibroblasts promote blood brain barrier repair and stroke recovery in a TIMP2 dependent manner

Authors: L. XU[1], ***Y. YAO**[2,1];
[1]Univ. of Georgia, Athens, GA; [2]Univ. of South Florida, Tampa, FL

Abstract: Recent studies identified Col1a1$^+$ fibroblast-like cells in large blood vessels in the CNS. To investigate the function of these cells, we generated mutants with reduced fibroblast-like cells (Col1a1-DTR) by crossing the Col1a1-Cre line with DTR mice, which contain a loxP-flanked STOP sequence before diphtheria toxin receptor. The Col1a1-DTR mutants failed to show obvious defects after diphtheria toxin injection, indicating a minimal role of Col1a1$^+$ fibroblast-like cells under homeostatic conditions. In an intracerebral hemorrhage (ICH) model, however, these mutants showed exacerbated blood-brain barrier (BBB) damage and aggravated brain injury with reduced fibroblast marker (Collagen-1 and PDGFRα) expression, highlighting a beneficial role of Col1a1$^+$ fibroblast-like cells in ICH. Echoed with this finding, fibroblast-conditioned medium (FM) significantly enhanced TEER and reduced FITC-Dextran leakage in an in vitro ICH model. Interestingly, FM up-regulated the expression of tight junction proteins (occludin, ZO-1, and claudin-5), which seal gaps between endothelial cells; but did not affect the expression of caveolin-1, a key protein in transcytosis, indicating a paracellular rather than transcellular mechanism. To investigate how exactly fibroblasts exert the beneficial role in ICH, we performed mass spectrometry analysis, and identified TIMP2 (a protein with known BBB-protecting function) in FM. Using both pharmacological and genetic approaches, we further demonstrated that ablating TIMP2 abolished the neuroprotective effect of FM. Together, these results suggest that Col1a1$^+$ fibroblast-like cells secrete TIMP2, which inhibits tight junction protein loss and paracellular leakage, promoting BBB repair and ICH recovery. These findings suggest that fibroblasts and TIMP2 may be targeted in the therapy of stroke.

Disclosures: L. Xu: None. **Y. Yao:** None.

Poster

P327. The Blood Brain Barrier in Ischemia and Hemorrhage

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P327.05

Topic: C.08. Ischemia

Support: 2021R1A2C2006110
2021M3E5D9021364
2019R1A5A2026045

Title: Renovascular hypertension induces subcortical white matter degeneration accompanied by disruption of the blood-brain barrier

Authors: *X. JIN[1,3,4], Y. OH[1,3], Y. CUI[5], J. CHOI[1,3,2], B. KIM[1,3,2];
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Abstract: Chronic impairment of cerebral blood flow due to vascular risk factors may underlie subcortical white matter stroke. To observe pathological changes in the white matter that are expected in human patients, it is important to have adequate animal models. However, very few studies have successfully established optimal animal models that exhibit unequivocal and consistent- ischemic subcortical white matter lesions. We have recently observed distinct white matter pathologies in rats with renovascular hypertension (RVHT) induced by two-kidney, two-clip (2K2C) procedure. In this model, we detected marked reduction in myelin basic protein and axonal degeneration exclusively within the corpus callosum and cingulum. A substantial extent of mature oligodendrocyte (OL) loss was evident accompanied by increased number of immature OLs, reactive astrogliosis, and microglial activation. We also investigated white matter hyperintensities on T2-weighted magnetic resonance imaging and decreased fractional anisotropy value on Diffusion Tensor Imaging. Furthermore, deposition of fibrinogen and IgG extravasation in the subcortical white matter indicated increased permeability of BBB during pathological progression of the white matter damage, which was approximately consistent with increased Hypoxyprobe-1 (pimonidazole) immunoreactivity. Expression of aquaporin 4 (AQ4), a water channel located in astrocytic end feet was markedly increased, while pericytes did not exhibit any sign of degeneration. In addition, we observed decreased endothelial tight junction proteins ZO-1 and increased matrix metallopeptidase 9 (MMP9) expression. MMP9 immunoreactivity was increased in a variety of cell types, including microglia, oligodendrocytes, astrocytes. Since MMP9 has been mechanistically linked to BBB compromise, we hypothesized that MMP9 inhibition would have a beneficial effect in ischemic white matter injury by reducing BBB disruption and improving behavioral outcomes. Therefore, we are now testing this hypothesis by an intracerebroventricular injection of MMP9 inhibitors (BB94) in rats with RVHT.

Disclosures: X. Jin: None. **Y. Oh:** None. **Y. Cui:** None. **J. Choi:** None. **B. Kim:** None.

Poster

P327. The Blood Brain Barrier in Ischemia and Hemorrhage

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P327.06

Topic: C.08. Ischemia

Support: Mäxi Foundation

Title: Characterization of the Blood Brain Barrier Disruption in the Photothrombotic Stroke Model

Authors: *R. Z. WEBER[1,2], L. GRÖNNERT[1], G. MULDERS[1,3], M. A. MAURER[1], C. TACKENBERG[1], M. E. SCHWAB[1], R. RUST[1];
[1]Inst. for Regenerative Medicine, Univ. of Zurich, Switzerland, Schlieren, Switzerland; [2]Neurosci. Ctr. Zurich, Univ. of Zurich and ETH Zurich, Zurich, Switzerland; [3]Dept. of Hlth. Sci. and Technology, ETH Zurich, Zurich, Switzerland

Abstract: Cerebrovascular physiology is maintained by the blood brain barrier (BBB), a dynamically regulated interface separating the central nervous system (CNS) from the systemic circulation. BBB disruption is an important pathophysiological feature in the course of ischemic stroke which significantly contributes to the development of severe brain injury. In this study, we investigated and characterized the extent of BBB leakage in the acute phase of the photothrombotic stroke model, a popular permanent occlusion model to study long term recovery following ischemia in rodents. We observed enhanced BBB permeability over a time course of 3 weeks in peri-infarct and core regions of the ischemic cortex. BBB permeability slightly increased in the contralesional cortex, hippocampus and the cerebellum at different time points, and in regions where lesion-induced degeneration of pathways is prominent. Severe damage of tight and adherens junctions and loss of pericytes was observed within the peri-infarct region. Overall, the resulting features observed in this model closely simulate human stroke. Thus, it represents a suitable model to study BBB damage and therapeutic approaches interfering with this process.

Disclosures: R.Z. Weber: None. **L. Grönnert:** None. **G. Mulders:** None. **M.A. Maurer:** None. **C. Tackenberg:** None. **M.E. Schwab:** None. **R. Rust:** None.

Poster

P327. The Blood Brain Barrier in Ischemia and Hemorrhage

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P327.07

Topic: C.08. Ischemia

Support: Tata Trusts Grant
CBR grant

Title: Structural changes at synapse due to Blood Brain Barrier leakage by recurrent vascular insult leads to memory defecits

Authors: *L. DIWAKAR, R. HUSSAIN, S. MAURYA, V. RAVINDRANATH;
Ctr. for Brain Research, Indian Inst. of Sci., Bengaluru, India

Abstract: Structural changes at synapse due to Blood Brain Barrier leakage by recurrent vascular insult leads to memory deficits. Authors: Latha Diwakar[1], Rehab Hussain[1], Shashank Kumar Maurya[1] and Vijayalakshmi Ravindranath[1], 1Centre for Brain Research, Indian Institute of Science, Bengaluru, India.Dementia is a concern worldwide as its incidence increases in aged population impacting social and economic factors. Many clinical studies have provided significant insights indicating the potential link of vascular dysfunction to dementia, particularly mixed dementia. Vascular dysfunction occurring as cerebral small vessel infract develops later to white matter hyperintensity (WMH), which has been linked to the onset of pathogenesis of vascular dementia. To understand the molecular drivers underlying the cognitive decline and disease progression, we developed a mouse model of vascular dementia. We implanted cannula into the lateral ventricles of C57 mice to produce repeated microinfarcts in brain using endothelin-1 (ET-1), a vasoconstrictor peptide. Recurrent vascular insults were given by 3-doses of ET-1 over 9 weeks and resulted in deficits in associative and spatial learning after 9 weeks of ET-1 injection. Further, there was prominent blood brain barrier (BBB) disruption after recurrent vascular insults observed as IgG deposition near the vessels. There was loss of pericytes coverage indicating changes in the integrity of BBB. To understand the consequence of BBB breach on structural changes of neurons, we performed Golgi staining and carefully analyzed apical and basal dendrites in CA1 region of hippocampus. We found significant loss in apical and basal dendrites corroborating memory deficits. This mouse model will be useful for understanding pathogenic mechanisms underlying vascular dementia.

Disclosures: L. Diwakar: None. **R. Hussain:** None. **S. Maurya:** None. **V. Ravindranath:** None.

Poster

P327. The Blood Brain Barrier in Ischemia and Hemorrhage

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Program #/Poster #: P327.08

Topic: C.08. Ischemia

Support: R15 AG059142
University of Mississippi Graduate Student Grant
Southern Regional Educational Board
UMMC-GTEC Smith Scholars Program

Title: Neuroendocrine modulation alters cellular protection in the neurogliovascular unit

Authors: *C. HAYES[1], B. ASHMORE[1], A. VIJAYASANKAR[1], J. POSEY[1], N. ASHPOLE[2]; [1]Univ. of Mississippi, Oxford, MS; [2]Dept. of BioMolecular Sci., Univ. of Mississippi, University, MS

Abstract: Insulin-like growth factor-1 (IGF-1) is a neuroendocrine modulator with autocrine and paracrine functions. IGF-I peaks during puberty but declines throughout adulthood and advanced ages, coincidentally when neurodegenerative diseases are more prevalent. Two of the most prominent contributors to disorders associated with neurological aging are oxidative stress and glutamate-induced excitotoxicity. Previous evidence highlights that exogenous IGF-1 treatments in animal models of dementia, cognitive impairment, and stroke acts directly on neurons protecting and preserving neuronal function. Interestingly, the IGF-1 receptor (IGF-1R) is expressed on all cells in the brain. Therefore, the systemic reductions in circulating IGF-1 likely contribute to the viability and function of the entire neuro-glio-vascular unit. We recently reported that reduced IGF-1 leads to impaired glutamate handling in astrocytes, indirectly impacting neurons. We hypothesized that IGFR deficiency within each component of the neuro-glio-vascular unit increases susceptibility to glutamate-induced toxicity and oxidative stress. To test this, we used pharmacologic and genetic interventions to reduce IGF-1R in specific cell types and assessed cellular toxicity, reactive oxygen species (ROS) production, and mitochondrial dysfunction in response to glutamate toxicity. The results confirmed that neurons are highly susceptible to excitotoxicity, unlike astrocytes or endothelial cells, and the reduction of IGFR does not lead to overt toxicity. Although astrocytes and endothelial cells are highly resistant to glutamate-induced toxicity when IGFR is reduced, their production of ROS in the hours following insult increases. Mitochondrial dysfunction in astrocytes is also apparent at maximal respiration in the previously described treatment. Reduction of IGF-1R signaling in astrocytes also increased neurotoxicity in co-cultures of neurons, astrocytes, and endothelial cells, suggesting maintenance of astrocytic IGF-1R signaling is critical for neuroprotection. Overall, our findings indicate that the excitotoxic response of each major cell type of the neuro-glio-vascular unit is regulated by IGF-1. Although astrocytes and endothelial cells are resistant to excitotoxic death, the alterations in oxidative stress signaling when IGF-1 is inhibited may inherently affect the survival and function of nearby neurons. Further studies are warranted to understand the selective reduction of IGF-1R in *in vivo* models of the specific neuro-glio-vascular cell types to delineate the mechanisms and processes by which IGF-1 signaling protects neuronal health and viability.

Disclosures: C. Hayes: None. **B. Ashmore:** None. **A. Vijayasankar:** None. **J. Posey:** None. **N. Ashpole:** None.

Poster

P341. Animal Models of Brain Injury: Molecular-Cellular Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P341.01

Topic: C.10. Brain Injury and Trauma

Support: NIH Grant 2R01-DA02912
NIH Grant 1R01-DA049737
NIH Grant 1R01-NS117906

Title: Antidiabetic drugs reduce tobacco smoke-promoted cerebrovascular damage after traumatic brain injury

Authors: *F. SIVANDZADE[1], F. ALQAHTANI[2], L. CUCULLO[1];
[1]Oakland Univ. William Beaumont Sch. of Med., Oakland Univ., Rochester, MI; [2]Pharmacol. and Toxicology, King Saud Univ., Riyadh, Saudi Arabia

Abstract: Traumatic Brain Injury (TBI) is among the main reasons for cerebrovascular and neurological damage. Premorbid conditions such as tobacco smoking (TS) can exacerbate post-TBI brain damage and impact recovery as a result of TS content of reactive oxygen species (ROS) and oxidative stress (OS) stimuli promoting vascular endothelial impairments. Additionally, due to common pathogenic modulators of BBB impairment between chronic TS and hyperglycemia (HG), antidiabetic drugs like metformin (MF) and rosiglitazone (RSG) were shown to reduce BBB damage promoted by the chronic TS exposure. This study evaluated antidiabetic drugs' therapeutic feasibility and effectiveness to prevent/reduce TS-promoted cerebrovascular damage after TBI and/or improve post-TBI recovery through an in vivo study. For this purpose, male C57BL/6J mice, age range 6-8 weeks, were exposed to TS for four weeks. RSG and MF were administered daily via intraperitoneal (IP) injections of dose levels of 10 or 20 mg/kg (RSG) and 100 or 200 mg/kg (MF) with dose volumes of 20 mL/kg. After four weeks, test animals were subjected to TBI by head weight drop so that a metal weight (30 g) was inserted into a pre-positioned vertical guide, whereas the weight free fell for a distance of 80 cm before reaching the target. Body weights of the mice were frequently measured and general activity (through open field test) of the mice were analyzed before, 1 hr, 24 hr and three days after TBI. Finally, mice were sacrificed to collect blood and brains samples for subsequent biochemical and molecular analysis. Western blotting and quantitative RT-PCR analyses were applied on homogenized brain tissues to assess the expression of Nrf2 (critical antioxidant transcription factor), its downstream effectors (HO-1, NQO-1) and tight junction proteins including ZO-1, Occludin, and Claudin-5. Levels of inflammatory markers including NF-kB,

PECAM-1, VCAM-1, IL-6, IL-10, and TNF-α were also evaluated. The results revealed that MF and RSG can effectively decrease the harmful effect of TS on body weight and OS generated in response to TS. RSG and MF also upregulated NRF2 expression and its downstream effectors NQO-1 and HO-1, decreased TS-induced endothelial inflammation and loss of barrier integrity. RSG and MF also decreased the TS-aggravated post-traumatic behavior in mice undergoing TBI. In conclusion**,** through the underlying linking mechanism, antidiabetic drugs could be repurposed to reduce BBB damage and subsequent severity of TBI outcomes caused by TS dysregulation of the cellular antioxidant response system.

Disclosures: F. Sivandzade: None. **F. Alqahtani:** None. **L. Cucullo:** None.

Poster

P341. Animal Models of Brain Injury: Molecular-Cellular Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P341.02

Topic: C.10. Brain Injury and Trauma

Support: CONACYT CB A1-S-55371

Title: Evaluation of acute neurological changes, mortality, body weight and immunofluorescence to NeuN and GFAP in rats with lateral traumatic brain injury induced by a novel hydropneumatic device

Authors: *F. V. VILLALPANDO VARGAS[1], L. G. MEDINA-CEJA[2], E. GONZÁLEZ HERRERA[1], T. D. HERNÁNDEZ SÁNCHEZ[1];
[1]Univ. of Guadalajara, Guadalajara, Mexico; [2]Univ. of Guadalajara, Zapopan, Jalisco, Mexico, Mexico

Abstract: Sequelae of traumatic brain injury (TBI) are showed in 20 to 50 million people, which represent a high economic and social cost. The experimental models of TBI are the best tool for studying the mechanisms underlying injuries, within which the lateral fluid percussion (LPF) is the most widely used. However, the main disadvantage of LFP is the complexity and limitation in biomechanical control. We evaluated lesions induced with a novel hydropneumatic device of TBI produced by MexBio Research Innovations, S.A. de C.V., which is controlled electronically and programmable. To this aim, male Wistar rats (190-250 g, n=24) divided in sham (n=12) and TBI (n=12) groups were used. Animals were anesthetized using isoflurane (58.2 ± 6.9 s) to realize a circular craniotomy (5.0 mm diameter; epicenter in -4.5 AP and 2.5 ML mm, in relation with bregma). To induce TBI, animals were placed in this device on a heating pad during and after injury; the pressure level was adjusted to 2.1 ± 0.05 atm (peak pressure) with duration of 25 ms. Occurrence of immediate mortality was observed in 8.33%, post-impact behavioral seizures were not presented. The acute neurological changes were evaluated such as duration of apnea

(was not presented), hind limb reflex (239.5 ± 148.9 s) and self-righting reflex (278.5 ±122s). After rats righted themselves, they were re-anesthetized to suture scalp incision. Body weight was daily monitoring, Sham group at 2 (n=12) and 7 days (n=6) presented a weight loss of 5.2% and of 4%, respectively; in TBI group at 2 (n=12) and 7 days (n=6) after TBI was 8.7% and 6%, respectively. After 2 or 7 days of injury (n=6 per group, per date), animals were intracardially perfused with paraformaldehyde 4%, cryopreserved with sucrose 15% followed by 30% and frozen with 2-methylbutane embedded in dry ice. Coronal sections of 20 um were obtained in cryostat and immunofluorescence (IF) was performed with NeuN and GFAP antibodies. Photos of CA1, CA3, dental gyrus (DG) and primary motor cortex in both hemispheres were obtained (objective 20x, two sections per rat). In TBI group, the NeuN IF showed neuronal loss in CA1 and DG of injured ipsilateral hemisphere compared to their own contralateral and with both hemispheres of sham group; The GFAP IF showed a reactive astrocyte response in TBI group of all regions analyzed of injured ipsilateral hemisphere compared to their own contralateral and both hemispheres of sham group. In conclusion, this novel hydropneumatic device induced mild TBI with the parameters used (peak and duration pressure). However, it is imperative to induce an insult with different parameters in order to identify which are the favorable conditions to induce different severities of TBI.

Disclosures: F.V. Villalpando Vargas: None. **L.G. Medina-Ceja:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); MexBio Research Innovations, S.A. de C.V. **E. González Herrera:** None. **T.D. Hernández Sánchez:** None.

Poster

P341. Animal Models of Brain Injury: Molecular-Cellular Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P341.03

Topic: C.10. Brain Injury and Trauma

Title: Brain-derived Neurotrophic Factor-Expressing Engineered Mesenchymal Stem Cells Restore Traumatic Brain Injury-Induced Tissue and Behavioral Deficit

Authors: *H.-J. KIM[1], B. CHOI[2], D. HONG[2], S. SUH[2], S. LEE[1];
[1]SL BiGen, Inc., Incheon, Korea, Republic of; [2]Dept. of Physiol., Hallym University, Col. of Med., Chuncheon, Kang Won Do, Korea, Republic of

Abstract: Traumatic brain injury (TBI) can cause serious damage of all races and ages, and in fact, it is estimated that 40-50 million people are affected worldwide each year. TBI causes not only primary damage but also secondary damage. Secondary damage is associated with molecular cascades including cellular excitatory toxicity, disruption of blood-brain barrier (BBB), ischemia, programmed cell death, and inflammation that appear after primary nerve

damage. Most scientific approaches focus on protection, repair, or regeneration in regenerative medicine, and stem cells has been studied in many disease and injury models using from a variety of sources and application at pre-clinical and clinical level. Brain-derived neurotrophic factor (BDNF) is one of the neurotrophic factors that play an important role in the differentiation, maturation, and survival of nerve cells. Here we show well-established BDNF-expressing engineered human mesenchymal stem cell (BDNF-eMSC) is an effective approach to treat TBI through intracerebroventricular (ICV) transplantation. For this, controlled cortical injury (CCI)-damaged 2-months old male rats were assessed for molecular, motor, and neurocognitive pathologies. Transplantation of BDNF-eMSCs successfully effects on neuronal cell death, innate brain inflammatory cell activation, motor and cognitive behavioral functions via dose-dependent manner. Thus, TBI-induced molecular, motor, learning and memory pathologies, which improve the transplantation of BDNF-eMSCs and represents toward for control of damage of TBI.

Disclosures: H. Kim: None. **B. Choi:** None. **D. Hong:** None. **S. Suh:** None. **S. Lee:** None.

Poster

P341. Animal Models of Brain Injury: Molecular-Cellular Mechanisms

Location: Virtual Only

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Program #/Poster #: P341.04

Topic: C.10. Brain Injury and Trauma

Support: NIH grant R43NS110343

Title: Biomarkers following repetitive mild head injury

Authors: M. STEVENS[1], K. FUETES[1], N. BENS[2], C. MURPHY[1], I. MOSLEY[1], S.-F. LU[1], P. KULKARNI[2], N. G. SIMON[1], M. BROWNSTEIN[3], *C. F. FERRIS[2];
[1]Lehigh Univ., Bethlehem, PA; [2]Northeastern University, Ctr. for Translational NeuroImaging, Boston, MA; [3]Azevan Pharmaceuticals, Bethlehem, PA

Abstract: Head injury is among the leading causes of disability and death in adults under 35 and over 65. There are 2.5 million emergency room visits annually for these injuries, with the vast majority diagnosed as “mild” with no specific treatment intervention. The number of mild head injuries is likely greater because many are unreported. There is increasing recognition that repeated mild head injury can result in long-term cognitive deficits and increase the risk for dementia later in life. In this pilot study, we assessed the effect of one or three mild head strikes on two proteins in plasma that are candidate biomarkers for blood-brain barrier (BBB) disruption, neurofilament light (NfL) and glial fibrillary acidic protein (GFAP), and on two markers of neuroinflammation, Iba1 and AQP4, in prefrontal cortex and midbrain by immunohistochemistry. The studies were approved by the Lehigh University IACUC (biomarkers) and the Northeastern University IACUC (immunochemistry). Mild concussion was

generated in adult rats anesthetized with isoflurane using a momentum exchange device with a pneumatic pressure drive, 50 g compactor at 7.4 m/s impact velocities. We have used this model to characterize the neuroradiological effects of a single and multiple mild head impact injuries in rats. The impact piston was directed to the top of the skull, midline, in the approximate area of Bregma. Animals are awake and moving within 5-7 min post-impact. There was no evidence of structural brain damage or contusion in any of the impacted rats based on MRI. The NfL and GFAP assays were conducted on samples taken 2, 4, 24, and 48 hrs after impact. The single impact injury had no effect on either marker. In the 3-hit animals, no change was detected in GFAP, while NfL levels were sharply elevated at 2 (100-fold) and 4 (60-fold) hrs post-strike in males. In females, NfL was elevated at 2 (40-fold) and 4 (8-fold) hrs after impact and remained elevated vs baseline for 48 hrs. Immunohistochemistry conducted 3 weeks after multiple head impacts showed elevated expression of Iba1 and AQP4, indicating sustained inflammation as a consequence of repetitive mild head injury. These data, while preliminary, suggest that repetitive mild head injuries result in BBB disruption and neuroinflammation that persists for weeks. While there is additional work needed to refine the blood biomarker assay as several studies have shown that plasma GFAP represents a marker for BBB disruption, the data suggest an approach that can help characterize the risks associated with repetitive mild head injury.

Disclosures: M. Stevens: None. **K. Fuetes:** None. **N. Bens:** None. **C. Murphy:** None. **I. Mosley:** None. **S. Lu:** None. **P. Kulkarni:** None. **N.G. Simon:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Azevan Pharmaceuticals. F. Consulting Fees (e.g., advisory boards); Azevan Pharmaceuticals. **M. Brownstein:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Azevan Pharmaceuticals. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Azevan Pharmaceuticals. **C.F. Ferris:** None.

Poster

P341. Animal Models of Brain Injury: Molecular-Cellular Mechanisms

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Program #/Poster #: P341.05

Topic: C.10. Brain Injury and Trauma

Support: CTSI Grant UL1TR002529

Title: Tbi on a chip: linking physical impact to neurodegeneration by differentiating primary and secondary injury mechanisms

Authors: *E. ROGERS[1], T. BEAUCLAIR[1], A. THYEN[2], L. PRACHT[1], R. SHI[1];
[1]Purdue, West Lafayette, IN; [2]Indiana Univ. Sch. of Med., Indianapolis, IN

Abstract: The rapidly increasing prevalence of post-traumatic brain injury (TBI) long-term neurological sequelae is an alarming trend with profound societal consequences. While clinical observations have confirmed a link between the development of neurodegenerative diseases (ND) and TBI, the underlying mechanisms remain elusive. Further, these complications are exacerbated by the initial, subclinical nature of mild-TBI (the most common) which can delay diagnosis past the prime window for intervention, after irreparable damage has been firmly established. In response, we have engineered a highly novel, primary neuronal culture-model to reproduce real-world physical trauma scenarios with unprecedented levels of interrogatory capabilities for mechanistic investigations into post-TBI neurodegeneration. Specifically, we can impart rapid acceleration injuries in clinically relevant g-force ranges to neuronal networks grown on multielectrode arrays with a novel pendulum-injury device, recapitulating TBI on a chip. We have coupled this high precision primary injury device with the post-impact monitoring of acrolein levels, a highly reactive aldehyde, established biomarker of post-TBI oxidative stress and neurodegeneration, and a likely convergence point between TBI and ND, to delineate impact-induced changes brought upon by secondary injuries.
Using immunocytochemical methods, we noted significant, force-dependent increases in acrolein (acrolein-lysine adducts) at 24hrs post-impact, indicative of post-impact neuronal degeneration. These changes were amplified by the following manipulations: exposure to higher g-forces (30-250 g, peak); the rapid (4-6 sec interval) application of multiple impacts (1, 3, 5 and 10x); and exposure to 40 mM EtOH for a 10 min duration immediately following impact. Further, we demonstrate the enhancement of injury-recovery as a function of increasing time intervals between repeated hits. In addition, conditioned media from maximumly-impacted cultures can cause intracellular acrolein elevation when introduced to non-impact, control networks, indicating acrolein's role as a diffusive-factor in post-TBI secondary injuries.
This model provides a unique level of cellular and temporal resolution not attainable with animal models. Further, it can separate primary and secondary injuries, while mechanistically untangling and evaluating relevant comorbidities. Combined with our available *in vivo* models, we expect to gain insight into the mechanisms underpinning TBI (acute and chronic) and its link to ND, helping to guide our translational laboratory endeavors and improving clinical diagnoses and treatments.

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Poster

P341. Animal Models of Brain Injury: Molecular-Cellular Mechanisms

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Topic: C.10. Brain Injury and Trauma

Support: NIH Grant NS077675
NIH-NINDS Center Core Grants 5P30NS047463
NIH-NCI Cancer Center Support Grants P30CA016059

Title: Axonal injury following mild traumatic brain injury is exacerbated by repetitive insult and is linked to the delayed loss of NeuN expression without concomitant neuronal death in the mouse

Authors: *Y. OGINO[1], T. BERNAS[1], J. GREER[2,3], J. T. POVLISHOCK[1];
[1]Anat. and Neurobio., [2]Neurosurg., Virginia Commonwealth Univ., Richmond, VA; [3]Surgery, Hunter Holmes McGuire Veterans Affairs Med. Ctr., Richmond, VA

Abstract: Traumatic brain injury (TBI) is a major health care problem worldwide. Mild TBI (mTBI) is the least severe type of TBI but it, even though, affects brain structure and function and can lead to persistent abnormalities. Repetitive mTBI exacerbates the acute phase response to injury but its long-term implications remain poorly understood. We aimed to elucidate the long-term implications of repetitive mTBI, particularly in the context of the chronic-phase burden of traumatic axonal injury (TAI), a major player in TBI-related morbidity, and its implications for retrograde neuronal perturbation or death. Murine neocortical tissue was investigated at acute (24-hours postinjury, 24hpi) and chronic time points (28-days postinjury, 28dpi) after singular or repetitive mTBI given by central fluid percussion injury (cFPI). Neurons immunofluorescently labeled with multiple markers, NeuroTrace and NeuN for detecting neurons, p-c-Jun for detecting axotomized neurons and DRAQ5 for detecting nuclei, were quantitatively assessed using automated 3D-image analysis. Single mTBI produced axotomy in 10% of neurons at 24hpi and the percentage increased after repetitive injury. The fraction of p-c-Jun+ neurons decreased at 28dpi but without neuronal loss (NeuroTrace), suggesting their reorganization and/or repair following TAI. In contrast, NeuN+ neurons decreased with repetitive injury at 24hpi while the corresponding fraction of NeuroTrace+ neurons decreased over 28dpi. This observed loss of NeuN expression was linked exclusively to non-axotomized neurons at 24hpi then extended to the axotomized at 28dpi, revealing a delayed response of the axotomized neurons. Collectively, we demonstrate an increased burden of TAI after repetitive mTBI, which is most striking in the acute phase response to the injury. Our finding of widespread axotomy in large fields of intact neurons rejects the premise that repetitive mTBI elicits progressive neuronal death, rather emphasizing the importance of axotomy-mediated change.

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Poster

P341. Animal Models of Brain Injury: Molecular-Cellular Mechanisms

Location: Virtual Only

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Program #/Poster #: P341.07

Topic: C.10. Brain Injury and Trauma

Title: Multiparametric Characterization of Physiology and Behavior in a Minipig Model of Pediatric Concussion

Authors: *A. H. NETZLEY[1], N. ARNOLD[2], K. A. MUNOZ[3], A. C. COLBATH[3], T. R. BUSH[2], A. MEODED[5], A. A. BADACHHAPE[5], G. PELLED[1,4];
[1]Biomed. Engin., [2]Mechanical Engin., [3]Vet. Med., [4]Radiology, Michigan State Univ., East Lansing, MI; [5]Radiology, Baylor Col. of Med., Houston, TX

Abstract: Children and adolescents who experience mild traumatic brain injury (mTBI), or concussion, often suffer from persistent cognitive and neurological deficits that can have a lasting negative impact on an individual's quality of life. Due to biomechanical considerations of the head impact, large animal models may accurately reproduce the unique consequences of pediatric concussion and provide a platform to identify biomarkers and test new therapies. We have developed a closed-head impact procedure capable of inducing concussion in juvenile (4 months old) Yucatan miniature pigs. We built a custom-engineered impact device with which we can generate closed-head injury with reproducible and controllable impact velocity, acceleration, and dwell time to deliver an impact that is clinically considered as a concussion. We conducted a comprehensive battery of tests designed to characterize post-injury behavioral and physiological function throughout maturation and development. We utilize traditional behavioral tests, such as open field and novel object recognition, as well as state of the art physiological assessment tools and wearable sensor technologies for gait analysis and activity tracking with the goal of evaluating multiple aspects of post-concussive impairment and recovery. Our preliminary results suggest that concussed pigs exhibit symptoms similar to those reported for post-concussive syndrome in humans. Specifically, we have found that pigs with concussion are less active throughout the day: Fitbit results show 50% fewer steps taken per day. We have also found that during a 10-minute open-field test the concussed pigs traveled twice the distance compared to control pigs. During a three-choice discrimination task, concussed pigs show waning interest in retrieving a food-reward, performing 33% slower over consecutive trials, whereas control pigs become 24% faster over trials. Additionally, in an executive function test where pigs had to remember location and retrieve treats, concussed pigs exhibit deficits of 57% in the month after injury. These data suggest that our injury model is capable of producing post-concussive symptoms in minipigs and can be used as a platform to identify new biomarkers to report on injury and to test new therapeutic strategies.

Disclosures: A.H. Netzley: None. **N. Arnold:** None. **K.A. Munoz:** None. **A.C. Colbath:** None. **T.R. Bush:** None. **A. Meoded:** None. **A.A. Badachhape:** None. **G. Pelled:** None.

Poster

P341. Animal Models of Brain Injury: Molecular-Cellular Mechanisms

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Program #/Poster #: P341.08

Topic: C.10. Brain Injury and Trauma

Support: WP911F-17-2-0222

Title: Acute Changes in Spine Phenotype and Dendritic Properties in the Porcine Cortex Following Impact

Authors: *B. HOFFE[1], H. TOMSON[2], R. BANTON[3], T. PIEHLER[3], O. PETEL[2], M. R. HOLAHAN[1];
[1]Neurosci., [2]Mechanical and Aerospace Engin., Carleton Univ., Ottawa, ON, Canada; [3]U.S. Army Res. Lab., Aberdeen Proving Ground, MD

Abstract: Dendritic spines are responsible for relaying excitatory input and regulating signalling between neurons and their morphology can give insight into synaptic function. Excitotoxicity brought on by brain injury results in synaptic dysfunction, as intracellular Ca^{2+} increases. This increase in intracellular Ca^{2+} over-activates various cellular kinases and proteases responsible for microtubule stabilization. Previous work in our lab has shown that after an impact, there is an increase in tensile strain within the depths of the porcine cingulate sulcus. Moreover, there was a marked decrease of the microtubule stabilizing protein MAP2 specifically within the apex of the sulcus. In our current work, we explored whether changes in spine morphology were associated with out impact model. The Golgi-Cox staining method was used to visualize dendritic spine morphology. Following impact, coronal slabs were placed into Golgi-Cox solution for 2 weeks. Cortical pyramidal neurons within the sulcus apex and arm were reconstructed. Preliminary data revealed a shift towards mushroom-type spines, compared to thin and stubby-type spines within the sulci of the brain after impact. These findings suggest a shift towards an excitatory synapse shortly after impact. Along with our previous findings of changes in MAP2 properties, the shift towards an excitatory synapse could implicate activation of kinases and proteases responsible for regulating MAP2 dynamics and altering spine morphology.

Disclosures: B. Hoffe: None. **H. Tomson:** None. **R. Banton:** None. **T. Piehler:** None. **O. Petel:** None. **M.R. Holahan:** None.

Poster

P341. Animal Models of Brain Injury: Molecular-Cellular Mechanisms

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Program #/Poster #: P341.09

Topic: C.10. Brain Injury and Trauma

Support: NIH 5R01NS034700
5R37NS077908
5R01NS040109
CURE W81XWH-15-2-0069

Title: Two-photon imaging of intra- and extracellular chloride in a porcine model of post-traumatic epilepsy

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Abstract: Post-traumatic epilepsy (PTE) results from up to 20% of civilian closed head injuries. There are currently no prophylactic treatments available to prevent post-traumatic epileptogenesis and PTE is frequently medically intractable. We hypothesize that injury-induced release of MMPs, alters the extracellular matrix, which alters chloride in the extracellular space and in nearby neurons, decreases GABA-mediated inhibition and increases network excitability in the injured tissue. In this study, we directly imaged chloride in the extracellular matrix or in neuronal cytoplasm following TBI in 4-6 month old Yucatan minipigs. Pigs were chosen because their gyrencephalic brains, containing gray and white matter, closely approximate human anatomy and deformational and inertial responses to external force. 30d prior to imaging, two burr holes were placed in the skull and corresponding durotomy was performed for each burr hole. Each hemisphere was then injected intraparenchymally with AAV encoding for either the genetically encoded chloride sensor SuperClomeleon (SClm, IC group) under control of the neuron-specific hSyn promoter or a control red fluorescent protein (EC group). The durotomy was then sealed with a transparent, flexible PDMS window. After 30d, the burr holes were exposed and EC pigs were injected bilaterally, through the PDMS membrane, with the extracellular chloride-sensitive dye ABP-dextran. The chloride-sensitive fluorophore was then imaged in both hemispheres using a custom-built large animal two-photon fluorescence lifetime microscope. One hemisphere was then injured using a cortical impactor. Both hemispheres were imaged again with two-photon microscopy. The animal then underwent an MRI, including diffusion weighted imaging (DWI). Finally, chloride was imaged again at a time point >4h post-injury. Four hours post-injury there was a mean decrease in ABP fluorescence lifetime of 25%, corresponding to an extracellular chloride increase of approximately 30mM. Similarly, the SClm YFP/CFP emission ratio decreased by 30%, corresponding to an increase in intracellular chloride. MRI DWI analysis is ongoing, but preliminary results show a mean 12% decrease in diffusivity in a subset of the lesion compared to the contralateral (un-injured) control. Together these preliminary findings are consistent with a model wherein TBI induces changes in the ECM that lead to secondary increases in both extracellular and intraneuronal chloride. If these findings are confirmed, this may point to a previously unknown mechanism of cerebral edema as well as pathological changes in GABAergic inhibition that contribute to post-traumatic epileptogenesis.

Disclosures: K.P. Lillis: None. **B.A. Costine-Bartell:** None. **L.F. Martinez-Ramirez:** None. **K.P. Normoyle:** None. **K.J. Staley:** None.

Poster

P341. Animal Models of Brain Injury: Molecular-Cellular Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P341.10

Topic: C.10. Brain Injury and Trauma

Support: NIH R01NS096225
Louisiana State University Research Council, and Joanna G. Magale Foundation

Title: Temporal changes of Protein arginine methyl transferase post-repetitive and mild traumatic brain injury

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Abstract: Traumatic brain injury (TBI) is defined as an impact to the head by an external force that causes brain alterations and subsequent long-term functional deficits. TBI contributes to an economic burden of $17 billion USD annually and is the leading cause of death and disability for individuals under 45. There are no current treatment(s) for repetitive and mild TBI (rmTBI), therefore, we sought to identify novel signaling molecules/pathways that could contribute to TBI. We employed a clinically relevant (non-surgical) closed-head impact model of engineered rotational acceleration (CHIMERA) that allows free rotation of the head upon impact generated from an air-compressed piston. *Our data suggest a novel role for PRMT7 (*protein arginine methyltransferases) *in the disease progression as indicated by the temporal decrease in protein expression post-rmTBI.* Our central hypothesis is that the loss of PRMT7, due to repetitive and mild TBI, mediates excitotoxicity, increases blood brain barrier (BBB) permeability, and contributes to behavioral deficits. PRMTs are novel targets that catalyze the methylation of arginine residues (a constitutive post-translational modification) involved in transcription, translation, receptor trafficking, and protein stability. There are currently 11 known PRMT isoforms (PRMT1-11), with PRMT7 gene deletion in human patients causing neurological deficits such as intellectual disability, microcephaly, and brachydactyly, along with hyperexcitability and impaired social behaviors in murine *in vivo* models. We measured PRMT7 protein levels post-rmTBI and found that it was decreased in the cortex (p=0.0005) and hippocampus (p=0.0309) 7-days post-rmTBI. Relative PRMT7 mRNA (via real-time qPCR) was enhanced in the cortex (0.040188) 1-day post-rmTBI. Using LC-MS, we measured excitatory neurotransmitters to suggest that glutamate was enhanced (P=0.05) in the hippocampus 3-day post-rmTBI. Tight junction proteins were decreased, such as JAMa (P=0.0023) and occluding

was modulated 1,3,7 days post-rmTBI. These findings were further supported by successively less evans blue dye 1,3 and 7 days post-rmTBI, altogether to suggest blood brain barrier permeability post-rmTBI. Finally, learning, working memory, and locomotor skills were significantly impaired as indicated by decreased alternation ratios (P=0.0271) via T-maze, novel object recognition (P=0.0041) and rotarod assessment (P=0.002103). PRMT7-related neurological deficits have been sparsely described in humans. Our current studies suggest a regulatory role of PRMT7 in TBI, which can also provide novel therapeutic insights into other neurodegenerative diseases.

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Poster

P478. Processing of Contrast, Form, and Color I

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Program #/Poster #: P478.01

Topic: D.07. Vision

Support: NIH Brain Inititative
NIH Eureka grant

Title: A distinct population of heterogeneously color tuned neurons in macaque visual cortex

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Abstract: Color constitutes an important property of the visual input to the brain. Color information in the natural environment is processed to guide behaviors such as food gathering, avoiding predators and social interactions that are crucial for the survival of higher vertebrates. An important characteristic of color information in natural scenes is the high degree of spatial diversity i.e., some patches in natural scenes contain a single color (homogeneous) whereas other patches contain multiple colors arranged close to each other. However, whether the receptive fields (RFs) of neurons in the visual cortex are adapted to this diversity in color features in the natural environment is unknown. We reasoned that for the visual cortex to construct an accurate representation of our environment, it should be able to differentiate between homogeneous and heterogeneous color patches. To test our hypothesis, we recorded neuronal populations in mid-level visual area V4 of awake macaques with chronically implanted Utah arrays while stimulating subfields (patches of size ~ 1º x 1º) within the RFs using equiluminant color stimuli (set of 16 colors). We discovered that in addition to neurons that preferred the same color at different spatial locations with their RFs (“homogeneous”), for a significant fraction of neurons,

color preference exhibited spatial variation within the RF ("heterogeneous"), with the most heterogeneous cells preferring up to an average of 6 unique colors. Heterogeneously tuned neurons exhibited higher latencies for peak color tuning compared to homogeneous neurons suggesting a possible role of local recurrent circuits in shaping tuning properties in addition to feedforward connections. Heterogeneous neurons produced significantly higher responses compared to homogeneous neurons when the entire RF was stimulated with a complex color patch containing the preferred colors of each sub-field. On the other hand, a uniform color stimulus covering the RF evoked weaker responses in the heterogeneous compared to homogeneous neurons. Our findings indicate a higher degree of complexity of color encoding mechanisms where homogeneous and heterogeneous neurons could together encode different aspects of chromatic information (uniform vs non-uniform) found in the natural environment.

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Poster

P478. Processing of Contrast, Form, and Color I

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Topic: D.07. Vision

Support: Swiss National Science Foundation Grant 320030_175616
National Research Foundation, Prime Minister's Office, Singapore under its Campus for Research Excellence and Technological Enterprise (CREATE) program (FHT)

Title: Modulation of visual contrast sensitivity with individualized transcranial random noise stimulation is time-dependent and specific for the primary visual cortex.

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Abstract: Transcranial random noise stimulation (tRNS) has been shown to significantly improve visual perception. Previous studies demonstrated that tRNS delivered over cortical areas acutely enhances visual contrast detection of stimuli when tRNS intensity is optimized for the individual. However, it is currently unknown whether tRNS-induced signal enhancement could be achieved within different neural substrates along the retino-cortical pathway and whether the beneficial effect of optimal tRNS intensities can be reproduced across sessions.
In 3 experimental sessions, we tested whether tRNS applied to the primary visual cortex (V1) and to the retina improves visual contrast detection (preregistration: https://osf.io/gacjw). We

first measured visual contrast detection threshold (VCT; N=24, 16 females) during tRNS delivery separately over V1 (no-tRNS, 0.75, 1, 1.5mA) and over the retina (no-tRNS, 0.1, 0.2, 0.3mA), determined the optimal tRNS intensities for each individual (ind-tRNS) and retested the effects of ind-tRNS within the sessions. We further investigated whether we could reproduce the ind-tRNS-induced modulation on a different session (N=18, 13 females). Finally, we tested whether the simultaneous application of ind-tRNS to the retina and V1 causes additive effects. We found that at the group level tRNS of 0.75mA decreases VCT compared to baseline when delivered to the V1 [$F_{(3, 69)}$=4.54, p=0.006]. Beneficial effects of ind-tRNS could be replicated when retested within the same experimental session [$t_{(23)}$=1.72, p=0.049] but not when retested in a separate session [$t_{(17)}$=-0.17, p=0.43]. Applying tRNS to retina did not cause a systematic reduction of VCT irrespective of whether the individually optimized intensity was considered [within $t_{(23)}$=1.05, p=0.15 and between sessions $t_{(17)}$=0.2, p=0.42] or not [$F_{(3, 69)}$=1.69, p=0.18]. We also did not observe consistent additive effects of V1 and retina stimulation [$t_{(17)}$=0.8, p=0.22].
Our findings demonstrate that V1 seems to be more sensitive than retina to tRNS-induced modulation of visual contrast processing. Moreover, the individual optimal tRNS intensity appears to vary across sessions.

Disclosures: W. Potok: None. **A. Post:** None. **M. Baechinger:** None. **N. Wenderoth:** None.

Poster

P478. Processing of Contrast, Form, and Color I

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Program #/Poster #: P478.03

Topic: D.07. Vision

Support: EY05253

Title: Contrast changes on-off dominance in human peripheral vision

Authors: *H. RAHIMI NASRABADI, V. MOORE-STOLL, J. JIN, M. W. DUL, J.-M. ALONSO;
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Abstract: All animals with form vision process light and dark stimuli with ON and OFF visual pathways that originate at the first retinal synapse. Central vision is dominated by the OFF pathway in primates and carnivores and humans are also more accurate at searching dark than light targets with foveal vision. Unlike for central vision, the relative dominance of ON and OFF pathways in peripheral vision remains unclear, which complicates the interpretation of visual deficits measured with traditional white-on-white perimetry in diseases such as glaucoma. To investigate ON-OFF dominance in human peripheral vision, we developed an ONOFF perimetry

test that requires subjects to signal whether a target embedded in noise is presented on the right or left hemifield. The target can be light or dark, be at 90 different positions of the visual field between 5 and 30 degrees eccentricity, and can have eight different luminance contrasts (from 5 to 20%). The targets are presented on a head mounted display equipped with an eye tracker that is fitted to the subjects with the appropriate optical correction (HTC VIVE Pro, refresh rate: 90 Hz, maximum luminance: 110 cd/m^2). All subjects recruited so far were able to perform the task monocularly while fixating on a central dot of the display (a task trial was aborted when the eyes deviated 2.5-3.5 degrees from fixation). Preliminary results from 13 eyes of 13 subjects (age range: 24-61) indicate that dark stimuli are perceived more accurately than light stimuli in peripheral vision (error percentage: 14.2 ± 10.1 for darks versus 22.5 ± 9.3, for lights, $p < 0.0001$, Wilcoxon test), and a significant light-dark difference in accuracy can be also demonstrated at the highest 20-30 degrees eccentricity (error percentage: 18.9 ± 17.7 for darks versus 22.1 ± 17.1 for lights, $p=0.0002$, paired t-test, $p < 0.0001$ normality test). However, decreasing the contrast reduced and sometimes inverted the light-dark difference in accuracy, as would be predicted from the greater contrast-response gain of ON than OFF pathways. For example, reducing the contrast from 20% to 5% made the light-dark difference in accuracy four times smaller (light-dark difference in percent errors: 11.0 ± 3.3 versus 2.7 ± 5.6, $p < 0.0001$, Wilcoxon test) and, at the highest eccentricity (20-30 degrees), it shifted the difference from slightly higher accuracy for darks to slightly higher accuracy for lights (light-dark percentage of errors 4.4 ± 35.7 versus -1.4 ± 68.7, $p= 0.031$, Wilcoxon test). These preliminary results indicate that the OFF pathway dominates human central vision within the central 30 degrees but the OFF dominance decreases and can reverse to ON dominance at low contrast.

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Poster

P478. Processing of Contrast, Form, and Color I

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Program #/Poster #: P478.04

Topic: D.07. Vision

Support: Hans-Goldmann-Stiftung, Bern, Switzerland

Title: Phase specific visual stimulation changes the contrast detection threshold in humans

Authors: *D. BRUEGGER, A. ZIMMERLI, M. ABEGG;
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Abstract: Introduction: Whether visual perception is continuous or organized in discrete cycles is debated. Recent evidence shows rhythmic changes in perception related to cortical oscillations

at various frequencies. Such rhythmic variations in visual perception open the possibility to influence perceptual performance by using phase-locked visual stimulation.
Methods: We applied phase specific visual stimulation in 20 healthy human participants and measured the contrast detection performance. For this, we briefly (48ms) presented Gabor patches with stripes in one out of four possible orientations close to the detection threshold. We recorded an occipital to frontal EEG signal with active surface electrode. We extracted the theta (4-8 Hz) oscillation to present visual stimuli at one out of four predefined theta phase angles (0°, 90°, 180° 270°). Phase estimation and prediction for the phase-specific stimulation is achieved with a custom developed system based on fitting of sine waves to the raw EEG signal. We tested 18 blocks consisting out of 50 stimuli in each participant. The stimuli within one block were always targeted at the same phase angle. The order of the blocks was randomized and the blocks targeting the positive and negative peaks of the oscillation (90°, 270°) have been shown 5 times each vs 4 times for the blocks at the through (0°, 180°).
Results: We found a significantly higher rate of correctly detected contrast stimuli in trials in which the stimuli were targeted at the 0° phase target ($\chi 2$ Test; p= 0.002; Odds = 2.16 1.87, 1.90, 1.83). In addition, post-hoc modelling showed significant influences of stimulus phase, orientation of Gabor patch, response latency and prestimulus interval on the detection probability.
Conclusion: We provide evidence that contrast detection in humans varies with the phase of theta oscillations. Due to the prospective nature of our experiment with phase-specific stimulation, we can avoid potentially confounding effects associated with retrospective analysis such as artefacts originating from evoked potentials and post-hoc signal processing steps.

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Poster

P478. Processing of Contrast, Form, and Color I

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Topic: D.07. Vision

Support: Leon Levy Fellowship in Neuroscience
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Simons Foundation

Title: Neuronal computations in macaque V2

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Abstract: The perception of visual form relies on processing within a ventral stream of visual cortical areas. Neurons in the first stage of this pathway, in primary visual cortex (V1), respond selectively to visual stimuli presented at specific locations, orientations, and spatial scales (spatial frequencies). Neurons late in this pathway, in inferotemporal (IT) cortex, respond selectively to complex objects regardless of changes in their location, size, and context. However, the representations of intermediate stages is not well understood. For example, neurons in V2 (but not in V1) are more strongly driven by images containing naturalistic texture than images containing spectrally matched noise, but the computation underlying this selectivity has not been characterized. We mapped receptive fields in areas V1 and V2 using a novel stimulus made of superimposed patches of gratings differing in location, orientation, and spatial scale. Responses of macaque neurons were recorded using two experimental preparations: single units were measured using single electrodes in an awake, fixating animal, and population responses were measured with dense multi-site linear probes (Neuropixels) in anesthetized animals. We used a two-stage model to fit the data. First, we simulated early cortical processing with a large, convolutional bank of filters varying in position, orientation, and scale. The responses of these filters, suitably rectified and pooled, approximate the responses of simple and complex V1 units. Second, we computed a weighted sum of the outputs of these filters and rectified them. We fit the weights and the form of the rectification to maximize likelihood of individual neural responses, using stochastic gradient descent. As expected, V1 neurons had simple receptive field structures, with receptive fields tightly localized in space, orientation, and spatial frequency. In contrast, V2 neurons had larger receptive fields often containing both excitatory and suppressive fields, each localized in the four dimensions of V1 response (spatial position, orientation, spatial frequency). These neurons effectively compute a difference of functionally localized groups of V1 afferents that can represent image features such as spatial gradients of orientation (e.g. curvature) or size (e.g. foreshortening), as well as local texture properties. Simulations of the responses of V2 neuronal ensembles show that such structures are well-suited to produce selectivity for naturalistic image structure relative to spectrally matched noise.

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Poster

P478. Processing of Contrast, Form, and Color I

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Program #/Poster #: P478.06

Topic: D.07. Vision

Support: IARPA D16PC00003

Title: Diverse set of highly exciting images reveals novel spatially localized phase invariance in mouse primary visual cortex

Authors: *Z. DING[1], D. TRAN[1], E. COBOS[1], T. MUHAMMAD[1], S. CADENA[2], A. ECKER[3], A. TOLIAS[1];
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Abstract: In order to decipher the algorithm of perception, it is important to characterize neural tuning functions and identify the directions of maximal tuning and invariance to stimulus features. To this end, simple parametric stimuli are typically used, but they make strong assumptions about the stimulus features that neurons are selective and invariant to. Recently, the "inception loop" paradigm was developed which provides a systematic characterization of neural tuning with minimal assumptions. This paradigm combines large-scale neuronal recordings with deep learning models which are trained to predict the responses of neurons to arbitrary stimuli. These models can then be used as *in silico* avatars to perform a large number of experiments and analyses, including ones that are practically impossible to perform in the real brain due to the limited experimental time available and the large space of stimuli and behaviors that must be explored. Inception loops have been used to find the most exciting inputs of neurons (MEIs) in mouse primary visual cortex (V1). These stimuli were verified back *in vivo*. The MEIs exhibited complex spatial features deviating strikingly from the textbook Gabor-like optimal stimuli for V1, challenging the decades-old dogma of V1 representation. Here, our work extends these deep learning optimization methods to study neural tuning invariance. We introduce "diverse exciting inputs" (DEIs), a model-based approach where we synthesize a set of diverse highly activating images. We found that the DEIs of many neurons in mouse V1 exhibited a novel type of invariance. Specifically, these neurons responded invariantly to phase changes of textures localized in only a portion of their receptive field. We propose a local-phase-invariant model that explains single-neuron invariances in mouse V1 which are not accounted for by the traditional Hubel & Wiesel simple-complex cell model. Taken together, our work extends deep learning methods to study sensory processing and reveals a novel type of single-neuron invariance.

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Poster

P478. Processing of Contrast, Form, and Color I

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The Natural Sciences and Engineering Research Council of Canada Grant RGPIN-2016-05352

Title: Learning a model of shape selectivity in V4 cells reveals shape encoding mechanisms in the brain

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Abstract: The mechanisms of local shape information transformation from V1 to more abstract representations in IT are unknown. Studying the selectivities in intermediate stages of transformation suggest plausible mechanisms. For example, findings in Macaque V4 responses suggest that these neurons are selective to convexities and concavities at a specific position in the stimulus (Pasupathy et al., 2001). Although such investigations reveal intermediate shape representations in the brain, they often do not suffice in capturing complex and long-range interactions within the receptive field due to imposing priors on tunings, e.g., fitting a single Gaussian to neuron responses. We propose a learning-based approach that eliminates the need for such strong priors. Specifically, we investigate shape representation in Macaque V4 cells and formulate shape tuning as a sparse coding problem according to previous findings of V4 neurons (Pasupathy et al., 2001). We emphasize that our goal is not to find a mapping from the stimulus to V4 responses but rather to study how V4 neurons combine responses of V2 neurons, in this case, curvature-selective V2 cells, to achieve their reported part-based selectivities. To this end, our proposed model takes responses of simulated curvature-selective V2 cells as input by combining two previously introduced hierarchical models 2DSIL (Rodríguez-Sánchez et al., 2012) and the RBO network (Mehrani et al., 2021). With simulated curvature signal as input, our algorithm learns a sparse mapping to V4 responses that reveals each Macaque V4 cell's tuning and the mechanism by which the tuning is achieved. Our results suggest that V4 cells combine shape parts from the full spatial extent of their receptive field with similar magnitudes of facilitatory and inhibitory contributions. Additionally, our approach suggests a mechanistic model on how object-centered representations in V4 are achieved, providing a better understanding of shape encoding mechanisms in the brain.
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Poster

P478. Processing of Contrast, Form, and Color I

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Topic: D.07. Vision

Support: C14/18/100

Title: High-resolution human recordings demonstrate diverse coding of the shape and category of visual objects

Authors: *V. BOUGOU, M. VANHOYLAND, A. BERTRAND, P. JANSSEN, H. OP DE BEECK, T. THEYS;
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Abstract: Recent noninvasive imaging studies have revealed multidimensional representations of objects and categories in the human brain. However, it is unknown which tuning curves at the level of single neurons and local neuronal clusters underlie these macroscopic representations at the population level. Invasive human recordings can capture both single-cell activity and the coherent activity of cell assemblies (LFPs - Local Field Potentials), and are therefore a very suitable and high-resolution approach to investigate selectivity for multiple dimensions such as shape and category in different brain areas. We used a combination of invasive recordings in the meso (high-density ECoG, SEEG) and micro (96-channel microelectrode Utah-array) scale in 3 neurosurgical patients, covering a wide range of brain areas (occipital, parietal and frontal cortex). We presented a set of 54 images in which shape and category were dissociated (Bracci et al., 2016). High-gamma power (60-120 Hz) per electrode was computed and averaged over time for 80-300 ms after stimulus onset. Per pair of conditions, we correlated the spatial multi-channel response pattern per grid/microarray. The resulting matrices were converted into dissimilarity matrices (1 - correlation) and were correlated with behavioral dissimilarity matrices for the shape and category dimensions. These multi-channel analyses confirmed previous fMRI data suggesting significant shape-based representations in the Lateral Occipital Complex. In contrast, the activity patterns measured over parietal cortex did not correlate with either of the two dimensions. However, with a further analysis of individual visually-responsive electrodes (two-way ANOVA), we observed significant selectivity for both shape and category. Single electrodes in LOC and in parietal cortex were either category-selective (2.06 % in LOC and 20 % in parietal cortex), shape-selective (13.4 % in LOC and 20 % in parietal cortex), or had interactions between shape and category (11.34 % in LOC). These results show that multichannel analysis offers a general description of the neural representation in an area but fails to capture the underlying neuronal selectivity and its diversity. Thus, invasive recordings in human visual cortex can provide a more detailed view on the shape and category representations, which complements data obtained with functional imaging.

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Poster

P482. Representation of Objects and Scenes I

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Tianqiao and Chrissy Chen Brain-machine Interface Center at Caltech
Boswell Foundation
Swartz Foundation

Title: Cognitive spatial representations in posterior parietal cortex

Authors: *J. GAMEZ[1], T. AFLALO[1], K. KADLEC[1], C. GUAN[1], K. W. PEJSA[1], E. ROSARIO[2], N. POURATIAN[3], R. A. ANDERSEN[1];
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Abstract: The posterior parietal cortex (PPC) has a privileged position in the flow of information through the brain, having access to data from sensory, motor, and association cortical areas. Different regions within PPC transform sensory information into body-centered egocentric representations using gain modulation by eye, head, and limb positions. However, the spatial position of an object can also be described in allocentric coordinates, coding the position with respect to visual landmarks in the visual scene. This representation requires a cognitive process to transform visual information that is initially coded as a retinal position into an abstract spatial representation. It is not known if the neural populations in human PPC and the motor cortex (MC) have access to this cognitive spatial information during visuomotor transformations and action planning and execution. To address this question, we recorded from the cortex of one human tetraplegic participant (N1), implanted with two 96-channel Blackrock arrays, one in PPC and one in MC, as part of a brain-machine interface clinical trial. The recordings were done while N1 performed a task where he had to decide if an object was to the left or the right of his gaze (egocentric condition) or a landmark (allocentric condition). N1 provided his answers using saccades or attempted thumb movements in a staged paradigm to dissociate the spatial decision from this motor response. We found that PPC can code the spatial position of the target object in both egocentric and allocentric coordinates. A population level across-condition analysis showed that egocentric and allocentric reference frames are coded in different neural subspaces with no positive correlation. This spatial coding was distinct during the decision and the motor planning/execution phases. Furthermore, this representation was independent of the effector used to execute the motor action. In contrast, we found no spatial cognitive coding in MC. In one variation of the allocentric condition, we switched the landmark and the object identities. This variation allowed us to establish that different landmarks share a common allocentric spatial representation, and further established that spatial encoding could not be attributed to low-level visual features of the task. These results suggest that PPC contains a neural population representation of cognitive spatial variables in addition to the egocentric variables, and that these cognitive variables are encoded independent of motor planning and execution.

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Poster

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Title: Decoding unfamiliar visual shapes from brain activity in human visual cortices

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Abstract: While decoding visual objects from functional brain activity when they fall into known categories, such as faces or places, is well established in humans, the multivariate decoding of novel, unfamiliar shapes is less studied. In order to fill this gap in knowledge, in the current work we presented six unknown shapes drawn from an old, now unused alphabet, to naive adult participants (N=17), performing a 1-back task. In a separate paradigm, the same participants also performed a 1-back task, yet on six distinct well-known visual objects (faces, body parts, places, words, numbers, manufactured objects). Participants performed similarly across these two types of stimuli. Indeed, high accuracy and fast reaction times in both paradigms indicated that participants could discriminate the six novel shapes even without knowing them *a priori*. We then built a multi-voxel pattern analysis (MVPA) decoder in the native space of each participant and for each experimental paradigm, using activation maps extracted from a trial-based general linear model. Our decoder considered the activation maps as input samples and performed supervised classification of the maps with respect to each of the 6 classes. Generalization error was assessed through nested cross validation, with the inner loop selecting the optimal hyper parameters by Tree-Parzen Estimator algorithm. By these means, we were able to decode both familiar (mean ± SEM over subjects: 60% ± 3%) and unfamiliar (46%. ± 3%) shapes with high accuracy, significantly above chance level. As expected, decoder performances were greater for well-known visual objects compared to unknown shapes. Our MVPA procedure started from the entire brain without prior biases towards specific brain regions. To ensure that relevant regions were selected and used by the decoder over the entire population, we computed probability maps, describing the stability of a given cluster of voxels to

perform classification across the whole population. Population-wide probability maps showed that all relevant classification clusters fell into visual areas. Our results demonstrate that novel, unfamiliar shapes can be accurately distinguished in the human visual cortex.

Disclosures: F. Guibert: None. **E. Pirondini:** None. **D. Bavelier:** None. **I. Altarelli:** None.

Poster

P482. Representation of Objects and Scenes I

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Program #/Poster #: P482.03

Topic: D.07. Vision

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Title: High-density neuronal recording reveals functional domains for shape selectivity across laminae in macaque V4

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Abstract: Previous studies have demonstrated that many neurons in macaque visual area V4 exhibit responses that are selective for the shape of an object; such shape selectivity can be well-described on the basis of the curvature of the object boundary at a specific angular position (APC model; Pasupathy and Connor, 2011). Recent studies using 2 photon calcium imaging and intrinsic optical imaging have reported functional domains for curvature encoding in the superficial layers of macaque V4 (Jiang et al., 2021; Hu et al., 2021). In our present work, we investigated the functional organization along the cortical depth and in high spatial resolution. We performed neuronal recordings in two fixating macaque monkeys as visual shape stimuli (120 two-dimensional shapes) were flashed (200 ms) separated by blank periods (200 ms) in random order, in the near-periphery. Each day we studied tens of well-isolated, nearby neurons using a Neuropixels probe. To quantify the similarity in shape preference for nearby neurons, we constructed a similarity matrix by computing the Pearson's correlation coefficient (r) between the shape responses for all pairs of simultaneously recorded neurons, Across many penetrations in both animals, we saw clear clusters of 5 or more neurons with high similarity in shape preference. These clusters were evident in both the deep and superficial layers. We next asked if the similarity in responses between neurons within the shape domains was based on similar tuning for boundary curvature. We fit an APC model for the responses of each neuron and compared the goodness of fit (GoF:correlation between observed and predicted responses) of the model for neurons within and outside the shape domains. We also compared the similarity matrices for each recording session based on responses predicted by the APC model versus

observed responses. Overall, we found that neurons within shape domains were better fit by the APC model and the responses predicted based on the APC model were also highly similar for neurons within the shape domains. Our preliminary results imply that V4 includes clusters of neurons with similar shape preferences and these neurons within shape domains may play a specialized role in the processing of boundary curvature.

Disclosures: T. Namima: None. **P. Zamarashkina:** None. **N. Owen:** None. **A.K. Pasupathy:** None.

Poster

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Title: Evidence of Object Based Warping in Retinotopic Cortex

Authors: *G. GURARIY[1], T. J. VICKERY[2], A. S. GREENBERG[1];
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Abstract: Objects are central to perception and arise from the organization of incoming visual signals. Although visual objects unfold over space, research has demonstrated that objects are capable of "warping" the very space over which they unfold - an effect known as Object-Based Warping (OBW; Vickery & Chun, 2010). This effect can be observed when two items placed within the contours of an object are perceived to be further apart relative to equidistant items viewed in the absence of an object. While numerous studies have quantified this effect behaviorally, the neural correlates of OBW have yet to be explored. In this study, we utilized fMRI to investigate the neural representation of OBW in human retinotopic cortex. First, a distance adjustment task performed outside the scanner was used to replicate previous findings and measure the strength of OBW in 22 participants. Next, a subset of these participants (n=7) who exhibited a behavioral effect of substantial magnitude completed an fMRI study in which they performed a letter discrimination task. The task included 3 conditions: (1) two letters enclosed within an object, (2) two letters without an object at the same spatial distances as in condition 1, and (3) two letters without an object at the "warped" distances (calculated from behavioral data). Separate localizers were used to identify retinotopic regions V1-V3 in addition to a spatial position localizer which was used to map retinotopic coordinates corresponding to the

spatial region in which the letters (real & warped) appeared. To identify the presence of warping, an MVPA classifier was trained on the two no-object conditions (conditions 2 & 3) and tested on the object-present condition (condition 1). Evidence of OBW was found in V1 & V2d, demonstrated by condition 1 (object present) being classified significantly above chance at matching condition 3 (object absent - warped). This warping effect in early visual cortex is likely driven by feedback from higher-level regions and suggests a fundamental change to visual perception caused by the presence of a simple object.

Disclosures: G. Gurariy: None. **T.J. Vickery:** None. **A.S. Greenberg:** None.

Poster

P482. Representation of Objects and Scenes I

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Program #/Poster #: P482.05

Topic: D.07. Vision

Support: NIH Grant R01 EB026943-01

Title: Understanding natural motion encoding in retinal populations

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Abstract: Over evolutionary time, the retina has been engineered to encode components of complex, natural environments that are most relevant for the organism. These components are essential for driving effective behavior. In this study, we use data from the salamander retina and a selection of different natural videos to train a deep neural network. We use this framework to explore and visualize what natural motion features are encoded in this early sensory neural population.

Because of sensory delays, whether finding prey, searching for a mate, or escaping from predators, animals must extrapolate natural motion trajectories to make effective interactions with their environment. We focus on the dynamic features of motion in natural scenes, to build on and differentiate from previous work that has mostly focussed on static features in the natural environment. This allows us to ask if the retinal encoding of natural motion reuses static representations.

We build a generative encoder-decoder network combining a cascade reconstruction network and variational autoencoders to reconstruct natural motion from the retinal population spike trains. Using this generative model, we observe that both the natural motion and static scene exhibit low dimensional structures. The low-dimensional structure of natural motion is distinct from the structure encoded by static scenes. Sampling from the low-dimensional structure of

natural motion, we identify features specific for natural motion of different directions (e.g., left to right, up to down), without scene details. Combining these motion features with specific scenes, we are able to synthesize naturalistic dynamic textures that mimic natural movies observed by the retina.

Disclosures: S. Wang: None. **O. Marre:** None. **M.J. Berry:** None. **S.E. Palmer:** None.

Poster

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Topic: D.07. Vision

Support: NRF-2019R1A2C4069863
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Title: Visual proportion sense in untrained deep neural networks

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Abstract: Visual proportion sense - the ability to estimate proportion between two visual magnitudes - is an essential cognitive function in animal survival (i.e. hunting, foraging, and mating), and it is widely observed in animals, humans, and even in infants without any prior knowledge (Jacob, 2012). Neurophysiological and neuroimaging experiments have found that several neurons in rhesus monkeys and voxels in adult humans show selective response to certain visual proportions (Vallentin, 2008; Jacob, 2009). However, how proportion sense can emerge without any learning is elusive, even in theoretical studies. A recent study showed that visual number sense can arise by hierarchical random feedforward connections in an untrained deep neural network (DNN), without any training (Kim, 2021). From this idea, we hypothesized that visual proportion sense can emerge in an untrained network without any training. We designed sets of images consisting of white (W) and black (B) dots, which represent specific proportions (i.e. W/(W+B) = 1/6), and fed these images into a randomly initialized DNN, AlexNet. In the designed images, the total number of dots can vary to 6, 12, or 18 dots, while they can represent the same proportion (i.e. W:(W+B) = 1:6, 2:12, and 3:18 dots = 1/6). In the deep layers of untrained AlexNet (Conv 3-5), we found proportion units in which responses are tuned to specific proportions invariant to the total number of dots in the images. These proportion units selectively and robustly respond to the preferred proportion, when the total number of dots varied from 2 to 30 (invariant to absolute number), and dot size, shape, or total area varied

(invariant to low-level image feature). Next, to examine the emergence mechanism of the proportion units, we tested two possible hypotheses - proportion units selectively receive input from 1) units that encode proportion in a specific total number of dots (i.e. 1/6 = W1 and B5, W2 and B10 ⋯) and 2) units that encode white/black increasing tendency - from the previous layers. From the weight analysis of the untrained network, we found that proportion units in layer 5 receive stronger inputs from white/black increment encoding units than number-specific proportion units in layer 4. In addition, ablation of increment units significantly diminished the emergence of the proportion units, whereas ablating number-specific units did not affect the emergence of the proportion units, supporting the latter hypothesis. In summary, our results suggest that visual proportion sense may arise without training and provide insights into how innate cognitive functions can emerge from a hierarchical network structure.

Disclosures: H. Lee: None. **W. Choi:** None. **S. Paik:** None.

Poster

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Topic: D.07. Vision

Support: NIH R01EY024678 (SK)

Title: Development of natural scene processing in primary visual cortex continues after the critical period for ocular dominance plasticity closes

Authors: *T. FUCHS, B. B. JEON, S. J. KUHLMAN;
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Abstract: The development of the visual system is known to be shaped by early-life experience. We recently demonstrated, using decoding methods, that mice raised without visual experience (dark-reared) have a deficit in natural scene processing (Kowalewski et al. 2021, Current Biology). In standard-reared mice, natural scene discriminability increases by 75% between the ages of 4 to 6 weeks. This increase in decoding accuracy is accompanied by a shift in neuronal preference towards natural scenes. Natural scene discriminability, therefore, continues to improve past the peak of the critical period for ocular dominance plasticity at postnatal day (p) 28. It is unknown whether there is a sensitive period for natural scene selectivity to mature, or whether continued visual experience is required for the maintenance of complex scene responses after their development. Here we hypothesize that natural scene representation in primary visual cortex (V1) becomes sparser with visual experience, and that visual experience continues to play a role in shaping natural scene responsiveness past the critical period for ocular dominance plasticity, well into adulthood. A sequential progression in which neurons first prefer simple

stimuli such as oriented bars of light (grating stimuli) prior to the development of selectivity for complex features may help ensure that the spatial organization of receptive fields is maintained as the system becomes refined to represent salient visual cues. We performed 2-photon calcium imaging of excitatory neurons in V1 in awake standard-reared mice at 4, 6 and 8 weeks of age and in an experimental group housed in darkness specifically between 4-6 weeks of age. Mice were presented with two classes of visual stimuli, 60 natural scenes containing complex features, and simple grating stimuli, spanning a range of spatial frequencies. We observed a shift in preference for complex features from 4 to 6 weeks, as well as 6 to 8 weeks in standard-reared mice. Visual deprivation specifically from p28 to p42 interfered with the full development of this preference shift and impaired natural scene decoding accuracy (Mann-Whitney U test, p=0.004). Our results indicate that in contrast to other aspects of visual processing, natural scene response maturation continues beyond the close of the critical periods for ocular dominance plasticity and binocular matching.

Disclosures: T. Fuchs: None. **B.B. Jeon:** None. **S.J. Kuhlman:** None.

Poster

P482. Representation of Objects and Scenes I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P482.08

Topic: D.07. Vision

Title: Feature representation in the Frontal Eye Field

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Abstract: Simultaneous expansion of the visual system and the Prefrontal Cortex in primate's brain can serve as evolution's clue on the link between these two. Given this idea, Prefrontal Cortex may be more involved in mapping the visual world into an internal representation than previously assumed. Frontal Eye Field is a visuomotor area of the Prefrontal Cortex. Since the discovery of FEF, much of the focus has been on its role in producing eye movements. Albeit, FEF's unique position of being extensively interconnected with visual cortical areas in both the dorsal and the ventral streams, along with its role in higher cognitive functions, makes it a probable candidate of an area with the ability to integrate the 'what' and the 'where' visual information. Given this supposition, we recorded the neural activity of FEF neurons of monkeys while they performed a Rapid Serial Visual Presentation task (RSVP). We calculated identity and category information of FEF neurons using a combination of encoding and decoding

methods. Our results suggest FEF neurons are in fact involved in representing both basic and complex features of objects.

Disclosures: A. Shahamati: None. **E. Rezayat:** None. **J. Noroozi:** None. **M. A. Dehaqani:** None.

Poster

P482. Representation of Objects and Scenes I

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Topic: D.07. Vision

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National Natural Science Foundation of China 31371111

Title: Orientation anisotropies in macaque visual areas V1 and V4

Authors: *C. FANG, X. CAI, H. D. LU;
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Abstract: In ferrets and cats, a larger number of neurons in V1 are devoted to cardinal (horizontal and vertical) orientations than those to the oblique orientations ("cardinal bias"). Other studies reported more neurons tended to prefer orientations connecting their RFs and the point of foveal fixation, a phenomenon called "radial bias". However, electrophysiological results from macaque monkey visual cortex were controversial on this aspect. Both isotropic and anisotropic orientation distributions have been reported. It is also unclear whether different visual areas along the visual processing have different orientation anisotropies. To investigate this, we analyzed orientation maps in a large set of existing V1 and V4 intrinsic signal optical imaging (ISOI) data. The stimuli we used were drifting gratings (1-1.5 c/deg) with 4 orientations, fully covered the visual field of the imaged cortex. Pixel-wise responses to 4 orientations were vector summed to obtain the preferred orientation for that pixel. For each case, the distribution of preferred orientation was calculated based on all V1 or V4 pixels in the images. We used a "cardinal + radial" combined model to fit the distributions and evaluated the amplitudes of the orientation anisotropies. We found that both V1 and V4 exhibited significant orientation anisotropies. However, their overrepresented orientations were very different: in V1, both cardinal and radial orientations were overrepresented, while in V4, only cardinal bias was presented. These findings suggest that different cortical areas have evolved to emphasize different features that suitable for their functional purposes, a factor that needs to be considered when efforts are made to explain the relationships between the visual environment and the cortical representation, and between the cortical representation and visual perception.

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Poster

P494. Visuomotor Processing: Behavior

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Program #/Poster #: P494.01

Topic: D.08. Visual Sensory-motor Processing

Support: ANII FCE 1 2019 1 155541
PEDECIBA

Title: The Novelty Response induced by visual stimuli in the weakly electric fish Gymnotus omarorum

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Abstract: Introduction. Electric fish of the family gymnotidae use self generated electric signals to explore their environment. Due to the uncoupling between fish movements and electrogenesis this is a paradigmatic reafferent sense. In *Gymnotus omarorum* electric organ discharge intervals (EODi) have a skew distribution with a small coefficient of variation. The skewness is due to novel sensory stimuli ('novelty response' NR) triggered by changes in reafferent electric images and also by mechanical, acoustical and electrical exafferent stimuli. These last improve, by means of increasing the sampling rate, the resolution of the main but short range sense of these fish. Here we characterize the effects of light on the EOD rate of *G. omarorum* particularly focusing on photically evoked NRs. **Methods.** In 15 fish EODi were recorded (after 1 hour of adaptation) using a differential amplifier and a Schmitt trigger circuit in a sound isolated cabin in which light was controlled. At noon and midnight (in each fish) the effects of continuous illumination and ligth-dark duty cicles were compared using non parametric statistics.**Results**. We found that: 1) during the day, the median, coefficient of variation and skewness of the EODi was smaller than during the night in both light and dark conditions; 2) continuos illumination decreased the EODi both at noon and midnight; 3) when studied using a 50-50% duty cycle with a 30 second period, on and off transition cause NRs that follow different time courses. Illumination onset were characterized by a decrease of the interval during the next 10 to 20 EODi. Darkening caused a slow decrease of the interval lasting 8 times more than lightening but reaching the same EODi reduction, both responses were followed by a slow relaxation lasting more than 100 EODi; no differences were found between day and night; 4) the amplitude of these NRs increased with the magnitud of the illumination increment; 5) no differences in the magnitude of the NRs were found when the duty stimulation cycle were altered in order to cause shorter or longer stimulation periods in steps of 100 EODs (approximately,4

s).**Conclusions**. Our data indicate variations of the visual sensitivity during the circadian cycle. On and off light transitions cause NRs with typical time courses, graded with the amplitude, but independent of the duty cycle of the stimuli and without day versus night difference suggesting a "wake up" effect.

Disclosures: A. Camargo: None. **A.A. Caputi:** None. **P.A. Aguilera:** None.

Poster

P494. Visuomotor Processing: Behavior

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Topic: D.08. Visual Sensory-motor Processing

Support: BRAIN 1UF1NS116377-01

Title: If a mouse can learn it, maybe a marmoset can, too: cross-species translation of the International Brain Laboratory task

Authors: *P. CHEN[1], D. ROWLEY[1], T. CZUBA[1], C. BADILLO[1], I. NAUHAUS[2], L. K. CORMACK[1], A. C. HUK[1];
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Abstract: The marmosets are quickly becoming a leading animal model in neuroscience for studying primate sensory processing and circuitry due to their small size and lissencephalic brains. However, there has been limited success with marmosets training for psychophysics, as they have not responded as well as other primates to strictly regimented tasks. The International Brain Laboratory (IBL) recently introduced a perceptual decision-making task that provides standardized and reproducible measurements of behavior in mice. Here we show that marmosets can learn an adapted version of the task in a controlled head fixed setting, though they require species-specific modifications.
In the IBL training task a Gabor patch with a randomized contrast is presented either on the left or right side of the center of gaze. The animal's task is to turn a wheel to move the visual stimulus from the periphery into the center. After a correct trial, a liquid reward is given, whereas a noise burst and extended intertrial interval follow an incorrect trial. We redesigned the steering-wheel chair used in the IBL task to suit the marmoset, scaling to body size and reorienting them to stand in the chair while holding the wheel.
Two common marmosets (callithrix jacchus) were able to learn the task at full and 50% contrast in only 3 or 4 consecutive training sessions. For habituation, the wheel was manually turned to help the marmosets associate the wheel's rotation with the motion of the stimulus, instead of passively viewing the stimulus jump to the center as in the mice task. A typical training session

was shorter with fewer completed trials and more trial timeouts than a mouse training session. Unlike mice, their performance did not drop when the contrast was lowered to 25%. The task was adapted to include more contrast levels below 6% to fully characterise their task performance.
The most immediate advantage to using this task is the cross-species comparisons that can be made by translating a widely used rodent behaviour task into the primate. These comparisons are key for understanding the degree of relevance that rodent literature has to the primate brain. This head-fixed task is also perfectly suited for use in conjunction with imaging and electrophysiological techniques. Finally, this task will serve as the initial primary training for more sophisticated psychophysical and cognitive tasks in projects to come and ultimately give us key insights into the processing of task-related signals.

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Poster

P494. Visuomotor Processing: Behavior

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Program #/Poster #: P494.03

Topic: D.08. Visual Sensory-motor Processing

Title: The effect of visual feedback and transcranial direct current stimulation (tDCS) on accuracy of ankle motor control

Authors: *M. CUMMINGS, A. DOSHI, F. IHMOUD, L. SHAH, S. MADHAVAN; Physical Therapy, Univ. of Illinois at Chicago, Chicago, IL

Abstract: Transcranial direct current stimulation (tDCS), a non-invasive neuromodulatory tool, has been shown to upregulate corticomotor pathways and improve motor learning and function in healthy individuals and those with stroke. Previous studies in adults with no neurological disorders have shown that the variability in ankle motor control is greater in conditions with no visual feedback than with visual feedback. In this study, our objective was to explore if application of tDCS over the primary lower limb motor cortex can enhance proprioceptive feedback and improve accuracy in motor control caused by the absence of visual feedback. We hypothesized that the accuracy of ankle motor control will decrease with lesser visual feedback, and that tDCS application will enhance accuracy in ankle motor control as visual feedback declines. Accuracy was examined by asking participants to perform ankle plantarflexion and dorsiflexion movements to match a computer-generated target sine wave using a custom-built ankle tracking device. The study was divided into two sessions, conducted a week apart, one with visual feedback (VF) and one with visual feedback and tDCS (VF-tDCS). Each session was divided into two blocks containing 3 trials of 4 randomized visual feedback

settings each. The visual feedback settings were: 1) 100% visual feedback: full visual feedback of the ankle position and target, 2) 50% visual feedback: visual feedback of the ankle position for the complete duration of the trial and target for half the time, 3) 0% visual feedback: visual feedback of the ankle position but not the target, and 4) no visual feedback: participants blindfolded with no visual feedback. During the VF-tDCS session, the first block included the application of 1 mA of tDCS for 20 mins. Accuracy was determined as the difference between ankle position and target, measured as the root mean square error, normalized to each participant's range of motion. Preliminary data from six healthy controls (25 participants to be collected) showed that accuracy of the ankle motor control task decreased with a decrease in visual feedback. Accuracy was 92% at 100%VF, 70% at 50%-VF, 62% at 0%-VF, and 61% for no-VF. During 100%, 50% and 0% VF-tDCS conditions, accuracy was demonstrated a trend to be higher than VF only. Accuracy was 93% at 100%VF-tDCS, 76% at 50%VF-tDCS, 65% at 0%VF-tDCS, and 60% at noVF-tDCS. Based on our preliminary results, tDCS may have the potential to improve accuracy of ankle motor control especially when visual feedback is not available. This could have implications in stroke populations where a decline in ankle proprioception maybe enhanced with neuromodulation using tDCS.

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Poster

P494. Visuomotor Processing: Behavior

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Program #/Poster #: P494.04

Topic: D.08. Visual Sensory-motor Processing

Title: M1 V1 - alpha/beta oscillations reflect task performance and movement variability during dynamic isometric pinch task

Authors: *C. HAN[1], L. D. ABRAHAM[2], M. MCMANIS[5], D. M. SCHNYER[3], J. S. SULZER[4], P. FERRARI[6,7,3];
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Abstract: Chungmin Han, Larry Abraham, David Schnyer, Mark McManis, James Sulzer, Paul Ferrari
Title: M1 V1 - alpha/beta oscillations reflect task performance and movement variability during dynamic isometric pinch task
Visuomotor control for precision pinch and grasp relies on an extensive cortical network for

which research has traditionally focused on frontal and parietal regions subserving executive and visuomotor integration functions, respectively. However, evidence suggests that cortical oscillations within the primary motor and visual cortices can reflect both low and high-level functions. Here we used MEG (MEGIN Oy, Ltd.) to non-invasively measure dynamic oscillatory activity in V1 and M1 to investigate their contribution to performance in an isometric pinch force task. A custom MEG-compatible sensor measured forefinger and thumb forces separately, which controlled the position of a cursor on the screen. The task required subjects to control finger forces normalized to <20% of maximum voluntary contraction (MVC) at increasing and decreasing magnitudes as they tracked a visual target along a 45-degree line. In an 'Independent' condition, forefinger and thumb forces were mapped to the cursor's x and y positions, respectively, in full 2D space. A 'Coupled' task was also performed where the cursor position was constrained along the diagonal task trajectory. A total of 14 subjects (7 females; mean 25.8 yrs) performed 120 trials of each condition. Cortical source activity was analyzed using a beamformer inverse solution using the Matlab toolbox Brainwave. As expected, subjects performed better for the coupled vs the independent condition (RMSE, $p<0.05$). However, force variability was greater in the coupled vs independent condition, creating a dissociation between motor output and visual feedback. We observed strong event-related beta-band desynchronization during the task, with transient beta-rebound synchrony during a change of movement trajectory in both motor and visual cortices. Visual alpha did not show a corresponding transient rebound during trajectory changes, indicating the visual cortex was mapping movement within a motor (beta) oscillation. Furthermore, an analysis of covariation across trials showed that for both M1 and V1, alpha band reflects fluctuations of visual feedback in real-time during the task ($F(1,14)=7.8$, $p=0.015$), while beta band is associated with the level of movement control to achieve high accuracy ($F(1,14)=4.8$, $p=0.047$). Results suggest that cortical oscillations in both M1 and V1 can dissociate task and movement parameters during dynamic pinch tasks and that they may share a common beta network for visuomotor control.

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Poster

P494. Visuomotor Processing: Behavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P494.05

Topic: D.08. Visual Sensory-motor Processing

Support: grant from the John Templeton Foundation
grant from the Fetzer Institute

Title: Brain correlates of visuomotor activation due to conscious and unconscious visual perception

Authors: *A. I. TRIGGIANI, S.-J. LEE, M. HALLETT;
Human Motor Control Section, Natl. Inst. of Neurolog. Disorders and Stroke, Bethesda, MD

Abstract: Subjectively, we have a sense that consciousness has a causal role in our voluntary movements. We do what we have decided to do. However, there is no formal proof that consciousness is causal. By using backwardly masked stimuli in a choice reaction time experiment, subjects made similar movements when they believed the movement was voluntary and when they believed it was a mistake. We explored the brain activity in these two situations.Fifteen healthy adults participated in a variation of a classic backward-masking task with electroencephalographic (EEG) recording. They had to press a button using the right or the left hand, according to an arrow on the screen. Arrows were presented at time S1, 50 ms later at time S2, or both times. When there were two arrows, the first was masked by the second, congruent or incongruent. When incongruent, they might respond to S1 or S2.When responding to S1 but seeing S2, they should believe that it was a mistake, as verified in a separate series of trials after the main experiment.Responses are to S1 for the situation of S1 alone, S1 masked and congruent, and S1 masked, incongruent, but with S1 response. In the first two, the experience is that it is a voluntary response. In the third situation, the experience is that the movement is not in accord with the consciousness of the stimulus, and it is a mistake. Indeed, in this last situation, the EEG confirms the subjectivity of the mistake with a manifestation of error-related negativity (ERN) followed by error-related positivity (ERP).The EEG showed a different cortical activation in all three situations, but, at a parietal level, the electrical activity in response to S1 alone was like S1 masked by a congruent S2. The parietal cortical activity related to the response to S1 in the incongruent condition was different from the other responses to S1. In the incongruent condition, despite similar primary sensory activation, the parietal EEG showed differences when the subject responded to S1 or S2 ($p<0.001$). The former was faster and more persistent, and the latter was higher, but slower and shorter.Results suggest that whether S1 is visible or masked in the congruent condition, the person sees and responds to S1. When S1 is masked in the incongruent condition, it is wiped out before being consciously perceived and replaced by S2. In this case, the S1 motor response is already released. This incompatibility of the motor response and the consciousness is revealed by the ERN/ERP and the behavioral test. Different processing of unseen stimuli seems to lead to the same motor outputs. Relating the brain activity to conscious experience will help answer the question of a causal role of consciousness in the choice of movement.

Disclosures: A.I. Triggiani: None. **S. Lee:** None. **M. Hallett:** None.

Poster

P494. Visuomotor Processing: Behavior

Location: Virtual Only

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Program #/Poster #: P494.06

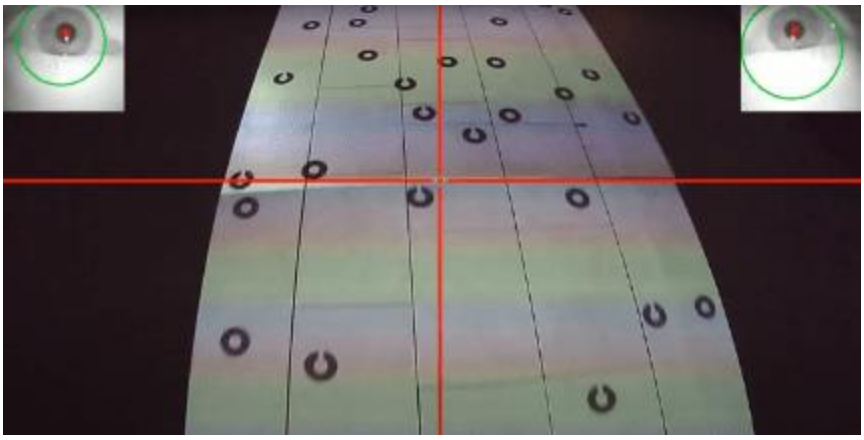
Topic: D.08. Visual Sensory-motor Processing

Support: NIH R00-EY028229

Title: Terrain complexity and the relationship between gaze and locomotion

Authors: *T. D. WIRTH, S. E. REINHART, R. L. HUYNH, J. S. MATTHIS;
Northeastern Univ., Boston, MA

Abstract: Successful human locomotion over complex terrain relies on interaction between the visual and motor systems. Investigations into recently accumulated datasets of visuo-locomotor behavior in natural environments (Matthis, Yates, & Hayhoe, 2018) demonstrates that the relationship between gaze and foot placement is tied to the complexity of the terrain (e.g. walking across flat ground vs a rocky creek bed). However, because these were quasi-observational experiments in a natural environment, it is unclear which aspects of the terrain were driving the relationship between gaze and locomotion. Examination of how the environment forms synergetic relationships between the visual and locomotor systems is critical in developing a basic-science understanding of visuo-locomotor behavior. Here, we present a newly developed paradigm which allows parametric control over an Augmented Reality Projected Groundplane (ARPG ~2x10m) stimulus, synchronized with full body kinematic motion capture (Qualisys) while binocular gaze data from Pupil Labs was spatiotemporally integrated via post-hoc calibration. Using eye tracking and full body motion capture, we will reconstruct the gaze-ground intersection to find where participants are looking at any given moment. In this experiment, participants (N=5) engage in a stepping stone task where they cross the 10m ARPG while stepping on only target Landolt C's and avoiding distractor O's as well as empty space. We manipulate the visual difficulty of the task by changing the similarity between the C's and O's (6mm vs. 36mm sized C-gap), and we manipulate the mechanical difficulty of the task by changing the ratio of presented targets to distractors (3:1, 1:1 or1:3). Each trial was repeated 20 times for each of the 6 conditions, resulting in 120 trials and 1600 steps per participant. Each condition was presented in randomized blocked order. With this new paradigm, we take our first step towards understanding how visual and mechanical terrain complexities interact to drive the connection between gaze and foot placement during locomotion over natural terrain.



Disclosures: T.D. Wirth: None. **S.E. Reinhart:** None. **R.L. Huynh:** None. **J.S. Matthis:** None.

Poster

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Program #/Poster #: P494.07

Topic: D.08. Visual Sensory-motor Processing

Support: NIH Grant R01NS053606

Title: An education-friendly tutorial for visuomotor experiments

Authors: *N. AGHAMOHAMMADI, M. DEMASI, A. ANAND, J. L. PATTON;
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Abstract: Targeted reaching experimentation is the bread and butter of motor control research, where subjects are typically asked to move the handle of a manipulandum or a robot, allowing analysis of error statistics, variability, adaptation, or other aspects. Despite their widespread use in research, they are not conducive to classroom education as they usually require sophisticated, expensive, and specialty tools such as robots. Such costly hardware dependence restricts widespread adoption and accessibility. Here we describe a proposed educational tool for neuroscience and biomedical engineering classrooms and laboratories. This paper presents a step-by-step visual-rotation adaptation reaching tutorial using open-access code, any computer, and any mouse or pointing device, and the freely accessible Unity® game-engine. By the end of the tutorial, the student will know how to set up a complete visuomotor adaptation experiment that demonstrates neuroplasticity in a hand-eye coordination task. Our work performed on this tool resulted in real evidence of motor learning, with significant after-effects of adaptation. This fundamental building block can be expanded to other experiments and lends itself well to online education, rehabilitation, and computational neuroscience.

Disclosures: N. Aghamohammadi: None. **M. Demasi:** None. **A. Anand:** None. **J.L. Patton:** A. Employment/Salary (full or part-time):; Shirley Ryan Ability Lab.

Poster

P494. Visuomotor Processing: Behavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P494.08

Topic: D.08. Visual Sensory-motor Processing

Title: Relationship Between Design Parameters of Head-Up Display and Human Visual Information Processing

Authors: *N. YAMADA, K. UEDA, S. TAKADA, M. NAKAO;
Univ. of Tokyo, Tokyo, Japan

Abstract: A head-up display (HUD) is a device that uses the windshield of a vehicle or other mobility device as a screen to display augmented reality images in the traffic environment. This is a type of driver-assistance technology and is expected to contribute to road traffic safety. To design a HUD that allows drivers to safely view images while driving, it is necessary to understand the visual information processing that occurs when humans view images. Visual information processing can be divided into three processes: "visual input" to input images from the eyes to the brain, "cognitive judgment" to understand the content of the images, and "motor output" to execute the actions directed by the images. In this study, we focused on two HUD design parameters, namely the optical image projection method and the image display distance, and investigated the effects of these two parameters on the three processes of visual information processing. Optical image projection methods involve real image projections or imaginary image projections. In the former, the screen was set at the same distance as the image, and in the latter, the screen was set at a closer distance than the image. There were two levels of distance-displaying images: 5 m and 10 m. In the experiment, the HUD image was sometimes displayed while a driving simulator was displayed 20 m away, and the participants answered which of the two patterns the image shape belonged to using two buttons on the steering wheel during driving. For the "visual input," the time required for the eye movement when viewing the HUD image was evaluated via eye movement measurement, and for the "cognitive judgment," the peak latency of P300 when understanding the shape of the HUD image was evaluated by electroencephalogram (EEG) measurement. In addition, the time taken for all the processes including the "motor output" was evaluated by the reaction time of the button press. Participants were divided into two groups: a younger group consisting of men in their 20s and an older group consisting of men in their 40s and 50s. In the younger group, there was no difference in the time required for each process of visual information processing for any of the HUD design parameters; however, in the older group, the time required for visual information processing was

prolonged when the projection method was the imaginary projection. In this presentation, we will discuss and report this difference based on the details of the gaze measurement and EEG data.

Disclosures: N. Yamada: None. **K. Ueda:** None. **S. Takada:** None. **M. Nakao:** None.

Poster

P494. Visuomotor Processing: Behavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P494.09

Topic: D.07. Vision

Title: Visual circuits modulating aggressive behavior in mice

Authors: *M. WAN[1], M. ARANDA[2], T. M. SCHMIDT[3];
[2]Dept. of Neurobio., [3]Neurobio., [1]Northwestern Univ., Evanston, IL

Abstract: VISUAL CIRCUITS MODULATING AGGRESSIVE BEHAVIOR IN MICE*Mingyu Wan, Marcos L. Aranda, Tiffany M. SchmidtDepartment of Neurobiology, Northwestern University, Evanston, IL*Animals must interact across a range of situations, some of which may lead to social interactions,and others which may lead to aggressive interactions. However, the role of the visual system inmodulating such interactions is not well understood. Intrinsically photosensitive retinal ganglioncells (ipRGCs) express the photopigment melanopsin (encoded by the Opn4 gene) and detectlight directly in absence of rods and cones. Brn3b-positive ipRGCs innervate brain areas involvedin social and aggressive behaviors, suggesting ipRGCs may modulate these types of behavior.The goal of this work was to identify the role of Brn3b-positive ipRGCs in social and aggressivebehaviors. To do this, we used the Resident-Intruder Aggression Test combined with deeplearning to compare social and aggressive behaviors in mice where Brn3b is conditionally deletedspecifically in ipRGCs ($Opn4^{Cre/+}$ $Brn3b^{cKO/cKO}$; referred to as Brn3bcKO). Surprisingly, weobserved a significant increase in aggressive behavior in Brn3bcKO mice compared to controllittermates, but no differences in social behavior. To identify the brain regions via which ipRGCsmay modulate aggression, we next measured expression of the immediate early gene c-Fosfollowing the Resident-Intruder Aggression Test and found greater c-Fos densities in the Dorsal-Medial Hypothalamus(DMH)the Lateral Hypothalamic Area (LHA), the medial amygdala(MEA),and the Basolateral Amygdala(BLA). Viral labeling of ipRGC axons showed that ipRGCs innervateboth the LHA and MEA, suggesting that these regions are candidates for modulation ofaggressive behaviors. Additionally, we observed a significant correlation between c-Fos densityin LHA with aggressive behaviors, suggesting an ipRGC-LHA circuit may modulate aggressivebehaviors. In order to test the sufficiency of LHA-projecting ipRGCs to modulate aggression levels,we chemo genetically excited LHA-projecting ipRGCs in $Opn4^{Cre/+}$ mice. We found that excitationof LHA-

projecting ipRGCs increased the duration of aggressive behaviors. These results suggestthe ipRGC-LHA circuit modulates aggression in mice.

Disclosures: M. Wan: None. **M. Aranda:** None. **T.M. Schmidt:** None.

Poster

P573. Motor Unit Recordings, Kinematics, and EMG I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P573.01

Topic: E.09. Motor Neurons and Muscle

Support: T32HD043730

Title: Trial-to-trial variability of multi motor unit activity accounts for worse endpoint control at low force levels

Authors: *S. DELMAS[1], Y. CHOI[1], B. YACOUBI KEYHANI[1], E. A. CHRISTOU[1,2]; [1]Applied Physiol. and Kinesiology, Univ. of Florida, Gainesville, FL; [2]Neurol., Norman Fixel Inst. of Neurolog. Disorders, Univ. of Florida, Gainesville, FL

Abstract: A robust observation in human motor control experiments is that the endpoint control of pre-planned (open loop) contractions is worse at low force levels relative to moderate or high force levels. Despite that the first observations originated in 1892, the underlying neuromuscular control that explains such robust behavioral differences remains unknown. Here, we use the abduction of the index finger, an experimental paradigm that requires control of a single muscle contraction (first dorsal interosseous; FDI), to study the multiple motor unit discharge characteristics that underlie this phenomenon. We recruited 15 young adults who participated in two distinct experiments that used goal-directed pre-planned contractions with the index finger. For both experiments, we quantified endpoint control as the relative endpoint error and variability in force and time and characterized the activity of multiple motor units in the FDI. For each contraction we summated the active motor units and quantified the following across 100 trials: 1) The average of the mean and coefficient of variation (CV) of the discharge rate (DR). 2) The trial-to-trial variability (CV) of the mean and coefficient of variation (CV) of the discharge rate (DR). 3) The average power from 3-8, 8-13, 13-33, and 33-60 Hz. 4) The trial-to-trial variability of power from 3-8, 8-13, 13-33, and 33-60 Hz. In the first experiment, participants aimed to match a force target either at 10% or 40% MVC while attempting to reverse their peak force at 160 ms. The greater force and time endpoint error at 10% relative to 40% MVC associated with greater force and time endpoint variability ($R2 = 0.34$, $p = 0.01$). The increased force ($R2 = 0.34$, $p = 0.01$) and time ($R2 = 0.23$, $p = 0.04$) endpoint variability at 10% relative to 40% associated only with increased trial-to-trial variability of the CV DR, which related to an increase in the variability from 13-33 Hz ($R2 = 0.56$, $p < 0.01$) and 33-60 Hz ($R2 = 0.53$, $p <$

0.01). In the second experiment, we examined if changing the speed of the contraction altered endpoint control and multi motor unit discharge characteristics. Thus, participants aimed to match a 40% MVC force target at either 160 or 280 ms. We found that the endpoint control and motor unit discharge characteristics were similar between the two contraction speeds. In summary, we provide novel findings showing that the worse endpoint control at low force levels relates to an inability to consistently repeat the motor unit discharge across trials.

Disclosures: S. Delmas: None. **Y. Choi:** None. **B. Yacoubi Keyhani:** None. **E.A. Christou:** None.

Poster

P573. Motor Unit Recordings, Kinematics, and EMG I

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Program #/Poster #: P573.02

Topic: E.09. Motor Neurons and Muscle

Support: NSERC Discovery Grant 180970
NSERC CGS-D 519372

Title: Motor unit firing rate trajectories in agonist muscles during elbow extension movements

Authors: *E. A. KIRK, C. L. RICE;
Western Univ., London, ON, Canada

Abstract: Voluntary movements ultimately are controlled by skeletal muscle contractions that create force in response to neural inputs from the spinal cord or brainstem. These inputs originate from lower motor neuron discharges that synapse onto innervated muscle fibres, defined as a motor unit (MU). Despite extensive experimental evidence during sustained isometric contractions, how the MU firing rate trajectory controls movement remains elusive. Our purpose was to measure MU firings rates as a function of time comparing firing rate trajectories of MU trains during sustained isometric contractions and elbow extension movements. In awake human participants, single MU activity was recorded by intramuscular electromyography from the anconeus (n = 15) and lateral head of the triceps brachii (n = 6) muscles during elbow extension contractions at various intensities, up to maximum efforts. From single MU trains, the change of firing rate was measured as a function of contraction time course using simple linear regression and was adjusted based on firing rate variance explained by elbow extension torque. For comparisons between contraction conditions, MU trains were unpaired. We hypothesized that MU firing rates would have greater changes (increase or decrease) during elbow extension movements that have greater effects of muscle shortening, as compared to sustained isometric contractions. Altogether, 42 MU trains during sustained isometric contractions (fixed 90 deg) and 222 MU trains during elbow extension movements (120 deg range of motion) were

measured. During increasing torque of isometric contractions, the firing rate trajectory was positive in 88% of measured MU trains. In contrast, the firing rate trajectory was significantly different (likelihood ratio test, chi-squared = 32±9.5; $p < 0.001$) and negative in 88% of measured MU trains during elbow extension movements at different targeted velocities. Through correlation analysis, the magnitude of the decreasing firing rate trajectory was related to both the targeted velocity (rho = -0.43, $p < 0.01$) and the MU recruitment threshold of related torque ($r = -0.35$, $p < 0.01$), that were different than during isometric contractions. These findings show that neural drive to the shortening muscle during voluntary movements decreases with contraction time course, suggesting a different pattern of motor command as compared to sustained isometric contraction. Supported by NSERC.

Disclosures: E.A. Kirk: None. **C.L. Rice:** None.

Poster

P573. Motor Unit Recordings, Kinematics, and EMG I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P573.03

Topic: E.09. Motor Neurons and Muscle

Support: NSERC: 18079

Title: Firing rate responses of human motor units during muscle potentiation

Authors: *A. M. ZERO, E. A. KIRK, C. L. RICE;
Univ. of Western Ontario, London, ON, Canada

Abstract: Post-activation potentiation (PAP) is the acute enhancement of submaximal contractile properties following a short duration high intensity contraction. During this enhanced contractile state (PAP) submaximal MUFRs are depressed, but the mechanisms or relationship for this response is unknown. Persistent inward currents (PICs) are long-lasting depolarizing intrinsic currents of the motoneuron and contribute to the net excitatory synaptic input of the motoneuron pool directly altering motor unit (MU) firing rate (FR) patterns. During small increments of voluntary isometric torque (proxy of increasing extrinsic synaptic input) MUs exhibit initial FR acceleration followed by saturation demonstrating non-linear responses to linear torque increases. PICs amplify synaptic input up to ~fivefold and thus during graded isometric contractions MUFRs will briefly accelerate following recruitment and then saturate. If there is a minimal PIC response or intrinsic synaptic amplification MUFRs will increase linearly. The purpose was to explore and compare the occurrence of MUFR saturation during isometric ramp contractions when the target muscle is either potentiated (PAP) or non-potentiated (control). We hypothesized that with PAP, MUFRs will be lower and will increase linearly, but under control conditions will briefly accelerate and then saturate. During dorsiflexion, graded

isometric ramp contractions up to 10% of maximal voluntary contraction (MVC) occurred over 10 seconds, with single MU potentials followed using two pairs of bipolar fine-wire electrodes inserted into the tibialis anterior muscle. The control condition consisted of only slow ramp contractions from rest, whereas the PAP condition was induced by a 5s ramp to MVC followed by a 5s hold, that was immediately followed by slow ramp contractions. To characterize the firing rate trajectory, each MUFR was fit to competing functions of linear regression and a negative exponential (i.e. saturation) in response to isometric torque (Fuglevand et al. 2015). From the 5 MUs obtained so far from 3 participants there was a significant decrease of 13% ($p<0.05$) in MUFRs during PAP (11.5 ± 2.7 Hz) compared to control (13 Hz ± 2.9 Hz). During the control condition MUs were better fit to the exponential (saturation) function (5/5; $p<0.05$), but under the PAP condition the same MUs were better fit to the linear function (5/5; $p<0.05$). These initial results indicate a smaller PIC or intrinsic motoneuron amplification response during PAP may contribute to the decrease of MUFRs. Supported by NSERC

Disclosures: A.M. Zero: None. **E.A. Kirk:** None. **C.L. Rice:** None.

Poster

P573. Motor Unit Recordings, Kinematics, and EMG I

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Program #/Poster #: P573.04

Topic: E.09. Motor Neurons and Muscle

Support: Supported by CUHK Faculty Innovation Award (FIA/2016/A/04) to VCKC.

Title: Neural representation of hindlimb muscle synergies in the mouse spinal cord revealed by spike-triggered averages of optogenetically elicited electromyographic activities

Authors: *B. HE[1], P. SALMAS[1], J. ZHANG[2], X. DUAN[3], V. C.-K. CHEUNG[1,4];
[1]The Chinese Univ. of Hong Kong, Hong Kong, China; [2]The James Franck Inst., Chicago, IL; [3]Col. of Future Technol., Peking, China; [4]The Chinese Univ. of Hong Kong and Kunming Inst. of Zoology of The Chinese Acad. of Sci., Hong Kong, China

Abstract: Movement execution requires the motor system to coordinate a large number of neuro-musculoskeletal elements. To simplify motor control, it has been proposed that spinal interneurons are organized into functional motor modules which activate different muscles concurrently as muscle synergies, and are recruited with distinct temporal activation profiles. Though previous studies using electrical micro-stimulation of the spinal cord delivered during electromyographic (EMG) and neural recordings have found that the spinal premotor interneurons underlie the muscle synergies, less is known about the specific interneuronal types that contribute to their organization. Here, we use optogenetics to selectively stimulate excitatory spinal interneurons in the anesthetized Thy1-ChR2 transgenic mice while simultaneously

recording intramuscular EMG (6 hindlimb muscles) and neural spike trains of spinal interneurons. Neural activities were recorded using soft carbon nanotube (CNT) electrodes. Since their shapes are not rigid, CNT electrodes do not require any holder for mechanical support at the recording sites, thus leaving the exposed spinal cord surface free of obstacles. This in turn allowed us to stimulate the lumbosacral spinal cord comprehensively by moving the laser in steps of 200-μm increment along the spinal surface. Using spike-triggered averages of EMG, the post-spike effect of a spinal interneuron on the recorded muscles was quantified as a muscle field. We found that the muscle fields of the recorded interneurons matched the muscle synergies extracted from EMG by nonnegative matrix factorization; the temporal activities of the interneurons also correlated highly with the synergies' activation coefficients. We argue that during locomotion, the hindlimb muscles are activated not randomly, but through recruitment of the muscle synergies with their temporal activities modified by the spinal interneurons. Furthermore, different synergies may result from different combinations of interneurons with distinct muscle fields. Nevertheless, the synergies' activation coefficients may be specified by a few specific interneurons responsible for precise temporal control of the muscle synergies.
Supported by CUHK Faculty Innovation Award (FIA/2016/A/04) to VCKC.

Disclosures: B. He: None. **P. Salmas:** None. **J. Zhang:** None. **X. Duan:** None. **V.C. Cheung:** None.

Poster

P573. Motor Unit Recordings, Kinematics, and EMG I

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Program #/Poster #: P573.05

Topic: E.09. Motor Neurons and Muscle

Support: NSERC Grant 18079

Title: Stimulated disruption of cortical or spinal activity during voluntary movement preparation

Authors: *A. D. PAISH[1], E. A. KIRK[1], C. L. RICE[1,2];
[1]Sch. of Kinesiology, Fac. of Hlth. Sci., [2]Dept. of Anat. and Cell Biology, Schulich Sch. of Med. and Dent., The Univ. of Western Ontario, London, ON, Canada

Abstract: Motor planning involves complex interactions of neural pathways to execute muscle contractions. In humans, transcranial magnetic stimulation (TMS) and cervicomedullary evoked potentials (CMEPs) are techniques used to investigate the neural contributions from supraspinal (cortical) and spinal portions, respectively. Moreover, these techniques can also be used to disrupt the timing of voluntary motor outputs, as prior studies have shown that supraspinal disruption using TMS results in delayed movement onset, while movement kinematics (i.e., torque, velocity and position) are otherwise unhindered. However, pre-movement disruption at

the spinal level (i.e., CMEP), or supraspinal levels (i.e., TMS) during maximal voluntary efforts have not been tested. Thusly, we assessed the electromyographical (EMG) and kinematic responses throughout reaction based elbow extension movements, with and without suprathreshold TMS or CMEP delivered during the preparatory time of voluntary movement. We hypothesized that a single TMS pulse to the primary motor cortex will both delay reaction time and decrease movement kinematic potency during maximal voluntary efforts, whereas, evoked CMEPs at the spinal level will have minimal effect. In five participants, muscle activity was recorded through surface and intramuscular EMG electrodes from the anconeus, triceps brachii and biceps brachii muscles. Before a visual go-cue, the contraction condition was randomized between submaximal and maximal elbow extension movements in an arm dynamometer. Prior to EMG activity, one of three stimulation conditions was randomly executed: TMS, CMEP or control (no stimulation). Each condition was repeated at least three times per participant. We found that the reaction time (triceps brachii EMG onset) was significantly increased ($\bar{x}\approx$107ms; +48%) with TMS compared to control, whereas the CMEP reduced the reaction time ($\bar{x}\approx$50ms; -18%). Peak torque and velocity, as well as, time to peak torque, velocity, and position were significantly lower with TMS (10%, 6%, 9%, 11%, and 6%, respectively), whereas the time to peak torque and velocity were quickened (11% and 6%, respectively) with a CMEP compared to control. These results indicate an inability to fully recover motor execution following cortical disruption (TMS) during maximal voluntary elbow extension movements. Yet, disruption at the spinal cord (CMEP), and presumably the motor unit pool, quickened movement outcomes.
Supported by NSERC.

Disclosures: A.D. Paish: None. **E.A. Kirk:** None. **C.L. Rice:** None.

Poster

P573. Motor Unit Recordings, Kinematics, and EMG I

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Program #/Poster #: P573.06

Topic: E.09. Motor Neurons and Muscle

Support: CUHK Group Research Scheme (NL/JW/rc/grs1819/0426/19hc)
HK Research Grants Council grant CUHK-R4022-18

Title: Mapping motor output units encoded in the human primary motor cortex with transcranial magnetic stimulation

Authors: *J. J. XIE[1], I. W. Y. CHOU[2], K. Y. S. LAU[1], B. HE[1], P. P. K. CHAN[3], O. N. W. LEUNG[2], R. T. H. CHEUNG[4], A. D. P. MAK[2], V. C. K. CHEUNG[1];
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Abstract: The diversity of voluntary movement originates from control mechanisms in the central nervous system (CNS). Previous studies have suggested that the CNS generates movement by translating motor intentions into commands for a small number of motor output units called muscle synergies that activate muscle groups involved in different behaviors. However, physiological evidence for the existence of muscle synergies in humans is lacking, and thus synergy-based movement generation has not been broadly accepted. Here, we aim to provide evidence to support the existence of lower-limb muscle synergies in the human CNS by applying focal, noninvasive transcranial magnetic stimulation (TMS) to the primary motor cortex (M1). Surface electromyographic signals (EMGs) were recorded from 16 lower-limb and trunk muscles of 3 healthy subjects during multiple voluntary locomotor tasks, and during the neuro-navigated TMS stimulations which were delivered with a double cone coil applied over the subject's scalp along with a grid corresponding to the leg region of M1. Muscle synergies were extracted from the behavioral EMGs and TMS-derived motor evoked potentials using nonnegative matrix factorization. The TMS-derived muscle synergies were then matched to the behavioral synergies. For each TMS synergy, its representation over M1 was reconstructed by mapping its activities to the spatial locations of the stimulated M1 loci. The blood-oxygen-level-dependent signals during motor imagery of locomotor tasks were also acquired through functional magnetic resonance imaging to build an activity map that could be compared with the muscle-synergy representation map from TMS. We found that across all subjects, 9 of 16 behavioral muscle synergies could be well matched to the TMS-derived synergies elicited from the same subject (scalar product >0.8). The topographic representations of different TMS-derived synergies overlapped substantially with each other over M1. Our result demonstrates that it is possible to access and map out muscle synergies with TMS, thereby revealing how muscle synergies for locomotion may be encoded in M1. The unmatched behavioral synergies may arise from spinal circuits. Our data provide direct evidence for the existence of at least a subset of locomotor muscle synergies in humans.

Disclosures: J.J. Xie: None. **I.W.Y. Chou:** None. **K.Y.S. Lau:** None. **B. He:** None. **P.P.K. Chan:** None. **O.N.W. Leung:** None. **R.T.H. Cheung:** None. **A.D.P. Mak:** None. **V.C.K. Cheung:** None.

Poster

P573. Motor Unit Recordings, Kinematics, and EMG I

Location: Virtual Only

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Program #/Poster #: P573.07

Topic: E.09. Motor Neurons and Muscle

Title: Fatigue-related changes in intermuscular coherence of rotator cuff and deltoid muscles

Authors: *X. YU[1], L. HAMMOND[2], H. ZHU[2], Z. WANG[3], L. GRIFFIN[4];
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Abstract: Maintaining shoulder joint stability requires coordination of the rotator cuff and deltoid muscles, neuromuscular fatigue may change these coordination patterns. EMG coherence between two muscles indicates that they have a common descending input, this common drive is important because it maintains joint stability. However, it has not been investigated during fatigue. Eight healthy individuals (mean age ± SD = 19.13 ± 0.99 years, 6 females, 2 males) participated in this study. Surface EMG activity was recorded from the anterior deltoid (AD), middle deltoid (MD), supraspinatus (SS) and infraspinatus (IS). Participants performed an isometric fatiguing contraction at 30° scaption at 25% maximal voluntary contraction (MVC) until endurance limit (the time at which the force deviated from the target by ± 5% MVC). Performance time was divided into 3 phases: initial, middle and final. As fatigue progressed, the EMG RMS amplitude of all muscles significantly increased, slopes of EMG median frequency of all muscles were significantly less than zero, with AD exhibiting the highest fatigue rate (-0.33 ± 0.02 %/phase, p=0.00). EMG coherence of each muscle pair (SS-IS, SS-AD, SS-MD, IS-AD, IS-MD) in the common drive (0-5 Hz), physiological force tremor (5-12 Hz), beta band (15-35 Hz), Piper band (35-60 Hz) and gamma band (60-90 Hz) in each fatigue phase were measured. A Friedman's and Wilcoxon signed-rank test revealed that coherence of SS-MD significantly increased with fatigue in all frequency bands except for the common drive band. SS-AD coherence exhibited a significant increase in the tremor and beta bands. Coherence of IS-MD and IS-AD also significantly increased in the tremor, beta and Piper bands. However, there were significantly lower SS-IS coherent oscillations in all frequency bands except the tremor band as the fatigue level increased. Thus, neuromuscular fatigue induced less common drive within the rotator cuff muscles than between rotator cuff and deltoid muscles. Greater coherence between rotator cuff and deltoid muscles allows these two muscle groups to contract in a more synergistic manner to maintain force output during neuromuscular fatigue.

Disclosures: X. Yu: None. **L. Hammond:** None. **H. Zhu:** None. **Z. Wang:** None. **L. Griffin:** None.

Poster

P573. Motor Unit Recordings, Kinematics, and EMG I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P573.08

Topic: E.09. Motor Neurons and Muscle

Support: NIH NINDS R01NS098509

NIH NINDS R01HD039343

Title: Estimates of persistent inward currents are reduced in upper limb motor units of older adults

Authors: *M. E. FAJARDO[1,2], A. S. HASSAN[2,3], M. CUMMINGS[6,2], L. M. MCPHERSON[7,8], F. NEGRO[9], J. P. A. DEWALD[2,3,4], C. J. HECKMAN[1,2,4,5], G. E. P. PEARCEY[1,2,4,5]; [1]Dept. of Physiol., [2]Dept. of Physical Therapy and Human Movement Sci., [3]Dept. of Biomed. Engin., [4]Dept. of Physical Med. and Rehabil., [5]Shirley Ryan AbilityLab, Northwestern Univ., Chicago, IL; [6]Dept. of Physical Therapy, Univ. of Illinois at Chicago, Chicago, IL; [8]Dept. of Neurol., [7]Washington Univ. Sch. of Med., St. Louis, MO; [9]Univ. degli Studi di Brescia, Dept. of Clin. and Exptl. Sci., Brescia, Italy

Abstract: Aging is a natural process that causes alterations in the neuromuscular system, which contribute to weakness and reduced quality of life. Reduced firing rates of individual motor units (MUs) likely contribute to weakness, but the mechanisms underlying reduced firing rates are not clear. Persistent inward currents (PICs) are crucial for the initiation, gain control, and maintenance of motoneuron firing, and are directly proportional to the level of monoaminergic input. Since the concentration of monoamines (i.e. serotonin and norepinephrine) are reduced with age, we sought to determine if estimates of PICs are reduced in older (>60 years old) compared to younger adults (<35 years old). We decomposed MU spike trains from high-density surface electromyography over the biceps brachii and triceps brachii during isometric ramp contractions to 20% of maximum. Estimates of PICs (i.e. ΔF) were computed using the paired MU analysis technique. Regardless of the muscle, peak firing rates of older adults were reduced by ~1.6 pulses per second (pps) ($P = 0.0292$), and ΔF was reduced by ~1.9 pps ($P < 0.0001$), compared to younger adults. We further found that age predicted ΔF in older adults ($P = 0.0261$), resulting in a reduction of ~1pps per decade, but there was no relationship in younger adults ($P = 0.9637$). These findings suggest that PICs are reduced in older adults, and, further, age is a significant predictor of estimates of PICs in older adults. Reduced PIC magnitude represents one plausible mechanism for reduced firing rates, weakness, and reduced function in older individuals.

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Poster

P573. Motor Unit Recordings, Kinematics, and EMG I

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Program #/Poster #: P573.09

Topic: E.09. Motor Neurons and Muscle

Support: NIH NINDS R01 NS098509

Title: Estimates of persistent inward currents are increased at short muscle lengths

Authors: S. T. JENZ[1,2], J. A. BEAUCHAMP[1,4], O. U. KHURRAM[1,4], C. J. HECKMAN[1,2,3,5], G. E. PEARCEY[1,2,3,5];
[1]Dept. of Physical Therapy and Human Movement Sci., [2]Dept. of Neurosci., [3]Dept. of Physical Med. and Rehabiliation, Feinberg Sch. of Medicine, Northwestern Univ., Chicago, IL; [4]Dept. of Biomed. Engin., McCormick Sch. of Engineering, Northwestern Univ., Chicago, IL; [5]Shirley Ryan AbilityLab, Chicago, IL

Abstract: Voluntary motor commands are comprised of excitation, inhibition, and neuromodulation. Persistent inward currents (PICs), which depend on the level of neuromodulatory drive, alter the intrinsic excitability of motoneurons and lead to amplification and prolongation of motoneuron discharge. PICs are reduced by antagonist muscle stretch, and changes in muscle length can impact an individual's maximum voluntary torque (MVT) due to mechanical properties of the muscle. It is not clear whether motoneuron excitability is altered with changes in static joint angle in humans, or whether reduced PICs contribute to reductions in MVT at short muscle lengths. To address these questions, we investigated whether changes in joint angle altered estimates of PICs and hypothesized that Ia input from the lengthening of the antagonist muscle would reduce estimates of PICs. High density surface electromyography was used to decompose individual motor unit (MU) spike trains from the tibalis anterior, soleus, and medial gastrocnemius. Participants (n = 13; 25.9 ± 3.09 years) performed isometric plantarflexion and dorsiflexion triangular ramp shaped contractions to a peak of 30% MVT. Contractions were performed at varying ankle joint angles (70° and 110°), representing short and long muscle lengths. PICs were estimated using the well-validated paired-MU analysis technique, which quantifies discharge rate hysteresis (ΔF) by comparing the discharge rate of a lower-threshold MU at the onset and offset of a higher-threshold MU. In agreement with previous studies, MVTs were lowest at joint angles that shortened the agonist muscle. A linear mixed effects model found that joint angle was a significant predictor of ΔF ($\chi 2$ (3) = 27.904, $p < 0.0001$). However, contrary to our hypothesis, ΔF was greater at short, compared to long, agonist muscle lengths for all muscles tested. These results suggest that motoneuron excitability is greater at short muscle lengths, despite the reduced force generating capacity at such angles. This may be due to altered monoaminergic input to motoneurons in order to compensate for biomechanically disadvantageous muscle lengths. These findings add to our basic understanding of neural control of movement at various muscle lengths and could shed light on remaining questions such as the source of muscle cramps at short muscle lengths.

Disclosures: S.T. Jenz: None. **J.A. Beauchamp:** None. **O.U. Khurram:** None. **C.J. Heckman:** None. **G.E. Pearcey:** None.

Poster

P573. Motor Unit Recordings, Kinematics, and EMG I

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Program #/Poster #: P573.10

Topic: E.09. Motor Neurons and Muscle

Support: NIH NINDS F31 NS120500
NIH NINDS R01 NS098509
NIH NICHD R01 HD039343

Title: The sombrero contraction: a novel task that highlights the prolongation of motoneuron discharge from persistent inward currents and their potential impediments in force control

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[1]Biomed. Engin., [2]Physical Therapy and Human Movement Sci., [3]Physiol., [4]Physical Med. and Rehabil., Northwestern Univ., Chicago, IL; [5]Shirly Ryan AbilityLab, Chicago, IL

Abstract: A central feature of normal motor function is the persistent inward currents (PICs) of motoneurons, the amplitude of which correlate with descending monoaminergic input. These PICs are important for amplifying and prolonging excitatory synaptic input to motoneurons and often manifest in their firing patterns, allowing for quantification. Here, we explored a novel isometric contraction paradigm that exploits the phenomenon of PIC induced prolongation, where PICs allow motoneurons to discharge at a lower level of excitatory input than was required at recruitment. Specifically, we asked human participants to perform isometric joint torques in the shape of a hat (i.e. a sombrero hat), comprised of a triangular contraction superimposed upon a steady low-level contraction. This joint torque profile demands two plateau regions of identical motor output before and after a triangular contraction, which allows for characterization of the motoneurons that are recruited by additional synaptic input in the triangular region and sustain discharge into the second plateau. To investigate this contraction profile, we asked participants (n = 7) to perform isometric plantarflexion and dorsiflexion tasks consisting of a slow increase to and hold at 10% of their maximum voluntary torque (MVT) for 10 s, a slow increase to 30% MVT, a decrease back to 10% MVT, another hold at 10% MVT for 10 s, and a slow decrease to rest. We collected high density surface electromyography from the tibialis anterior, medial gastrocnemius, and soleus for all contractions and decomposed signals into individual motor unit (MU) spike trains using convolutive blind source separation. To examine the effects of higher threshold MUs firing into the second plateau, we quantified the percentage of MUs recruited in the triangular region that sustained discharge into the second plateau and the variability in torque during the two plateau regions. Initial analysis revealed sustained discharge in all muscles, with ~50% of MUs recruited in the triangular region firing to the end of the second plateau phase. Furthermore, torque variability was highest during the second plateau, with the coefficient of variation in torque exhibiting a 1.5 fold increase from the first to the second plateau. This suggests that, once activated, PICs may be a hindrance to static precision tasks, and points to a potential limitation in human static torque control introduced by PICs.

Disclosures: J.A. Beauchamp: None. **G.E. Pearcey:** None. **O.U. Khurram:** None. **J.P. Dewald:** None. **C.J. Heckman:** None.

Poster

P573. Motor Unit Recordings, Kinematics, and EMG I

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Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P573.11

Topic: E.09. Motor Neurons and Muscle

Title: Augmented motor unit discharge rates underlie acute intermittent hypoxia-induced increases in volitional elbow strength in persons with chronic incomplete spinal cord injury

Authors: *G. PEARCEY[1], B. AFSHARIPOUR[2], A. HOLOBAR[3], W. Z. RYMER[4], M. S. SANDHU[5];
[1]Northwestern Univ., Chicago, IL; [2]Univ. of Alberta, Edmonton, AB, Canada; [3]Univ. of Maribor, Maribor, Slovenia; [4]Shirley Ryan AbilityLab, Chicago, IL; [5]Sensory Motor Performance Program, ShirleyRyan Ability Lab., Chicago, IL

Abstract: Acute intermittent hypoxia (AIH) is an emerging technique for facilitating neural plasticity in individuals with neurological deficits. It utilizes a brief, rapid reduction of oxygen concentration in the inspired air, which stimulates the serotonergic system, a potent neuromodulator of α-motoneurons. In humans with incomplete spinal cord injury (iSCI), a single 15 dose sequence of AIH enhances ankle, hand grip, and elbow strength for up to 90 minutes. We have recently observed that changes in torque are associated with an increased spatial distribution of muscle activation over the muscle surface and an overall increase in root mean squared EMG amplitude of the biceps and triceps brachii, but mechanisms underlying the altered EMG maps are not clear. Herein, we examined how AIH contributes to improved muscle strength by decomposing individual motor unit spike trains from high-density surface electromyography during maximal elbow flexion and extension torques, recorded prior to and 60 minutes after AIH. Seven individuals with chronic iSCI at the cervical level visited the laboratory on two different occasions, at least a week apart, and received AIH and Sham AIH interventions in a randomized order. Participants performed maximal voluntary isometric elbow flexion and extension contractions prior to and 60 minutes after either intervention. AIH consisted of 15 brief (~60s) periods of low oxygen (fraction of inspired O_2 = 0.09) interspersed with 60s periods of normoxia, whereas Sham AIH consisted of repeated exposures to normoxia. After AIH, elbow flexion and extension torque increased by 104.1 ± 30.56% ($p = 0.016$) and 71.7 ± 30.34% ($p = 0.031$) from baseline, respectively, whereas there was no change (i.e. < 10% change) after Sham AIH ($p > 0.2$ in both cases). We were able to track individual motor units pre-to-60 minutes post intervention and found that the discharge rates during maximal contraction increased by ~20% following AIH. These findings show that a single session of AIH

enhances elbow flexion and extension strength in individuals with chronic iSCI. Furthermore, our preliminary motor unit analysis suggests that excitability and/or the activation of spinal motoneurons are augmented after AIH, providing an underlying mechanism to explain the observed strength increases induced by AIH.

Disclosures: G. Pearcey: None. **B. Afsharipour:** None. **A. Holobar:** None. **W.Z. Rymer:** None. **M.S. Sandhu:** None.

Poster

P575. Muscle Physiology and Pathology

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Program #/Poster #: P575.01

Topic: E.09. Motor Neurons and Muscle

Support: NSF DBI-2015317
NIH R01 NS118606

Title: Characterization and creation of a linear Hill-type muscle model of the Aplysia californica I1/I3 retractor muscle complex

Authors: Q. CHEN[1], R. SUKHNANDAN[2], S. PAO[1], J. P. GILL[1], H. J. CHIEL[3], *V. A. WEBSTER-WOOD[2];
[1]Case Western Reserve Univ., Cleveland, OH; [2]Carnegie Mellon Univ., Pittsburgh, PA; [3]Case Western Res. Univ., Cleveland, OH

Abstract: Understanding and predicting how neural patterns generate behavior requires both neural and biomechanical models. One model of interest to study multifunctionality is feeding behavior in the marine mollusk *Aplysia californica* [1] which generates (1) biting, an unsuccessful attempt to grasp food, (2) swallowing, successfully grasping and ingesting food, and (3) rejection, egestion of inedible food. Although many neurons involved have been identified, a Hill muscle model has only been reported for the I2 protractor muscle [2]. In all three behaviors, the I3 retractor muscle plays a critical role. This study aimed to characterize the I1/I3 retractor muscle complex and create a linear Hill muscle model. Half sections of the cylindrical I1/I3 complex were isolated from fully anesthetized adult *Aplysia*, maintaining innervation by Buccal Nerve 2 (BN2). Isolated muscles were mounted to a length-controlled servo-motor along the I3 contraction direction. A stimulation suction electrode was applied to BN2, and a recording hook electrode was applied proximally along the nerve. Muscles were maintained at 14-20 °C in an aerated mixture of 50% *Aplysia* saline and 50% low calcium, high cobalt saline containing glucose to reduce spontaneous firing. Three experiments were performed as follows. **Force-Frequency:** Muscles were held at rest length and stimulated for 5 seconds at increasing stimulation frequencies (2-50 Hz, in 4 Hz increments) once every three minutes. The

optimal stimulation frequency was used in subsequent experiments. **Force Stability:** Muscles were held at rest length and stimulated 5 seconds every three minutes to test force stability over time. **Length-Tension:** Length-tension characterization was performed using three-minute episodes in increments of 0.5 mm. Active tension was determined by stimulating the muscle for 5 seconds at each length following passive force relaxation. **Linear Hill Muscle Model:** To create a muscle model, force-frequency and length-tension results were normalized to the maximum active tension in the length-tension study. Normalized force-frequency was fit to a saturating exponential model [2]. Normalized passive force was fit to a piecewise quadratic model and active force to a Gaussian function [3,4], resulting in a linear Hill-type muscle model. In the future, we will use this model in neuromechanical models of *Aplysia* feeding and expand our characterization to include muscle force-velocity.
References: [1] Webster-Wood et al. *Biol Cybern*. 2020. [2] Yu et al. *Biol Cybern*. 1999;81:505-513. [3] Romero, et al. *Mechanical Sciences* 7, no. 1 (Jan. 18, 2016): 19-29. [4] Thelen, Darryl G. *J. Biomech. Eng.* 125, no. 1 (Feb. 1, 2003): 70-77.

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Poster

P575. Muscle Physiology and Pathology

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Program #/Poster #: P575.02

Topic: E.09. Motor Neurons and Muscle

Support: NCN grant 2018/31/B/NZ4/02789; NCN grant 2020/37/N/NZ4/04136; Statutory grant for the Nencki Institute

Title: Early changes in ultrastructure of NMJs in Soleus and Tibialis Anterior muscles after spinal cord transection and spinal AAV-BDNF overexpression in adult rat

Authors: *A. GLOWACKA[1], A. A. SZCZEPANKIEWICZ[2], B. JI[3], M. H. SKUP[3]; [1]Group of Restorative Neurobio., [2]Lab. of Electron Microscopy, Nencki Inst. of Exptl. Biol., Warszawa, Poland; [3]Group of Restorative Neurobio., Nencki Inst. of Exptl. Biol., Warsaw, Poland

Abstract: Overexpression of brain-derived neurotrophic factor (BDNF) in rats with spinal cord transection (SCT) was shown by us and others to increase function of the lumbar spinal network associated with improvements in assisted locomotor performance detectable as early as 1-2 weeks postlesion, but led also to functional deterioration in time (Boyce et al., 2012, Ziemlinska et al., 2014). Elevated neurotransmitter markers and symptoms of hyperexcitability in BDNF-treated rats prompted us to search for their consequences at the level of peripheral synapse. We

used transmission electron microscopy (TEM) imaging to examine the neuromuscular junctions (NMJs) of motoneurons controlling extensor (Soleus) and flexor (Tibialis anterior) muscles of the ankle joint. We hypothesized that their ultrastructure is changed to different extent, because denervation of extensor and flexor motoneurons (MNs) develops to varying degrees in time after spinalization (Skup et al., 2012, Grycz et al., 2019).Three groups of adult male Wistar rats were used: intact control (n=3), spinalized at Th10, with intraspinal injection of phosphate-buffered saline (SCT-PBS, n=3), and spinalized, with intraspinal injection of AAV-BDNF (SCT-BDNF, n=3) at lumbar L1-2 segments. At 2 weeks postlesion rats were perfused with 4% PFA, and single muscle fibers were isolated from Soleus and Tibialis anterior muscles. NMJs were identified using nicotinic ACh receptor binding of fluorescent conjugate of alpha-bungarotoxin. Areas of fibers containing labeled NMJs were localized under the microscope (obj. 20x), 1-2 mm long blocks were cut out and post-fixed in 2.5% glutaraldehyde. Six NMJs per group were analyzed.TEM imaging showed differences in NMJs ultrastructure between groups, but not muscle types. Control junctions had MN boutons tightly packed with vesicles, pre- and postsynaptic areas almost devoid of loose filaments, regularly arranged myofibers with repeating dublets of mitochondria assisting the majority of sarcomeres. In NMJs from SCT-PBS rats less mitochondria and synaptic vesicles were present in MN terminals, perijunctional area and axoplasm were rich in filaments, and myofiber structure was loose. In SCT-BDNF rats, mitochondria located between motor endplate and myofibers revealed regular arrangement, and the structure of myofibers was tighter and more regular than in SCT-PBS rats. Clearly higher abundance of mitochondria in the perijunctional area in SCT-BDNF group might be connected to increased activity of motor units.

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Poster

P575. Muscle Physiology and Pathology

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Topic: E.09. Motor Neurons and Muscle

Support: NIH Grant/1R15NS112920-01A1

Title: Use It or Lose It? Maybe Not: Enhanced excitability and maintenance of force output in breathing muscle after a month-long apnea in frogs

Authors: *S. KIHUWA-MANI, J. SANTIN;
Univ. of North Carolina at Greensboro, Greensboro, NC

Abstract: Skeletal muscle requires ongoing activity to maintain healthy physiological properties. This is clear in several clinical scenarios, such as mechanical ventilation, spinal cord injury, and bedrest, whereby muscle structure and function degrades due to muscle disuse. The motor system that produces breathing in American Bullfrogs is interesting because breathing muscles are inactive during underwater hibernation. In contrast to other animals, frogs do not experience observable changes to breathing behavior after months of inactivity, which is surprising because highly active skeletal muscles typically atrophy rapidly in most other vertebrate animals. Using *in vitro* muscle force assessments, we tested the hypothesis that skeletal muscle involved in breathing from hibernating frogs resists atrophy. To test this hypothesis, we measured function of a breathing muscle of the lower jaw (*m. intermandibularis*) in control (n=5) and hibernating frogs (n=6). Using direct muscle stimulation, we found that specific twitch force, tetanic force, and recovery from fatigue are maintained despite 1 month of disuse during hibernation ($p>0.05$). In addition, the time between stimulus and twitch force development (twitch latency) was reduced, indicative of enhanced muscle excitability($p=0.01$; two-tailed t-test). Finally, twitch relaxation time was reduced relative to controls ($p=0.008$; two-tailed t-test), suggesting improved sarcoplasmic Ca^{2+} handling following hibernation. These results indicating maintained or improved function are remarkable since this muscle is typically highly active but was unused for one month..These results suggest a diverse set of physiological and biochemical compensatory mechanisms which act to ensure breathing resumes appropriately when animals emerge from hibernation. Ongoing follow-up studies aim to repeat these experiments using nerve stimulation to infer function of the neuromuscular junction, as well as quantitative PCR to measure the expression of candidate genes that may contribute to enhanced muscle excitability and Ca^{2+} handling. Uncovering the mechanisms that allow this muscle to maintain function despite inactivity, which may uncover new insights for muscle disorders in humans.

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Poster

P575. Muscle Physiology and Pathology

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Topic: E.09. Motor Neurons and Muscle

Support: Brown University Carney Graduate Award
NIH 2T32MH20068-16
NIH 1U01NS064295

Title: Retrograde signaling at the neuromuscular junction: The MuSK-BMP pathway regulates Collagen XIII expression and synapse stability

Authors: *L. A. FISH[1,2], M. EWING[3], D. JAIME[4], L. A. MADIGAN[4], J. R. FALLON[3,2]; [1]Neurosci. Grad. Program, [2]Carney Inst. for Brain Sci., [3]Dept. of Neurosci., [4]Mol. Biology, Cell Biol. & Biochem. Grad. Program, Brown Univ., Providence, RI

Abstract: Synaptic stability is essential for information processing and storage. The neuromuscular junction (NMJ) is a genetically and structurally accessible synapse that is amenable to precise mechanistic dissection of synapse stability. We recently reported that postsynaptic NMJ component muscle-specific kinase (MuSK), in addition to its role as an agrin-LRP4 receptor, is a bone morphogenetic protein (BMP) co-receptor. MuSK binds BMPs and their Type I receptors and regulates BMP-mediated transcription (Yilmaz et al, 2016). To investigate the role of the MuSK-BMP pathway at the NMJ, we created a ΔIg3-MuSK mouse model and cell lines that lack the BMP-binding MuSK Ig3 domain. MuSK-BMP signaling is attenuated in ΔIg3-MuSK cells as assessed by BMP-induced Smad1/5/8 phosphorylation and transcriptional output, but agrin-induced AChR clustering is preserved. To probe the role of the MuSK-BMP pathway in NMJ development and stability across different synapse types, we first conducted a morphological analysis of NMJs in the fast sternomastoid (STM) and slow soleus (SOL) muscles, which show differential susceptibility to denervation in ALS and other disorders. Analysis of the STM showed that NMJs in ΔIg3-MuSK mice form normally but are unstable as indicated by endplate fragmentation, which is observed as early as P21. Interestingly, when compared to WT, NMJ areas in ΔIg3-MuSK muscle are larger in immature, but smaller in mature animals. Preliminary ultrastructural analysis indicated that ΔIg3-MuSK NMJs have sparser and shallower junctional folds that are more variable in spacing and depth than WT. In SOL, NMJ area is also reduced in mature ΔIg3-MuSK animals. However, the axonal branches at the SOL NMJ are strikingly thicker compared to WT or to ΔIg3-MuSK STM. To gain insight into the mechanisms by which the MuSK-BMP pathway regulates NMJ structure, we performed RNA-seq analysis of ΔIg3-MuSK SOL. Notably, we observed a reduction in the expression of Collagen XIII, a component of the synaptic basal lamina known to be required for NMJ stability and structure. Multiple transcripts involved in glutamatergic and GABAergic synaptic signaling are also upregulated. Unexpectedly, we also observed increased expression of transcripts encoding the presynaptic components snap25, synaptotagmin-1 and piccolo. Together, our data support a model in which postsynaptic MuSK-BMP signaling regulates the expression of Collagen XIII, which in turn acts as a retrograde signal that is required for synaptic stability, normal pre- and postsynaptic structure, and branch size. The MuSK-BMP pathway could present new therapeutic targets to promote synaptic resiliency in neuromuscular disorders.

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Poster

P575. Muscle Physiology and Pathology

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Topic: E.09. Motor Neurons and Muscle

Support: Guangzhou Science and Technology Program (Grant no. 201904010256)

Title: Sex differences in intrinsic hand muscle size and echo intensity measured by ultrasonography

Authors: *C. S. KLEIN, H. LUI, C. ZHAO, Z. MENG;
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Abstract: It is generally accepted that the average female has smaller muscles than the average male adult. However, this may not necessarily be the case in the intrinsic hand muscles given the conflicting, albeit limited, findings in previous studies. [1, 2] The intrinsic hand muscles are a unique and convenient model for the study of sex differences in muscle morphology as they are free of the confounding effects of weight bearing. In the present study we used ultrasound imaging to quantify sex differences in muscle cross-sectional area (CSA), muscle thickness, and echo intensity (EI, an indicator of fat and connective tissue in muscle). Height, weight, and various hand and digit dimensions (i.e., finger and thumb length, diameter) were recorded to assess whether any sex differences in muscle morphology are entirely explained by anthropometric (body size) differences. Ultrasonography of the flexor pollicis brevis (FPB), abductor pollicis brevis (APB), opponens pollicis (OP), abductor digiti minimi (ADM), and first dorsal interosseous (FDI) was completed in 15 healthy females (28.1 ± 6.9 y, mean ±SD) and 14 males (31.1 ± 5.4 y). We found that the mean female FPB, APB, OP, ADM and FDI thickness was 86%, 73%, 84%, 80% and 81% of the male means , and APB, ADM, and FDI CSA were 71%, 64%, and 71% ($P < 0.01$). EI was about 5-10% higher in some female than male muscles ($P < 0.05$). Total CSA (APB+ADM+FDI CSA) was correlated with body height in females ($r = 0.79$) and males ($r = 0.69$), and was stronger than the correlations with hand or digit dimensions. After accounting for differences in height, about 80% of the sex difference in total CSA can be attributed to factors other than body size. Intra-rater and inter-rater reliability of thickness and CSA was high; intraclass correlation coefficient (ICC) > 0.8. These results indicate that intrinsic hand muscles are 20-30% smaller in females than males. The findings suggest that factors other than body dimensions, such as neural or systemic factors, may account for much of the sex differences in muscle morphology. 1. Lee H, Jee S, Park H, Ahn SC, Im J, Sohn MK. Quantitative Muscle Ultrasonography in Carpal Tunnel Syndrome. *Annals of rehabilitation medicine* 2016; **40**(6)**:** 1048-1056. 2. Simon NG, Ralph JW, Lomen-Hoerth C, Poncelet AN, Vucic S, Kiernan MC *et al.* Quantitative ultrasound of denervated hand muscles. *Muscle & nerve* 2015; **52**(2)**:** 221-30.

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Poster

P575. Muscle Physiology and Pathology

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Julie Bonasera Fund for ALS and Neuromuscular Diseases
William White Family Fund for ALS

Title: Als-associated kif5a mutation causes delayed and decreased recovery following sciatic nerve crush.

Authors: *K. A. RICH, H. HARRIS, A. FOX, M. G. PINO, M. L. JONES, W. ARNOLD, M. YALVAC, S. J. KOLB;
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Abstract: Background: *KIF5A* encodes kinesin protein KIF5A, which is involved in anterograde transport of cargos in eukaryotic cells. C-terminal mutations in *KIF5A* have been identified in patients with amyotrophic lateral sclerosis (ALS). The mechanism linking *KIF5A* mutations and ALS pathogenesis is unknown. Peripheral nerve injury (PNI) is an established paradigm to study mechanisms of axon repair, including axon transport, in rodent models. Complete recovery is observed in young mice, though recovery diminishes with age. Following PNI, electrophysiology can be used to monitor recovery *in vivo* and is a clinically relevant measure of motor unit integrity. Degradation of the motor unit is a primary biological feature of ALS. To investigate this, we created a novel mouse with an ALS-associated C-terminal *Kif5a* mutation (c.3005+1G>A).
Methods: PNI was induced via sciatic nerve crush (wildtype, *Kif5a*$^{+/c.3005+1G>A}$ and *Kif5a*$^{c.3005+1G>A/c.3005+1G>A}$) at 14 weeks or 35 weeks. Before crush and throughout recovery, mice were monitored via weekly assessments of electrophysiology (compound muscle action potential [CMAP] and estimation of motor unit number [MUNE]). At 16 weeks post-recovery, Nissl staining of the L3-L4 region of the lumbar spinal cord was used for motor neuron analysis (motor neuron area and number). Cross-sectional electrophysiological evaluations were also performed in uninjured mice.
Results: *Kif5a*$^{+/c.3005+1G>A}$ mice demonstrated delayed and incomplete recovery of MUNE compared to wildtype, and recovery was further impaired in *Kif5a*$^{c.3005+1G>A/c.3005+1G>A}$ mice. CMAP did not differ between genotypes. Pathologically, motor neuron area was reduced in mutants on the injured side of the spinal cord. There appeared to be an influence of age on latency to recover, as mice with PNI at 35 weeks had an even more delayed and incomplete electrophysiological recovery compared to those injured at 14 weeks.
Conclusions: Our findings suggest that *Kif5a* C-terminal mutations result in a loss of normal KIF5A function, impacting motor unit recovery. Decreases in nerve crush recovery in older mice suggests age-related nerve degeneration may lead to a more severe phenotype. This work

establishes a model system to develop disease-modifying therapies and understand basic principles of *KIF5A* biology.

Disclosures: K.A. Rich: None. **H. Harris:** None. **A. Fox:** None. **M.G. Pino:** None. **M.L. Jones:** None. **W. Arnold:** F. Consulting Fees (e.g., advisory boards); La Hoffmann Roche, Cadent Therapeutics, Novartis, and Genentech. **M. Yalvac:** None. **S.J. Kolb:** F. Consulting Fees (e.g., advisory boards); AveXis, Biogen and Genentech.

Poster

P575. Muscle Physiology and Pathology

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P575.07

Topic: E.09. Motor Neurons and Muscle

Support: CONACYT grant under projects: Salud-2007-01-69989
SIP-IPN: 20194947, 20170579

Title: Spinal manipulation therapy with activator IV induces no change in the excitability of the spinal cord on dancers and control subjects.

Authors: A. R. FRAGOSO[1], B. MARTÍNEZ[1], E. HERRERA[1], J. J. SALDAÑA[1], M. E. CEBALLOS-VILLEGAS[2], ***J. LOMELI**[3];
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Abstract: Introduction: The Hoffmann reflex is obtained by the electrical stimulation of afferent fibers Ia that innervate the muscle spindles of a muscle under study. The excitability of the spinal cord is in correlation with the Hoffmann reflex amplitude. Spinal manipulation can modify the spinal cord excitability, in this work, we made ten spinal manipulations with the activator IV as part of a treatment to solve biomechanical problems of the spine, it was to seek to reduce sub-dislocations in patients. After complete therapy of ten sessions, we analyzed the amplitude of Hoffmann reflex. **Objective:** Measure the amplitude of the Hoffmann reflexes recorded on soleus muscles of subjects who practice regional dance and compare them with the records obtained in control subjects, before and after spinal manipulation using the IV activator. **Method:** we had a sample of 7 regional dance subjects, as well as 7 control subjects. The protocol was approved by the local ethics committee. Participants underwent medical history, they were asked to sign an informed consent form, they were taken radiographs. Then we applied current constant stimuli with DS5 and DS7 Digitimer stimulators to record Hoffmann reflexes on bilateral soleus muscles of both groups. **Results:** The ratios H_{max}/M_{max} for control subjects were 0.56±0.15 before and 0.55±0.19 after (p=0.47), on the dancers were 0.43±0.35 before and

0.31±0.23 after (p=0.27). **Conclusion:** A complete chiropractic therapy of 10 sessions using activator IV does not produce differential effects on spinal excitability on dancers neither control subjects.

Disclosures: A.R. Fragoso: None. **B. Martínez:** None. **E. Herrera:** None. **J.J. Saldaña:** None. **M.E. Ceballos-Villegas:** None. **J. Lomeli:** None.

Poster

P575. Muscle Physiology and Pathology

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P575.08

Topic: E.09. Motor Neurons and Muscle

Title: Vasti Motor Units are Less Synchronized in Adults with Patellofemoral Pain

Authors: *Z. WANG[1,2], A. JOHNSON[3], L. GRIFFIN[2];
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Abstract: The vastus medialis oblique (VMO) and vastus lateralis (VL) are important patellar stabilizers. Imbalanced activation of these muscles can lead to knee injury and pain. The purpose of this study was to investigate differences in the correlations of the motor unit firing rates of the two muscles between healthy adults and adults with patellofemoral pain (PFP). Twenty healthy participants (10 females and 10 males) and 21 participants with PFP (11 females and 10 males) performed a sustained isometric knee extension of 20% maximal voluntary contraction until task failure. Intramuscular fine-wire electrodes were used to record single motor unit discharge rates. Fifty-four motor units of the VMO and 51 motor units of the VL were recorded. VMO motor unit firing rates were significantly higher in healthy males (11.2 ± 0.6 Hz) than in healthy females (8.7 ± 0.7 Hz, $p = 0.006$), males with PFP (8.9 ± 0.6 Hz, $p = 0.011$) and females with PFP (7.5 ± 0.6 Hz, $p < 0.001$); whereas VL motor unit firing rates were similar across the four groups (healthy females: 9.6 ± 0.5 Hz; healthy males: 9.2 ± 0.6 Hz; PFP females: 9.1 ± 0.5 Hz; PFP males: 10.3 ± 0.6 Hz). Motor unit firing rates were higher in the VMO than in the VL only in healthy males ($p = 0.011$). VMO motor unit firing rates were lower than the VL in females with PFP ($p = 0.002$) and similar to the VL in healthy females and males with PFP. Motor unit firing rate synchronization within motor unit pairs was calculated using spike cross-correlograms from 106 motor unit pairs across the VMO and VL of both the healthy and PFP groups. VMO-VL motor unit firing rate synchronization was highest in healthy males (0.457 ± 0.027 Hz) compared to healthy females (0.350 ± 0.029 Hz, $p = 0.043$), males with PFP (0.322 ± 0.029 Hz, $p = 0.006$) and females with PFP (0.273 ± 0.031 Hz, $p < 0.001$). Only healthy males exhibited fatigue-induced increases in VMO-VL synchronization ($p = 0.031$). This study shows that the reduction of VMO motor unit firing rates and weak common drive among motor unit pairs of

vasti synergists in individuals with PFP may be neural mechanisms underlying patellofemoral degeneration.

Disclosures: Z. Wang: None. **A. Johnson:** None. **L. Griffin:** None.

Poster

P620. Blood Flow I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P620.01

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: Wellcome Trust/Royal Society Sir Henry Dale Fellowship 105586/Z/14/Z

Title: Characterisation of neurovascular function in old mice

Authors: K. SHAW[1,2], L. LEE[3], L. W. BOORMAN[3], P. S. SHARP[3], J. BERWICK[3], ***C. HOWARTH**[3];
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Abstract: Neurovascular coupling (NVC) enables increasing neuronal energy demands to be met by local increases in cerebral blood flow. This mechanism is not only important for normal brain function but also underlies functional imaging techniques, which are commonly used as surrogate measures of neural activity in humans. NVC dysfunction has been previously reported in aging, and such changes in NVC may be associated with age-related cognitive and motor impairments[1]. As the global population is aging, it is therefore of growing importance to characterise how neurovascular function may be altered during aging. Therefore, in adult (6-8 months old) and old (18 -25 months old) mice, we applied 2-dimensional optical imaging spectroscopy (2D-OIS), with and without electrophysiology, to assess the effect of age on cortical haemodynamic responses to whisker stimulation and hypercapnia in anaesthetised mice. Hypercapnia-evoked responses were reduced in old mice as compared to adult mice. However, in contrast to previous studies[2], the amplitude of sensory-evoked haemodynamic responses were unaltered with age. As these experiments were performed using non-invasive procedures, differences between our and previous findings may reflect differences in experimental procedures[3]. In separate experiments, electrophysiological recordings suggested that neural activity evoked by whisker stimulation was similar in adult and old mice. In these experiments, insertion of the electrode caused a cortical spreading depolarisation (CSD), which affects NVC. We hypothesise that the effects of trauma associated with acute procedures differs with age. The characterisation of different aspects of neurovascular function in old mice will not only elucidate how aging affects NVC but may also highlight neurovascular biomarkers of aging. 1.Tarantini S,

et al. (2017) Exp Gerontol. 94:52-58 2.Park L, et al. (2007) JCBFM 27:1908-1918 3.Sharp, PS, et al. (2020) JCBFM 40:2289-2303

Disclosures: K. Shaw: None. **L. Lee:** None. **L.W. Boorman:** None. **P.S. Sharp:** None. **J. Berwick:** None. **C. Howarth:** None.

Poster

P620. Blood Flow I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P620.02

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIH Grant R01NS078168
NIH Grant R01NS079737

Title: A cellular substrate for ultra-slow bilateral hemodynamic correlations

Authors: *K. L. TURNER[1], M. HOSSAIN[1], Q. ZHANG[2], K. W. GHERES[3], P. J. DREW[2]; [1]Dept. of Biomed. Engin., [2]Dept. of Engin. Sci. and Mechanics, Pennsylvania State Univ., University Park, PA; [3]Huck Inst. of the Life Sci., Pennsylvania State Univ. Univ. Park, University Park, PA

Abstract: Hemodynamic signals in the brain are routinely used to infer local neural activity through a relationship referred to as neurovascular coupling (NVC). Understanding which neurons modulate the arousal-state-dependent strength of NVC and functional correlations in the hemodynamic signals between brain regions is of fundamental importance. Previous work has shown that the activity of neuronal nitric oxide synthase (nNOS) expressing neurons play an important role in NVC and setting the basal diameter of arterioles. To explore the role of these neurons in NVC and bilateral hemodynamic correlations, we selectively removed a subset of neuronal nitric oxide (nNOS) interneurons, known as type-I nNOS neurons, from the whisker-related region of somatosensory cortex of mice using a targeted ablation technique. We concurrently measured cerebral blood volume and neural activity in these mice during sleep and wake states. We also measured the dynamics of pial and penetrating arteriole diameter changes using 2-photon microscopy. Ablation of these neurons had no effect on sensory-evoked and fidgeting-evoked neural and hemodynamic responses, but did decrease the bilateral correlations in gamma-band power and blood volume in the somatosensory cortex at ultra-low frequencies (~0.003-0.1 Hz). Additionally, ablation of type-I nNOS neurons reduced the coherence between gamma-band power and hemodynamic signals in the ultra-low frequency band. Our results suggest that type-I interneurons may play a role in synchronizing neural and vascular signals across hemispheres at ultra-low frequencies.

Disclosures: K.L. Turner: None. **M. Hossain:** None. **Q. Zhang:** None. **K.W. Gheres:** None. **P.J. Drew:** None.

Poster

P620. Blood Flow I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P620.03

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: Wellcome Trust and Royal Society Sir Henry Dale Fellowship [grant number 105586/Z/14/Z]

Title: The role of nitric oxide in nNOS interneuron and sensory-evoked neurovascular coupling

Authors: *L. LEE[1,2], O. SHABIR[2,3], J. BERWICK[1,2], C. HOWARTH[1,2];
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Abstract: To meet the resultant demand for oxygen and glucose, changes in neural activity are tightly coupled with local cerebral blood flow (CBF), through a process termed neurovascular coupling (NVC). NVC relies on different populations of neural and glial cells mediating CBF through several vasoactive messengers, including K^+, prostaglandin E2 and nitric oxide (NO)[1]. Of these molecules, NO has been suggested to contribute up to 64% of the NVC response[1]. We have recently shown that activation of cortical neuronal nitric oxide synthase (nNOS)-expressing interneurons evokes localised haemodynamic responses[2]. While such evidence points to NO having a primary involvement in NVC, it's role remains unresolved, with recent studies suggesting that NVC is NO-independent[3]. Therefore, we combined optogenetic and pharmacological approaches to investigate the contribution of NO in the haemodynamic responses to sensory stimulation and direct activation of nNOS-interneurons. Using two-dimensional optical imaging spectroscopy (2D-OIS) and electrophysiology, we recorded the cortical surface haemodynamics in response to interleaved 2s photostimulation (473nm LED) and 2s mechanical stimulation of whiskers in lightly anaesthetised nNOS-CreERT x ChR2-EYFP mice (n=12, 6 male 6 female, 5-10 months old, [Jax 014541[4] x Jax 024109[5]]). Experiments were performed in the absence and presence of the nonspecific NOS inhibitor, N(ω)-nitro-L-arginine methyl ester (LNAME), to block the production of NO. We demonstrate that, when NOS is inhibited, the initial haemodynamic response to a mechanical whisker stimulation is unaltered. Conversely, specific activation of nNOS-expressing interneurons evokes an initial reduction in blood volume followed by a delayed increase. Inhibition of NOS affected the haemodynamic return to baseline regardless of type of stimulation. These results indicate that, unlike those evoked by nNOS-interneuron activation, the initiation of sensory-evoked haemodynamic responses is NO-independent. Thus, different vasoactive pathways may be

involved in sensory and nNOS-interneuron evoked NVC. Experiments in which photo- and whisker- stimulation were simultaneously applied further support this idea. In summary, we demonstrate that NO is not involved in the initiation of sensory-evoked NVC, suggesting that the role of NO in NVC is more complex than simply as a direct mediator.
1) Hosford PS & Gourine AV; (2019) Neurosci Biobehav Rev 96: 174. 2) Lee L et al; (2020) Cerebral cortex 30: 2452. 3) Zambach SA et al; (2021) PNAS 118: e2023749118. 4) Taniguchi H et al; (2011) Neuron 71: 995. 5) Madisen L et al; (2012) Nat Neurosci 15: 793

Disclosures: L. Lee: None. **O. Shabir:** None. **J. Berwick:** None. **C. Howarth:** None.

Poster

P620. Blood Flow I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P620.04

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: R01NS107383
R01GM112715
R01NS119251

Title: The participation of local field potentials and single neuronal activity in the regulation of brain tissue oxygen

Authors: *D. P. AKSENOV[1], E. D. DOUBOVIKOV[1], A. V. DMITRIEV[2], N. A. SERDYUKOVA[2], A. DROBYSHEVSKY[1], D. A. GASCOIGNE[1], R. A. LINSENMEIER[2]; [1]NorthShore Univ. HealthSystem, Evanston, IL; [2]Northwestern Univ., Evanston, IL

Abstract: The brain is a metabolically demanding organ, and sufficient oxygen must be delivered to brain regions to prevent global or local hypoxia. As the many pathological consequences of hypoxia have become better understood, the need to characterize the relationship between neuronal oxygen supply and demand has grown. Several distinct systems regulate brain tissue oxygen (PO_2). Oxygen levels throughout the brain are maintained by cerebrovascular autoregulation in response to fluctuations in arterial pressure. Local increases in oxygen demand, in response to sensory signals, motor output, etc., are supported by transient increases in cerebral blood flow via the hemodynamic response (i.e., functional hyperemia). In the resting state, PO_2 oscillations are generated by changes of vascular tone (i.e., vasomotion) at the level of arterioles. PO_2 is therefore the product of oxygen delivery through local blood flow and oxygen consumption, primarily by neurons. Presently, the exact underlying mechanisms of PO_2 regulation remain a point of contention. Previous studies have primarily used acute recording of vasomotion and neuronal activity (e.g., local field potentials). Instead, to address the hypothesis that chronic implantation of both oxygen electrodes and neuronal activity electrodes

for simultaneous recordings can characterize their relationship in a novel manner, we implanted rabbits and recorded these signals from awake subjects. For PO_2 recordings, electrodes were polarized and connected to an ammeter to record current. The current was then converted to voltage, notch and low-pass filtered (30 or 50 Hz), and lastly amplified. For neuronal activity recordings, the signals from the microwires were fed through a miniature preamplifier to a multichannel differential amplifier system (Neuralynx Inc., Bozeman, MT, USA). The signals were then amplified, band-pass-filtered (300Hz to 3 kHz for single units and 1Hz-100Hz for local field potentials), and digitized (32 kHz/channel) using a Neuralynx data acquisition system. Our results confirmed previous observations that vasomotion follows a specific band (gamma) of local field potentials. Beyond this, we expanded the results to characterize the interaction between changes in the behavior of individual neurons and PO_2, which in turn, will help to further the understanding of the physiological basis for fMRI techniques. Our study suffered an obvious limitation; due to the limited number of electrodes used, we were not able to interrogate the relationship between PO_2 and the entire local neuronal network.

Disclosures: D.P. Aksenov: None. **E.D. Doubovikov:** None. **A.V. Dmitriev:** None. **N.A. Serdyukova:** None. **A. Drobyshevsky:** None. **D.A. Gascoigne:** None. **R.A. Linsenmeier:** None.

Poster

P620. Blood Flow I

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Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P620.05

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: R01NS115401
U24EB028941
R01MH111359
R01NS091230

Title: Circadian rhythm induces changes in the cerebral microvascular blood flow in the awake mice

Authors: B. LI[1,2,3], I. SENCAN[1], J. LENG[2,3], R. OHTOMO[1], H. TAKASE[1], J. E. PORTER[1], Q. PIAN[1], M. THUNEMANN[4,1], S. R. ADAMS[5], J. YANG[1], B. FU[1], M. A. YASEEN[1,6], C. RAN[1], D. A. BOAS[4], A. DEVOR[4,1], C. AYATA[1], K. ARAI[1], E. T. MANDEVILLE[1], E. ESPOSITO[1], E. H. LO[1], ***S. SAKADZIC**[1];
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Abstract: We have recently shown that effectiveness of the neuroprotection strategies used to improve the focal ischemia outcome is circadian rhythm dependent. While understanding the cerebral blood flow regulation is critically important to improve the treatment strategies, a significant gap remains in our understanding of how circadian rhythm influences both global and microvascular blood flow across brain regions. In this work, we applied two-photon microscopic imaging to investigate the circadian rhythm associated changes in the microvascular blood flow and intravascular oxygenation in both middle-cerebral-artery (MCA) and adjacent watershed cortical territories in head-restrained C57BL/6 mice, through a sealed chronic cranial window. The same group of mice was imaged in the awake state, at rest, during the night (mice active phase) and the daytime (mice inactive phase). The results showed that both the baseline capillary red-blood-cell (RBC) flux and blood flow in the surfacing venules were higher during the inactive phase than during the active phase. Importantly, the relative decrease of both capillary RBC flux and venular blood flow between the inactive and the active phase was larger in the watershed region than in the adjacent MCA territory. Finally, whisker stimulation revealed the difference in the transient blood flow response between the active and inactive phase. Our preliminary results imply that both resting-state blood flow and its response to functional activation are circadian cycle dependent, and that effect of the circadian rhythm may be larger in the more vulnerable watershed territories. These results will pave the way towards better understanding of the blood flow regulation and neuroprotective treatment strategies.

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Poster

P620. Blood Flow I

Location: Virtual Only

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Program #/Poster #: P620.06

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: R01NS108407
R01NS078168
R01NS101353

Title: The cellular architecture of microvessels, pericytes and neuronal cell types in organizing regional brain energy homeostasis in mice

Authors: *Y.-T. WU[1], H. BENNETT[1], U. CHON[3], D. VANSELOW[2], **Y. KIM**[1], P. J. DREW[4], K. CHENG[2], Q. ZHANG[5], P. OSTEN[6], R. MUÑOZ-CASTAÑEDA[6];

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Abstract: The cerebrovascular network and its mural cells must meet the dynamic energy demands of different neuronal cell types across the brain, but their spatial relationship among these cells is largely unknown. Here, we apply brain-wide mapping methods to create a comprehensive cellular-resolution resource comprising the distribution of and quantitative relationship between microvessels, pericytes, and glutamatergic and GABAergic neurons including neuronal nitric oxide synthase-positive (nNOS+) neurons and their subtypes in mice. Leveraging these data, we discovered region-specific signatures of vasculature and cell type compositions across cortical and subcortical areas, including strikingly contrasting correlations between the density of vasculature, pericytes, glutamatergic neurons and parvalbumin-positive interneurons versus nNOS+ neurons in the isocortex. We also found surprisingly low vasculature and pericyte density in the hippocampus, and distinctly high pericyte to vasculature ratio in the hypothalamus. These findings suggest that vascular density and mural cell composition is finely tuned to maintain regional energy homeostasis.

Disclosures: Y. Wu: None. **H. Bennett:** None. **U. Chon:** None. **D. Vanselow:** None. **Y. Kim:** None. **P.J. Drew:** None. **K. Cheng:** None. **Q. Zhang:** None. **P. Osten:** None. **R. Muñoz-Castañeda:** None.

Poster

P620. Blood Flow I

Location: Virtual Only

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Program #/Poster #: P620.07

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIH grant NS092388
NIGMS 5U54GM104942-03

Title: Exposure to dim light at night during recovery from cardiac arrest affects behavior and brain vasculature

Authors: *O. MELÉNDEZ-FERNÁNDEZ[1,3], J. C. WALTON[1,3], W. H. WALKER, II[1,3], J. R. BUMGARNER[1,3], J. A. LIU[1,3], N. ZHANG[1,3], A. DEVRIES[1,3,2], R. J. NELSON[1,3];
[1]Neurosci., [2]Medicince, Div. of Oncology/Hematology, West Virginia Univ., Morgantown, WV; [3]Rockefeller Neurosci. Inst., Morgantown, WV

Abstract: Electrical lighting and devices are now ubiquitous, but the negative effects of exposure to light at night (LAN) is becoming uncovered. Exposure to LAN disrupts endogenous circadian biological clocks, and consequently, has many downstream effects. Mice exposed to dim LAN (dLAN) decrease expression of vascular endothelial growth factor-A (VEGF), increased microglial expression, and reduced brain derived neurotrophic factor. These results suggest that LAN alters brain physiology and behavior. In the present study, the effects of dLAN on mouse behavior and neurovasculature after cardiac arrest (CA) were examined. We hypothesized that exposure to dLAN after CA impairs behavior and hippocampal vascularization in mice. Male and female Swiss Webster mice (8 wks of age; n=160) underwent a cardiac arrest/cardiopulmonary resuscitation (CA/CPR) or sham procedure and were exposed to either dark nights [14 h light 125 lux):10 h dark (0 lux) n=40m/40f)] or dim white LAN conditions [14 h light (125 lux):10h dim light (5 lux) (n=40m/40f)] for one week. Following that week, all mice were returned to dark night conditions for 1 week. Behavioral testing and tissue collection were conducted at the end of this week. Anxiety-like behaviors were assessed using the elevated plus maze (EPM). Prior to euthanasia, mice were administered tomato lectin via a tail vein injection to visualize and quantify vasculature. Females that underwent CA and were exposed to dLAN reduced anxiety-like behaviors in the EPM compared to their counterparts in dark nights. Males exposed to dLAN reduced their latency to enter open arms in the EPM compared to their counterparts, however, no differences in total open arm entries were observed. In the hippocampus, females exposed to dLAN reduced vascularization in the CA2 and dentate gyrus regions of the hippocampus, which was exacerbated by CA; however, dLAN elevated vascularization in the CA1 and CA3 regions of Sham females. In males, dLAN elevated vascularization in the dentate gyrus regardless of procedure. Overall, we observed sex differences in vascularization across all regions assayed. These data suggest that re-exposure to dark nights following one week of recovery from CA while exposed to dLAN may not be sufficient to reverse the effects of dLAN. Moreover, these effects may occur in a sex-dependent manner.

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Poster

P620. Blood Flow I

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Program #/Poster #: P620.08

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: Washington University Institute for Clinical and Translational Sciences

Title: Biophysical model of autoregulatory feedback generating slow oscillations in hemodynamics, intracranial pressure, and electrophysiology

Authors: *M. LOE[1], M. MORRISSEY[2], R. LANDRE[3], S. TOMKO[2], R. M. GUERRIERO[4], S. CHING[1];
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Abstract: We recently identified a novel millihertz modulation of the EEG of very young pediatric patients with acquired brain injuries, which we termed macroperiodic oscillations (MOs). During MOs the EEG power attenuates in a gradual and cyclic manner with a periodicity of 2-5 minutes. The mechanisms underlying this phenomenology remain unknown. However, we observed that during periods of MOs, fluctuations in ICP and HR occurred with the same 2-5 minute periodicity, synchronized with the EEG modulation. Prior studies of ICP oscillations in critically ill patients have not shown a relationship with brain electrical activity and HR. The multi-modal synchronization we observed is suggestive of a broader physiological mechanism underlying MOs. To investigate this mechanism, we generated and characterized a computational model of the relationship between cerebral hemodynamics, ICP, and electrophysiology.
We augmented existing models of ICP biophysics and integrated these with novel biophysical models of cerebral hemodynamics, by coupling a chemoreceptor modulation coefficient to the mechanosensitive autoregulation and cerebral blood flow dynamics. We then coupled equations modeling electrical activity in the brain to hemodynamic autoregulation, wherein partial pressures of CO_2 and O_2 were a proxy for substrate availability, similar to BOLD hemodynamic response functions. Feedback between substrate availability and hemodynamics were governed by the chemoreceptor modulation coefficient. Physiological parameters were initialized based on documented measurements. Our developed model recapitulates ICP oscillations and demonstrates their ability to impact electrophysiology and in certain parameter regimes, induce coupled slow modulation commensurate with MOs. The modulation factor modeling physiological autoregulation through partial pressures in cerebral blood is the key mechanistic mediator linking the three different physiological processes (ICP, hemodynamics, and electrophysiology).
We generated a model with physiological correlates for key parameters to study MOs, a regime of aberrant hemodynamic and electrophysiological oscillations observed in patients with brain injuries. The brain is not a closed system, and this work reveals ways in which physiological processes interact with brain electrical activity during injuries with aberrant autoregulation, building on existing modeling of metabolic processes and neural activity. Our findings suggest a possible mechanism for MOs, paving the way for analysis of potential interventions at various feedback points within the system.

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Poster

P620. Blood Flow I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P620.09

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIA R00 AG059893
Alzheimer's Association 2019-AARG-641299

Title: Impairments in vascular function in a mouse model of cerebral amyloid angiopathy

Authors: *M. G. KOZBERG, L. P. MUNTING, B. J. BACSKAI, S. M. GREENBERG, S. J. VAN VELUW;
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Abstract: Cerebral amyloid angiopathy (CAA) is a small vessel disease in which amyloid-β is deposited in the walls of leptomeningeal and cortical blood vessels. CAA leads to several hemorrhagic and ischemic pathologies and is a leading cause of intraparenchymal hemorrhage in the elderly. The mechanisms leading to vascular amyloid-β deposition and subsequent vascular injury in CAA remain largely unknown, but it has been suggested that impairments in the clearance of amyloid-β may play an important role in CAA pathogenesis. Prior work from our group has shown that spontaneous vasomotion, slow oscillations (~0.1 Hz) in vessel diameter, can aid in paravascular waste clearance and is impaired in advanced CAA (van Veluw, Neuron 2020). In this study, using longitudinal, awake two-photon microscopy, we tracked changes in vascular structure and function in the APP23 mouse model of CAA preceding and immediately following initial amyloid-β deposition. This model was chosen given its propensity for spontaneous macro and microbleeds, similar to human CAA. Chronic cranial windows and headposts were implanted in transgenic mice (n = 4) and wildtype littermates (n = 4) and individual vessels in each mouse were imaged monthly from 7 - 12 months of age. We observed amyloid-β deposition in the walls of pial arteries starting at ~8-10 months of age in transgenic mice. Spontaneous vasomotion significantly declined with age in transgenic mice but remained intact in wildtype littermates. Vessel pulsatility, i.e. low-amplitude fluctuations in vessel diameter corresponding to each heartbeat, was observed to increase with age in transgenic mice. Additionally, coinciding with amyloid-β accumulation, pial arteries were observed to increase in diameter by 12 months of age in transgenic mice. Given the importance of smooth muscle cells to maintaining vascular tone and mediating larger changes in vessel diameter as in vasomotion, we hypothesize that impairments in smooth muscle cell function occurring with initial amyloid-β deposition may drive these changes in vascular function. Further work is underway to assess smooth muscle cell function in detail over this age range in the APP23 mouse model. Early impairments in vasomotion (and potentially underlying loss of smooth muscle cell function) may be an important therapeutic target in CAA; reduced vasomotion may lead to a feed-forward

mechanism in which vessels with amyloid-β deposits are less able to clear soluble amyloid-β, leading to extension of the pathology.

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Poster

P649. Feeding and Body Weight: Neuropeptide Regulators

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P649.01

Topic: F.09. Food and Water Intake and Energy Balance

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Title: Assessing neuronal ciliary localization of melanin concentrating hormone receptor (MCHR1) in vivo

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Abstract: Assessing neuronal ciliary localization of melanin concentrating hormone receptor 1 (MCHR1) *in vivo*Kathryn M. Brewer[1], Tisianna K. Kamba[1], Staci E. Engle[1], Ruchi Bansal[1], and Nicolas F. Berbari[1,2,31]Department of Biology, Indiana University-Purdue University Indianapolis, Indianapolis, IN USA.[2]Stark Neurosciences Research Institute, Indiana University, Indianapolis IN, USA.[3]Center for Diabetes and Metabolic Diseases, Indiana University School of Medicine, Indianapolis, IN, USA.
Dysfunctional primary cilia are associated with a broad range of clinical features, including an array of neural developmental and behavioral phenotypes. Primary cilia on neurons throughout the central nervous system localize a diverse set of G protein-coupled receptors. Several of these receptors dynamically localize to cilia in heterologous expression *in vitro* approaches. Here, we sought to investigate whether a ciliary GPCR, melanin concentrating hormone receptor 1 (MCHR1), dynamically localizes to cilia *in vivo* under physiological conditions in which MCHR1 signaling has been implicated such as feeding behavior, obesity, and circadian rhythm. Cilia were detected in images of several mouse brain regions using artificial intelligence driven software followed by analysis of MCHR1 positive cilia frequency as well as cilia length and fluorescence intensity. We found significantly different ciliary lengths across brain regions. However, the percentage of MCHR1 positive cilia, cilia length, and intensity did not change. Further analysis of MCHR1 localization in relation to other ciliary markers as well as a detailed

analysis of MCHR1 localization outside of cilia is ongoing. While these results suggest that MCHR1 does not dynamically localize, it does not rule out the possibility that dynamic ciliary localization could occur *in vivo* under other conditions or for other GPCRs. A better understanding of the subcellular localization dynamics of ciliary GPCRs could reveal mechanisms regulating behavior and potential opportunities to therapeutically target these ciliary receptors.

Disclosures: K.M. Brewer: None. **T.K. Kamba:** None. **S.E. Engle:** None. **R. Bansal:** None. **N.F. Berbari:** None.

Poster

P649. Feeding and Body Weight: Neuropeptide Regulators

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

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Topic: F.09. Food and Water Intake and Energy Balance

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H2020-WF-01-2018-867459P-APPETITE

Title: Marmite, a new conserved neuropeptide mediates protein-specific satiety

Authors: *A. P. FRANCISCO[1], I. TASTEKIN[1], G. EZRA-NEVO[1], B. DEPLANCKE[2], A. M. GONTIJO[3], C. RIBEIRO[1];
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Abstract: Neuropeptides are a diverse class of signaling molecules produced in the nervous system, with different spatial and temporal scales of action, ranging from local to systemic effects. They are involved in almost all aspects of animal's physiology and behavior, from metabolism to feeding. While we know that animals can modify their food choices in response to the lack of specific nutrients, how that is done is still not clear. *Drosophila melanogaster* is an attractive model to explore the role of neuropeptides in feeding and metabolism due to its molecular similarities to vertebrate systems as well as its genetic toolkit and its complex behavioral repertoire. We have identified *marmite*, an uncharacterized gene whose expression is upregulated in amino acid-rich diets, and downregulated in amino acid-poor diets. This gene is

predicted to encode a secreted peptide with unknown function in *Drosophila*. Bioinformatic analyses suggest that the encoded peptide is a possible orthologue of vertebrate neuropeptides which have previously been proposed to affect feeding behavior. To characterize the role of *marmite* in mediating feeding decisions, we used the flyPAD technology, an automated capacitance-based setup that enables the quantitative analysis of feeding in flies choosing to eat from either yeast (protein-rich source) or sucrose. Using this technology, we found that *marmite* knockdown in the nervous system changes feeding decisions, leading to an increase in yeast intake in satiated flies. We have also used a CRISPR-based approach to generate mutants and knock-in driver lines for this gene. *marmite* mutants also showed an increase in yeast but not sucrose intake, confirming the knockdown results, and suggesting a role in protein homeostasis. A *marmite*-specific Gal4 line driving GFP showed expression in only one neuron in each brain hemisphere and neuronal silencing of *marmite*-expressing neurons also leads to an increase in yeast feeding, recapitulating the loss of function phenotype. Moreover, activation of these neurons is sufficient to decrease yeast feeding in a *marmite*-dependent manner, suggesting that *marmite* acts as a satiety signal. To better understand the evolution of this new family of peptides, we are also exploring the functional conservation of *marmite* orthologues. Our experiments show that neuronal expression of the human orthologue is sufficient to modulate feeding in flies. Thus, we have identified a novel class of conserved neuropeptides that play a key role in nutritional homeostasis and feeding decisions across phyla.

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Poster

P649. Feeding and Body Weight: Neuropeptide Regulators

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P649.03

Topic: F.09. Food and Water Intake and Energy Balance

Support: RF1MH120144

Title: Glp-1 signaling regulates energy homeostasis in the paraventricular-dorsal vagal complex circuit

Authors: *L. WANG, M. BERNABUCCI, I. SINGH, A. E. OUAAMARI, Z. P. PANG; Neurosci., Child Hlth. Inst. of New Jersey and Dept. of Neurosci. and Cell Biol., NEW BRUNSWICK, NJ

Abstract: Glucagon-like Peptide-1 (GLP-1) controls feeding and influences glucose homeostasis. GLP-1 Receptor (GLP-1R)-expressing neurons in paraventricular nucleus of the hypothalamus (PVN) mediate food intake suppression induced by GLP-1. The PVN downstream

circuit mechanisms remain unclear. Here we define a PVN-to-dorsal vagal complex (DVC) circuit as an essential pathway that the PVN GLP-1R neurons mediate signaling in regulating feeding and glucose homeostasis. We found that the PVN GLP-1R-expressing neurons send robust projections to the DVC, composed of the nucleus tractus solitarius (NTS), the dorsal motor nucleus of the vagus nerve (DMX), and the area postrema (AP), and release glutamate. The release of glutamate from the PVN GLP-1R neurons is enhanced by GLP-1 signaling, suggesting GLP-1 regulates energy homeostasis, at least partially via this circuit. Inactivation of the PVN-to-DVC projecting neurons causes overeating, obesity, and diabetes. Activation of this neuronal pathway suppresses food intake. These data suggest that the PVN GLP-1R neurons projecting to the DVC play a pivotal role in regulating energy metabolism.

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Poster

P649. Feeding and Body Weight: Neuropeptide Regulators

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Program #/Poster #: P649.04

Topic: F.09. Food and Water Intake and Energy Balance

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Title: The role of NPFFR2 on hypothalamic insulin signals and food consumption

Authors: *Y.-T. LIN[1,2], K.-H. WU[3], J.-J. JHANG[4], Z. YU[4], H.-Y. LI[5], J.-C. CHEN[1,2,6]; [1]Grad. Inst. of Biomed. Sci., [2]Hlth. Aging Res. Ctr., [3]Dept. of Med. Biotech. and Lab. Sci., [4]Dept. of Med., Chang Gung Univ., Tao-Yuan, Taiwan; [5]Biology-Health Sci. Program, Dept. of Natural Sci., Oregon Tech., Klamath Falls, OR; [6]Neurosci. Res. Ctr., Chang Gung Mem. Hosp., Tao-Yuan, Taiwan

Abstract: Neuropeptide FF (NPFF) belongs to a FMRF-NH2 peptide family and is recognized as an opioid modulating peptide. NPFF involves in different physiological functions including the regulation of analgesic effect of opioids, and controls food consumption and cardiovascular function through its interaction with two cognate receptors, NPFFR1 and NPFFR2. NPFFR2 has higher binding affinity towards NPFF than NPFFR1. Central control of food intake is mainly regulated in the hypothalamus through the arcuate nucleus (ARC), which regulates the feeding behaviors by receiving the peripheral signals, which including insulin and leptin. NPFF is highly expressed in the hypothalamus and has been reported to regulate feeding behavior. However, its downstream cellular mechanism remains unclear. In the current study, we explore the role NPFFR2 in food intake through regulating the hypothalamic insulin downstream signals. Our

evidences suggest that the activation of hypothalamic NPFFR2 inhibit the insulin-regulated satiety signals. The knockdown of NPFFR2 in mice is benefic to metabolic health.

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Poster

P649. Feeding and Body Weight: Neuropeptide Regulators

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Topic: F.09. Food and Water Intake and Energy Balance

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Title: Estrogen drives melanocortin signaling to increase spontaneous activity and reduce sedentary behavior

Authors: *W. C. KRAUSE[1], R. RODRIGUEZ[1], B. GEGENHUBER[5], N. MATHARU[2], A. N. RODRIGUEZ[1], A. M. PADILLA-ROGER[3], K. TOMA[4], C. B. HERBER[1], S. M. CORREA[6], X. DUAN[4], N. AHITUV[2], J. TOLLKUHN[7], H. A. INGRAHAM[1];
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Abstract: Sex differences in metabolism and physiology depend heavily on estrogen receptor alpha (ERα) in hypothalamic neurons that control energy expenditure and intake. In response to changes in circulating estrogens, such as 17β-estradiol (E2), these neurons shift homeostasis to prioritize energy utilization and thereby meet the metabolic demands and behavioral outputs required for mating and reproduction. In both humans and rodent models, loss of ERα signaling promotes a hypometabolic state that includes physical inactivity, unhealthy adipose accumulation, and increased risk of metabolic and cardiovascular disease. The ventrolateral division of the ventromedial hypothalamic nucleus (VMHvl) contains a key module of ERα-expressing neurons that promotes energy expenditure by increasing spontaneous physical activity; however, the mechanisms and exact VMHvl neurons that carry out this function remain unclear. Here, we identified the melanocortin-4 receptor (MC4R) as a direct ERα target gene that

is upregulated in the VMHvl by endogenous or exogenous E2. Chemogenetic activation of this MC4R-VMHvl module in mice greatly increases spontaneous activity and is sufficient to decrease sedentary behavior following ovariectomy. Consistent with E2-dependent *Mc4r* transcription, specifically restoring the functional *Mc4r* allele to the VMH of otherwise null mice improves hypoactivity and body weight phenotypes only in females. In contrast, increasing *Mc4r* expression in the VMHvl of adult wild type mice using CRISPR-mediated activation (CRISPRa) sidesteps the E2 requirement and results in a long-lasting increase in physical activity levels in both sexes.
Collectively, our data indicate that VMHvl neurons integrate steroid hormone and central metabolic signals to optimize energy mobilization across the reproductive cycle. That both chemogenetic stimulation of MC4R-VMHvl neurons and CRISPRa augmentation of *Mc4r* expression increases activity in males suggests that the organization of this hormone-sensitive behavioral module is conserved in both sexes. Our findings now explain the pre-ovulatory surge in spontaneous activity observed in ruminants and in rodents first reported nearly a century ago. We suggest that the central intersection of ERα and MC4R signaling accounts for the stronger phenotypes noted in both female rodent/human MC4R mutants. Finally, we predict that age-related decline in circulating estrogens limits MC4R signaling in the VMHvl, and potentially other brain regions, which negatively impact physical activity behaviors and metabolism.

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Poster

P649. Feeding and Body Weight: Neuropeptide Regulators

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P649.06

Topic: F.09. Food and Water Intake and Energy Balance

Title: Modified starch from cassava (Manihot esculenta) and sweet potato (Ipomoea batatas) and its metabolic effects in rats on high sucrose diet

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Abstract: Introduction: Starch is the main source of carbohydrates in human diet, it allows many innovations in food industry due to its functional properties, however, modifications are done with the purpose of improving its functionality. Pyrodextrinization, is one of them and confers the property of being almost or totally indigestible, so allows it to reach the colon to be fermented by microbiota (resistant starch, RS), consequently, affect some other functions of the central nervous system though its ability to produce short-chain fatty acids, which can act by triggering a series of physiological mechanisms regulated by the hypothalamus involving appetite and satiety neurotransmitters. Obesity have developed around the world, which is related to chronic non transmissible diseases, however, RS consumption has been associated with a risk reduction of developing them. Several studies remark that starch is found in most food products because it has a low production cost and high acceptance by most of the population, due to its mixed with other ingredients facility. Cassava (C) and sweet potato (SP) are tubers with considerable starch yields, more profitable than corn plus have an important production in south of Mexico. **Objective:** To modify cassava and sweet potato starch and to evaluate their metabolic effects by consumption in a rat model with a high sucrose diet (HSD). **Methods:** This study consisted of two steps, starting with the modification of cassava and sweet potato starch by pyrodextrinization. For the evaluation of the metabolic effect of pyrodextrins intake, the experiment was divided in two periods: HSD induction (24 weeks) and treatment period (4 weeks). 7 groups were formed CN (normal diet, control), CP (control), HM (hi-maize, 1 g/kg), PY1 (C pyrodextrin, 1 g/kg), PY2 (C pyrodextrin, 1.5 g/kg), PC1 (SP pyrodextrin, 1 g/kg) and PC2 (SP pyrodextrin, 1.5 g/kg). **Results:** C pyrodextrin have 24.42 % RS and SP pyrodextrin 47.15 %. After 20 weeks of HSD, glycemia of CP was (96.50 ± 3.756 mg/dL), CN (71 ± 3.076 mg/dL), HM (66.13 ± 2.69 mg/dL), PY1 (80.63 ± 2.95 mg/dL), PY2 (86.38 ± 11.89 mg/dL), PC1 (97.86 ± 3.52 mg/dL) and PC2 (94.13 ± 1.40 mg/dL). After treatments, glycemia of CN was 72.38 ± 1.91 mg/dL), HM (78.25 ± 3.63 mg/dL), PY1 (81.63 ± 2.40), PY2 (85 ± 2.29 mg/dL), PC1 (65.86 ± 2.91 mg/dL) and PC2 (69 ± 2.84 mg/dL) ($p<0.05$). **Conclusion:** Cassava pyrodextrins may not be able to decrease glycemia due its lower content of RS, on the contrary, sweet potato pyrodextrins show decrease of glycemia, it may be due its higher content of RS.

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Poster

P649. Feeding and Body Weight: Neuropeptide Regulators

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P649.07

Topic: F.09. Food and Water Intake and Energy Balance

Title: Identification of Ixodes Neuropeptides and GPCRs Related to Feeding

Authors: *C. A. KNOX[1], L. L. KYLE[1], L. M. SZCZESNIAK[2], R. J. H. WOJCIKIEWICZ[2], B. E. KNOX[1];
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Abstract: Identification of *Ixodes* Neuropeptides and GPCRs Related to FeedingHard ticks transmit a wide variety of pathogenic microorganisms during hematophagy. Central tothe regulation of feeding and seeking are neuropeptide (NP) systems. A major gap in Ixodidneurobiology is knowledge about the NP-GPCR receptors that control feeding behaviors. Thereare many arthropods GPCRs that impact feeding, with three well-studied examples being theshort NPF, CCHamide and sulfakinin receptors. To address this problem, we have characterizedthese NP-GPCRs and their related preproneuropeptides from *Ixodes scapularis*.**Methods**: Phylogeny and comparative sequence analysis were used to identify potential NP-GPCRs and preproneuropeptides cDNAs. RNA was isolated from adult ticks and RT-PCR orcommercial DNA synthesis was used to obtain full length cDNAs. *I. scapularis* G-protein alphasubunits were identified and full-length cDNAs were synthesized commercially. Peptidesencoding putative *Ixodes* neuropeptides were synthesized commercially. Full-length cDNAs forthe NP-GPCRs were expressed in human 293T and GNAQ/GNA12 mutant 293T (Alvarez-Curto*et al*., (2016) J. Biol. Chem. 291, 27147-27159) cells by transient transfection. Activation of NP-GPCRs coupled to the endogenous Gαq/PLCβ signaling pathway was monitored by a calciumrelease fluorescence-based assay with a FlexStation3 Multimode Plate Reader.**Results:** *Ixodes* CCHamide receptor was activated by a CCHamide containing an internaldisulfide bond with an EC50 of 12 pM (CI: 11, 14). However, a scrambled CCHamide peptide withan internal disulfide bond weakly activated the receptor with an EC50 >>1 x 10^{-6} M. TheCCHamide receptor did not elicit calcium changes in 293 cells with GNAQ/GNA12 disruptions.Human but not *Ixodes* GNAQ could restore the peptide responsiveness when co-transfected withthe CCHamide receptor into GNAQ/GNA12 mutant 293 cells. One of the *Ixodes* NPY-likereceptors (1A, Dinglasan et al., (2013) PLoS Negl Trop Dis 7, e2486) responded to the *Ixodes*sNPF peptide with an EC50 of 1.9 nM (CI: 1.7, 2.1) while the other (1B) did not respond, evenwith concentrations up to 0.1 mM. The *Ixodes* sulfakinin receptor was activated by a sulfatedsulfakinin peptide with an EC50 of 220 pM (CI: 191, 260) but not with the non-sulfated peptide.**Conclusion:** The preproneuropeptides and their receptors for CCHamide, sNPF, and Sulfakininhave been isolated from *Ixodes scapularis.* All three receptors are able to activate a Gq pathwayin 293T cells when exposed to peptides in the pM to nM range. Our experiments open the wayfor *in vivo* characterization of their function.

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Poster

P649. Feeding and Body Weight: Neuropeptide Regulators

Location: Virtual Only

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Program #/Poster #: P649.08

Topic: F.09. Food and Water Intake and Energy Balance

Support: ANID FONDECYT REGULAR 1200578

Title: Regulation of motivation for sucrose by orexin and dynorphin peptides: Analysis using an open-source operant lickometer

Authors: C. SANDOVAL[1], L. LUARTE[1], J. TESKE[2], ***C. PEREZ-LEIGHTON**[1];
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Abstract: The orexin and dynorphin (DYN) peptides regulate motivated behaviors, including intake of palatable foods. In the paraventricular nucleus (PVN), the opioid peptide DYN-A_{1-13} increase intake of palatable foods, an effect blocked by co-treatment with the orexin-A peptide (Nutritional Neuroscience 5:1-10). However, whether orexin and DYN-A_{1-13} in the PVN regulate motivation to work for palatable food remains unknown. To answer this question, we built an open-source operant lickometer using 3D printing with open-source electronics and software. Our device is low cost, easily constructed and can provide operant access to two different liquids. We validated this device in male C57BL/6J mice. First, we successfully measured the circadian rhythm of water intake (n = 3, licks/h active period: 216.5 ± 83.8; inactive period: 83.8 ± 1.8; $P<0.01$). Next, we trained mice to work for a solution of sucrose 5% w/v (~10 μL sucrose/delivery event) in two reinforcement schedules (1 h daily sessions during their active period): a fixed ratio 5 (FR5, 5 licks / delivery event) or a progressive ratio (PR, exponential increase in licks/delivery) and access to water in a FR5 schedule in both schedules of sucrose access. After 6 sessions of FR5, mice worked significantly more for sucrose compared to water (n = 6; delivery events; sucrose: 21.2 ± 4.8; water: 4.5 ± 1.4; $P=0.01$) and after 7 sessions of PR, mice reached 1,000 ± 202 licks for sucrose compared to 24.7 ± 8.5 licks for water ($P<0.01$). Next, a different set of mice (n=10) were cannulated aiming at the right PVN and trained to work for sucrose in the same FR5 and PR schedules (1 h, 7 sessions each). Next, mice were injected with DYN-A_{1-13} (0, 1, 3, 6 nmol) into PVN before PR tests using a repeated measures design. Compared to vehicle, DYN-A_{1-13} (3 nmol) significantly increased sucrose but not water licks during the first 30 min (sucrose: 225 ± 78.9; water: ± 2.3; $P<0.05$) without differences in the second half of the session ($P=0.69$). These data suggest that in the PVN, DYN-A_{1-13} increases motivation for sucrose. Ongoing experiments will determine whether this effect is blocked by orexin-A or the kappa opioid receptor antagonist norBNI. The operant lickometer device and these data open new avenues of research to determine the contribution of orexin and opioid DYN peptides to motivated behavior.

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Poster

P649. Feeding and Body Weight: Neuropeptide Regulators

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Support: NIH Grant DK 118000
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Title: Paraventricular hypothalamic melanin-concentrating hormone signaling and food intake control

Authors: M. SPAULDING, J. HOFFMAN, M. LORD, *E. NOBLE;
Univ. of Georgia, Athens, GA

Abstract: Melanin concentrating hormone (MCH) is a neuropeptide produced in the lateral hypothalamic area (LHA) and zona incerta (ZI) that promotes weight gain by both elevating food intake and reducing energy expenditure. The receptor for MCH (MCH1R) is expressed throughout the brain and is a target for novel obesity pharmacotherapy development, however, the site(s) of action for MCH-mediated alterations in energy balance are largely unknown. We first set out to determine putative sites of action for MCH-mediated feeding effects by injecting MCH or vehicle (CTL) into the lateral ventricle of rats either given a meal of 5 grams of standard chow or in the absence of food (4 groups: MCH with food, MCH without food, VEH with food, VEH without food). At 60 or 90 minutes post injection, animals were sacrificed for immunohistochemical analyses of cFOS expression. Results revealed elevated levels of cFOS in the paraventricular hypothalamus (PVH) of animals injected with MCH in the presence of food. The PVH is a critical orchestrator of energy homeostasis, and MCH1R is expressed in several neuronal populations in the PVH known to regulate body weight. Therefore, we investigated whether MCH signaling in the PVH impacts eating behavior. Male and female Sprague Dawley rats were implanted with chronic indwelling cannula bilaterally targeting the PVH. MCH (0, 0.25 µg, 0.5µg) was injected in a within-subjects design and food intake was monitored. Results revealed that MCH injection into the PVH increases food intake at one hour post injection in both male and female rats. We further investigated whether MCH in the PVH impacts food motivated behavior using a progressive ratio task. MCH injection into the PVH had no effect on motivated responding for palatable sucrose pellets. Given that MCH alters both food intake and energy expenditure, we next investigated whether chronic MCH 1R knockdown in the PVH affects energy balance in rodents using shRNA targeting MCH1R gene expression. Bilateral injection of an AAV1-GFP-U6-r-Mchr1-shRNA, or AAV1-GFP-U6-r-scram-shRNA (control) were injected into the PVH of a small pilot cohort of male rats (n=6 per group), and daily food intake and body weight were monitored. MCH1R knockdown had no impact on food intake but tended to reduce body weight gain on an obesogenic high fat/ high sugar diet. We conclude that the PVH is a brain region in which MCH signaling modulates food intake and possibly energy balance. Future experiments will build on these findings to determine the endogenous relevance

of PVH MCH1R signaling to energy balance regulation and susceptibility to diet induced obesity in both male and female rodents.

Disclosures: M. Spaulding: None. **J. Hoffman:** None. **M. Lord:** None. **E. Noble:** None.

Poster

P649. Feeding and Body Weight: Neuropeptide Regulators

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P649.10

Topic: F.09. Food and Water Intake and Energy Balance

Support: Texas Tech University
National Science Foundation IOS #1656734

Title: The influence of CRF on evoked calcium influx in the optic tectum of the South African clawed frog, Xenopus laevis.

Authors: *C. WANG, P. KEYEL, J. CARR;
Texas Tech. Univ., lubbock, TX

Abstract: As the primary visual center of the non-mammalian vertebrate brain, optic tectum contains CRFR1 receptors that modulate feeding and predator avoidance in part. Tectal CRF neurons respond to stressors and food deprivation, using an intrinsic CRF signaling system, but the signaling pathway is poorly understood. Since gamma-aminobutyric acid (GABA) is one major inhibitory neurotransmitter in the optic tectum, GABAergic neurons might act downstream of CRFR1. This study tested the hypothesis that CRF acts on tectal CRFR1 to inhibit basal and K^+-evoked calcium flux in tectal neurons via GABAergic signaling. We performed three sets of experiments using in-vitro tectal slices from *Xenopus laevis* adults. We tested the following questions: 1) Does CRF dose-dependently inhibit evoked calcium influx in tectal neurons? 2) Do any inhibitory effects of CRF on evoked calcium influx require CRFR1? 3) Does GABA inhibit evoked calcium influx in tectal neurons? These questions were tested using 300 μm brain slices perfused with artificial cerebrospinal fluid containing 2.5 mM K^+ (basal) or 30-60 mM K^+ (depolarization). Peak evoked calcium influx, the latency to peak influx, and the decay in evoked influx were measured as dependent variables. We found subtle but unreproducible effects of CRF at single doses on peak and latency to peak evoked calcium. The water-soluble and selective CRFR1 antagonist NBI 35965 hydrochloride did not change evoked calcium influx. Importantly, there were no substantial inhibitory effects of GABA on calcium influx evoked with either 30 or 60 mM K^+. These findings suggest that peak calcium influx evoked with high K^+ primarily reflects the rapid opening of voltage-gated calcium channels in presynaptic nerve endings upstream of tectal GABA-responsive cells. Whether these findings reflect a lack of CRF-mediated presynaptic inhibition needs to be confirmed using electrophysiological methods.

Disclosures: **C. Wang:** A. Employment/Salary (full or part-time):; Texas Tech University. **P. Keyel:** A. Employment/Salary (full or part-time):; Texas Tech University. **J. Carr:** A. Employment/Salary (full or part-time):; Texas Tech University. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; National Science Foundation.

Poster

P654. Threat/Fear: Memory Modification

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P654.01

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH Grant MH069558
NIH Grant F32MH120938

Title: Fear Devaluation and Extinction Rely on Distinct Molecular Mechanisms but Produce a Similar Behavioral Outcome

Authors: ***S. TRASK**[1], J. NELSEN[2], S. ORSI[2], H. DURAISWAMY[2], K. GRISALES[1], T. J. JAROME[3], F. J. HELMSTETTER[4];
[1]Univ. of Wisconsin - Milwaukee, Milwaukee, WI; [2]Virginia Polytechnic Inst. and State Univ., Blacksburg, VA; [3]Virginia Tech., Blacksburg, VA; [4]Deptartment of Psychology, Univ. of Wisconsin Milwaukee Dept. of Psychology, Milwaukee, WI

Abstract: Exposure to weak shock following contextual fear conditioning with a strong shock can have opposing effects on fear behavior depending on the number of weak-shock exposures. While 2 context-weak shock pairings will increase fear responding, 10 context-weak shock pairings reduces responding. While we hypothesized that the 10-shock procedure worked by modifying and weakening the original UCS memory, the 10-shock group received more chamber exposure than other groups suggesting this might instead be an extinction-like effect which relies on creation of a new inhibitory memory. The current experiments aimed to directly compare devaluation to a more straightforward extinction procedure. Following contextual fear conditioning with a strong (1.0mA) shock, animals were given one of three interference procedures: updating (2x .3mA shocks), devaluation (10x .3mA shocks), or extinction. Updating increased fear relative to both the devaluation and extinction groups who did not differ from each other. While devaluation and extinction both reduced spontaneous recovery, only devaluation reduced reinstatement. We also found that, like extinction, the effects of devaluation are context specific. In separate animals, brain tissue was collected 60-min following updating, devaluation, or extinction. Phosphorylated CREB was upregulated in the lateral amygdala following devaluation, but downregulated following extinction. Only devaluation produced an upregulation

of K48 polyubiquitination in the synapse, a marker of plasticity associated with memory reconsolidation and modification. Together, these results suggest that devaluation represents a unique interference paradigm that weakens fear responding by targeting the original acquisition memory rather than through the creation of a new memory, as is the case in extinction. Further work will need to examine the circuit-level mechanisms of devaluation as it compares to extinction, which is heavily dependent on cortical regions like the infralimbic cortex for expression.

Disclosures: S. Trask: None. **J. Nelsen:** None. **S. Orsi:** None. **H. Duraiswamy:** None. **K. Grisales:** None. **T.J. Jarome:** None. **F.J. Helmstetter:** None.

Poster

P654. Threat/Fear: Memory Modification

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P654.02

Topic: G.01. Fear and Aversive Learning and Memory

Support: Muhlenberg College Collaborative Summer Research Program Grant

Title: Habituation and disruption of memory with the use of a sedative in Physarum polycephalum

Authors: *E. MULLER, A. DUSKIC, G. GOTTHARD;
Muhlenberg Col., Allentown, PA

Abstract: Memory has typically been reserved for multicellular organisms with a central nervous system, and yet *Physarum polycephalum*, which is a unicellular, non-neuronal organism, can hold some form of memory. Two experiments were conducted to test the strength of *Physarum*'s memory. In Experiment 1A, *Physarum* was exposed to 5% white vinegar, which is a known mold killer yet unstudied in a lab setting, to assess the organism's ability to habituate to the deterrent (from 3-5 days). *Physarum* was placed at one end of a 1 cm x 2 cm vinegar/agar bridge (experimental group; n=35) or an agar only bridge (control group; n=36), with its food source at the opposite end of the bridge (adapted from Boisseau et al., 2016). The speed of *Physarum* to traverse the bridge and reach the food source (i.e., oats) was recorded using infrared cameras. In Experiment 1B, disruption of *Physarum*'s habituation memory was tested. Once habituation was established, the *Physarum*'s speed to cross the vinegar/agar bridge (experimental group; n=28) or the agar only bridge (control; n=29) was challenged using a sedative, Valerian root extract (300µL), which was confirmed by Adamatzky et al. (2011) to slow down the oscillation and, potentially, the movement and transfer of information throughout the organism, which may disrupt the organism's ability to "remember" what it learned. For Experiment 1A, repeated measures ANOVA revealed a significant main effect of trial showing time to cross

significantly decreased from the first to the last habituation trial [$F(3,207)=17.382$, $p<0.001$, $\eta p^2=0.201$]. There was a significant interaction between groups for speed to cross and showed that the slime exposed to the vinegar took longer to cross than the control [$F(3,207)=2.586$, $p=0.054$, $\eta p^2= 0.036$]. In Experiment 1B, a significant main effect of Valerian was observed, with all *Physarum* crossing at a decreased speed [$F(1,55)=43.986$, $p<0.0001$, $\eta p^2=0.444$]. Additionally, a significant interaction was observed; Valerian disrupted vinegar-habituated "memory" resulting in increased speed to cross the bridge when compared to controls [$F(1,55=8.243$, $p=0.036$, $\eta p^2= 0.078$]. Although the mechanism underlying *Physarum* "memory" is still unknown, our findings provide support for memory disruption via Valerian root in an organism without a brain.

Disclosures: E. Muller: None. **A. Duskic:** None. **G. Gotthard:** None.

Poster

P654. Threat/Fear: Memory Modification

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Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P654.03

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH-R01 MH093320

Title: Optogenetic photo-stimulation of basolateral amygdala neurons activated during fear memory formation promotes anxiety-like behaviors

Authors: *R. HAMMACK, N. CLAUSS, L. C. DAWS, G. M. TONEY;
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Abstract: Fear-related neuropsychiatric disorders such as posttraumatic stress disorder (PTSD) often present with symptoms of anxiety. Although fear and anxiety are often considered distinct both in their behavioral manifestations and their mediating neuronal pathways, to what extent re-activation of neurons activated during fear memory formation elicit anxiety-like behaviors is unknown. Since activity of basolateral amygdala (BLA) neurons is key to formation of fear and anxiety, we hypothesized that photo-stimulated re-activation of neurons activated during fear memory formation will elicit anxiety-like behaviors. To test this hypothesis, we used the 2nd generation of Targeted Recombination in Active Populations (TRAP2) transgenic mice injected bilaterally with adeno-associated virus encoding Cre-dependent enhanced channelrhodopsin (ChR2) in BLA. Mice were instrumented with fiber optic cannulae above the injection sites. Nine days later mice were habituated to the fear conditioning chamber for 5 days (3 min/day) to decrease novel exploration induced BLA activation. The following day, mice underwent fear conditioning (5, 0.75 mA foot shocks in 6 min) and immediately (<1 min) after were fear-

TRAPed with 4-hydroxytamoxifen (4-OHT) to induce Cre recombinase expression of ChR2 driven by *c-fos* promoter/enhancer elements. Three weeks post TRAPing, we validated functional ChR2 expression using brain slice electrophysiology. At this same time point, mice (n=4/group) underwent an open field test (OFT) while being photo-stimulated either with dummy (no light) or blue light (473 nm, 20 Hz, 5 ms) for 5 min. One hour following this test, mice underwent elevated plus maze (EPM) testing, again with or without photo-stimulation. Compared to dummy pulses, photo-stimulation of fear-TRAPed BLA neurons increased time that mice spent around the edges of the open field (blue light: 268 ± 9 s, no light: 132 ± 28 s, unpaired t test P=0.0040) and decreased time spent in the open arms of the EPM (blue light: 43 ± 16 s, no light: 206 ± 12 s, unpaired t test P=0.0018). Findings indicate that optogenetic re-activation of BLA neurons that were *c-fos* activated (TRAPed) during fear memory formation promotes anxiety-like behaviors in two test paradigms. We conclude that fear-related BLA neurons could contribute to subsequent anxiety-like behaviors. Further investigation into cellular and circuit mechanisms underlying these observations is warranted.

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Poster

P654. Threat/Fear: Memory Modification

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Program #/Poster #: P654.04

Topic: G.01. Fear and Aversive Learning and Memory

Support: NRF grant funded by the Korea government (No. 2020R1A2C2102134)

Title: Manipulations of fear memory induced by a non-invasive activation of dopamine neurons

Authors: *Y. JO[1], G. PYEON[1], L. S. ZWEIFEL[2];
[1]Sch. of Psychology, Korea Univ., Seoul, Korea, Republic of; [2]Pharmacol., Univ. of Washington, Seattle, WA

Abstract: Recent evidence indicates that optogenetic stimulation of dopamine cells during fear conditioning significantly decreases fear generalization. The optical dopamine manipulation during extinction training also facilitates fear extinction. Here we tested that instead of the optogenetic intervention, whether a non-invasive activation of dopamine cells induced such effects on fear memory. *Slc6a3*-cre mice (DAT-cre) first underwent a Pavlovian reinforcement learning where an auditory conditioned stimulus (CS) was associated with the delivery of a food pellet (rewarding CS; re-CS) and the other CS was not (nonrewarding CS; nonre-CS). Electrophysiological results showed that dopamine cells preferentially responded to re-CS over 8 days of Pavlovian learning. Then the mice received a probabilistic version of fear conditioning in which a high-probability CS (highCS) was paired with a footshock 7 out of 10 trials, whereas a

low-probability CS (lowCS) was paired with the other three trials. When highCS was presented during fear conditioning, either re- or nonre-CS was also played as a compound cue to excite dopamine cells for 1 s. Mice with nonre-CS failed to discriminate between highCS and lowCS as they exhibited comparable levels of freezing to both CSs. However, mice with re-CS showed an improvement in discriminatory freezing behavior. To demonstrate that the better performance requires dopamine activation by re-CS, another group of DAT-cre mice expressing Jaws, an inhibitory opsin, in dopamine cells was included. The Jaws group went through the same behavioral procedures, except that dopamine neurons were inhibited by red light (1 s) during the presentation of re-CS. Dopamine inactivation prevented discriminatory fear memory. Next, we examined whether the non-invasive excitation of dopamine neurons facilitated the extinction of fear memory. Mice received Pavlovian reward association for 8 days, followed by fear conditioning where a CS was paired with a footshock 4 times. During the training of fear extinction, CS-alone trials were presented 40 times. In each trial, either re-CS or nonre-CS was turned on for 1 s at the end of the fear-associated CS. Compared to mice with nonre-CS, mice with re-CS showed significantly less freezing behavior during the extinction training. Overall, these results indicate that dopamine activation evoked by a reward-predicting cue can facilitate fear discrimination and extinction as effectively as optical dopamine activation. The present study also suggests a possibility that fear- and reward-related compound cues can help to diminish generalized fear responses in human patients suffering anxiety-related disorders.

Disclosures: Y. Jo: None. **G. Pyeon:** None. **L.S. Zweifel:** None.

Poster

P654. Threat/Fear: Memory Modification

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Program #/Poster #: P654.05

Topic: G.01. Fear and Aversive Learning and Memory

Support: 5R01MH116445
5R01MH105414
5R21MH114170

Title: Hippocampal somatostatin interneurons mediate fear extinction retrieval

Authors: *A. F. LACAGNINA, S. KHAN, R. L. CLEM;
Neurosci., Icahn Sch. of Med. at Mount Sinai, New York, NY

Abstract: Fear extinction is a process where defensive responses to a conditioned stimulus diminish when the stimulus no longer predicts a threat. While plasticity in prefrontal-hippocampal-amygdala circuits has been implicated in extinction learning, much less is known about the mechanisms of extinction retrieval. We first identified brain regions associated with

extinction retrieval by mapping expression of the activity-related gene c-Fos in mice retrieving a contextual fear extinction memory. We found that extinction retrieval was associated with elevated c-Fos expression in the stratum oriens layer of ventral hippocampal area CA1 (vCA1) predominantly in interneurons co-expressing somatostatin (SST-INs) compared to fear retrieval or in unshocked mice. An extinction-specific recruitment of SST-INs in vCA1 was confirmed using an intersectional genetic strategy to fluorescently tag SST-INs active during fear conditioning or extinction retrieval. Optogenetically activating the extinction-tagged SST-INs attenuated fear relapse but did not affect extinction retrieval or context discrimination. Additionally, silencing excitatory projections from vCA1 to prelimbic medial prefrontal cortex did not affect fear retrieval but prevented extinction retrieval. Our results suggest extinction retrieval is mediated by activity of vCA1 SST-INs, which we hypothesize regulate the output of hippocampal ensembles to promote extinction-related behaviors in part through projections to the prefrontal cortex.

Disclosures: A.F. Lacagnina: None. **S. Khan:** None. **R.L. Clem:** None.

Poster

P654. Threat/Fear: Memory Modification

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Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P654.06

Topic: G.01. Fear and Aversive Learning and Memory

Support: Roche Postdoc Fellowship
Synapsis Foundation
Swiss National Science Foundation
Marie Heim Vögtlin SNF

Title: Social buffering extinguishes fear by oxytocin triggering sustained changes in neuronal activity in the central amygdala

Authors: C. HEGOBURU[1], S. GHOSH[2], R. TRIANA DEL RIO[2], I. SALGADO[2], M. ABATIS[2], R. NIU[2], E. H. VAN DEN BURG[2], C. GRUNDSCHOBER[3], ***R. STOOP**[4];
[1]Univ. of Lausanne, Ctr. for Psychiatric Neurosci., Prilly, Lausanne, Switzerland; [2]Ctr. For Psychiatric Neuroscience, Univ. Lausanne, Prilly, Lausanne, Switzerland; [3]Roche Pharma Res. and Early Develop., Roche Innovation Ctr. Basel, Basel, Switzerland; [4]Univ. of Lausanne, Prilly, Lausanne, Switzerland

Abstract: Background: In fear extinction therapy, social interaction can help to realize successful and long-lasting decrease of fear. In rat models of fear-conditioning, the presence of a conspecific during the recall can attenuate fear responses. The mechanism underlying this social buffering of fear (SBF) is not fully understood. One candidate to mediate SBF is oxytocin (OT),

a neuropeptide produced by the paraventricular nucleus of the hypothalamus (PVN) and known for its anxiolytic effects in the central amygdala (CeA). We hypothesize that OT, through its projections from the PVN to the CeA, plays a role in socially-induced acute and long-lasting decrease (extinction) of fear.
Methods: We developed a fear conditioning paradigm for rats, in which the recall of a threat was delivered in the presence of a conspecific. Using pharmacological, optogenetic, chemogenetic and electrophysiological approaches, we assessed the role of OT signaling in the PVN to CeA pathway during SBF.
Results: We found that social interaction reduced the freezing responses immediately and long-lastingly and that OT release from the PVN into CeA plays a crucial role in this process. Multi-unit *in vivo* electrophysiological recordings in the CeA in combination with optogenetic stimulation showed that these behavioral responses were accompanied by acute and long-lasting changes in spiking patterns in OTergic neurons in the PVN and three different types of fear-conditioned responding neurons in the CeA.
Conclusion: Our findings provide insight in the neurobiological mechanisms of OT-signaling underlying the short, and long-lasting effects of social buffering of fear.

Disclosures: C. Hegoburu: None. **S. Ghosh:** None. **R. Triana Del Rio:** None. **I. Salgado:** None. **M. Abatis:** None. **R. Niu:** None. **E.H. van den Burg:** None. **C. Grundschober:** A. Employment/Salary (full or part-time):; Roche Pharma Research. **R. Stoop:** None.

Poster

P654. Threat/Fear: Memory Modification

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Program #/Poster #: P654.07

Topic: G.01. Fear and Aversive Learning and Memory

Support: MH117426
UT Catalyst Grant

Title: Fear Extinction Training Suppresses Reactivation of Fear Ensembles Throughout the Hippocampus and Induces Specific Single-Cell Transcriptomes

Authors: *A. ZUNIGA[1], J. HAN[2], I. MILLER-CREWS[1], C. ROSSMEISSL[2], H. A. HOFMANN[1], *M. R. DREW[1];
[1]Univ. of Texas At Austin, Austin, TX; [2]Univ. of Texas at Austin, Austin, TX

Abstract: Learned fear often relapses after extinction, suggesting that extinction training generates a new memory that coexists with the original fear memory. Recent work from our lab has identified the hippocampal dentate gyrus (DG) as a region where such fear and extinction memories are generated. Specifically, using activity-dependent neural tagging in mice, we

discovered that extinction training suppresses reactivation of contextual fear engram cells in DG while activating a second ensemble, a putative extinction engram. Here we investigate (1) whether these effects of extinction training on fear ensembles are limited to the DG or exist in other regions of the hippocampus and parahippocampal cortex, and (2) whether fear and extinction ensembles in DG exhibit distinct patterns of gene expression. ArcCreErt2-eYFPflx mice injected with tamoxifen immediately after contextual fear conditioning, thereby inducing expression of eYFP in neurons active during fear acquisition. Mice then received extinction training or were left undisturbed in their home cages. After a test of conditioned fear in the training context, we first assessed reactivation of eYFP-tagged fear acquisition neurons using immunohistochemistry against Arc and Fos proteins. Although the total number of eYFP+, Fos+ and Arc+ neurons in the dorsal DG did not differ between Extinction and No-Extinction groups, extinction training suppressed reactivation of YFP-tagged fear acquisition neurons in the DG, replicating results of Lacagnina et al. (2019). This extinction-induced suppression of reactivation of eYFP+ fear acquisition neurons was observed in dorsal and ventral DG, dorsal CA1, and dorsal and ventral lateral entorhinal cortex. In DG there was considerable overlap between Arc and c-Fos expression, and expression of both immediate-early genes in DG was suppressed by extinction training. Next we used single-nuclei RNA Sequencing (snRNA-seq) to characterize gene expression profiles in eYFP+ and eYFP- cells in the dorsal DG of mice from Extinction and No-Extinction groups. We recovered the canonical DG cell types, examined how the transcriptomes of eYFP+, Fos+ and Arc+ fear acquisition neurons changed after extinction training, and identified the transcriptomes of extinction-activated neurons. Our findings show that extinction training has anatomically and molecularly robust effects on hippocampal fear representations.

Disclosures: A. Zuniga: None. **J. Han:** None. **I. Miller-Crews:** None. **C. Rossmeissl:** None. **H.A. Hofmann:** None. **M.R. Drew:** None.

Poster

P654. Threat/Fear: Memory Modification

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Program #/Poster #: P654.08

Topic: G.01. Fear and Aversive Learning and Memory

Support: IBRO travel grant

Title: The dynamic nature of a fear engram in extinction and reconsolidation memories

Authors: *J. GRIEBLER LUFT, L. DE OLIVEIRA ALVARES;
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Abstract: Neurons active in memory formation will be re-engaged again during retrieval. Those cells are called engrams or neuronal ensembles. However, an important question about engrams remains unknown: Does the fear engram remain the same after extinction or memory reconsolidation in Basolateral Amygdala (BLA)? Using Fos-LacZ transgenic rats, we aimed to investigate if the reconsolidation and extinction of memories would maintain the same pattern of neurons that were active during its consolidation. With this technique, we can silence neurons that express the immediate early gene c-fos at the moment of the infusion of the drug Daun02. We first tested the role of the BLA engram in the reconsolidation of an aversive memory and demonstrated that silencing the neurons active during reconsolidation caused an impairment of the fear memory. However, silencing BLA neurons active during an extinction memory recall did not disrupt the extinction memory as seen by the low fear expression presented by the animals during the test. Our findings suggest that reconsolidation and extinction memory engrams have distinct dynamics in BLA, meaning that reconsolidation memories use the same subset of neurons, while extinction memories do not.

Disclosures: J. Griebler Luft: None. **L. de Oliveira Alvares:** None.

Poster

P654. Threat/Fear: Memory Modification

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P654.09

Topic: G.01. Fear and Aversive Learning and Memory

Title: Synaptic correlates of associative fear memory in the lateral amygdala

Authors: *D. CHOI, J. KIM, H. LEE, J.-I. KIM, Y. SUNG, J. CHOI, P. PARK, H. JUNG, B.-K. KAANG;
Sch. of Biol. Sci., Seoul Natl. Univ., Seoul, Korea, Republic of

Abstract: Successful adaptation to the environment requires accurate responding to external threats by recalling specific memories. Memory formation and recall requires engram cell activity and synaptic strengthening among activated neuronal ensembles. However, elucidating the underlying neural substrates of associative fear memory remained limited without a direct interrogation of extinction-induced changes of specific synapses that encode a specific auditory fear memory. Using dual-eGRASP (enhanced Green Fluorescent Protein Reconstitution Across Synaptic Partners), we found synapses among activated neuronal ensembles or activated synaptic ensembles showed a significantly larger spine morphology at auditory cortex (AC) to lateral amygdala (LA) projections after auditory fear conditioning in mice. Fear extinction reversed these enhanced synaptic ensemble spines while re-conditioning with the same tone and shock restored the spine size of the synaptic ensemble. Taken together, we suggest that synaptic ensembles encode and represent different fear memory states.

Disclosures: D. Choi: None. **J. Kim:** None. **H. Lee:** None. **J. Kim:** None. **Y. Sung:** None. **J. Choi:** None. **P. Park:** None. **H. Jung:** None. **B. Kaang:** None.

Poster

P654. Threat/Fear: Memory Modification

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P654.10

Topic: G.01. Fear and Aversive Learning and Memory

Support: University of Zurich's Clinical Research Priority Program for the CRPP "Synapse & Trauma"

Title: Forget the threat: Effects of the matrix metalloproteinase inhibitor doxycycline on human trace fear memory.

Authors: *J. WEHRLI[1], Y. XIA[1], D. BACH[2,1];
[1]Univ. of Zurich, Zürich, Switzerland; [2]Univ. Col. London, London, United Kingdom

Abstract: Multiple studies have tried to interfere with the consolidation of fearful memories by pharmacological means, however this has proven difficult in humans. The matrix metalloproteinase (MMP) inhibitor doxycycline has been shown to attenuate the consolidation of aversive memory in delay fear conditioning when ingested before the acquisition phase (Bach et al., 2018). Delay fear conditioning, where a conditioned stimulus (CS) co-terminates with an unconditioned stimulus (US), is widely used to model traumatic memory; however, in real-life memory cues that elicit trauma often are not present during the trauma but separated in time. To model this situation, we sought to replicate the previous finding in a trace fear conditioning paradigm. Crucially, this difference in experimental timing leads to additional involvement of hippocampus, but currently it is unclear whether doxycycline can block memory consolidation when it is hippocampus dependent. We tested the impact of Doxycycline on trace fear memory consolidation in a randomised double-blind placebo-controlled study with 97 participants: n = 49 (25 females) received placebo, n = 48 (24 females) received doxycycline 3.5 hours before conditioning. We used two visual CS and electric shock as US with 100% reinforcement of the CS+ and a 15-s trace interval. The first acquisition session (day 0) encompassed 40 trials (20 CS+ and 20 CS-). A recall session (day +7) included 30 trials (15 CS+, 15CS-) without reinforcement. We used startle-eye blink responses (SEBR) - elicited by a white noise burst - to measure memory retention. Beforehand, we ensured in two independent samples trace fear memory retention in this paradigm can be measured after 7 days. We found that the placebo group retained fear memory indicated by a significant CS+/CS- difference, while the Doxycycline group showed no memory retention. However, we did not observe a significant group difference. As a side finding it is worth noting that, within the placebo group, only females but not males retained fear memory.

Disclosures: J. Wehrli: None. **Y. Xia:** None. **D. Bach:** None.

Poster

P664. Memory Modification

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Program #/Poster #: P664.01

Topic: G.02. Reward and Appetitive Learning and Memory

Support: MOST Grant 107-2320-B-182 -019 -MY3

Title: Infralimbic prefrontal glutamate projection in extinction of methamphetamine conditioned place preference

Authors: *J.-C. CHEN, T.-Y. WU, H.-C. CHANG, Y.-T. LIN;
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Abstract: Exposure to contextual cues frequently triggers relapse of addictive drug use, even after prolonged abstinence. To eliminate this drug-cue association, strengthening the extinction process would be a practical manner to effectively suppress the devastating drug relapse. Cortical glutamate efferent projects to many midbrain regions and this projection pathway has been demonstrated to play a critical role in the extinction of fear conditioning (aversive). Whether prefrontal glutamate plays a similar role in appetitive conditioning remains unclear. To delineate if infralimbic mPFC (IL-mPFC) glutamate, via activation of inhibitory GABA neurons in the VTA, would modulate the METH-CPP extinction process, we adopted optogenetics along with specific Cre mice to unwire this neural circuitry. We found photo-inactivation (AAV5/Efla-DIO-eNpHR3.0-mCherry) of IL-mPFC glutamate projection in the VTA of vGluT2::Cre mice during the extinction training suppresses the METH-CPP extinction. Photo-inactivation of GABA interneurons in the VTA of Parvalbumin::Cre mice during the extinction training also blocks the METH-CPP extinction. Nevertheless, photo-inactivation of GABA projection neuron, with NpHR delivered in the NAc-shell but optical fiber implanted in the VTA of GAD2::Cre mice, displayed normal METH-CPP extinction after CPP re-test. In addition, animals that received mutant GluR1/S831A/S845A or GluR1/S845A exhibited normal acquisition of METH-CPP but could not extinguish the CPP, suggesting that disrupts the glutamate response at VTA GluR1 level prevents the formation of METH-CPP extinction. In addition, extinction of METH-CPP could also be blocked by systemic pretreatment of AMPA-R antagonist, NBQX during METH-CPP extinction training. These data suggest that disrupts the glutamate response at VTA GluR1 level prevents the formation of METH-CPP extinction. Overall, we hypothesize that during extinction training of METH-CPP, IL-PFC consistently sends an excitatory signal to the VTA GABA interneuron, in turn, suppresses VTA dopamine activity hence results in a suppression of METH-CPP. Thus, photo-inactivation of IL-PFC glutamate during this critical extinction

training stage would totally disrupt the memory extinction process and animals remain in the acquisition stage of METH-CPP.

Disclosures: J. Chen: None. **T. Wu:** None. **H. Chang:** None. **Y. Lin:** None.

Poster

P664. Memory Modification

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P664.02

Topic: G.02. Reward and Appetitive Learning and Memory

Support: BBRF Young Investigator Grant
NIH 5R01MH104602-08

Title: Coding of social odors in the hippocampal CA2 region as a substrate for social memory

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Abstract: The ability to store, recall, and update social information about individuals is critical for lasting social relationships. The computational demands for the brain to support those features are challenging as multiple parallel streams of information have to be woven into a coherent representation, which at the same time has to be flexible enough to cope with the dynamic nature of social behavior. Research over the last decades established a crucial role of the hippocampus in the formation of social recognition memory, however, the underlying computations are unknown. Here we took inspiration from scent marking as a crucial feature for individual recognition in many mammalian species and paired the presentation of the volatile fraction of urine from individual mice via a custom-built olfactometer with large-scale, high-resolution calcium imaging in the hippocampus of awake head fixed mice. We focused on area CA2, whose crucial role in social recognition memory is well established on a behavioral level, and report network computation features underlying social as well as non-social odor representation. We show found that during passive experience of individual novel social odors, CA2 uses a sparse code to form specific and robust neural representations that allow for stimulus decoding with high accuracy. Moreover, these representations are highly associative and incorporate valence in a flexible manner when tested in an associative reward learning task. Optogenetic silencing of CA2 impairs the learning of odor-reward associations, supporting a causal role of CA2 in social odor discrimination. To further explore the logic by which CA2 encodes odor stimuli, we trained mice in the reward learning task using two novel social odors and two non-social odors, in which one each of the social and non-social odors was rewarded. After learning, we found that CA2 activity can distinguish all four odors in a manner consistent with a geometry of neural representations allowing for it providing abstract representations of

social versus non-social odors. Together these results argue strongly that CA2 forms a hub that classifies and distinguishes multiple forms of olfactory stimuli, which both enhances the learning of olfactory-reward associations and is poised to contribute to social memory storage.

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Poster

P664. Memory Modification

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Topic: G.02. Reward and Appetitive Learning and Memory

Support: DA047981
DA007262

Title: Viral inhibition of HDAC3 in the infralimbic cortex during Pavlovian extinction eliminates Pavlovian-to-instrumental transfer

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Abstract: The extinction of learned (conditioned) behaviors is an essential process for functioning adaptively in an ever changing environment. In the context of Pavlovian processes, extinction of conditioned responses (CRs) occurs when an unconditioned stimulus (US) is omitted from presentations of an acquired conditional stimulus (CS). Although extinction results in the disappearance CRs, the original associative CS-US memory is preserved and can be revealed with several experimental procedures. Most of the approaches that test the preservation of the original association focus on the return of CRs themselves (e.g., spontaneous recovery, reinstatement, and contextual renewal), however one particularly powerful approach is a test for Pavlovian-to-instrumental transfer (PIT). In this approach presentation of an acquired CS invigorates an independently acquired instrumental behavior (e.g, lever pressing for a US). Although extremely useful for understanding behavioral mechanisms of extinction, PIT has been underutilized for the purposes of studying the extent and efficacy of extinction training. Here we evaluate the effects of viral manipulation of histone deacetylase 3 (HDAC3) in the infralimbic (IL) cortex of male Long Evans rats on Pavlovian extinction. Normally functioning HDAC3 keeps chromatin in a repressive state and reduces transcription; in contrast, inhibition of HDAC3 shifts chromatin to a permissive state, facilitating transcription and enhancing learning and memory. The IL has been identified as a key structure meditating the preservation of associative memories in the face of extinction training. In this study, following initial instrumental training and Pavlovian conditioning, rats received IL injections of a point mutant HDAC3 virus to

interfere with HDAC3 function, a wild type HDAC3 to enhance HDAC3 activity, or null virus to serve as control. After recovery rats underwent Pavlovian extinction followed by PIT testing. During extinction training CRs (food cup entries) disappeared with only subtle differences in the rate of extinction among groups. However, during PIT testing only null and wild type transduced groups demonstrated PIT, pointing to a preservation of the original CS-US association. In contrast, PIT was notably absent in the point mutant transduced group, suggesting a degradation of the original CS-US association. In a final tests for contextual renewal of CRs, testing in a new context revived conditioned food cup entries only in the null transduced group. These data identify HDAC3 as one underlying mechanism by which the IL acts to preserve original memories.

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Poster

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Program #/Poster #: P664.04

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Support: NIH R00-MH105549
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Rising STARs Award from the University of Texas System
NARSAD Young Investigator

Title: Repeated morphine exposure alters prelimbic cortex activity and induces risk-taking behavior in rats

Authors: *A. M. VASQUEZ[1], C. B. QUAVE[1], E. P. BORA[1], C. L. CHIDOMERE[1,2], G. AQUINO-MIRANDA[1], D. S. ENGELKE[1], F. H. DO MONTE[1];
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[2]Dept. of Biomed. Engin., Univ. of Houston, Houston, TX

Abstract: Opioid addiction is associated with impaired risk-related decision-making. However, it remains unclear how the brain processes risk in the pursuit of opioid use to alter motivated behavior during conflict situations. The prelimbic area of the medial prefrontal cortex (PL) has been associated with executive functioning and behavioral responses to rewarding and aversive stimuli. We therefore hypothesized that repeated opioid exposure would suppress activity in PL and increase risk-taking behavior. To test this hypothesis, we designed an approach-avoidance conflict model that involves the presentation of an innate fear-inducing predator odor. Using a biased opioid conditioned place preference (CPP) model, we injected adult male Long-Evans rats with saline (n = 36) or morphine (n = 24) and exposed them to their least-preferred side of the

chamber on alternating days over a 10-day period. After 72 h of forced abstinence, morphine-treated rats showed a higher preference for the drug-paired side when compared to the saline group ($p < 0.001$). Immediately after preference testing, we added predator odor (cat saliva) to the previously drug-paired side to produce a motivational conflict. While the saline group showed avoidance to the cat odor during the conflict test, the morphine group continued to enter the drug-paired side, demonstrating that morphine conditioning induces an increase in risk-taking behavior. Single-unit recordings from PL neurons revealed a significant number of cells with suppressed firing rates after acute administration of morphine (22 %, 19/86), but not saline (6 %, 5/64; Fisher exact test, $p = 0.011$). On the last conditioning day, morphine administration did not induce changes in PL firing rates as compared to saline ($p = 0.397$), suggesting that PL neurons adapt to the effects of repeated morphine over time. During the conflict test, distinct populations of PL neurons were either excited ($n = 28$ neurons, $p = 0.011$) or inhibited ($n = 14$ neurons, $p = 0.0061$) when saline rats entered the cat odor-paired side as compared to when they entered the same side during the preference test. Interestingly, this change in neural activity was not observed in the morphine group during the conflict test. Taken together, our results demonstrate that opioid-induced CPP is associated with reduced inhibition of PL activity following repeated administrations of morphine. In addition, after repeated opioid exposure, PL neurons failed to respond to threat stimuli, and this pattern of neuronal activity occurred alongside increased risk-taking behavior during approach-avoidance conflict.

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Poster

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Rising STARs Award from the University of Texas System
NARSAD Young Investigator

Title: Dynamics of prelimbic cortex activity encode the predictive value of reward-associated cues.

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Abstract: Behavioral adaptation to shifts in food availability is critical for maintaining efficiency during foraging. Neurons in the prelimbic prefrontal cortex (PL) display changes in firing rates in response to food-associated cues. However, it is unknown whether PL activity encodes fast changes in food availability (e.g. reward omission). We hypothesized that PL neurons show modified responses to reward availability and associated cues according to variations in reward predictability**.** Male Long-Evans rats implanted with a single-unit electrode in PL were trained to press a lever to obtain a reward (single sucrose pellet) in a nearby dish. Reward availability was limited to the duration of 15-second audiovisual cues. Rats were then subjected to an intercalated omission session (20 rewarded and 20 omitted trials semi-randomly interspersed) followed by a repeated omission session (18 rewarded followed by 20 omitted trials) to compare the neuronal correlates of unexpected vs. expected reward omission, respectively. PL firing rates were analyzed during both the onset of cues and when the animal's head entered the dish after cued lever presses. During unexpected reward omission, rats exhibited "frustrative behaviors" characterized by higher lever press rate, greater number of burst presses, and increased locomotion speed (ANY-Maze automated video tracking system). However, these behaviors were not observed when reward omission was expected. Recordings from PL neurons (n = 233 cells from 16 rats) revealed a subset of cells that altered their firing rates (16% excited, 14% inhibited) when rats detected the presence of reward in the dish. Interestingly, 58% of these cells did not respond when reward was omitted, suggesting that PL neurons encode loss of a previously available reward. During expected reward omission, a subset of cells emerged exhibiting increased firing rates exclusively upon repeated detection of reward omission (8%), indicating that some PL neurons are recruited when reward omission becomes predictable. In addition, 13% of PL cells showed reductions in firing rates upon repeated reward cue presentation, half of which did not respond to repeated omission cues. It is possible that this progressive loss of inhibition in PL neurons may encode the reduced predictive value of cues when reward omission becomes an expected outcome. Together, our findings suggest that separate populations of PL neurons signal immediate vs. persistent reward loss and that real-time updating of PL activity is associated with dynamic changes in the predictive value of reward cues.

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Poster

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Topic: G.02. Reward and Appetitive Learning and Memory

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Rising STARs Award from University of Texas System
NARSAD Young Investigator

Title: Activity in the prelimbic prefrontal cortex encodes individual differences in approach-avoidance conflict in rats

Authors: *G. AQUINO-MIRANDA[1], J. A. FERNANDEZ-LEON[1], D. S. ENGELKE[1], A. GODSON[2], F. H. DO MONTE[1];
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Abstract: Neurons in the prelimbic subregion of the prefrontal cortex (PL) respond to both reward- and threat-associated cues. However, whether PL activity regulates the animals' decision to approach or avoid such cues as remains unknown. To address this question, male Long-Evans rats with single-unit recording electrodes implanted in PL were initially trained to press a bar for sucrose during the presentation of audiovisual cues. Next, rats were fear conditioned by pairing a neutral odor with electrical footshocks. During the test session, rats were placed in an arena and exposed to three phases: only audiovisual cues (reward), only odor cues (fear), or both simultaneously (conflict). During the reward phase, animals pressed the bar immediately after the onset of the food cues. In the fear phase, animals showed robust defensive behaviors characterized by high levels of freezing, avoidance, and risk-assessment responses. Interestingly, two different behavioral phenotypes emerged during the conflict phase: rats that continued searching for food (*pressers)* vs. rats that remained in the hidden area and showed suppression of food-seeking responses (*non-pressers*). PL recordings during the reward phase revealed a greater number of reward-cue responses in *pressers* (~33% of 237 neurons), when compared to *non-pressers* (~21% of 89 neurons; Fisher Exact Test, $p = 0.042$*)*. Notably, the magnitude of reward-cue responses observed during the conflict phase was similar to the reward phase for *pressers,* but was drastically reduced for *non-pressers*. Using a combination of recordings and optogenetics for photoidentification of distinct subtypes of PL neurons in *pressers*, we found that ~31% of PL glutamatergic (PL^{GLUT}) neurons inhibited their spontaneous firing rate during the reward phase, when compared to baseline. Presentation of the conditioned odor during the fear phase disinhibited ~67% of these PL^{GLUT} neurons, whereas the introduction of reward cues during the conflict phase restored the inhibition to the same levels as the reward phase. In contrast, changes in spontaneous firing rate of PL GABAergic neurons were similar across the different phases. Moreover, photoactivation of PL^{GLUT} neurons during the onset of the reward cues reduced reward-seeking responses in *pressers,* whereas photoinhibition of PL^{GLUT} neurons in non-pressers reduced defensive responses and biased animals' behavior toward the food area during conflict (Wilcoxon test, all p values < 0.05). Together, our results establish a role for PL in the regulation of approach-avoidance conflict by demonstrating that reduced activity in PL^{GLUT} neurons is associated with increased risky sucrose-seeking behavior.

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Poster

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Topic: G.02. Reward and Appetitive Learning and Memory

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Rising STARs Award from University of Texas System
NARSAD Young Investigator

Title: Shifts in the activity of prelimbic cortex and paraventricular thalamic neurons correlate with changes in food-seeking strategies in a predator-associated context

Authors: *D. S. ENGELKE[1], G. AQUINO-MIRANDA[1], M. CATTANI[2], F. H. DO MONTE[1]; [1]Neurobio. and Anat., The Univ. of Texas Hlth. Sci. Ctr. at Houston, Houston, TX; [2]Tufts Univ., Medford-Somerville, MA

Abstract: In nature, animals live in dynamic environments where availability of food and risk of predation constantly change. Updating a contextual memory is important to resume foraging behavior when a threatening environment becomes safe again, but which neural circuits regulate this dynamic process remains unclear. Rats exposed to predator odor form a contextual fear memory and express defensive behaviors when returned to the same environment. Here, we designed a contextual fear conflict model in which rats have to forage for food in a context previously associated with predator odor (cat saliva). Adult male Long-Evans rats were initially trained to press a lever for food during the presentation of audiovisual cues. Next, rats were exposed to a context with cat odor and in the following day reexposed to the same chamber for a contextual fear conflict test. To investigate whether foraging behavior changed across the session, we compared food-seeking responses during the first 18 food cues (early) vs. the last 18 food cues (late). Although cued lever presses remained the same across the session, the latency to press reduced from early to late cues, suggesting behavioral flexibility over time. Recently, we demonstrated that cat odor exposure suppresses food-seeking responses, and this effect is mediated by increased neuronal activity in the paraventricular nucleus of the thalamus (PVT). The PVT is reciprocally interconnected with the prelimbic cortex (PL), a region directly involved with behavioral flexibility and contextual information. To test whether shifts in the activity of PL and PVT neurons correlate with the dynamic changes in lever-press latency observed during the test, we performed single-unit recordings from PL (223 cells, 14 rats) and PVT (161 cells, 13 rats) neurons in two different cohorts. We observed that a higher number of PL neurons changed

their spontaneous activity from early to late session (37 % excited, 42 % inhibited) when compared to PVT neurons (16 % excited, 20 % inhibited, $p < 0.001$). When we time-locked neuronal activity with the onset of the food cues, both PL and PVT neurons showed fast excitatory food cue responses at the early phase, with sustained excitatory food cue responses emerging only at the late phase when rats exhibited shorter latencies to press the lever. In contrast, fast inhibitory food cue responses in PL and PVT neurons were observed at both phases, but sustained inhibitory responses were exclusively observed in PVT neurons at the early phase. Dynamic shifts in PL and PVT activity may serve to update information about the predator context and food cues, thereby allowing changes in foraging strategies in a conflicting environment.

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Poster

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Rising STARs Award from UT System
NARSAD Young Investigator

Title: Computational-designed training protocols enhance associative learning in rats

Authors: *X. O. ZHANG[1], Y. ZHANG[1], D. S. ENGELKE[1], C. E. CHO[2], P. D. SMOLEN[1], J. H. BYRNE[1], F. H. DO MONTE[1];
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Abstract: Associative learning requires the activation of protein kinases with distinct temporal dynamics. Learning protocols with computationally designed inter-trial intervals (ITIs) that maximize the interaction between fast-activated protein kinase A (PKA) and slow-activated extracellular signal-regulated kinase (ERK) enhance memory acquisition in *Aplysia*. Here, we tested whether an optimized learning protocol, predicted computationally to increase the overlap between PKA and ERK signaling in rat hippocampus, would enhance memory formation. On day 1, male adult Long-Evans rats were exposed to an auditory fear conditioning session (Context A) with multiple trials of a conditioned stimulus (CS, 3 kHz tone, 30 s) that co-

terminated with an unconditioned stimulus (US, footshock, 0.7 mA, 0.5 s). Rats were randomly assigned to 3 groups: i) full conditioning (FC, n = 7), which received 8 CS-US pairings with 4.5 min ITIs; ii) partial conditioning (PC, n = 6), which received only 4 CS-US pairings with 4.5 min ITIs; and iii) enhanced partial conditioning (EPC, n = 7), which received only 4 CS-US pairings, but with ITIs of 8, 8 and 16 min. After conditioning, all groups exhibited high levels of freezing (~70 %) and reduced locomotion. On day 2, rats were exposed to a retrieval test consisting of two CS presentations in a novel context (Context B). Rats in the FC and EPC groups showed reduced locomotion speed when compared to the PC group, suggesting stronger memory retrieval. On day 3, rats were returned to Context A for extinction training (12 CS presentations only). Interestingly, EPC and FC groups showed impaired extinction learning characterized by sustained freezing levels within the session, compared to the PC group which showed successful extinction as indicated by reduced freezing and increased locomotion. Next, we tested whether the same optimized ITIs would enhance fear extinction. A separate group of rats that were previously trained to press a lever for sucrose were fear conditioned, and then randomly assigned to three groups: i) full extinction (FE, n = 12), which received 12 CS presentations with 2.5 min ITIs; ii) partial extinction (PE, n = 13), which received only 4 CS presentations with 2.5 min ITIs; and iii) enhanced partial extinction (EPE, n = 11), which received only 4 CS presentations, but with ITIs of 8, 8 and 16 min. Compared to FE and PE groups, the EPE group showed higher lever press motivation during a spontaneous recovery test on day 28, suggesting an enhancement in the extinction of contextual fear memory. Together, these results demonstrate that computationally designed learning protocols based on PKA and ERK dynamics may enhance associative learning in mammals.

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Poster

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Program #/Poster #: P664.09

Topic: G.02. Reward and Appetitive Learning and Memory

Title: Sex differences in reward processing: A focus on incentive contrast

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Abstract: Research on sex differences for preferences of reward outcomes, such as flavors, show mixed results between male and female rats. Some consumption studies suggest that sexual dimorphism of the brain is contributing to female's consistent performance at higher rates for sucrose concentrations than males; however, these finding are not reliably shown between

studies. The current study examined sex difference in motivation for sucrose reward and focused on relative reward effects as they interact with basic motivation. Inducing hunger through food restriction is a method that is used in the present study to examine its influence on reward processing. This study uses three levels of sucrose solution concentrations as rewards between 12 male and 12 female Sprague-Dawley rats. This study is focused on determining if sexual dimorphism of the brain is reflected by performance differences of processing reward information. Specifically, we are examining the differences between the sexes on their ability to change performance on the original concentration (control) given when an up-shift or down-shift occurs. If differences in responses to the same outcome occurs, then it is labeled as an incentive contrast (IC). MED-PC IV software was used to record nose-pokes, licking rates, and latency data, while consumptions were taken directly from sucrose syringes (ml) after trials. Results support few to no sex differences in reward discrimination or IC, but large sex differences in sucrose consumption. Moreover, significant effects were found for the influence of motivational state and behavioral context. Animals expressed incentive contrast when food restricted and in the context of their home cage. These effects were diminished to absent when animals ate freely or were tested in an operant task for relative reward effects. Overall, the findings point to diverse influences on sexual dimorphism for reward processing. Other implications include brain behavior relations such as memory consolidation, motivation, and drug and alcohol use/abuse.

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Poster

P687. Motivation: Social Communication and Behavior in Mice

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Topic: G.03. Motivation

Support: NRF-2017M3C7A1026959
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Title: Impact of social isolation on the prefrontal-nucleus accumbens circuit critical for social recognition in mice

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Abstract: It is known that social isolation in early life causes prolonged deficit in cognitive functions. Previous studies have shown that social isolation in the postnatal critical period altered functions of various medial prefrontal cortical (mPFC) within a variety of related behaviors including social preference in mice. However, it is still elusive how other aspects of social functions and which downstream targets for the mPFC are affected by the insubstantial social experience in young age. In this study, we observed that socially isolated mice in their postnatal developmental period showing an impairment in social behavior which was not rescued by following re-socialization period. Interestingly, we found that the functional connectivity between mPFC and the shell part of nucleus accumbens (NAc shell) is altered after the social isolation in both electrophysiology and c-fos immunohistochemistry. Consistently, *in vivo* Ca^{2+} imaging using fiber photometry also revealed an increased activity of NAcSh-projecting IL neurons when the subject mouse encountered a familiar mouse only in normal mice, but not in singly housed mice. Chemogenetic inhibition of NAcSh-projecting IL neurons caused a significant impairment in the social recognition while activation of such neuron in single isolated mice exhibited a rescued social recognition. This result suggests that early social experience of mice can have a lifelong-lasting impact on their social behavior, which is critically modulated by mPFC-NAc shell circuit.

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Poster

P687. Motivation: Social Communication and Behavior in Mice

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Topic: G.03. Motivation

Support: NIH Grant 1R01MH101377
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Title: Dopamine release in nucleus accumbens core during social behaviors in mice

Authors: *B. DAI[1], F. SUN[2], A. KUANG[1], X. TONG[1], Y. LI[2], D. LIN[1];
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Abstract: Social behaviors are among the most important and rewarding motivational behaviors. How dopamine, a “reward” signal, releases in the nucleus accumbens (NAc) during social behaviors has become a topic of interest for decades. However, due to limitations in temporal resolution, long-term stability and sensitivity of early recording methods, a complete understanding of moment-to-moment dopamine responses during social behaviors remains lacking. Here, we employ a genetically encoded dopamine sensor, $GRAB_{DA2h}$, to record dopamine activity in the NAc core in mice and find acute changes in extracellular dopamine

levels during all three phases of social behaviors: approach, investigation and consummation. We find that dopamine releases during approach and investigation correlate with animal's motivation and familiarity towards the social target respectively whereas its release during consummatory phase signals the valence of the experience. Negative and positive social experiences evokes opposite dopamine release patterns at both the onset and offset of the experience. Furthermore, dopamine release during sexual and aggressive behaviors are sexually dimorphic with an overall higher level in males than in females. Taken together, our results reveal complex information encoded by NAc dopamine activity during social behaviors and beyond.

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Poster

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Topic: G.03. Motivation

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Title: Role of parvalbumin-expressing neurons in the prelimbic cortex in social bonding

Authors: *A. PUśCIAN[1], A. BRYKSA[1], M. WINIARSKI[1], J. BOROWSKA[1], J. SADOWSKA[1], S. ŁęSKI[2], E. KNAPSKA[1];
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Abstract: Classifying individuals as members of an in-group, that is social circles with whom we identify, is a rapid and unconscious process. Consequently, very little is required to discriminate against the ones we identify as out-group, which happens on a similarly prompt and arbitrary basis, and oftentimes results in an unconscious bias. Arguably, it is because classifying individuals as either "us" or "them" is a hard-wired neuronal mechanism present in many mammalian species**.** Now more than ever, understanding brain mechanisms underlying the formation of attachment and resulting social bonding, seems crucial for our ability to address aggravating health and societal issues. As studies indicate a crucial role of the prefrontal cortex in social bonding, we studied the effects of the cell-specific inhibition in its prelimbic part (PL). To investigate various aspects of social behavior group-housed mice were tested in both, automated, ecologically relevant assays, which closely follow murine ethology, and standard

tests of social affiliation. Specifically, we used Eco-HAB, a system measuring spontaneous social interactions to assess sociability and exploration of social scents, and IntelliCage, a system measuring cognition to investigate animals' ability to socially learn from their conspecifics about aversive and appetitive stimuli. Additionally, standard social familiarity assays were used to assess the propensity to spend time with cagemates after separation. Using genetically modified mice and the Pharmacologically Selective Actuator Module-based chemogenetic approach, we performed time-constrained manipulation of the PV-expressing neurons in the PL and tested subjects' behavior. We show that activation of the PV-expressing neurons in the PL impairs animals' sociability expressed towards familiar conspecifics. Specifically, it reduces spontaneous social interactions with cagemates, diminishes animals' interest in familiar social olfactory stimuli, and decreases the propensity to spend time with a recently separated cagemate. On the other hand, such manipulation of neuronal activity does not influence the exploration of novel social stimuli. Moreover, it has no impact on social learning. Taken together, our findings indicate distinctive neural mechanisms influencing bonding with familiar conspecifics, but not other forms of social behavior. Our research reveals a specific role of the PV-expressing cells in the PL in the maintenance of the social structure and in sustaining ongoing interest in familiar conspecifics. Thus, presented data lays the foundation for understanding the neural underpinnings of social attachment.

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Poster

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Title: Opposite activity dynamics of two midbrain circuits encodes social novelty

Authors: *S. MOLAS, T. G. FREELS, R. ZHAO-SHEA, A. R. TAPPER;
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Abstract: Social investigations represent innate motivational drives critical for species survival and community wellbeing. Individuals engage social contacts to fulfill needs of internal states and these contacts are strongly associated with the overall salience of the social stimuli. For instance, novel (or unfamiliar) social cues are more attractive than familiar ones and lead to higher levels of arousal/attention. This enhanced response to social novelty is necessary to assess

the potential benefits of sociability, as well as its threatening aspects such as aggression. Social novelty responses are significantly disrupted in numerous neurodevelopmental and neuropsychiatric disorders including autism and schizophrenia, although to date, the neurobiological substrates underlying novel social investigations in a healthy brain, and how they are affected in neuropathological conditions remain unclear. Accumulating evidence indicates that dopaminergic (DAergic) midbrain circuits, including the mesolimbic pathway, promote exploratory assessment and guide approach/avoidance behavior to novel social stimuli. In addition, we previously demonstrated that the interpeduncular nucleus (IPN) of the midbrain represents a neuroanatomical substrate for familiarity signaling and the preference for novelty. The IPN is a major component of the habenulo-interpeduncular pathway that connects the limbic forebrain with midbrain/hindbrain areas, integrating cognitive with emotional and sensory processing. However, how DA responses habituate as novel stimuli become familiar and the interplay between DAergic ventral tegmental area (VTA) and IPN GABAergic neuron activity during these transitions is poorly understood. Here, we have implemented fiber photometry combined with biosensors to record activity dynamics from VTA DAergic neurons, IPN GABAergic neurons, and striatal DA release in awake behaving mice during social novelty tests and during novel social stimuli transitions to familiarity. Fiber photometry recordings reveal that activity dynamics of IPN GABAergic subpopulations inversely correlates with the DA response to novelty. In addition, we have mapped activity and functional connectivity of IPN GABAergic--hindbrain projecting neurons that convey social novelty information. The present study unravels a new circuitry within the novelty network that may be key in understanding disrupted social novelty response observed in numerous neurological disorders.

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Poster

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Topic: G.03. Motivation

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Title: Medial preoptic area mediates anxiety and parental behavior

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Abstract: Anxiety is a negative emotional state that is overly displayed in anxiety disorders and depression. Although anxiety is known to be controlled by distributed brain networks, key components for its initiation, maintenance and coordination with behavioral state remain poorly understood. Here, we report that anxiogenic stressors elicit acute and prolonged responses in glutamatergic neurons of the mouse medial preoptic area (mPOA). These neurons encode extremely negative valence and mediate the induction and expression of anxiety-like behaviors. Conversely, mPOA GABA-containing neurons encode positive valence and produce anxiolytic effects. Such opposing roles are mediated by competing local interactions and long-range projections of neurons to the periaqueductal gray. The two neuronal populations antagonistically regulate anxiety-like and parental behaviors: anxiety is reduced, while parenting is enhanced and vice versa. Thus, by evaluating negative and positive valences through distinct but interacting circuits, the mPOA coordinates emotional state and social behavior.

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Poster

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Title: Neural control of prosocial touch in comforting behavior

Authors: Y. WU, J. DANG, L. KINGSBURY, M. ZHANG, F. SUN, R. HU, *W. HONG; UCLA, Los Angeles, CA

Abstract: The ability to help and care for others fosters social cohesiveness and is vital to the physical and emotional well-being of social species including humans. Affiliative social touch, such as allogrooming, is a major type of prosocial behavior that provides comfort to others. It serves to establish and strengthen social bonds throughout a wide range of social species, such as birds, bats, rodents, canids, equids, and primates, and can help console distressed conspecifics.

However, neural circuits that promote prosocial touch have remained elusive. Here, we show that mice exhibit affiliative allogrooming behavior toward distressed partners, providing a consoling effect. This phenomenon occurs in response to different types of stressors and can be evoked by olfactory cues from distressed partners. We record neural activity of the medial amygdala (MeA) and find that single-neuron and ensemble activity in the MeA respond differentially to stressed vs. unstressed animals and encode allogrooming behavior. Through functional manipulations and intersectional genetic targeting, we establish a direct causal role of a select subpopulation of MeA neurons in controlling affiliative allogrooming. Our study uncovers a crucial node that controls affiliative social touch during prosocial interaction and opens up new avenues for interrogating the neural basis of prosocial behavior.

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Poster

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Title: Leptin enhances social motivation and reverses chronic stress-induced social anhedonia during adolescence

Authors: Y. LEI[1], D. WANG[2], Y. BAI[1], J. NOUGAISSE[1], N. L. WEINTRAUB[1], M. GUO[2], X.-Y. LU[1];
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Abstract: Social anhedonia, a loss of interest and pleasure in social interactions, is a common symptom of depression as well as other psychiatric disorders. Depression can occur at any age, but typically emerges in adolescence or early adulthood, which represents a sensitive period for social interaction that is vulnerable to stress. In this study, we evaluated social interaction reward using a conditioned place preference (CPP) paradigm in adolescent male and female mice. Adolescent mice of both sexes exhibited a preference for the social interaction-associated context. Chronic unpredictable stress (CUS) impaired the development of CPP for social interaction, mimicking social anhedonia in depressed adolescents. Conversely, administration of

leptin, an adipocyte-derived hormone with antidepressant-like properties, enhanced social interaction-induced CPP in non-stressed control mice and reversed social anhedonia in CUS mice. By dissecting the motivational processes of social CPP into social approach and isolation avoidance components, we demonstrated that leptin treatment increased isolation aversion without an overt social reward effect. Further mechanistic exploration revealed that leptin stimulated oxytocin transcription in the paraventricular nucleus of the hypothalamus, while blockade of oxytocin receptors abolished the leptin enhancement of socially-induced CPP. These results establish a behavioral model to study social anhedonia in adolescent mice and provide evidence that leptin's effects on social motivation are mediated by oxytocin.

Disclosures: Y. Lei: None. **D. Wang:** None. **Y. Bai:** None. **J. Nougaisse:** None. **N.L. Weintraub:** None. **M. Guo:** None. **X. Lu:** None.

Poster

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Title: Evaluation of voluntary physical exercise as a strategy to reverse the cognitive impairments induced by high-fat diet

Authors: *S. HERNÁNDEZ-RAMÍREZ[1], P. SALCEDO-TELLO[1], D. OSORIO-GÓMEZ[2], B. BERMÚDEZ-RATTONI[2], G. PACHECO-LÓPEZ[1], G. FERREIRA[3], P. LAFENETRE[3], K. R. GUZMÁN-RAMOS[1];
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Abstract: The worldwide obesity rate in adolescents has been on the rise in recent years. Previous research has reported that the consumption of high calorie diets can affect the proper development of structures in the brain during adolescence. As a result, cognitive alterations, mood, personality and eating disorders can manifest. On the other hand, it has been widely established that physical exercise is beneficial for various cognitive functions and has a positive

influence on certain social behaviors under stress conditions, which could counteract the effect of high-fat diets (HFD). In this study, we evaluated the effect of voluntary physical exercise in c57BL6 mice exposed to a HFD (consisting of 45% of fat) for a period of 24 weeks, a running wheel was added after 3 months of diet exposure. Four groups of animals were used: normal chow (NC), normal chow + running wheel (NC-R), high-fat diet (HFD) and high-fat diet + running wheel (HFD-R). After 24 weeks of diet exposure, behavioral tasks were evaluated: memory recognition (Novel Object Recognition Task [NOR]), spatial memory (object location memory [OLM]) and social memory (three-chamber test [3CT]). Glucose tolerance test and the body weight of all groups of animals were measured.We observed that HFD animals are not able to recognize between two objects previously presented, and we observed an improvement in the recognition index of HFD-R animals when we compare normal and treated groups. Results of OLM test indicated an impairment in the spatial memory on HFD fed animals which could not discriminate between the displaced and non-displaced object. However physical activity improved this memory in the HFD-R group.. 3CT test showed and impairment in the social memory in the HFD animals, that could not recognize the conspecific that were in contact with previously. Voluntary physical activity also improves recognition of a previously known conspecific. These results suggest a palliative effect of exercise when HFD exposure has affected brain function, restoring both metabolic profile and cognitive performance even after a long exposure started at weaning.

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Poster

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Topic: G.03. Motivation

Support: F31 1F1MH122094-01A1)

Title: Altered social behavior following conditional genetic deletion of the mu opioid receptor (Oprm1) from nucleus accumbens neurons

Authors: *C. TODDES[1], C. RETZLAFF[1], E. LEFEVRE[1], B. L. KIEFFER[2], P. E. ROTHWELL[1];
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Abstract: The mu opioid receptor regulates naturally rewarding behaviors, including social interaction and drug reward. The constituitive mu opioid receptor knockout mouse has been shown to display impaired reciprocal social interaction and social reward alongside an attenuation in response to the rewarding aspects of opioid drugs. Our group recently identified changes within the microcircuit of the nucleus accumbens following constitutive mu opioid receptor deletion that specifically affected perisomatic inhibitory connections onto D2 receptor expressing medium spiny neurons (Toddes et al., *J Neurosci*, in press). The current study sought to expand upon these findings by conditionally deleting mu opioid receptors from targeted cell types within the nucleus accumbens microcircuit and evaluating the resultant effect upon social related behaviors. We deleted mu opioid receptors from cells within the nucleus accumbens by injecting AAVdj-hsyn-cre-GFP, AAV2retro-hSyn-Cre-P2A-tdTomato, or AAV8-mdlx-icre-tdTomato into equal numbers of male and female mice with a floxed OPRM1 gene, or by crossing our floxed mouse line with a line expressing Cre in either the D1 receptor expressing or D2 receptor expressing medium spiny neurons. We found a significant attenuation in reciprocal social behaviors of male and female mice injected with either local or retro-Cre and thus focused our attention on neurons specifically within the nucleus accumbens. Interestingly, mice lacking mu opioid receptors in either D1- or D2-medium spiny neurons displayed no changes in reciprocal social behaviors. However, male and female mice with a conditional deletion of mu opioid receptors from only GABAergic interneurons (AAV8-mdlx-icre-tdTomato) demonstrated a robust attenuation of reciprocal social interactions that recapitulated the phenotype seen in the constitutive knockout model. Responses to the analgesic, hyperlocomotive, and rewarding properties of opioid drugs were also tested in these mouse models with a change in response only demonstrated in mice lacking mu opioid receptors on both D1- and D2-medium spiny neurons where there was a significant attenuation of hyperlocomotion. These results indicate that inhibitory interneurons within the nucleus accumbens may be a key target for mu opioid receptor modulation of social interaction in male and female mice.

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Poster

P691. Human Emotion II

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Program #/Poster #: P691.01

Topic: G.04. Emotion

Title: An fMRI study of grandmaternal neural responses to grandchild stimuli

Authors: *J. RILLING, A. GONZALEZ, M. LEE;
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Abstract: In many societies, grandmothers are important caregivers, and grandmaternal investment is often associated with improved grandchild well-being. Here, we present the first study to examine grandmaternal brain function. We recruited 50 grandmothers with at least one biological grandchild between 3 and 12 years old. Brain function was measured with fMRI as grandmothers viewed pictures of their grandchild, an unknown child, the same-sex parent of the grandchild, and an unknown adult. Grandmothers also completed questionnaires to measure their degree of involvement with and attachment to their grandchild. After controlling for age and familiarity, viewing grandchild pictures activated areas involved with emotional empathy (insula and secondary somatosensory cortex) and movement (motor cortex and supplementary motor area). Grandmothers who more strongly activated areas involved with cognitive empathy (temporo-parietal junction and dorsomedial prefrontal cortex) when viewing pictures of the grandchild desired greater involvement in caring for the grandchild. Finally, compared with results from an earlier study of fathers, grandmothers more strongly activated regions involved with emotional empathy (dorsal anterior cingulate cortex, insula, and secondary somatosensory cortex), and motivation (NAc, ventral pallidum and caudate nucleus). All in all, our findings suggest that emotional empathy may be a key component of grandmaternal responses to their grandchildren.

Disclosures: J. Rilling: None. **A. Gonzalez:** None. **M. Lee:** None.

Poster

P691. Human Emotion II

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Topic: G.04. Emotion

Support: MOST 108-2410-H-007-044
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Title: Sex differences in the creative brain

Authors: *J.-Y. YANG, Y.-C. CHAN;
Natl. Tsing Hua Univ., Hsinchu City, Taiwan

Abstract: Creative thought relies on an associative mode of processing, which involves novelty and usefulness. Executive control network (ECN) and default-mode network (DMN) are involved in creative processes when generating and evaluating ideas (Beaty et al., 2015; Kleinmintz et al., 2019). The present study employed a fMRI paradigm to explore sex differences in appreciating alternative uses production. Forty healthy participants (20 males and 20 females) were right-handed and no history of neurological or psychiatric disease. The stimuli were presented across conditions for the 32 creative objects and 32 common objects (baselines).

The present study used a 2 × 2 mixed factorial design with sex (males and females) and stimulus type (creativity and common) as factors. Imaging was performed using a 3 T Siemens Magnetom Prisma scanner using a standard 20-channel head coil at the Imaging Center for Integrated Body, Mind and Culture. The fMRI images were pre-processed and analyzed using Statistical Parametric Mapping (SPM12) implemented in Matlab. A flexible factorial analysis was performed for brain activations. Creative appreciation elicited greater activation in males than females in the left precuneus, left middle frontal gyrus (MFG), left middle temporal gyrus (MTG), and right posterior cingulate gyrus (PCC), while females showed greater activation than males in the left midbrain, left MTG, right superior frontal gyrus (SFG), right temporoparietal junction (TPJ), left dorsal anterior cingulate gyrus, right precuneus, left PCC, left MFG, right insula, and left inferior parietal lobule (IPL) associated creative appreciation. These finding provide the mechanisms underlying the processing of creative appreciation for the sex differences in cognitive control, attention, evaluative processes, and feelings of pleasure.

Disclosures: J. Yang: None. **Y. Chan:** None.

Poster

P691. Human Emotion II

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Program #/Poster #: P691.03

Topic: G.04. Emotion

Title: Idiosyncratic emotional responses evoked by music

Authors: S. SPIVACK[1], A. POINCOT[2], G. IENNER[2], *P. WALLISCH[1];
[1]Data Sci., [2]Psychology, New York Univ., New York, NY

Abstract: We are interested in emotional responses evoked by music. Specifically, we wondered whether these responses are idiosyncratic or whether they can be predicted by the sonic qualities of the music. To investigate this question, we exposed a large sample of research participants to a diverse musical selection and recorded their emotional responses as well as subjective appraisal and arousal. We then related these emotional responses to the sonic characteristics of the music as well as to each other, between participants. We found that music evokes strong emotional responses, but that appraisal and emotional responses are highly idiosyncratic and not well determined by sonic properties of the music, in contrast to arousal. We model these responses and conclude that personal features of the individual are reflected in their “emotional fingerprint” in response to music.

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Poster

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Topic: G.04. Emotion

Support: NSF grant#1020906

Title: How do I love thee?: Using psychophysiology to validate a relationship satisfaction theory.

Authors: *E. LEAVER[1], D. PHILLIPS[2], A. MIANO-BURKHARDT[1], S. CURTIS[1], M. P. W. DONELLY[3], S. FURMAN[1];
[1]Salisbury Univ., Salisbury, MD; [2]Psychology, Univ. of South Carolina Aiken, Aiken, SC; [3]Menomie, Menomie, WI

Abstract: This is the first study that has used psychophysiological measures to investigate the validity of Gary Chapman's (1992) relationship satisfaction theory, *The Five Love Languages.* This theory proposes that individuals have a preferred means of communicating love. Heart rate, skin conductance, and respiration rate were recorded while 89 college students listened to guided imagery scripts representing one of the five love languages (LL). Using a within-subjects design, a repeated-measures ANOVA was run on each physiological measure. A composite score of the physiological responses indicated a significant increase in arousal for when participants listened to their primary LL as compared to their non-preferred LL ($t(88)=-2.3$, $p=.02$, $d=.37$). For skin conductance ($t(88)=-2.03$, $p<.05$, $d=.35$) and heart rate ($t(88)=-2.15$, $p=.03$, $d=.35$), we observed a significant difference in physiological response found for the preferred versus non-preferred LL. This study gives the first evidence that Chapman's five love languages may be reflected in human physiology, suggesting some degree of construct validity for this relationship typology theory. Interestingly, whether an individual received their preferred love language from their significant other or not had no impact on their relationship satisfaction nor their commitment. Unresolved questions include the nature of the relationship between LLs, relationship satisfaction, and commitment.

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Poster

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Wharton Behavioral Lab

Title: Subjective Well-Being Predicts Foraging Behavior in Humans

Authors: *V. U. LUDWIG[1,2], V. GUPTA[6], D. L. CRONE[3], A. RAMAKRISHNAN[6], M. L. PLATT[1,2,4,5];
[1]Dept. of Neurosci., [2]Wharton Neurosci. Initiative, [3]Positive Psychology Ctr., [4]Marketing Department, Wharton Sch. of Business, [5]Dept. of Psychology, Sch. of Arts and Sci., Univ. of Pennsylvania, Philadelphia, PA; [6]Indian Inst. of Technol., Kanpur, India

Abstract: Foraging is a fundamental behavioral challenge all brains evolved to solve. The marginal value theorem (MVT) specifies the optimal solution to determining when to abandon a depleting resource and search for a new one: leave the current patch when the instantaneous return rate falls below the environmental average. Although all animals tested thus far conform on average to the MVT, systematic differences are observed across species adapted to rich or poor environments as well as in mental health disorders including anxiety (Ramakrishnan et al., SFN 2018, 2019), problem gambling (Addicott et al. 2015), and Parkinson's disease (Rutledge et al., 2009). These observations suggest that conditions characterized by altered patterns of decision making may reflect systematic variation in the function of circuits that collectively carry out MVT computations.

Although variation in foraging behavior, as well as its underlying neural mechanisms, have been clearly linked with mental health disorders, we know little about how they vary with positive mental states such as well-being. Here we tested whether individual differences in satisfaction with life (SWL) and positive emotion (PE) are linked to individual variation in foraging behavior. Participants played a virtual foraging game in which they collected berries, which were redeemable for money at the end of the game. Harvest rates per patch declined over time within a patch, and participants could choose to travel to a new patch at any time. The time needed to travel between patches varied across blocks (3 or 10 sec). Participants also completed a series of questionnaires, including Diener's SWL Scale and the PE subscale of the PERMA-profiler by Butler and Kern (2015).

Study 1 tested a cross-cultural sample (N = 416; 214 in India, 42 female; 202 in the US, 85 female) using the MTurk online platform. Study 2 was pre-registered and tested a representative US sample using the Prolific online platform (N = 488, 247 female).

We used binomial regression to model trial-by-trial choices and found significant effects of SWL and PE. Participants scoring high in SWL, as well as PE, persisted significantly longer in patches

than participants scoring low in SWL or PE. One potential explanation for these differences is that people who have high well-being experience higher subjective reward rates within patches thus diminishing motivation to explore. These findings connect well-being to well-described, mathematically formalized mechanisms regulating the trade-off between exploitation and exploration, and suggest people manage these decisions along a computational continuum systematically set by both internal and external conditions.

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Poster

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Title: Anger, Exuberance and Incentive Reward

Authors: *T. L. WHITE[1], M. A. GONSALVES[2], C. ZIMMERMAN[2], H. JOYCE[3]; [1]Behavioral and Social Sci., [2]Neurosci. Grad. Program, [3]Undergraduate Program in Cognitive Neurosci., Brown Univ., Providence, RI

Abstract: Anger is a powerful, negatively valenced emotion with significant impact on human health and well-being. As such, there is potentially great value in better understanding the behavioral phenotypes associated with anger and their underlying neural substrates. We present a novel incentive theory of anger derived from assessments of emotion, personality, task-based functional magnetic resonance imaging (fMRI), and pharmacological challenge in healthy young adults. Two experiments were conducted to evaluate tiered hypothesis regarding the relationship between anger, exuberance, reward and approach. Experiment I found self-reports of anger to frustrative non-reward and exuberance to success events rose jointly in an incentive challenge task (*N*=39). Anger and exuberance responses were also positively associated with scores on the personality trait of social potency (SP), a measure of agentic extraversion that relates to the sensitivity to reward. Experiment II used task-based fMRI and found anger was positively related to (a) task-induced exuberance, (b) trait SP and (c) fMRI BOLD response in right nucleus

accumbens to *d*-amphetamine, a psychostimulant with dopaminergic and neurometabolic effects (*N*=10). Our findings are consistent with mechanistic contributions of dopamine to anger, indicating anger is an incentive state that facilitates the approach and acquisition of rewards that entail risk. Implications for neural mechanisms of anger, exuberance and its relevance to reward-motivated action and risk-taking are discussed.

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Title: Emergence of emotion selectivity in deep neural networks trained to recognize visual objects

Authors: P. LIU[1], K. BO[3], L. CUI[1], Y. CHEN[1], C. T. TRAN[2], M. DING[1], ***R. FANG**[1]; [1]Dept. of Biomed. Engin., [2]Dept. of Electrical and Computer Engin., Univ. of Florida, Gainesville, FL; [3]Dept. of Physiol. and Brain Sci., Dartmouth Col., Hanover, NH

Abstract: Affective images with pleasant, unpleasant, and neutral content are differentially represented in the human visual cortex. How these emotion-specific neural representations arise is debated. Here we show that emotion selectivity can arise spontaneously in a deep neural network (DNN) model of the visual cortex trained to recognize visual objects. Adopting a VGG16 network pre-trained on the ImageNet data, the 1000 neurons in the last object recognition layer were replaced by 2 neurons, which were trained to perform binary emotion recognition: pleasant versus non-pleasant, neutral versus non-neutral, or unpleasant versus non-unpleasant. Affective images from two datasets: the International Affective Picture System (IAPS) and the Nencki Affective Picture System (NAPS) were used for training and testing. The emotion selectivity of each neuron was defined according to the largest normalized mean response among the three emotion types. The functionality of these neurons was tested using lesion and attention enhancement methods. The results showed that (1) in all layers of the DNN model, there were artificial neurons that responded consistently and selectively to neutral, pleasant, or unpleasant images, with later layers showing higher selectivity than earlier layers

and (2) lesioning the emotion selective neurons leads to decrease in emotion recognition performance whereas implementing feature attention in these neurons leads to increase in emotion recognition performance. These results suggest that emotion selectivity can arise intrinsically in the visual cortex and the functional importance of emotion selective neurons in emotion perception.

Disclosures: P. Liu: None. **K. Bo:** None. **L. Cui:** None. **Y. Chen:** None. **C.T. Tran:** None. **M. Ding:** None. **R. Fang:** None.

Poster

P691. Human Emotion II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P691.08

Topic: G.04. Emotion

Support: NIMH Grant ZIA MH002588-26

Title: Taste and emotion semantics are linked by the shared attribute of valence across human languages

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Abstract: ‘Parting is such sweet sorrow’. Sensory metaphors, particularly from the sense of taste, provide a rich vocabulary for describing emotional experience. This may be partly due to the common neural substrates shared by taste and emotion processing, such as the insular cortex and amygdala. With such a close functional connection in the human brain, we might expect that these cognitive domains are tightly linked within the conceptual structure of human thought. One way we can explore this is by examining the semantic similarity of concepts revealed in the structure of language. To do this, we used a behavioral ‘odd-one-out’ task (Hebart et al., 2020) in an online sample of 1059 participants on Amazon Mechanical Turk whom we asked to compare concepts related to emotion, taste, and color. Using the data from this task, we generated a semantic similarity matrix of these concepts. We then compared the similarity of emotion and taste concepts to the similarity of emotion and color concepts, another rich source of sensory metaphors. Consistent with our brain-based prediction, we found that the semantic similarity of emotion and taste concepts was greater than the similarity of emotion and color concepts ($t = 7.0$, $p < 0.001$). Importantly, the similarity of taste and emotion concepts was related to the similarity in hedonic valence between these concepts ($r = 0.55$, $p < 0.001$), mirroring a recent finding about valence as a source of universal structure of emotion semantics (Jackson et al., *Science*, 2019). As in this previous study, we also looked at patterns of colexification - where semantically related concepts are named with the same word - using a concept database containing cross-

linguistic polysemies aggregated from over 2400 languages (Rzymski et al, *Scientific Data*, 2020). Interestingly, we also observed that the similarity of colexification patterns between taste and emotion concepts across world languages was significantly greater, and thus less variable, than the colexification patterns within emotion concepts themselves ($t = 4.9$, $p < 0.001$). These findings suggest that, across human cultures, we use hedonic valence as a way of bridging the divide between concepts about our shared sensory environment and concepts which relate to our internal emotional experience.

Disclosures: J.A. Avery: None. **A.G. Liu:** None. **M. Carrington:** None. **A. Martin:** None.

Poster

P691. Human Emotion II

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Program #/Poster #: P691.09

Topic: G.04. Emotion

Support: ERC 295673
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ERC-synergy 856495
MBIC funding

Title: Interaction of the semantic and action observation networks in the brain underlies subjective understanding of actions and emotions

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Abstract: Studies on the brain basis of action and emotion perception use predefined categories. How we subjectively generate an understanding of other people's bodily actions and emotions is not well understood. In this 7T fMRI study with a large stimulus set, alongside conventional univariate/multivariate analyses with predefined categories, we examined the representational geometry of bodily action- and emotion-understanding by mapping individual subjective reports with word embeddings. For the behavioral reports, dimensionality reduction (principle component analysis) revealed that the representations for perceived action and emotion were high dimensional, each correlated to but were not reducible to the predefined action and emotion categories. For brain activity, searchlight representational similarity analysis showed that the left middle superior temporal sulcus and left dorsal premotor cortex showed systematic correspondence with the subjective action and emotion representations. Furthermore, using task-residual functional connectivity and hierarchical clustering, we found that areas in the action observation network and the semantic/default-mode network were functionally connected to

these two seed regions and showed similar representations. Our study illustrates the feasibility of subjective report analyses and provides direct evidence that both networks were concurrently involved in subjective action and emotion understanding.

Disclosures: M. Zhan: None. **R. Goebel:** None. **B. de Gelder:** None.

Poster

P691. Human Emotion II

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Program #/Poster #: P691.10

Topic: G.04. Emotion

Support: NIH Grant MH112558

Title: Effects of Emotional Distractors on the Processing of Motion Stimuli

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Abstract: Images containing affective scenes are thought to be highly potent distractors. We tested the behavioral and neural effects of three categories of affective images from the International Affective Picture System (IAPS): pleasant (erotic couples), neutral (workplace people), and unpleasant (bodily mutilations) on task-relevant visual processing. Functional magnetic resonance imaging (fMRI) were recorded while participants detected instances of coherent motion in a random dot kinematogram (primary task) overlayed on IAPS pictures (distractors). Applying support vector machines to multivariate BOLD response patterns in the ventral visual cortex (VVC) and MT we found the following results. First, the behavioral accuracy on the primary task was not significantly different whether the distracting scenes were pleasant, neutral, or unpleasant, all being ~60%. Second, in VVC and MT, the decoding accuracy of pleasant vs neutral scenes and unpleasant vs neutral scenes were significantly higher than chance level, suggesting that distractor information was represented in the motion processing regions of the brain. Third, the aforementioned decoding accuracy and the behavioral accuracy on the primary task were significantly negatively correlated, suggesting that stronger neural representations of distractor information are more detrimental for task-relevant visual processing. These results reveal that valence is a not crucial factor in determining distractor potency and the competition between the primary task and distractors underlies the behavioral outcome on the primary task.

Disclosures: C. Xiong: None. **K. Bo:** None. **N.M. Petro:** None. **A. Keil:** None. **M. Ding:** None.

Poster

P712. Human Studies and Therapeutic Approaches: Focus on PTSD

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Program #/Poster #: P712.01

Topic: G.07. Post-Traumatic Stress Disorder

Support: VHA1I21RX002588

Title: Classification of Self-Reported Stress Levels from fMRI data in Veterans with Posttraumatic Stress Disorder

Authors: *T. TSE[1], R. GOEL[2], B. NAYLOR[3], L. SMITH[5,6], D. RESS[3], W. WILLIAMS[6], R. SALAS[3,4,6,7];
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Abstract: *Objective:* Posttraumatic stress disorder (PTSD) is a significant burden among combat Veterans returning from active warzones in Iraq and Afghanistan. While empirically supported treatments such as exposure therapy have demonstrated reductions in PTSD symptomatology, many Veterans still meet clinical definitions of PTSD after treatment and there remains a need to improve treatment effectiveness. Real-time functional magnetic resonance imaging (fMRI) neurofeedback has emerged as an innovative treatment to ameliorate PTSD symptom severity. Virtual reality (VR) approaches have also shown promise as a means of exposing patients with trauma-related events to controlled stimuli, thus hopefully increasing desensitization, treatment compliance, and outcomes. To facilitate fMRI neurofeedback-associated therapies, it would be advantageous to be able to accurately and quickly classify internal brain stress levels while Veterans are exposed to trauma-associated VR imagery. *Methods:* Initially, male combat Veterans with PTSD symptoms (N = 8) were screened to ascertain their stress responses to various video stimuli, enabling design of audiovisual stimuli designed to evoke variable amounts of stress; stimuli were created using a VR tool called “BraveMind” that was specifically designed to provide VR exposure therapy to Veterans. Next, we use two sessions of fMRI to collect neural responses to the VR stimuli. Veterans reported their self-perceived stress level on a scale from 1-8 every 15 seconds throughout the fMRI sessions. In our proposed framework, we precisely sample the fMRI data on cortical gray matter at the 2x2x2-mm fMRI spatial resolution, blurring the data along the gray-matter manifold to reduce noise and dimensionality while preserving maximum information. Then, three machine learning (ML) algorithms were independently applied to these fMRI data collected across two sessions, separately for each Veteran, to build individualized ML models that predicted their internal brain states (self-reported stress responses). *Results:* We were able to classify the 8-class self-reported stress responses accurately with a mean (± standard error) root mean square error of 0.6 (± 0.1) across

all Veterans using the best ML approach. *Conclusion:* The findings demonstrate the predictive ability of ML algorithms applied to cortical fMRI data collected during individual Veteran sessions. *Significance:* The framework we have developed to pre-process cortical fMRI data and train ML models across sessions should provide a valuable tool to enable individualized real-time fMRI neurofeedback during VR exposure therapy for PTSD.

Disclosures: T. Tse: None. **R. Goel:** None. **B. Naylor:** None. **L. Smith:** None. **D. Ress:** None. **W. Williams:** None. **R. Salas:** None.

Poster

P712. Human Studies and Therapeutic Approaches: Focus on PTSD

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Program #/Poster #: P712.02

Topic: G.07. Post-Traumatic Stress Disorder

Support: Department of Veterans Affairs: CX000715-04

Title: Resting state functional connectivity predicts postconcussive symptoms in trauma-exposed veterans

Authors: *M. MARVIN[1], J. BOMYEA[1,2,3], A. SPADONI[1,2,3], A. SIMMONS[1,2,3];
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Abstract: Post-Traumatic Stress Disorder (PTSD), mild traumatic brain injury (mTBI), and depression are commonly diagnosed in veterans exposed to combat stress. These conditions are difficult to diagnose accurately due to the overlap in diagnostic criteria. Previous neuroimaging research suggests there may be overlapping and distinct functional connectivity which may aid in distinguishing PTSD, mTBI, and depression. The present study utilized a data driven approach to investigate a longitudinal dataset of combat veterans' resting state functional connectivity and its utility as a predictor of clinical outcomes. Seventy-six Veterans with combat-related trauma exposure (mean age = 33.7, SD = 7.6) completed questionnaires and a resting state functional magnetic resonance imaging (fMRI) scan at three time points over 18 months. Participants completed the self-report PTSD checklist (PCL), Neurobehavioral Symptom Inventory (NSI), and Patient Health Questionnaire-9 (PHQ-9), to assess PTSD, postconcussive, and depression symptom severity, respectively. Based on a recent meta-analysis of areas often associated with PTSD, twelve regions of interest (ROIs) were defined to be used in the Subgroup Group Iterative Multiple Model Estimation (S-GIMME) algorithm. S-GIMME is a data driven approach that estimates the directed functional connectivity of fMRI BOLD signal between predefined brain ROIs at the individual, group, and subgroup level. The unsupervised S-GIMME produced two subgroups. Subgroup 1 (n-48) had fewer connections, including the right insula to right medal

prefrontal cortex and right inferior frontal gyrus to left supramarginal gyrus. Subgroup 2 (n=28) had connections between the left supramarginal to right IFG and right insula, right Heschl's gyrus to right caudate and right calcarine gyrus, right fusiform gyrus to left supramarginal gyrus, right calcarine gyrus, and right Heschl's gyrus. Associations between the neural subgroups and symptom trajectories revealed no associations between subgroup and PTSD or depression severity, but there was a significant association between subgroup and postconcussive symptom severity over time. Subgroup 2's postconcussive symptoms increased over time whereas Subgroup 1's symptoms began higher but were relatively stable. Results indicate that S-GIMME could effectively identify data-driven subgroups of veterans with heterogeneous networks of functional connectivity during a resting state fMRI scan. The brain-derived patterns and subgroup membership may be predictive of the clinical course of mTBI and indicative of the mechanisms that underlie postconcussive symptoms.

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Poster

P712. Human Studies and Therapeutic Approaches: Focus on PTSD

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Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P712.03

Topic: G.07. Post-Traumatic Stress Disorder

Title: Ayahuasca-based psychedelic therapy for PTSD and its effects on the microbiome: a discovery cohort

Authors: *L. R. KESSLER[1], L. M. DAWUD[1], J. D. STERRETT[1], A. I. ELSAYED[1], B. WEISS[2], J. GOULD[3], K. PATE[3], C. A. LOWRY[1];
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Abstract: Posttraumatic stress disorder (PTSD) is a mental health condition triggered by a traumatic event, associated with symptoms such as flashbacks, nightmares, depression, and anxiety, which can persist for years after the trauma experience. Although prolonged exposure therapy is an effective first-line treatment for PTSD, approximately 20-30% of individuals who complete treatment continue to have a PTSD diagnosis. An even smaller number of individuals respond to first-line pharmacological treatments, such as selective serotonin reuptake inhibitors (SSRIs), highlighting a need for novel interventions. Recent studies have evaluated the therapeutic potential of psychedelics for treatment of various mental health conditions. In this study, we investigated how the ceremonial use of ayahuasca to treat PTSD in United States Veterans can affect the microbiome. Ayahuasca is a decoction of plants such as *Psychotria viridis* which contains *N,N*-dimethyltryptamine (DMT), and the *Banisteropsis caapi* ayahuasca vine, which contains monoamine oxidase inhibitors (MAOIs) required for DMT to be orally

active. This brew has a long history of ceremonial use among indigenous peoples of Brazil and the Amazonian basin of South America. Participants of the current study included 14 US Veterans, 12 male, who attended one of two different week-long retreats where they participated in traditional ayahuasca ceremonies. While previous analysis of psychological survey data from this cohort revealed a healing experience among participants, little is known of the effects on the microbiome. This is of particular interest since the microbiome has been implicated in the etiology of other mental health conditions such as depression and anxiety. Additionally, ingestion of the brew is known to cause emesis and other gastric distress during the ceremony, a potential cause of instability in the host microbiome. Fecal samples were collected from participants at two timepoints, before and after ayahuasca-based treatment. Two retreats (n=9, n=5) were processed using 16S rRNA gene sequencing. Changes in personality traits conceived by the Five-Factor Model (FFM) were observed, with a significant decrease in neuroticism (a trait characterized by emotional instability in the FFM). Analysis revealed a trend toward lower alpha diversity in post-treatment samples. Overall, while there was a decrease in neuroticism, we did not detect changes in diversity of the microbiome in this cohort.

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Poster

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Program #/Poster #: P712.04

Topic: G.07. Post-Traumatic Stress Disorder

Support: R01MH103287-01

Title: Resting-state functional connectivity predicts posttraumatic stress disorder symptoms in recent trauma survivors.

Authors: *Y. LOKSHINA[1,2], J. SHEYNIN[2], T. NICKELSEN[2], S. AHRARI[2], E. R. DUVAL[3], M. ANGSTAD[3], J. N. KEYNAN[4,5], Z. BEN-ZION[4,5], R. ADMON[8], M. HALEVI[6,4], N. GREEN[6,4], T. HENDLER[6,5,4,7], A. Y. SHALEV[9], I. LIBERZON[1,2];
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Abstract: Patterns of resting-state functional connectivity (rsFC) following a traumatic event might reflect underlying neurobiological processes that play a role in the pathogenesis of and recovery from posttraumatic stress disorder (PTSD). In the current study, we examined rsFC in recent trauma survivors who either developed sustained PTSD symptomology or showed recovery from early acute stress symptoms. Specifically, we tested the association between rsFC and PTSD symptoms (both absolute severity and relative change in severity) at one, six and 14 months following trauma exposure (T1, T2 and T3, respectively). Functional MRI (fMRI) and clinical data were collected from n=158 adult civilians recruited from the emergency department of Tel-Aviv Sourasky Medical Center at T1 (50% male; mean age = 33.5 years), n=119 at T2 (47.05% male; mean age = 34.6 years) and n=109 at T3 (44.9% male; mean age = 34.4 years). PTSD severity was assessed by Clinician-Administered PTSD Scale for DSM-5 (CAPS-5) and intrinsic connectivity during a resting-state scan was examined using a seed-based whole-brain approach. We focused on seed regions of Default-Mode Network (DMN; hippocampus, ventromedial prefrontal cortex and posterior cingulate cortex) and Salience Network (SN; anterior insula, amygdala and dorsal anterior cingulate (dACC)), which have been reported to be altered in PTSD. Results showed that functional connectivities between insula and hippocampus, between insula and temporal gyrus, as well as between amygdala and inferior frontal gyrus at T1 were associated with PTSD symptoms at T3. We also found that connectivities between amygdala and insula, between hippocampus and insula and between hippocampus and dACC at T1 were associated with change in symptoms from T1 to T3. Lastly, connectivity between insula and dACC at T3 was associated with symptom severity at T3 (all $p < .050$, FWE corrected). Overall, this study suggests that specific connectivity patterns between SN and DMN, within SN and between SN and attention networks early after trauma, could potentially be used as biomarkers to predict PTSD symptom severity and recovery following a traumatic event. (YL and JS contributed equally to this work.)

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Poster

P712. Human Studies and Therapeutic Approaches: Focus on PTSD

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Program #/Poster #: P712.05

Topic: G.07. Post-Traumatic Stress Disorder

Support: US Department of Defense grant W81XWH-15-1-0645 to NGS
Azevan Pharmaceuticals, Inc.
NIH UL1TR002384

Title: A proof-of-concept randomized crossover clinical trial of a first-in-class vasopressin 1a receptor antagonist for PTSD

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Abstract: About 8 million Americans suffer from PTSD annually and the lifetime prevalence of the disorder is 8%. Drug therapies for PTSD are repurposed antidepressants and anxiolytics, which are minimally effective and produce undesirable side effects. The CNS vasopressin system, through the V1a receptor subtype, represents a novel target for pharmacological intervention. Preclinical imaging studies in a conditioned fear paradigm in rats showed that treatment with a V1a antagonist blocked hyperarousal to the memory of fear. In a human experimental medicine imaging study, V1a antagonist treatment significantly and selectively reduced BOLD signal in fear/threat response circuitry. We tested the primary hypothesis that treatment with a V1a antagonist, SRX246 (from Azevan Pharmaceuticals, Inc.), would result in a clinically meaningful 10-point reduction in mean CAPS score compared to placebo. Secondary hypotheses included testing for tolerability and safety as well as changes in other well-known scales (e.g., PTSD Symptom Checklist, Beck Depression Inventory-II, Pittsburgh Sleep Quality Index, Sheehan Disability Scale). The study was approved by the Weill Cornell IRB and the DoD Human Research Protection Office. Participation was open to 18-65 year-olds with a primary diagnosis of PTSD as defined by DSM-V for at least 3 months and a minimum CAPS-5 score of 32. All participants received SRX246 (160 mg BID) for 8 weeks and placebo for 8 weeks in an 18-week proof-of-concept randomized crossover clinical trial. Participants were reassessed every 2 weeks. Two hundred sixty-three subjects were screened, 89 were consented and assessed, and 33 were randomized. Substance abuse was the primary rule-out criterion during the baseline assessment. Randomization was balanced by sex and comorbid major depression. The primary outcome, CAPS-5 Severity, decreased significantly over the course of the study regardless of treatment order. Although the mixed effects model did not show significant treatment differences, examination of effect sizes adjusted by covariates suggested superiority for SRX246, with an effect about 30% higher than placebo. SRX246 was safe and well tolerated; the type and number of AEs did not differ from placebo; there were no SAEs. Regarding secondary endpoint scales, the PSQI findings suggest that SRX246 may show some stabilizing effects on sleep quality; no other effects or trends were found. These preliminary findings suggest SRX246 may be an effective treatment for PTSD. If confirmed through additional clinical studies, SRX246 would represent a breakthrough in pharmacotherapy for PTSD.

Disclosures: J. Difede: B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Azevan

Pharmaceuticals, Inc.. **L. Spielman:** None. **A. McAleavey:** None. **M. Emerich:** None. **K. Wyka:** None. **J. Kocsis:** None. **M. Olden:** None. **A. JIck:** None. **J. Gordon-Eliot:** None. **D. Itzkowitz:** A. Employment/Salary (full or part-time):; Azevan Pharmaceuticals, Inc. **S. Lu:** A. Employment/Salary (full or part-time):; Azevan Pharmaceuticals, Inc. **E. Damiano:** A. Employment/Salary (full or part-time):; Azevan Pharmaceuticals, Inc. **M. Brownstein:** A. Employment/Salary (full or part-time):; Azevan Pharmaceuticals, Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Azevan Pharmaceuticals, Inc. **N. Simon:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Azevan Pharmaceuticals, Inc. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Azevan Pharmaceuticals, Inc. F. Consulting Fees (e.g., advisory boards); Azevan Pharmaceuticals, Inc..

Poster

P712. Human Studies and Therapeutic Approaches: Focus on PTSD

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P712.06

Topic: G.07. Post-Traumatic Stress Disorder

Title: Dorsolateral Prefrontal Cortex Interneuron Deficits in PTSD

Authors: *K. A. YOUNG[1,2,3], M. J. GIRGENTI[4], H. ZHAO[5], D. JI[6], D. A. CRUZ[7,8], B. R. HUBER[9,10], D. E. WILLIAMSON[7,11,8], J. WANG[6], J. H. KRYSTAL[4], M. J. FRIEDMAN[12,13]; [1]Psychiatry and Behavioral Sci., Texas A&M Col. of Med., Bryan, TX; [2]Central Texas Veterans Hlth. Care Syst., Temple, TX; [3]Psychiatry, Baylor Scott & White Med. Ctr., Temple, TX; [4]Yale Univ. Sch. of Med., New Haven, CT; [5]Computat. Biol. & Bioinformatics, [6]Dept. of Biostatistics, Yale Sch. of Publ. Hlth., New Haven, CT; [7]Psychiatry & Behavioral Sci., Duke Univ., Durham, NC; [8]Natl. PTSD Brain Bank Durham, Durham, NC; [9]VA Boston Healthcare, Boston, MA; [10]Natl. PTSD Brain Bank Boston, Boston, MA; [11]Durham VAMC, Durham, NC; [12]Psychiatry, Geisel Sch. of Med. at Dartmouth, Hanover, NH; [13]NCPTSD, White River Junction, VT

Abstract: The search for pathophysiological changes in PTSD has been facilitated by the establishment of the National PTSD Brain Bank (NPTSDBB). In the first postmortem anatomical study of PTSD, we described deficits in dendritic spine density in PTSD frontal cortex (FCTX) which were associated with increased transcript levels of the chaperone FKBP5 (PMID: 26844242). In a larger cohort, our NPTSDBB research group recently reported higher levels of FKBP5 and alterations in both neuronal and glial transcripts (PMID: 33349712). Among the most robustly affected were transcripts selectively expressed in interneurons.

Interestingly, a deconvolution analysis found that the proportion of interneuron cells was not altered in PTSD, suggesting that the affected transcripts in PTSD were down-regulated. In the present re-analysis of the combined sex data, we examine the distribution of interneuron-specific transcripts in WGCNA modules. Interneuron-specific transcripts were enriched in the Blue module (VIP, CRH, DLX1, DLX2) and in a small module (Coral2) composed almost entirely of interneuron transcripts (SST, CORT, GAD1, GAD2, PNOC, RELN, ELFN1, OSTN, TAC1). All of the above transcripts were at least nominally down-regulated in PTSD dlPFC. We generated signed p-value ranked GSEA lists of the Blue and Gray (unassigned transcripts) modules and probed these with the Hallmark gene sets, designed to investigate cellular stress responses. In the Gray analysis, no Hallmark sets were significantly enriched, while the Blue module recapitulated almost all of the up-regulated associations found in the entire dlPFC dataset. Up-regulated enriched sets in the Blue module (FDRq<0.01) included HYPOXIA, TNFA_SIGNALING_VIA_ NFKB, IL6_ JAK_ STAT3_ SIGNALING, EPITHELIAL_ MESENCHYMAL_ TRANSITION (FDRq<0.05), IL2_STAT5_SIGNALING, COAGULATION, INTERFERON_ GAMMA_ RESPONSE, INFLAMMATORY_RESPONSE, MTORC1_ SIGNALING, XENOBIOTIC_METABOLISM and APOPTOSIS. When we repeated the analysis in the sgACC, stress response signals such as HYPOXIA were less robust and interneuron-specific transcripts were less consistently down-regulated than in the dlPFC. These data support the presence of significant modulators of cell stress such as hypoxia in PTSD cortex, with the effect(s) being especially prominent in the dlPFC. A subset of interneuron transcripts (VIP, CRH) were co-regulated with cellular stress modules, but others (SST, CORT, GAD1, GAD2) were not. Investigation into the forces driving these transcriptomic changes have the opportunity to provide insight into how they may be reversed, potentially contributing to the development of novel treatments for chronic PTSD.

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Poster

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Topic: G.07. Post-Traumatic Stress Disorder

Support: NIGMS 1U54GM115458-01
Center for Brain and Behavior Research
South Dakota Governor's Center for Genetics and Behavioral Health

Title: Role of Inflammation and Immune Markers on Psychological Health

Authors: *J. L. SCHOLL[1], Z. KING[2], K. POTTER[3], D. DECINO[4], D. HERTEL[4], K. PEARSON[5], E. GRAACK[1], L. A. BAUGH[1], G. L. FORSTER[6];
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Abstract: There has been a growing emphasis on the relationship between inflammatory responses, gut microbiota and the immune system, and their influence on mental health outcomes. Stress can influence the composition of gut microbiota which, in turn, influences the central nervous systems response to future stressors. C-Reactive Protein (CRP) is a biomarker for inflammation that has been linked to depression, anxiety disorders and PTSD that is released in response to pro-inflammatory cytokines. The goal of this study was to test a direct association between behavior profiles such as anxiety and depression, the composition of the gut microbiota, basal levels of CRP and cytokines, cortisol and genetic predisposition. Measures of anxiety, depression, PTSD, substance use and overall health were obtained from adults with no recent illness or surgery, or exposure to antibiotics. Biological measures focused on pro- and anti-inflammatory cytokines from plasma, cortisol levels from hair, and gut microbiome; as well as single nucleotide polymorphisms (SNPs) were assayed. Using factor analysis we determined a shared mental health panel including depression, anxiety and PTSD measures that was negatively correlated to related cytokines. Regression modeling was used to related quantitative traits to demographic and genetic factors, revealing significant relationships between inflammatory markers and mental health. Understanding these relationships will help to determine direct future research in assessing causal direction between all of these factors that can promote mental well-being, and could in the future be used as biomarkers for vulnerability to mental illness, and even help re-think how we treat mental health disorders.

Disclosures: J.L. Scholl: None. **Z. King:** None. **K. Potter:** None. **D. DeCino:** None. **D. Hertel:** None. **K. Pearson:** None. **E. Graack:** None. **L.A. Baugh:** None. **G.L. Forster:** None.

Poster

P712. Human Studies and Therapeutic Approaches: Focus on PTSD

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P712.08

Topic: G.07. Post-Traumatic Stress Disorder

Support: Herman Dana Foundation

Title: Long term neuroendocrine immune alterations accompanying chronic Post-Traumatic Stress Disorder following exposure to suicide bomb terror incidents during childhood.

Authors: *R. SEGMAN[1], T. GOLTSER[2], A. SHALEV[3], F. BENARROCH[3], D. PEVZNER[3], L. CANETTI[4], C. SALONER[4], E. GALILI-WEISSTUB[3];
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Abstract: Background and Aim: Long term neuroendocrine immune alterations have been proposed to play a mechanistic role in Post-Traumatic Stress Disorder (PTSD) as well as in its associated increase in physical morbidity and mortality. Better characterization of altered immune function may help identify diagnostic and prognostic biomarkers and potentially targets for preventive intervention. **Methods:** As part of a prospective cohort ongoing study, we conducted a preliminary case control comparison of resting mononuclear cell profile between terror victims treated in childhood at the Hadassah Medical Center emergency department over the previous decade, who developed chronic PTSD upon long term follow up or recovered, and healthy non traumatized controls. **Results:** Our results point to childhood trauma related childhood long term alterations in the expression of genes related to cortisol reactivity, including the glucocorticoid receptor as well as its trans-activator Spindle And Kinetochore Associated Complex Subunit 2 (SKA2) among resting mononuclear cells in young adults. **Conclusion:** Chronic resetting of the neuroendocrine reactivity of immune cells may participate in inflammatory activation, as well as modulate immune - neuro-endocrine feedback. Better characterization and understanding of these findings may point to diagnostic and prognostic biomarkers and potentially elucidate mechanistic involvement of immune activation in childhood trauma related chronic PTSD.

Disclosures: R. Segman: None. **T. Goltser:** None. **A. Shalev:** None. **F. Benarroch:** None. **D. Pevzner:** None. **L. Canetti:** None. **C. Saloner:** None. **E. Galili-Weisstub:** None.

Poster

P712. Human Studies and Therapeutic Approaches: Focus on PTSD

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P712.09

Topic: G.07. Post-Traumatic Stress Disorder

Support: DARPA ElectRX

Title: Translating a novel neuromodulation therapy to treat PTSD

Authors: *D. PRUITT[1], H. L. CAREY[2], J. D. EPPERSON[2], N. BLEKER[2], S. A. HAYS[3], R. L. RENNAKER[5], J. WIGGINTON[6], M. POWERS[7], M. P. KILGARD[4];
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Abstract: Prolonged exposure therapy (PE) is a powerful treatment for PTSD. However, roughly 15% of patients do not respond, 20% dropout of treatment, and 60% are not fully recovered (remission/loss of diagnosis). Preclinical studies suggest vagus nerve stimulation (VNS) augmentation of extinction may enhance efficacy and tolerability by enhancing synaptic plasticity in brain networks (efficacy) and engaging parasympathetic activation (tolerability). Based on these promising preclinical studies, we have begun a first-in-human clinical trial to evaluate the ability of VNS paired with prolonged exposure therapy to reduce symptoms in patients with PTSD. This study is funded by the DARPA ElectRx program. Here, we present the initial findings from this ongoing clinical study. Participants with a diagnosis of PTSD were enrolled and implanted with a VNS device on the left vagus nerve. All participants then underwent six weeks of prolonged exposure therapy with a licensed therapist. During in-office sessions, the therapist used a software app to record the session and trigger brief bursts of VNS coincident with prolonged exposure therapy exercises. Participants then used the software app to perform homework sessions, during which VNS bursts were automatically triggered matching the timing of the in-office session. Safety was assessed throughout the course of the study. Functional outcome measures were assessed prior to and after the conclusion of therapy. Enrollment commenced in early March 2021 and the initial implantations occurred in late March 2021. Findings from the initial participants indicate that the therapy, including implantation and stimulation, is safe and can be feasibly delivered. We expect to have completed up to 10 subjects by the end of summer 2021. We will present up-to-date data on safety and functional outcome measures at the meeting. Together, these preclinical and initial clinical findings demonstrate that VNS paired with extinction training represents a novel strategy to enhance prolonged exposure therapy for patients with PTSD. Pending successful results of the first-in-human study, VNS therapy should be evaluated in a larger double-blind, multi-site study.

Disclosures: D. Pruitt: None. **H.L. Carey:** None. **J.D. Epperson:** None. **N. Bleker:** None. **S.A. Hays:** None. **R.L. Rennaker:** None. **J. Wigginton:** None. **M. Powers:** None. **M.P. Kilgard:** None.

Poster

P728. Alcohol: Potential Therapeutic Strategies

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Program #/Poster #: P728.01

Topic: G.09. Drugs of Abuse and Addiction

Support: NIAAA AA006420

NIAAA AA026999
NIAAA AA028549
NIAAA T32 AA007456
Instituto de Salud Carlos III (M-AES)

Title: Blockade of CRF_1 receptors in the infralimbic cortex prevents stress-induced reinstatement of alcohol seeking in male Wistar rats: Evidence of CRF/orexin interaction.

Authors: *F. J. FLORES-RAMIREZ[1], A. MATZEU[2], L. SANCHEZ-MARIN[4], R. MARTIN-FARDON[3];
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Abstract: Alcohol use disorder (AUD) remains one of the most treatment-elusive medical conditions globally. Although major advances have been made in the management of other health conditions, few pharmacological therapies have emerged for AUD. Indeed, a major problem in the management of AUD is the high vulnerability to relapse, even after long periods of abstinence. One explanation for this phenomenon is that chronic alcohol use dysregulates responsivity to stress, which is mediated by corticotropin-releasing factor (CRF). Thus, with repeated cycles of alcohol use, the hypothalamic-pituitary-adrenal axis becomes hyporesponsive, rendering individuals vulnerable to relapse during stressful episodes. Orexin (Orx) plays a well-established role in regulating diverse physiological processes, including stress, and interacts with CRF. The present study tested the CRF/Orx interaction in the infralimbic cortex (IL). We administered the CRF_1 receptor antagonist CP154,526, dual Orx-A/Orx-B receptor antagonist TCS1102, and their combination before testing the stress-induced reinstatement of alcohol-seeking behavior. Male Wistar rats were trained to self-administer 10% alcohol (w/v) for 3 weeks. The rats then underwent 2 weeks of extinction training (identical to the alcohol self-administration sessions, but alcohol was withheld). The day after the last extinction session, the rats received an intra-IL injection of CP154,526 (0.6 µg/0.5 µl/side), TCS1102 (15 µg/0.5µl/side), or their combination and then tested for the footshock stress-induced reinstatement of alcohol-seeking behavior. CP154,526 significantly prevented reinstatement, but TCS1102 did not produce any effect. Interestingly, the co-administration of TCS1102 with CP154,526 reversed the effect of CP154,526 alone, thus demonstrating a functional interaction between Orx receptor and CRF_1 receptor signaling and suggesting that CRF_1 receptor antagonism may ameliorate stress-induced alcohol craving and relapse.

Disclosures: F.J. Flores-Ramirez: None. **A. Matzeu:** None. **L. Sanchez-Marin:** None. **R. Martin-Fardon:** None.

Poster

P728. Alcohol: Potential Therapeutic Strategies

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Program #/Poster #: P728.02

Topic: G.09. Drugs of Abuse and Addiction

Support: DA041560
DA050896
internal funds from Rowan University and Cooper Medical School of Rowan University

Title: Is the dopamine D4 receptor a pharmacotherapeutic target for alcohol use disorder?

Authors: C. DONEGAN[1], S. URIBE[1], H. BUECHLER[1], M. SUMI[1], F. DIGIORGIO, Jr.[1], E. DIDOMENICO[1], A. ANTONIO JUAREZ[1], C. SIDE[1], Y. TAPIA[1], J. CONVISER[1], R. NAMBIAR[1], A. ACOSTA[1], I. MITHRA MADHURANTHAKAM[1], C. A. BOATENG[2], B. D. FISCHER[3], ***T. M. KECK**[1];
[1]Rowan Univ., Glassboro, NJ; [2]Basic Pharmaceut. Sci., High Point University, Fred Wilson Sch. of Pharm, High Point, NC; [3]Cooper Med. Sch. of Rowan Univ., Camden, NJ

Abstract: Alcohol use disorder (AUD) affects more than 15 million people in the United States. Current pharmacotherapeutic treatments for AUD are only modestly effective, necessitating the identification of new targets for medications development. The dopamine D4 receptor (D4R) is a target of interest in the development of medications for psychostimulant addiction, but has been largely unexplored for AUDs. In this study, we investigated the effects of the classic D4R antagonist L-745,870 and the novel, high-affinity D4R-selective antagonist CAB01-019 in rodent models of alcohol use disorder using adult male CD-1 mice. L-745,870 and CAB01-019 were tested for effects in operant food and ethanol self-administration tasks. Food-restricted mice were trained to nose poke for delivery of palatable food (vanilla Ensure) or alcohol rewards (8% w/v in water). L-745,870 (1.5 and 3.0 mg/kg, i.p.) did not significantly attenuate ethanol or food self-administration compared to vehicle. Additional testing with L-745,870 pretreatment during conditioned place preference (CPP) training did not affect the rewarding value of 2.0 g/kg ethanol using a three-compartment CPP apparatus. Similar tests with CAB01-019 are ongoing. These results, and future studies exploring whether D4R antagonism affects relapse-like responding in reinstatement models, will provide a comprehensive analysis of whether D4R antagonism affects alcohol-taking and -seeking behaviors.

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Poster

P728. Alcohol: Potential Therapeutic Strategies

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Topic: G.09. Drugs of Abuse and Addiction

Support: The Academy of Finland Grant 1317399
The Finnish Foundation for Alcohol Studies
The Finnish Cultural Foundation

Title: Lysergic acid diethylamide acutely reduces intermittent ethanol intake without effects on sucrose consumption or intracranial stimulation reward in mice

Authors: L. V. ELSILÄ, J. HARKKI, E. ENBERG, A. MARTTI, A.-M. LINDEN, ***E. R. KORPI**;
Dept. of Pharmacol., Univ. of Helsinki, Helsinki, Finland

Abstract: Small clinical studies have raised great expectations for the treatment of alcohol use disorders using psychedelics, such as lysergic acid diethylamide (LSD). To get more mechanistic insight, we studied the action of LSD on alcohol drinking and reward in male C57BL/6J mice. We aimed to test if a single dose of LSD decreases binge-like ethanol drinking in mice, both acutely and in a prolonged fashion, and if the observed acute effects are due to reward attenuation and generalized to a more natural reinforcer, sucrose. The ethanol consumption was monitored using intermittent two-bottle choice design (n=27) with ethanol available for 2 h on 4 consecutive days a week. The drug treatment was given on the fourth drinking day. The same design was used with sucrose (n=29). Effects on reward were tested using discrete-trial current threshold method of intracranial self-stimulation (ICSS) with electrodes in the medial forebrain bundle (n=9). In all experiments, LSD was administered i.p. immediately before the start of the measured behaviour. LSD doses of 0.05 and 0.1 mg/kg were tested in the drinking experiments, and additional doses of 0.2 mg/kg LSD and 3.0 mg/kg D-amphetamine were used in the ICSS. Statistical significance was tested using either one-way or repeated measures ANOVA and Bonferroni-corrected multiple comparisons with alpha level 0.05, as appropriate. Both tested LSD doses induced a decrease in 2-h ethanol drinking relative to control during the day of administration (0.05 -32%, $p>0.05$; 0.1 -83%, $p<0.05$), but the drinking returned to the control level in the next session. No changes in water consumption were observed. In the sucrose consumption experiment, minor, non-significant reductions were observed with both LSD doses (0.05 -12%, $p=0.4$; 0.1 -8%, $p=0.2$, relative to control). In the ICSS, the LSD doses tested failed to affect the response threshold acutely ($p=0.2$) or at 24 h after the treatment ($p=0.4$). When administered together with 3.0 mg/kg D-amphetamine, 0.1 mg/kg LSD did not modify the amphetamine-induced decrease in the threshold (-20% from the baseline) to either direction (sal vs. amph: $p=0.03$, sal vs. amph-LSD: $p=0.02$). LSD acutely decreased intermittent ethanol self-administration in mice but failed to induce prolonged effects. The present results imply that the decrease does not stem from effects on the animals' ability to drink as the mice continued to drink water. The ICSS results imply that the decrease is not due to LSD being aversive, nor does LSD cause any general attenuation of reward by an addictive drug. Further studies on LSD effects on consummatory behaviour are warranted.

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Poster

P728. Alcohol: Potential Therapeutic Strategies

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Program #/Poster #: P728.04

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH
NIDA IRP
DA000069

Title: Exposure to a Ketogenic Diet Alters Metabolic Brain Activity, Alcohol Intake and Withdrawal in Rats

Authors: *J. L. GOMEZ, S. K. ELVIG, L. F. VENDRUSCOLO, M. MICHAELIDES; Natl. Inst. on Drug Abuse, Baltimore, MD

Abstract: A ketogenic diet (KD) is comprised of a high-fat, adequate-protein, low-carbohydrate diet. In contrast to typical Western diets, individuals on a KD utilize more fatty acids and ketone bodies for energy metabolism and less glucose. Current studies have examined the effects of the diet on various disease models, such as diabetes, cancer, Alzheimer's, and alcohol misuse. Specific to alcohol misuse, individuals acutely intoxicated metabolize less glucose and heavy alcohol drinkers switch from using glucose to ketones as the main energy source in the brain. Using positron emission tomography (PET), we examined whole brain metabolic uptake of [^{18}F]Fluorodeoxyglucose (FDG) following exposure to a KD, alcohol exposure, and withdrawal. Rats on a KD for 5-weeks showed a global reduction in FDG uptake compared to regular chow diet. After alcohol exposure, rats on a KD had fewer changes in FDG uptake and rats on a regular chow diet had increased metabolic activity during acute withdrawal. These changes persisted after 6-weeks of protracted withdrawal from alcohol in rats on the chow diet. Here, we show the potential neural circuitry that may be involved in the beneficial effects of a KD on treating alcohol use disorders. In conclusion, the advantageous effects previously reported on KD reducing alcohol craving and intake, while ameliorating negative withdrawal symptoms during detoxification are reflected in the metabolic brain activity seen in this study.

Disclosures: J.L. Gomez: None. **S.K. Elvig:** None. **L.F. Vendruscolo:** None. **M. Michaelides:** None.

Poster

P728. Alcohol: Potential Therapeutic Strategies

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Program #/Poster #: P728.05

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant R01OD023700
NIH Grant R21AA027172
NIH Grant R01DA047785
NIH Grant R01DA043361

Title: Reducing Alcohol and/or Cocaine-induced Reward and Toxicity via an Epidermal Stem Cell-Based Gene Delivery Platform

Authors: *Q. KONG[1], L. RIEDY[1], Y. LI[2], J. YUE[2], X. WU[2], M. XU[1];
[1]Dept. of Anesthesia and Critical Care, [2]Ben May Dept. for Cancer Res., The university of Chicago, CHICAGO, IL

Abstract: Alcohol use disorder (AUD) is one of the foremost public health problems. Alcohol is also frequently co-abused with cocaine. There is a huge unmet need for the treatment of AUD and/or cocaine co-abuse. We recently demonstrated that skin grafts generated from mouse epidermal stem cells that had been engineered by CRISPR-mediated genome editing could be transplanted onto mice as a gene delivery platform. Here, we show that expression of the glucagon-like peptide-1 (GLP1) gene delivered by epidermal stem cells attenuated development and reinstatement of alcohol-induced drug-taking and seeking as well as voluntary oral alcohol consumption. GLP1 derived from the skin grafts decreased alcohol-induced increase in dopamine levels in the nucleus accumbens. In exploring the potential of this platform in reducing concurrent use of drugs, we developed a novel co-grafting procedure for both modified human butyrylcholinesterase (hBChE)- and GLP1-expressing cells. Epidermal stem cell-derived hBChE and GLP1 reduced acquisition of drug-taking and toxicity induced by alcohol and cocaine co-administration. These results imply that cutaneous gene delivery through skin transplants may add a new option to treat drug abuse and co-abuse.

Disclosures: Q. Kong: None. **L. Riedy:** None. **Y. Li:** None. **J. Yue:** None. **X. Wu:** None. **M. Xu:** None.

Poster

P728. Alcohol: Potential Therapeutic Strategies

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Program #/Poster #: P728.06

Topic: G.09. Drugs of Abuse and Addiction

Support: NIAAA R01AA025591

Title: Adolescent intermittent ethanol exposure followed by free-choice drinking in adulthood: with and without environmental enrichment

Authors: *J. I. WOODEN, J. MELBOURNE, K. NIXON;
Pharmacol. & Toxicology, The Univ. of Texas at Austin, Austin, TX

Abstract: Binge drinking in adolescence increases likelihood of problematic alcohol use in adulthood. In rats, this has been modeled with intermittent ethanol (AIE) exposure in adolescence followed by testing 2-bottle free-choice (2BC) alcohol drinking in adulthood. Rats are single-housed during 2BC, although single housing can enhance development of some addictive-like behaviors. Environmental enrichment (EE) in the home cage of single-housed rats is often used to prevent stress from isolation. Here, we conducted 2 experiments that utilized AIE vs. adolescent intermittent saline (AIS) in male Sprague-Dawley rats, followed by 2BC. AIE was 8 *i.p.* injections of 2 g/kg EtOH (20% w/v or 0.9% saline for AIS) in a 2 days on/2 days off manner during PND 30-43. After 44 days, rats were single-housed with access to 2 bottles in the home cage: one with EtOH in water, the other plain water. For Exp 1, rats (AIE n = 8, AIS n = 8) had EE in cages (PVC pipe), and were offered increasing concentrations of EtOH every 3 days ranging 3% to 9% (PND 87-111). For Exp 2, rats (AIE n = 8, AIS n = 8) did not have EE and were offered an identical 3% to 9% range of EtOH concentrations (PND 87-111), then examined up to 20% EtOH (PND 112-121). Rats in Exp 2 were subjected to abstinence for 2 wks and the alcohol deprivation effect examined: 2BC was returned with 20% EtOH (PND 135-138). We did not see the expected effect of escalated adult drinking following alcohol exposure in adolescence. There was a main effect of EtOH concentration (p = 0.04 for Exp 1, $p < 0.0001$ for Exp 2), reflecting increased consumption across 3-9% EtOH, but no main effect of group (AIS/AIE) for either experiment nor statistically significant interaction of adolescent treatment group and EtOH concentration (3-9% or 3-20%) for 3-day average of g/kg consumed. The 3-day average of g/kg consumed was not different between Exp 1 (1.0±0.2) and Exp 2 (1.0 ±0.1) for concentrations of 3-9% ethanol, indicating EE did not influence consumption. For Exp 2, both AIE and AIS rats displayed increased consumption during the first day of post-abstinence reinstatement testing compared to baseline ($t(7)$=3.610, p = 0.0086 for AIE, $t(7)$=4.236, p = 0.0039 for AIS), although consumption was similar between groups. BECs measured 1h after the last *i.p.* injection did not differ between experiments (Exp 1 = 154.6±13.9 mg/dL, Exp 2 = 190.9±14.2 mg/dL; p = 0.09). Thus, we were not able to model escalation of drinking following adolescent ethanol exposure using this AIE-2BC paradigm. Additional stressors or isolation in the adolescent period may be necessary to increase alcohol consumption in adulthood in some lab environments.

Disclosures: J.I. Wooden: None. **J. Melbourne:** None. **K. Nixon:** None.

Poster

P728. Alcohol: Potential Therapeutic Strategies

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Program #/Poster #: P728.07

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant P60AA011605-24
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Title: Role of nociceptin receptor antagonist and nociceptin-containing neurons of the lateral septum in alcohol drinking behavior in mice.

Authors: *H. HAUN, T. KASH;
UNC- Chapel Hill, Chapel Hill, NC

Abstract: Excessive alcohol drinking in individuals diagnosed with an Alcohol Use Disorder (AUD) has seen a marked increase in recent years further reinforcing the need for novel therapeutic interventions aimed at curbing chronic, uncontrolled alcohol consumption. Antagonists targeting the nociceptin receptor (NOPR) have shown promise in attenuating excessive alcohol drinking, garnering much interest in this system as a druggable target for the treatment of AUD. However, little is known about the mechanisms by which NOPR antagonists influence drinking behavior nor the endogenous nociceptin-expressing populations that drive this effect. We have identified the lateral septum (LS) as rich in nociceptin-expressing neurons (LS^{Pnoc}) and this structure has been implicated in alcohol drinking and withdrawal. Thus, the present studies were designed to screen the effect of a NOPR antagonist (SB-612111; 10 mg/kg) on alcohol drinking and other behaviors, and determine the contribution of LS^{Pnoc} to alcohol drinking. Male C57BL/6J were given access to alcohol (20% v/v) in a 2-hr drinking session, 3-hrs into the dark cycle (N=24). SB-612111 treatment 30-min prior to drinking reduced alcohol intake compared to vehicle (2.8 vs 1.9 g/kg; $p< 0.001$). Next, the effect of SB-612111 was tested on the expression of alcohol-induced conditioned place preference (CPP) to determine if NOPR contribute to the rewarding properties of alcohol (N= 20). SB-612111 treatment 30-min prior to testing was without effect on CPP expression (72 vs 62.6%; $p> 0.05$). Subsequently, NOPR contribution to the aversive properties of alcohol was tested in an alcohol-induced conditioned taste aversion (CTA) test. Mice were given access to 1% sucrose for a 30-min drinking session and alcohol (2 g/kg) was administered immediately after (N=20). SB-612111 administration concurrent with alcohol treatment attenuated the expression of CTA the following day (-72.2 vs -50.1%; $p< 0.05$) suggesting that NOPR contribute to the aversive properties of alcohol. In a final experiment, we selectively targeted LS^{Pnoc} by infusing AAV8-hSyn-DIO-hM4Di (0.3uL/side) into the LS of male Pnoc-IRES-Cre mice (N=14). Challenge with clozapine-*N*-oxide (CNO; 3 mg/kg) or vehicle 30-min prior to drinking resulted in a reduction of alcohol drinking (2.6 vs 2.1 g/kg, $p< 0.05$). Together, these data support previous findings that NOPR antagonists are effective in reducing alcohol intake and suggest that this effect is mediated by influencing the aversive properties of alcohol. We then found that silencing of LS^{Pnoc} reduced alcohol drinking

pointing to this structure as an endogenous source of nociceptin that contributes to drinking behavior.

Disclosures: H. Haun: None. **T. Kash:** None.

Poster

P774. Mechanisms of Attention: Primate

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Program #/Poster #: P774.01

Topic: H.01. Attention

Support: R01EY027402
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P30EY008126
F31EY031293
T32EY007135
S10OD021771

Title: Laminar profile of feature selectivity in V4 and its rhythmic enhancement with exogenous attention

Authors: *J. A. WESTERBERG[1], E. A. SIGWORTH[2], J. D. SCHALL[3], A. MAIER[1]; [1]Dept. of Psychology, [2]Dept. of Biostatistics, Vanderbilt Univ., Nashville, TN; [3]Departments of Biol. and Psychology, York Univ., Toronto, ON, Canada

Abstract: Visual search is a ubiquitous attention task necessitating rapid processing of sensory information. Attentional selection has been linked to altered cortical sensory responses and feature preferences (i.e., tuning). However, attentional modulation of feature selectivity during visual search is largely unexplored. Here we map the spatiotemporal profile of feature selectivity during a pop-out visual search task. Monkeys performed search where a single ("pop-out") feature determined the target of attention. We recorded laminar neural responses across 30 recording sessions from mid-level visual area V4 in two macaque monkeys performing a color-based pop-out visual search task. We first identified "feature columns" on a session-by-session basis which showed preference for individual colors. These columns had a consistent receptive field along cortical depth. Correctly performed trials where the pop-out stimulus appeared in the columnar receptive field were considered exogenously attended and trials when the pop-out was outside the receptive field were considered unattended. In the unattended condition, superficial layers of V4 feature columns showed significantly greater feature selectivity than middle and deep layers. Attending a stimulus increased feature selectivity along all layers, but not equally. Feature selectivity increased most in middle and deep layers, leading to a normalization of

feature selectivity across the cortical column. This attention-induced enhancement was rhythmically gated in phase with the beta-band local field potential. Beta power dominated both extragranular laminar compartments, but current source density analysis pointed to an origin in superficial layers, specifically. While this activity was present regardless of attentional state, feature selectivity was only beta-gated with attention. Neither the beta oscillation nor its gating of feature selectivity were predicated on microsaccades. Importantly, beta modulation of neural activity predicted response times, suggesting a direct link between attentional gating and behavioral output. Together, these findings suggest beta-range synaptic activation in V4's superficial layers rhythmically gates attentional enhancement of feature tuning in a way that affects the speed of attentional selection.

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Poster

P774. Mechanisms of Attention: Primate

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Program #/Poster #: P774.02

Topic: H.01. Attention

Support: NIH-NEIR00 EY025768
Brain & Behavior Research Foundation #28174
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NIH-NEIR01 EY02881

Title: Distinct attention modulation on noise correlation of V4 neurons during the absence and presence of visual stimuli.

Authors: *S. LIU[1], A. C. SNYDER[2];
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Abstract: The ability to selectively attend to more behaviorally relevant locations of the surrounding environment, known as spatial selective attention, is crucial to survival. The neural mechanism of this cognitive function has been vastly studied from the perspective of neural processing of visual stimuli. Typically, correlated variability, so-called noise correlation, between pairs of neurons in the visual cortex, has been observed to generally decrease with attention during stimulus response, which can improve stimulus encoding by reducing dependence of noise within a population. However, spatial attention can also be engaged during anticipation of visual information, when stimuli are absent. How spatial attention modulates neural activity during such anticipation periods was relatively poorly understood. In this study, we aim to understand how co-variability of V4 neurons change with spatial attention during

anticipation and whether it is distinct from when visual stimuli are present.
V4 neural activity was recorded when two rhesus macaque monkeys were performing an orientation change-detection task where location of one of the two gratings were cued to change with higher probability, encouraging selective attention. We computed noise correlation between pairs of V4 neurons using their response to repeated visual stimuli or their spontaneous activity during inter-stimulus-interval. When stimuli were absent, noise correlations were also modulated by covert spatial attention, in a way distinct from when stimuli are present. Overall, noise correlations decreased when attention was directed towards the receptive field of V4 neurons and when stimuli were present, but increased when stimuli were absent (i.e., the opposite effect). This suggests absence/presence of stimulus should be considered when we develop theory for attention neural mechanisms.
Studies have shown all directions of co-variability affect task performance. To explore whether such attention modulation on co-variability was task-relevant, we identified directions along which change of pair-wise activity of V4 neurons is meaningful to solve the task, which we call task vectors. Theoretically, high variability along them is harmful for task performance. By computing the variability of neural activity along the task vector direction and other directions, we found that variability is alway highest along the task vector. And the above noise correlation results are mainly contributed by change of variability along the task vector. This suggests that the considering context of stimulus and task is important for understanding why noise correlations change under attention.

Disclosures: S. Liu: None. **A.C. Snyder:** None.

Poster

P774. Mechanisms of Attention: Primate

Location: Virtual Only

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Program #/Poster #: P774.03

Topic: H.01. Attention

Support: NSERC Discovery Grant
CIHR Project Grant
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Title: A neural mechanism for enhanced visual processing in peri-hand space

Authors: *H. RAMEZANPOUR[1,2,3], D. H. KEHOE[1,2,4], C. J. PERRY[1,2,3], M. FALLAH[1,4,2,3,5]; [1]Ctr. for Vision Res., [2]VISTA: Vision Sci. to Application, [3]Sch. of Kinesiology and Hlth. Science, Fac. of Hlth., [4]Dept. of Psychology, Fac. of Hlth., York Univ., Toronto, ON, Canada; [5]Dept. of Human Hlth. and Nutritional Sciences, Col. of Biol. Sci., Univ. of Guelph, Guelph, ON, Canada

Abstract: Previous studies have provided indirect evidence that visual processing is enhanced in space near the hand, so called “peri-hand space”. It is hypothesized that these alterations are result of deploying attention to peri-hand space, like how the oculomotor system enhances visual processing at the endpoint of a saccade. This similarity notwithstanding, the underlying neural mechanisms that would drive altered visual processing near the hand are, as yet, not well understood. More specifically, it is not clear whether peri-hand attention might be solely due to vision of one own’s hand, proprioceptive feedback to visual cortex, or a combination of both signals. It has been already established that parietal neurons integrate sources of visual and proprioceptive information for visually guided reach movements. Hence, we hypothesize that enhanced visual processing in peri-hand space is a result of feedback from parietal to early visual areas. To unravel the neural underpinnings of enhanced visual processing in peri-hand space and specifically the nature of these feedback signals, we recorded from individual neurons in area V2 of rhesus monkeys, engaged in a passive fixation task in which an oriented bar was presented in the receptive field of V2 neurons in the presence or absence of a nearby hand. When the hand was placed near, yet outside the receptive field, responses to the preferred orientation were significantly enhanced no matter if the hand was visible to the animal or occluded. This result which resembled attentional gain modulation in the oculomotor system, diminished when the animal’s hand was far from the visual stimulus. These findings, together with the fact that the hand was not the target of future saccades (animals had to keep fixation to get rewarded) support the notion that there exist parallel effector-based mechanisms for deploying spatial attention which might ease decoupling of eye and hand movements, and the feedback signals responsible for deploying these attentional signals are proprioceptive in nature.

Disclosures: H. Ramezanpour: None. **D.H. Kehoe:** None. **C.J. Perry:** None. **M. Fallah:** None.

Poster

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Topic: H.01. Attention

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Title: Effects of feature attention on local field potential oscillations vary systematically across the visual processing stream

Authors: *A. UMOREN[1], D. MENDOZA-HALLIDAY[1], R. DESIMONE[2];
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Abstract: Visual feature attention selectively modulates the spike rates of feature-selective neurons in multiple areas of the primate visual stream, with increasing strength as one progresses downstream. While some studies have shown that feature attention also modulates the power of local field potential (LFP) oscillations, no study has systematically examined and compared, under the same experimental conditions, the effects of feature attention on LFP power across multiple visual processing stages. To examine this, we trained two macaque monkeys to perform a spatially-global feature attention task in which they attended to one of two full-screen overlapping random dot surfaces (tests) moving in opposite directions, cued by a prior surface (sample). We recorded neuronal activity simultaneously from 5 laminar probes in several areas across multiple processing stages of the visual stream – MT, MST, LIP, MIP and LPFC. Feature attentional modulation was quantified as the effect size from the statistical comparison between the LFP power when monkeys attended to one or the other test surface. Feature attention modulated broadband LFP power in all areas, with modulation strength being lowest upstream (MT) and increasing in a downstream progression. Furthermore, attention most strongly modulated the lower frequencies in upstream areas, and progressively higher frequencies in downstream areas. Among all areas, attentional modulation strength showed two peaks along the frequency spectrum, one in the low (below 20 Hz) and one in the high (between 40 and 80 Hz) frequencies. In LIP and MIP, LFP sites were functionally segregated into two types showing attentional modulation exclusively in the low or high frequencies. Remarkably, we further found that a fraction of LFP sites showed an inverted feature attentional effect: an increase in LFP power when attending to the antipreferred direction compared to the preferred. The incidence of such effects was highest upstream and systematically decreased downstream. Our results show that attention modulates the oscillatory activity of neuronal populations across multiple visual processing stages, and that such effects have distinct characteristics that vary systematically across stages in a downstream progression.

Disclosures: A. Umoren: None. **D. Mendoza-Halliday:** None. **R. Desimone:** None.

Poster

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Program #/Poster #: P774.05

Topic: H.01. Attention

Support: EY029666

Title: Large-scale bilateral optogenetic inactivation of the lateral prefrontal cortex decreases feature attentional effects in MT and MST neurons and impairs attentional performance

Authors: *D. MENDOZA-HALLIDAY, H. XU, F. AZEVEDO, R. DESIMONE;
McGovern Inst. for Brain Res., MIT, Cambridge, MA

Abstract: Previous studies have suggested that feature attention is mediated in part by feedback from LPFC, which is thought to enhance processing of attended features at multiple visual processing stages. To test the causal role of LPFC, we developed an optogenetic method for large-scale bilateral inactivation of LPFC with temporal specificity limited to the task period of attentional allocation. We replaced the native dura with a transparent artificial dura over LPFC bilaterally and expressed the inhibitory opsin *Jaws* across a ~100 mm^2 area via 168 viral injections. We designed a spatially-global feature attention task in which monkeys were cued to attend to one of two full-screen overlapping random dot surfaces moving in opposite directions (tests) in order to detect a small randomly-located speed change. We recorded the activity of motion direction-selective neurons simultaneously in bilateral LPFC and visual areas MT, MST and LIP. To measure the effects of feature attention, we statistically compared neurons' firing rates between trials in which monkeys attended to one test surface vs. the other. In half of the trials, both LPFCs were optogenetically inactivated during the test presentation using 635 nm lasers located above the cortical surface. We found that feature attentional modulation was weakest in area MT and increased downstream. Remarkably, half of the modulated neurons in MT showed an inverse effect: a significant firing rate increase with attention to the anti-preferred direction compared to the preferred. The fraction of neurons with inverse effects progressively decreased in downstream areas. These results suggest the need to update current attention models that predict attentional enhancements exclusively aligned to neurons' feature preference. Moreover, bilateral LPFC optogenetic inactivation impaired task performance and caused a reduction in the neuronal effects of feature attention not only locally but also in MT and MST. Our results serve as evidence that LPFC plays a causal role in the modulation of visual cortical responses during feature attention, and that this role is necessary for attentional performance.

Disclosures: D. Mendoza-Halliday: None. **H. Xu:** None. **F. Azevedo:** None. **R. Desimone:** None.

Poster

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Program #/Poster #: P774.06

Topic: H.01. Attention

Title: Dynamic coding of task rules by neuronal ensembles in the primate lateral prefrontal cortex

Authors: *A. ROUZITALAB[1,2], C. BOULAY[2], J. C. MARTINEZ-TRUJILLO[3], A. J. SACHS[4];
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Physiol. and Pharmacol. and Psychiatry, Brain and Mind Institute, Univ. of Western On, London, ON, Canada; [4]Ottawa Hosp., Ottawa, ON, Canada

Abstract: Dynamic coding of task rules by neuronal ensembles in the primate lateral prefrontal cortex Alireza Rouzitalab[1,2], Chadwick B Boulay[2,3], Julio Martinez Trujillo[4], and Adam J Sachs[2,31]Electrical and Computer Engineering Department, The University of Ottawa, Ontario, Canada[2]The Ottawa Hospital Research institute, Ontario, Canada[3]The University of Ottawa Brain and Mind Research Institute, Ontario, Canada[4]Department of Physiology, McGill University, Quebec, Canada

Introduction: One of the cortical areas heavily involved in multiple stages of the perception-to-action cycle is the lateral prefrontal cortex (LPFC). Neurons in LPFC encode sensory and motor signals, movement planning and execution, working memory, attention, reward expectation and task rules. Moreover, the LPFC is one of the regions of the cortex in which changes in the selectivity of neurons during learning of new task rules have been documented.***Task:*** Two macaque monkeys are presented with cue information while fixating on a central point on a screen. After a delay period, based on the received information, they should make a saccade towards one of eight possible targets uniformly arranged around the perimeter of the screen on a circle (Figure 1). During a random number of trials, only two of the three possible colors will appear while keeping the same possible two targets (Figure 2).***Methods and Results:*** We passed the single unit activities obtained from learned and not-learned trials to a neuronal generalized additive model tool (neuroGAM), to confirm the coding of task rules in learned trials and the presence of saccade information in not-learned trials (figure 3).A deep neural network decodes the rule information from the neuronal activities. Figure 4 shows the distinction of rules in a PCA/t-SNE space using the firing rates vs. the deep model.Furthermore, we decoded the saccade intention of monkeys from the pre-saccadic activities in different sessions using a deep model which compared to conventional methods [1], has improved significantly (Figure 5).***Discussion:*** We explored the existence of task rule information in population activities of LPFC neurons. While the monkey learns a rule presented in the task, the neuronal activities encode this information quickly. We also improved the decoding accuracy of saccade intentions using the pre-saccadic activities in LPFC neurons.***References:***[1] C. B. Boulay, F. Pieper, M. Leavitt, J. Martinez-Trujillo, and A. J. Sachs, “Single-trial decoding of intended eye movement goals from lateral prefrontal cortex neural ensembles,” *J Neurophysiol*, vol. 115, pp. 486–499, 2016.

Figure 1Figure 2

Figure 3

Figure 4Figure 5

Disclosures: A. Rouzitalab: None. **C. Boulay:** None. **J.C. Martinez-Trujillo:** None. **A.J. Sachs:** None.

Poster

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Title: Fixational saccades occur along a theta rhythm under covert visual spatial attention

Authors: *K. DOUGHERTY, B. WU, B. MOREA, R. CHEN, R. BOSHRA, S. KASTNER; Princeton Neurosci. Inst., Princeton Univ., Princeton, NJ

Abstract: The rhythmic theory of attention (Fiebelkorn and Kastner, 2019) proposes that attention fluctuates over time along a theta rhythm, with periods of enhanced perceptual sensitivity followed by periods of diminished perceptual sensitivity in which shifts of attention and/or gaze may occur. This theory is based both on behavioral data showing that detection of a visual target fluctuates at theta frequency, as well as neural data indicating cortical visuo-motor neurons show theta-nested activity that is predictive of behavior during a visual attention task. One prediction from this theory is that small fixational saccades (< 2 dva), known to occur in covert attention tasks and thought to index attention shifts, are more likely to occur during periods when sensory processing is diminished. In other words, the theory predicts that the probability of fixational saccades fluctuates at a theta rhythm under covert visual spatial attention. To test this prediction, we recorded gaze position with an infrared-based eye tracking system while two macaques performed a visual spatial attention paradigm, known as the Egly-Driver task. Following initial fixation, two vertically-oriented or horizontally-oriented parallel bars appeared in the periphery on either side of the central fixation point. An informative (80% valid) cue appeared at one of the corners of the bar locations, indicating the likely position of an upcoming visual target. Following a variable delay (500-1700 ms), a target appeared at one of four possible locations, and the animal released a lever following onset of the target to receive juice reward. With gaze position data, we detected the onset of fixational saccades and their durations. We observed that the probability of a fixational saccade was not uniformly distributed during the cue-target interval, but instead waxed and waned with a theta-range rhythm. This fluctuation in oculomotor behavior, together with fluctuations in visual detection performance, suggest a dynamic interplay between sensory and motor processing during attention.

Disclosures: K. Dougherty: None. **B. Wu:** None. **B. Morea:** None. **R. Chen:** None. **R. Boshra:** None. **S. Kastner:** None.

Poster

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Support: NIH Grant EY017292
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Title: Neural mechanisms of attentive object processing in primates structured by microsaccades

Authors: *K. SRINIVASAN[1,2], E. LOWET[3,2], B. GOMES[4,2], R. DESIMONE[2,1];
[2]McGovern Inst. for Brain Res., [1]MIT, Cambridge, MA; [3]Dept. of Biomed. Engin., Boston Univ., Boston, MA; [4]Inst. de Ciências Biológicas, Univ. Federal do Pará, Belém-Pa, Brazil

Abstract: Covert spatial attention is usually attributed to be directed without any eye movements. However, we had previously shown that even during visual fixation periods in a sustained attention task, the neural firing rates and stimulus representations in primate visual cortical areas V4 and the inferotemporal cortex (IT) are enhanced only following microsaccades (MS) towards the attended target locations (Lowet et.al., Neuron, 2018). Here we present evidence that the first MS towards the intended (cued) target location kickstarts the attentional modulation of neural firing rates typically observed in area V4. That is, at the neural level, the timing of the first MS structures the process of attention which continues for the remainder of the trial. Subsequent MS during the trial modulate the underlying attentional enhancement. Furthermore, the neural modulations triggered after the MS onsets have significant effects in shaping the object representation, tuning, and selectivity at the target locations. Decoding analyses of V4 firing rates for objects present in the target location confirms that the performance of the decoder is significantly increased for the MS towards intervals towards the attended stimulus compared to intervals following MS directed away from the attended stimulus. The type of MS, towards or away from the target, gives rise to distinct temporal regimes of representation, selectivity, and decoding performance. Such mechanisms are usually obscured when analyzing the neural activity during the full trial periods without taking MS into account. These results thus reveal a major link between spatial attention and its coordination with eye movements. We thus suggest that selective visual attention is not merely a covert cognitive process, but has visuomotor components that shape our ability to dynamically attend, recognize, and act on the objects in the world.

Disclosures: K. Srinivasan: None. **E. Lowet:** None. **B. Gomes:** None. **R. Desimone:** None.

Poster

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Support: National Eye Institute Intramural Research Program at the National Institutes of Health (ZIA EY000511)

Title: Microsaccades are not necessary for attention-related modulation

Authors: *G. YU, J. P. HERMAN, L. N. KATZ, R. J. KRAUZLIS;
Lab. of Sensorimotor Res., Natl. Eye Inst., Bethesda, MD

Abstract: The allocation of visual spatial attention is associated with both the enhancement of neuronal activity at the attended location and the tendency to make microsaccades towards the covertly attended stimulus while fixating. A recent study found that cortical neurons displayed attention-related modulation only after microsaccades directed towards the attended stimulus, raising important questions about how to interpret neuronal data obtained during fixation tasks commonly used in the study of visual attention. Here, we tested the relationship between microsaccades and attention-related modulation in the superior colliculus (SC), a brain structure crucial for allocating attention. We collected neuronal activity (64 units) and eye movement data in two monkeys (Macaca mulatta) during a covert spatial attention task. Our analyses focused on the 'delay period' when SC neurons displayed classic sustained attention-related modulation. First, we found that attention-related modulation in SC emerged over a similar time course during the attention task, regardless of microsaccades generation; Second, subsets of neuronal data with no microsaccades in 400-ms windows selected to match the microsaccades timing distribution still displayed substantial attention-related modulation when microsaccades were completely absent; Third, when microsaccades did occur, there was a relationship between attention-related modulation and microsaccade direction, however, we determined that the relationship was already present before the onset of microsaccades and persisted unchanged after the occurrence of the microsaccade. Together, our results suggest that microsaccades are not necessary for neuronal modulation during sustained attention. Instead, microsaccades provide an additional marker for the state of attention.

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Poster

P774. Mechanisms of Attention: Primate

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Topic: H.05. Working Memory

Support: NIH-NEIR00 EY025768
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NIH-NEIR01 EY02881

Title: Ensemble-level neural correlates of visual working memory from the local field potentials recorded from the lateral prefrontal cortex of non-human primates

Authors: *D. Y. JUNG, M. A. CONLEY, A. C. SNYDER;
Univ. of Rochester, Rochester, NY

Abstract: Visual working memory (VWM) allows transient maintenance of relevant visual information (hereafter, "items") for near-term goals. In non-human primates, individual neurons in the lateral prefrontal cortex (LPFC) significantly participate in item maintenance through ensemble-level interactions. But the ensemble-level interactions are mostly inferred from pseudo-populations of neurons pooled from separate recording sessions, an approach that potentially neglects *real-time* ensemble dynamics. Local field potentials (LFPs), related to summed electrical activity of an ensemble near a recording electrode(s), provide a means to indirectly study large ensemble dynamics at high temporal resolution. Through measuring high-density LFPs from the LPFC of monkeys, we studied ensemble-level neural dynamics especially during the *delay period* of a VWM task, when the item maintenance occurs. Spontaneous LFPs during the delay period, however, lack item-dependent features, which makes inferring items in VWM difficult. Therefore, we introduced a "perturbing" visual stimulus impulse that is task-irrelevant but can drive LFP responses in the LPFC, which we hypothesized to be dependent on the underlying LPFC ensemble dynamics. To be precise, we hypothesized that the LPFC ensemble maintains items in VWM and therefore LPFC LFP responses to the impulse stimulus would be dependent on the ensemble, which will allow us to infer (decode) items from the LFPs and reveal large neural ensemble interactions during item maintenance. We recorded high-density LFPs from LPFC of monkeys performing a memory-guided saccade (MGS) task. Critically, the impulse stimuli (i.e., task-irrelevant checkerboard stimuli) during the delay periods on half of the trials enabled us to decode the item. Importantly, checkerboard presentation did not improve decoding performance when the identical paradigm was presented to an untrained monkey, indicating that the item decodability from the LPFC LFPs is not due to passive visual processing. Interestingly, we failed to decode the item from LFPs collected extrastriate cortex (V4), a possible area participating in (visual) item maintenance. Our results, therefore, show the specificity of PFC's role in transient item maintenance and provide an approach to probe states of large neural dynamics of cognitive processes from the LFP responses to the impulse stimuli.

Disclosures: D.Y. Jung: None. **M.A. Conley:** None. **A.C. Snyder:** None.

Poster

P803. Cortical Mechanisms in Working Memory

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Program #/Poster #: P803.01

Topic: H.05. Working Memory

Title: Memory guided saccades in marmosets: behavior and electrophysiology in posterior parietal cortex

Authors: *H. CARNEY, E. HART, J.-O. MUTHMANN, A. C. HUK;
Univ. of Texas at Austin, Austin, TX

Abstract: Introduction: Persistent neural activity is a long-studied phenomenon thought to underlie working memory, but the detailed circuitry that supports it has been largely inaccessible in the primate. Marmosets represent an exciting opportunity to deploy powerful techniques, but it is unclear whether this species can be trained to perform memory-guided saccades (MGS), nor whether they have cortical areas functionally analogous to those studied in this context in macaques.
Methods: We developed marmoset-specific training techniques for the MGS task, and mapped neural activity in posterior parietal cortex during performance of this task using chronically implanted 3D "N-form" multielectrode arrays. We then compared the behavior and the neurophysiology to comparable data from macaques. All marmosets (N=3) learned the task and performed at levels acceptable for physiological study, with delay periods exceeding 1000ms.
Results: Marmoset memory-guided saccades were somewhat less accurate and consistent than macaque MGS, but marmosets were willing to perform significant numbers of trials per session (averaging 473 trials per session). In the first monkey's dataset, 490 individual units were identified across 78 sessions spanning a ten month period, and an average of 34 units were identified per session. Neural activity in marmoset PPC reflected an often-multiplexed combination of sensory, motor, and delay-period activity, both time-locked to relevant task events and with spatial selectivity. Of 490 identified units, 12% (n=61) of the recorded units had obvious spatial selectivity for visual and/or motor aspects of the task. The majority of non-spatially selective units were still modulated by visual or saccadic task components. 91% (n=56) of units with spatial selectivity were spatially selective during both visual and motor epochs. A similar amount (93% n=57) of spatially selective units exhibited distinct persistent activity during the delay period. This activity was similar to activity classically reported in macaque PPC during memory-guided saccades. The one clear difference to macaque PPC was that marmoset firing rates were considerably lower.
Discussion: Marmosets can be trained to perform the MGS task using shaping approaches not typically used with macaques, and activity in their PPC suggests functionally analogous signals to those observed in macaque LIP, including single neurons exhibiting persistent activity.

Disclosures: H. Carney: None. **E. Hart:** None. **J. Muthmann:** None. **A.C. Huk:** None.

Poster

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Topic: H.05. Working Memory

Support: CIHR
BrainsCAN
NeuroNex

Title: Persistent activity in the prefrontal cortex of the common marmoset during a delayed-response task

Authors: *R. K. WONG, J. SELVANAYAGAM, K. D. JOHNSTON, S. EVERLING; Physiol. and Pharmacol., Western Univ., London, ON, Canada

Abstract: The common marmoset (*Callithrix jacchus*) has attracted attention in neuroscience research because of its small size, high reproductive efficiency and compatibility with gene editing techniques. This species' relatively lissencephalic cortex offers practical advantages for laminar recording, two-photon and calcium imaging. Marmosets have been established as a model for visual neuroscience, but its suitability for investigating higher order cognitive functions, such as working memory and its underlying neural circuitry, remain to be determined. The neurophysiological basis of working memory has been investigated in detail in the prefrontal cortex (PFC) of the macaque monkey. PFC neurons have been shown to exhibit sustained activity during the delay period of a delayed-response task in the absence of a stimulus. This persistent firing during the delay period is regarded as the neuronal correlate of a mnemonic event. It is unknown whether PFC neurons exhibit delay-period activity in the marmoset, as working memory in marmosets has been examined primarily at the behavioural level. Here, we addressed this gap by carrying out single neuron recordings in a common marmoset monkey performing a delayed-match-to-location (DMTL) task with varying delay lengths (2-8s) on a touchscreen attached to the home cage. Untethered extracellular electrophysiological recordings were obtained from a 96-channel Utah array (4x 4 mm, 1mm electrode length, 400μm pitch) implanted in areas 46d, 46v, and 8Ad. We recorded 706 units, of which 564 units were modulated during the task. Of these 564 units, 269 units (38.1%) displayed delay period activity, 167 units (23.7%) displayed cue-related activity and 513 (72.7%) units displayed touch-related activity. Of the 269 delay units, 90 units (33.5%) were excited and 179 units (65.5%) were suppressed, when compared to baseline firing rate. These data show that prefrontal cortex neurons in marmosets exhibit persistent activity during the delay period of a delayed-response task, and that marmosets are a suitable model for investigating the neurophysiological circuitry underlying working memory.

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Poster

P803. Cortical Mechanisms in Working Memory

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Topic: H.05. Working Memory

Support: Canadian Institutes of Health Research
Natural Sciences and Engineering Research Council of Canada
Neuronexus Brain Initiative

Title: Neuronal ensembles of primate lateral prefrontal cortex encode spatial working memory in different frames of reference

Authors: *R. LUNA[1,2], M. ROUSSY[1,2,3], B. W. CORRIGAN[1,2], R. A. GULLI[4], G. DOUCET[5], A. J. SACHS[5], S. TREUE[6], J. C. MARTINEZ-TRUJILLO[3,1,2,7];
[1]Physiol. and Pharmacol., Schulich Fac. of Medicine, Univ. of Western Ontario, London, ON, Canada; [2]Robarts Res. Institute, Univ. of Western Ontario, London, ON, Canada; [3]Brain and Mind Institute, Univ. of Western Ontario, London, ON, Canada; [4]Zuckerman Mind Brain Behavior Inst., Columbia Univ., New York, NY; [5]The Ottawa Hosp. Res. Inst., Ottawa, ON, Canada; [6]German Primate Ctr., Goettingen, Germany; [7]Lawson Hlth. Res. Inst., London, ON, Canada

Abstract: Neurons in the lateral prefrontal cortex (LPFC) of macaques encode spatial information during working memory (WM) tasks. Yet it is poorly understood whether ensembles of neurons could represent memorized locations on different frames of reference (FOR) - such as space centered frame. To tackle this question, we trained two rhesus monkeys on an oculomotor delayed response task with variable fixation and target positions. In a single trial, animals fixated a dot displayed at one of sixteen different positions on the screen and a target transiently appeared for 1 second at another one of the remaining positions. Animals maintained fixation for another second and upon extinction of the fixation dot they made a saccade to the remembered target location. Reward was delivered for correct trials. This task design allowed data analysis for remembered locations relative to the fixation dot (retinotopic FOR), and relative to the target location (spatiotopic FOR). We recorded and spike sorted the extra-cellular activity by implanting multi-electrode arrays in LPFC dorsal and ventral banks (areas 8a and 9/46) of each animal (3039 multi- and single units in the dorsal array and 2410 in the ventral array). Trials were pooled by visual quadrants of each FOR. We computed the average firing rate during the first 1-second interval of the WM delay for each unit and determined their spatial selectivity (Mutual information; 1000 permutations; $p < 0.05$). We found 820 (27%) spatial WM-encoding selective units in the retinotopic FOR, 334 (11%) in the spatiotopic FOR, and 169 (5.5%) for both FORs, in the dorsal array, as well as 681 (28.3%) in the retinotopic FOR, 325 (13.5%) in the spatiotopic FOR, and 175 (7.3%) for both FORs, in the ventral array. We then constructed neuronal ensembles of different sizes (n=1, ⋯, 50) and trained linear classifiers to decode the memorized quadrant out of each ensemble (five-fold cross validations; 20 decoding iterations; balanced conditions). On average, ensembles of units from dorsal and ventral arrays yielded decoding accuracies of 69%

and 79% in the retinotopic FOR, respectively, and 55% and 52% in the spatiotopic FOR (chance performance ~25%). Notably, decoding accuracies reached asymptotic values at ensembles of 4 to 18 units. Furthermore, neurons selective for either or both reference frames were sparsely distributed in the dorsal or ventral array, although distances between units were generally smaller than predicted by chance. Neuronal ensembles in the primate LFPC could encode locations during spatial WM in the retinotopic and spatiotopic reference frames, also showing some degree of grouping of informative units in the surface covered by the arrays.

Disclosures: R. Luna: None. **M. Roussy:** None. **B.W. Corrigan:** None. **R.A. Gulli:** None. **G. Doucet:** None. **A.J. Sachs:** None. **S. Treue:** None. **J.C. Martinez-Trujillo:** None.

Poster

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Title: Different neural codes serve long and short-term memory functions in primate Hippocampus and Lateral Prefrontal Cortex during virtual navigation

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Abstract: Neurons in the primate Hippocampus (HPC) and the lateral prefrontal cortex (LPFC) integrate high level information about objects, features and space during behavior. Previous lesions studies have suggested that while the HPC plays a role in the formation of long-term declarative memories, the LPFC plays a role in maintaining short term memories. These two functions have different underlying mechanisms. Here we hypothesize that neural codes in the HPC have evolved to support short-term synaptic plasticity while in the LPFC codes have evolved to support persistent firing in large neuronal ensembles without necessarily triggering

plasticity. We use a virtual reality task in which animals navigated through a maze using a joystick and selected one of two targets in the arms of the maze according to a learned context-color rule. We found that neurons and neuronal populations in both regions were similarly selective for the different task periods. However, HPC and PFC neurons differed substantially in the temporal structure of their spike trains. Using statistical analyses and linear classifiers, we demonstrated that while HPC neurons and populations 'packed' information into bursts of action potentials, their PFC homologues fire more sparsely. We propose that the HPC, a structure with limited connectivity, enhanced plasticity and known to encode episodic memories, uses a code that facilitates temporal summation of synaptic events. The PFC, a more widely connected structure and known to encode short-term memories uses predominantly a code with spikes temporally distributed across many neurons which allows encoding and broadcasting information to other brain regions without inducing short term plasticity in individual synapses. The latter may also allow encoding of short term memories.

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Poster

P803. Cortical Mechanisms in Working Memory

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Title: Fast spiking neurons from non-human primate prefrontal and visual cortex: Electrophysiological and morphological characterization

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Abstract: INTRODUCTION: Fast spiking (FS) parvalbumin (PV) expressing cells are one of the three major subpopulations of GABAergic interneurons. They show a consistent set of electrophysiological features across brain areas that are thought to be crucial for fast inhibition and generation of network oscillations associated with various cognitive phenomena. However, previous studies have shown a wide range of diversity in morphological and intrinsic electrophysiological properties between neocortical neurons across different species. In this study, we compare the somatodendritic configuration and subthreshold intrinsic properties of putative FS neurons from macaque primary visual area (V1) and dorsolateral prefrontal cortex (DLPFC, areas 9/46), and cells from multiple areas of mice and humans (Allen Institute Cell Type Database). METHODOLOGY:The electrophysiological properties of 195 cells from NHP were obtained from whole cell patch clamp recordings from the DLPFC and V1 of 5 male Macaca mulatta and 9 Macaca fascicularis (4 male, 5 female). Cells were filled with biocytin for morphological characterization. Putative FS neurons from NHP and human were identified using waveforms via a machine learning procedure trained using data from genetically labeled neurons obtained from the Allen Institute Cell Type Database. RESULTS AND CONCLUSION: We found that NHP/human neurons were more alike, having higher excitability and sag amplitude than those of mice. Compared to humans NHP cells showed higher excitability and sag than human cells. No significant difference was observed between subthreshold properties of DLPFC and V1 putative FS cells in NHP. Our results suggest that an increase of cortical surface in macaques and humans relative to mouse is accompanied by changes in the input resistance and somatodendritic configuration of PV+ interneurons. Such changes in FS cells may accommodate the demands of the local network, providing fast inhibitory inputs to pyramidal cells and facilitating a rich temporal dynamics that may be required for complex cognitive function.

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Poster

P803. Cortical Mechanisms in Working Memory

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Title: Non-linear mixed selectivity in prefrontal and posterior parietal cortex during working memory

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Abstract: Neurons in the prefrontal cortex (PFC) and posterior parietal cortex (PPC) respond differently to the same stimuli depending on task context. Conjunctive representations of multiple stimulus and task features can be found on single-neuron activity, a phenomenon known as nonlinear mixed selectivity (NMS). Here we measured nonlinear mixed selectivity in PFC and PPC in a working memory task that required monkeys to observe two consecutive stimuli with an intervening delay period and make a judgment on whether they are the same or not. Neurophysiological recordings were performed in areas 8a and 46 of the dorsolateral prefrontal cortex, and areas 7a and lateral intraparietal cortex (LIP) of the posterior parietal cortex. We analyzed 404 PFC neurons and 671 PPC neurons with sufficient data from two male rhesus monkeys (*Macaca mulatta*). We identified neurons that exhibited NMS with respect to spatial tuning for the same stimulus when it appeared as first (cue) or second (match) in the trial. The percentage of NMS neurons in PFC was higher than in PPC (8.7% of neurons in PFC vs. 4.4% of neurons in PPC). In our task, the subjects' behavioral performance improved within a behavioral session as they learned the session-specific statistics of the task. Our analysis revealed that in PFC the proportion of neurons that exhibited NMS also increased as a function of time within a single session, while this was not the case in PPC. However, we did not find appreciable differences in the percentage of neurons that exhibited NMS between correct and error trials. Analysis of the combined population of neuronal activities showed that the distance between first and second stimulus in state-space was greater for the PFC than PPC, however for both areas the two stimuli could be projected in the same subspace, with little rotation. Our results provide evidence of greater plasticity and flexibility of neuronal representations in PFC than PPC in a task requiring maintenance of precise working memory information, suggesting a cortical specialization for processes representing.

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Poster

P803. Cortical Mechanisms in Working Memory

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Program #/Poster #: P803.07

Topic: H.05. Working Memory

Title: Geometry of neural computations unifies working memory and planning

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Abstract: The ability to maintain information in working memory forms one of the key building blocks of cognition. Traditionally, computational models of working memory have been conceived as a form of extended sensory perception that preserves prior information to inform current computation. In this study, we recast working memory representations as an active part of ongoing goal-oriented computations. Using both Recurrent Neural Networks (RNNs) and human subjects performing a set of logical tasks, encompassing several known working memory paradigms, we demonstrate how such working memory representations may help to minimize computational complexity of cognitive operations.
The contextual delayed logic (CDL) task, is a novel delayed decision task in which subjects use a rule context to flexibly modulate their response to a pair of binary stimulus inputs separated by a mnemonic delay. On each trial the subject sees one of ten Boolean logical operations (e.g. 'AND'), must hold trial variables in memory across a delay, and then upon presentation of the second stimulus, must apply the stated logical operation. The different retrospective and prospective demands between rules allow us to disentangle memory, planning, computational or rule specific influences on representations and behavior.
We trained RNNs to perform the CDL task, analyzing resulting optimized networks and state trajectories. We found that delay epoch representations were organized computationally and prospectively with overlapping sub-problems sharing network resources (e.g. fixed points, transitions). Further, delay states were geometrically arranged within a stable mnemonic subspace to linearize the overall stimulus-output classification problem.
Intriguingly the resulting representations recapitulate known properties of prefrontal delay task firing patterns recorded in primate delay tasks. First we find that units represent task variables in a mixed rather than pure encoding structure, and can explain this mixing as a function of the underlying prospective tuning utilized by our models. Second, our network demonstrates behavior reflective of dynamic tuning, where the transition from retrospective encoding to prospective encoding from the stimulus to delay epoch leads to an apparent shift in the stimulus coding direction when each task is analyzed independently.
Lastly we designed a human version of our CDL task paradigm identifying behavior that is consistent with our models, demonstrating evidence of pre-computation during mnemonic delays and prospective representational organization.

Disclosures: D.B. Ehrlich: None. **J.D. Murray:** None.

Poster

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Title: A method for identifying cell types in mammalian neocortex during in vivo and ex vivo electrophysiological experiments

Authors: *E. KUEBLER[1], M. D. FEYERABEND[1], S. A. MESTERN[2], M. S. JIMENEZ-SOSA[2], J. K. SUNSTRUM[2], J. M. D. POULIN[2], B. W. CORRIGAN[2], M. ROUSSY[2], R. LUNA[1], B. MAHMOUDIAN[2], S. TREUE[4], S. EVERLING[1], W. INOUE[1], J. C. MARTINEZ-TRUJILLO[3];
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Abstract: Electrophysiological single cell recordings during in vivo and in vitro experiments have been the backbone of system neurophysiology allowing for exploration of the functional properties of neurons in different brain areas. However, so far, the in vivo techniques have provided limited insight on the identity of the cell types recorded from. Linking response patterns to neuronal identities is fundamental for revealing the role of different cell types in the neural computations underlying information processing and behavior in the brain. Here, we use data from three species, mouse, macaque monkey and human to identify features of electrophysiological recordings that are common to in vivo and in vitro experiments and map them into genetically and histologically identified cell types in neocortex using computational techniques. We elaborate on a probabilistic classification of four main cell types, namely: Pyramidal, Parvalbumin, Vasointestinal Peptide, and Somatotastin positive from electrophysiological recordings in both in vivo and in vitro experiments. Our results provide electrophysiologists with a tool to probabilistically identify individual circuits components during functional exploration of the neocortex.

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Poster

P803. Cortical Mechanisms in Working Memory

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Title: Understanding Cognition through Connectivity Analyses: Examples from Working Memory Research

Authors: *D. PINOTSIS[1], E. K. MILLER[2];
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Abstract: Cognition arises from coordinated activity of different neuronal populations that form brain networks. The activity in each of these populations is the result of input from all other populations connected to it. Therefore connectivity is a central notion in describing the function of brain networks and understanding cognition. In this presentation, we will show how the analysis of connectivity patterns provides novel insights for understanding the mechanisms underlying an important cognitive function, working memory. We will present two studies of working memory. The first focused on a change detection task while the second focused on a spatial delayed saccade task. Both studies exploit modern machine learning algorithms, biophysical modeling and high resolution electrophysiology data.
First, in the change detection task, the number of items that the subject had to remember (memory load) was varied (1-3 items in the same visual hemifield). We will show that changing memory load resulted in changes in feedforward and feedback coupling in the network of brain areas supporting memory function (memory network). We will also show that feedback connectivity broke down when the cognitive capacity limit was exceeded (when more than 2 items were presented in the same hemifield, the animal's behavioral performance decreased dramatically).
Second, in the spatial delayed response task, the location of the item that had to be remembered was varied (six possible locations in the visual field). We will show that coding different locations resulted in different spatial organization of neural activity within each brain area in the memory network. Different neural ensembles (groups of neurons that coded locations) were formed. We will also provide a mechanistic explanation of a well-known behavioral effect, called the oblique effect (performance is better for stimuli on than off the horizontal axis) and link it to the structure of the underlying neural ensembles.

To sum up, in this presentation we will show that a combination of modern machine learning algorithms, biophysical modeling and high resolution electrophysiology data sheds new light to the mechanisms underlying working memory and explains important behavioral effects.

Disclosures: D. Pinotsis: None. **E.K. Miller:** None.

Poster

P803. Cortical Mechanisms in Working Memory

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Title: Specialized prefrontal circuits explain population dynamics during working memory encoding and maintenance

Authors: *N. POLLÁN HAUER[1], B. PESARAN[2], A. COMPTE[3], K. WIMMER[4];
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Abstract: Neuronal population activity recorded from primate prefrontal cortex carries information about the presented stimulus during working memory tasks. During cue presentation and in the beginning of the delay period the population code is dynamic, before it stabilizes and remains stable throughout the rest of the delay period. It is an open question how individual neurons support the dynamic and stable population codes and what are the underlying circuit mechanisms. Here, we show that a spiking network model composed of three specialized attractor circuits can explain the experimentally observed transition from cue to delay dynamics. Each conceptual ring circuit represents a specialized PFC sub-population (encoding, storage and

readout neurons). Based on experimental findings we structured the model such that the external input excites the encoding population while it suppresses the storage population. When a stimulus is presented, an activity bump forms in the encoding population. Recurrent connections are not strong enough to sustain persistent activity in the encoding ring, so that, after stimulus offset, the bump fades in the encoding units. At the same time, the strongly recurrent storage ring is released from inhibition and a stable bump emerges in the storage units. The readout circuit receives input from the other two populations and is thus active during cue and delay periods even without strong recurrency. We found that when we randomly select a subpopulation of the entire network, cross-temporal decoding analysis fails to generalize across cue and delay periods because different neuronal subsets are most informative during the respective epochs. We confirmed similar cross-temporal decoding patterns in prefrontal population recordings from a visual and memory task. Finally, from a functional point of view, the network model shows increased robustness to distractors once the activity bump has formed in the storage population, a prediction that can be tested experimentally. In sum, our findings suggest that the presence of a highly dynamic cue to delay transition originates mainly from different neuronal subpopulations. After this initial transient, a stable state is reached, and memory maintenance is achieved through attractor dynamics.

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Poster

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Title: Distinct neural codes for content and ordinal structure in human auditory sequence memory

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Abstract: Two forms of information - frequency (content) and ordinal position (structure) - have to be stored when retaining a sequence of auditory tones in working memory (WM). However, the neural representations and coding characteristics of content and structure, particularly during WM maintenance, remain elusive. Here, in two electroencephalography (EEG) studies in human participants performing a delayed-match-to-sample task with a retrocue, by transiently

perturbing the ‘activity-silent’ WM retention state and decoding the reactivated WM information, we demonstrate that content and structure are stored in a dissociative manner with distinct characteristics throughout WM process. First, each tone in the sequence is associated with two codes in parallel, characterizing its frequency and ordinal position, respectively. Second, during retention, a structural retrocue successfully reactivates structure but not content, whereas a following neutral white noise triggers content but not structure. Meanwhile, a content retrocue is able to reactivate both content and structure information, while a subsequent neutral visual impulse successfully makes maintained structure information detectable. Third, structure representation remains stable whereas content code undergoes a dynamic transformation through memory progress. Finally, the neutral-impulse-triggered content and structure reactivations during retention correlate with WM behaviors on frequency and ordinal position, respectively. Overall, our results support distinct content and structure representations in auditory WM and provide efficient approaches to access the silently stored WM information (both content and structure) in the human brain.

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Poster

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Title: Neuronex exploration: how does the brain generate the mental representations that underlie working memory and abstract thought?

Authors: *J. MARTINEZ-TRUJILLO;
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Abstract: How does the brain generate the mental representations that are the foundation of abstract thought? Mental representations that can be dissociated from incoming sensory inputs and ongoing motor actions are a step up in brain evolution. They exist ‘virtually’, so that the brain can load and keep online information acquired in the past to solve complex problems, e.g., “simulate” situations and “predict” outcomes without the need to immediately act. When and

how did mental representations arise during brain evolution? Current evidence indicates that they may have arose in anthropoid primates, with the development and expansion of the granular, dorsolateral prefrontal cortex (dlPFC).Studies in macaques performing spatial working memory tasks have found neurons in dlPFC with strong persistent firing resilient to distracter signals and representing a remembered location. This capability may arise from layer 3 microcircuits, containing neurons with abundant recurrent excitatory connections, extensive dendritic branching, and unique molecular regulation. However, the foundation of this hypothesis rests on data from disparate labs/approaches with poor integration across levels. In our NEURONEX we address the following questions. What are the features of neurons and circuits that allow mental representations to arise in the dlPFC?Are these features particularly salient in old world primates relative to new world primates?Are there unique molecular characteristics, e.g. in dopamine, NMDAR and calcium signaling, that are necessary to support mental representations?Answering these questions will reveal how evolution shaped primate cognition and vulnerabilities to mental disease. Our emerging data demonstrate that dlPFC neurons in macaque encode working memory representations in naturalistic environments, in both macaques and marmosets. In vitro electrophysiology shows neurons in dlPFC have distinct intrinsic properties than those in sensory cortices and the parietal lobe. The latter is supported by transcriptomic data. ImmunoEM and in vivo recordings show that dlPFC pyramidal cells have magnified calcium signaling near NMDAR synapses, and a focal location for dopamine D1R. Computational modeling shows gradients of neuronal and receptor types and distinct circuit motifs across the cortical hierarchy, consistent with the emergence of persistent firing capabilities resilient to distracting sensory and motor events. Our data suggest complex mental representations resulted as an emergent property of complex brains evolution due to changes in the make-up features of the primate neocortical mantle.

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Poster

P807. Working Memory: Circuits

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Title: Spontaneous blink-related BOLD-fMRI delay activity correlates with working memory performance

Authors: *T. ELLMORE[1], C. REICHERT PLASKA[3], B. A. GOMES[4], J. ORTEGA[2]; [1]Psychology, [2]The City Col. of New York, New York, NY; [3]City Univ. of New York, New York, NY; [4]Neurosurg., Cedars-Sinai Med. Ctr., Los Angeles, CA

Abstract: Spontaneous blinks are typically considered a nuisance in cognitive neuroscience studies with instructions to inhibit blinks to minimize signal artifact. Recent findings, however, demonstrate the functional significance of spontaneous blink-related oscillations[1] and document a relationship between spontaneous blink rate during the working memory delay period and subsequent task accuracy[2]. Together these results motivate an investigation of the neural substrates of spontaneous blinking and its role in working memory. We studied 7 adults (mean age 27.8, 3 males) during performance of a Sternberg scene working memory task. Whole-brain 3 tesla BOLD-fMRI (2 mm^3, 2 sec TR) and continuous in-bore camera-based eye tracking data were collected. Single subject imaging data were preprocessed using *fmriprep* followed by exploratory voxelwise group analyses in *AFNI* using *3dMVM* with blink event-related responses during the working memory delay period and non-delay periods as separate conditions and working memory task accuracy as a covariate of interest. We found a cluster of voxels located within the inferior portion of right hemisphere central sulcus encompassing portions of the pre- and post-central gyrus (MNI: x=+54, y=-5, z=+23 mm] but also extending posteriorly to parietal cortex whose BOLD fMRI activity evoked by spontaneous blinks during the delay period correlated with working memory task accuracy (r=+0.88, p=0.008). We examined activity in this region during spontaneous blinks occurring at other times during the imaging runs and found these non-delay period evoked responses to be unrelated to task performance (r=-0.19, p=0.68). The spontaneous blink rate has been linked to dopamine function with higher and lower rates in Schizophrenia and Parkinson's disease, respectively. These disorders also present with deficits in working memory, with perseveration and compromised updating. The finding of the present study, that activity evoked by spontaneous blinks during the working memory delay period correlates with task accuracy, supports the idea that spontaneous blinking may serve a functional role in working memory. The localization of spontaneous blink related evoked activity to a region that is consistent with the location of eyelid/eyeball on the sensorimotor homunculus and including parietal cortex supports the idea that temporary memory may emerge from the coordinated interplay of circuits that include sensory-motor regions.
1. Liu et al, NeuroImage. 2020 Sep 1;218:116879
2. Ortega et al, bioRxiv 2021.03.15.435433; doi: https://doi.org/10.1101/2021.03.15.435433

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Poster

P807. Working Memory: Circuits

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Title: Measuring the spatiotemporal scale of prefrontal population codes for visuospatial working memory

Authors: *J. XIANG[1,2,3,6,7], M. ROUSSY[2,4,6,7], B. CORRIGAN[2,6,7], M. H. MOFRAD[3,6], R. LUNA[2,6,7], L. MULLER[3,6,7], J. MARTINEZ-TRUJILLO[2,6,7], M. MUR[1,5,6];
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Abstract: The lateral prefrontal cortex (LPFC) plays a key role in working memory (WM). Electrophysiological studies in monkeys show that LPFC neurons robustly encode visuospatial WM content. However, fMRI studies in humans report weak WM signals in LPFC. We hypothesize that the contradiction arises because the spatial topography of prefrontal neural population codes for WM is too fine to be effectively resolved by standard fMRI (2x2x2 mm^3, 1-2 s resolution).
To test this hypothesis, we analyzed spike data from two microelectrode arrays (Utah array, 4x4 mm^2, 10x10 electrodes spaced 0.4 mm apart) implanted on the left LPFC areas 9/46 of two adult male rhesus monkeys performing a visuospatial WM task in a virtual reality environment. During the task, the animals had to remember one of nine target locations. To assess the spatiotemporal scale of neural population codes, we applied 13 degrees of spatial (2D Gaussian kernels, 0.2-4.4 mm full-width-at-half-maximum [FWHM]) and 12 degrees of temporal smoothing (1D Gaussian kernels, 2-2048 ms sigma) to the data and assessed how this affected decoding of WM content. We estimated decoding accuracy using linear support vector machines with five-fold cross validation across trials within each measurement session. We performed statistical inference using t-tests and ANOVA with measurement session as random effect (11 sessions in animal B, 8 sessions in animal T; $p < 0.05$ after correction for multiple comparisons). Decoding accuracy for the ventral array was above chance (chance performance = 0.11) for all levels of spatial and temporal smoothing with mean accuracy up to approximately 0.5 (mean ± sd from 0.13 ± 0.003 to 0.49 ± 0.05 for animal B, and from 0.14 ± 0.009 to 0.34 ± 0.06 for animal T). Both spatial and temporal smoothing affected decoding accuracy, with larger effect sizes for temporal smoothing. Effects of spatial smoothing were more pronounced at higher levels of temporal smoothing. Best decoding accuracies were observed for low levels of spatial smoothing (0.4 mm FWHM) and high levels of temporal smoothing (> 512 ms sigma). Similar effects were observed for the dorsal array. The results indicate that coding of remembered stimuli is stable across time for at least a few hundred milliseconds, but not stable across space - channels with similar selectivity do not seem to cluster together.
Our findings suggest that fMRI signals integrating LPFC information across voxels of several millimeters will reach suboptimal performance. We suggest fMRI will be most sensitive to WM

signals encoded in LPFC population activity measured with voxel sizes in the submillimeter range. Future studies could experimentally confirm this using high-field fMRI.

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Poster

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Program #/Poster #: P807.03

Topic: H.05. Working Memory

Title: Sequence structure reorganizes memory items in orthogonal planes of working memory network and leads to recency effect

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Abstract: Abstract: When retaining a sequence of items in working memory, human subjects typically demonstrate recency effect in behavior, i.e., better memory performance for recently presented item compared to early items. Our recent studies further demonstrate that items located at different ordinal positions in the sequence tend to be stored in different latent states of the memory network and are represented in different neural formats. Furthermore, when subjects do not need to retain the sequence order of items, the recency effect is largely disrupted. Here we built and trained a recurrent network model to perform sequence working memory tasks, aiming to examine i) how memory items in the sequence are represented in different task contexts and ii) how recency effect would emerge from the neural representations. **Methods:** A 256-unit LSTM recurrent network was built and trained to perform working memory tasks. Specifically, a list of items were first serially loaded in the network, and in the sequence task, the network was trained to memorize items as well as their ordinal positions, whereas in the non-sequence task, the network only needs to memorize items without sequence requirements. Moreover, we examined the multivariate neural representations of items in the principal component space (the first three PCs) of the network, in sequence and non-sequence tasks, respectively. **Results:** (1) The recurrent network model shows recency effect in sequence task and disrupted recency effect in non-sequence task, consistent with human behavior. (2) In sequence task, memory items that are located at different ordinal positions of the sequence are stored in different PC planes that are orthogonal to each other, and recent item is in planes with higher variation. (3) In non-sequence task, although memory items are represented in different PC planes during the encoding epoch, they start to transform to the same PC planes during the maintaining epoch. **Conclusion:** When sequence structure needs to be retained in working memory (sequence task), each item in the

sequence is stored in distinct and orthogonal PC planes of the neural network, from which the recency effect emerges. When sequence structure is not needed to be retained (non-sequence task), items in the sequence are maintained in the same PC planes, leading to the disruption of recency effect.

Disclosures: T. Tang: None. **Y. Mi:** None. **H. Luo:** None.

Poster

P807. Working Memory: Circuits

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P807.04

Topic: H.05. Working Memory

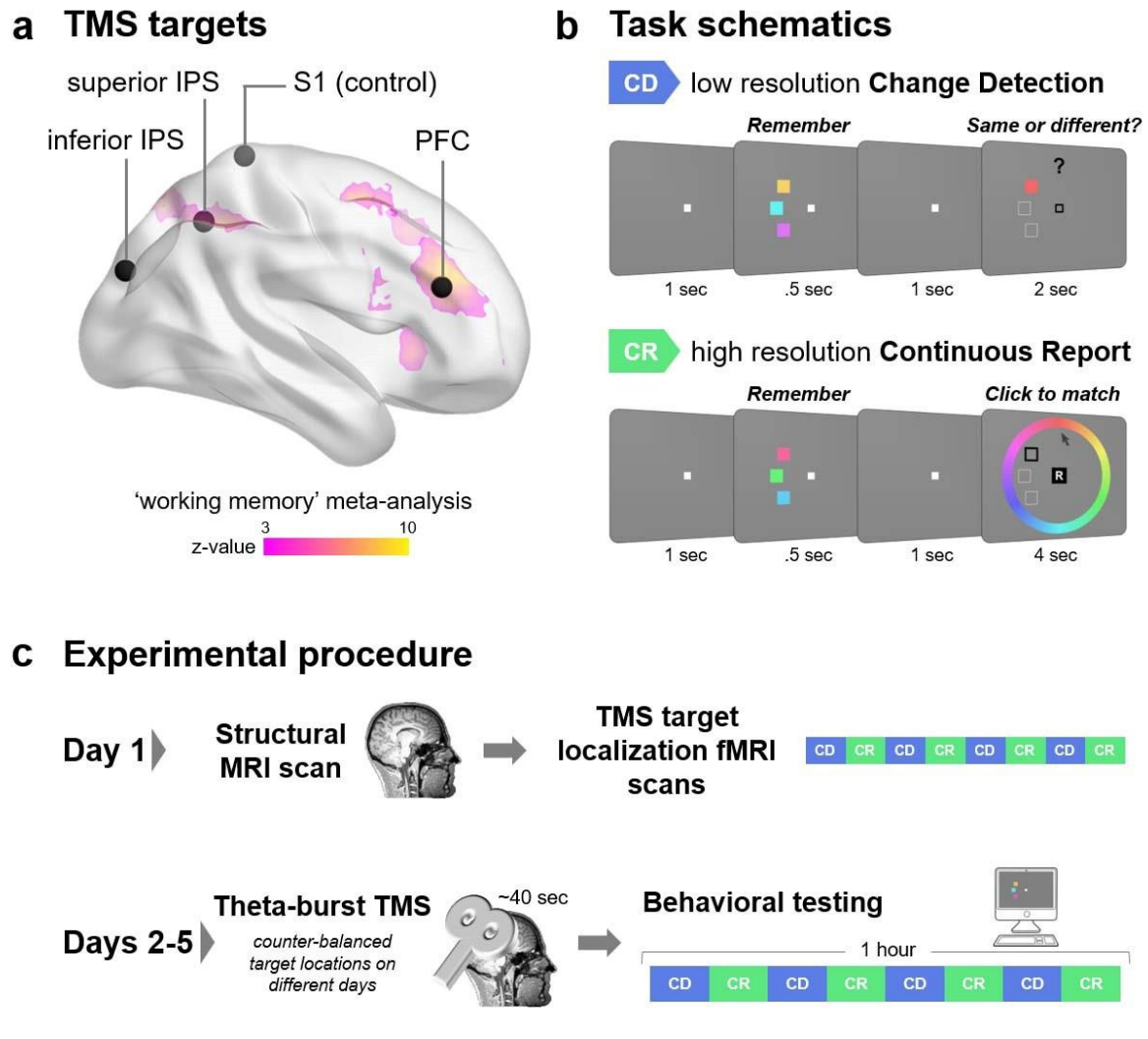
Support: NIH Grant MH063901
NIH Grant MH111204

Title: Dissociating the causal contributions of frontal and parietal cortex to working memory capacity

Authors: *J. M. SCIMECA, A. KIYONAGA, M. D'ESPOSITO;
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Abstract: Goal-directed cognition and behavior require the ability to keep information temporarily activated in mind. Decades of neuroimaging and neurophysiology research indicate that fronto-parietal (FP) cortex supports this working memory (WM) function. However, this research has been inconsistent as to the precise roles that these regions play in WM, and patients with FP damage often present clinically with relatively spared WM performance. To address these inconsistencies, we carried out a large-scale, multi-modal Registered Report using fMRI, TMS, and quantitative modeling of behavior. On Day 1, we used fMRI to localize subject-specific regions within frontal and parietal cortex (Fig 1a; N=120) that are involved in the performance of two canonical visual WM tasks (Fig 1b). On Days 2-5, we used transcranial magnetic stimulation (TMS) to perturb these regions and assessed the impact on several measures of WM function (Fig 1c; N=52). On each day, we applied continuous theta-burst TMS to either a baseline control site or one of three target sites: dorsolateral prefrontal cortex (PFC), superior intraparietal sulcus (IPS), or inferior IPS. Following TMS, subjects completed interleaved blocks of the two WM tasks. In interim analyses, we found that TMS to PFC and IPS largely spared WM function when task demands were low. However, with greater task demands and increased memory load, TMS produced greater overall impairment and a dissociable pattern across sites: IPS stimulation was more detrimental in a low-resolution (change detection) WM task while PFC stimulation was more detrimental in a high-resolution (continuous report) WM task. This causal manipulation therefore reveals distinct contributions of FP regions that only

emerge under heightened task demands. This study employs the rigorous research standards of the Registered Report format to help resolve ongoing debates about fronto-parietal cortex and WM function. Furthermore, this project provides a large, publicly available dataset of fMRI and TMS measures that can be leveraged to query the neurocognitive mechanisms of WM function.



Disclosures: J.M. Scimeca: None. **A. Kiyonaga:** None. **M. D'Esposito:** None.

Poster

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Program #/Poster #: P807.05

Topic: H.05. Working Memory

Support: NIH Grant R01MH063901

Title: Long-term learning transforms prefrontal cortex and medial temporal lobe activity patterns during working memory

Authors: *J. MILLER[1], A. TAMBINI[2], A. KIYONAGA[3], M. D'ESPOSITO[1];
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Abstract: The lateral prefrontal cortex (lPFC) is reliably active during working memory (WM) maintenance across human and animal models, but there is ongoing debate regarding the role of the lPFC in successful WM. For instance, non-human primate electrophysiology research finds that lPFC circuitry maintains WM representations (Leavitt et al., 2017), while human neuroimaging suggests that WM content is instead stored in sensory cortices (Serences, 2016). These seemingly incompatible accounts for WM are often confounded by differences in the amount of task training and stimulus exposure across studies, suggesting that long-term learning may influence the role of lPFC function in WM maintenance. Moreover, prior theories posit that WM function directly interfaces with representations and associations from long-term memory (LTM) (Fukuda & Woodman, 2017; Oberauer, 2009), predicting that such long-term training may bolster interactions between lPFC and medial temporal lobe (MTL) structures to facilitate WM. To test whether long-term learning influences (1) WM representations in lPFC and (2) the involvement of MTL in WM, we implemented a longitudinal functional MRI (fMRI) protocol in each of three human participants. Across three months, each participant was trained on (1) a serial reaction time (SRT) task, wherein complex fractal stimuli were embedded within probabilistic sequences, and (2) a delayed recognition task probing WM for trained or novel stimuli. This allowed us to track WM activity patterns over time and ask whether or how representations in WM are shaped by long-term associative learning. Participants showed learning benefits in the WM task for trained, but not novel, fractals. Pattern similarity analyses of WM delay activity demonstrate that across learning, item-level representations emerged within lPFC but not in sensory cortices or MTL. Single-item WM delay activity patterns in lPFC also reflected sequence information present in the SRT task even while not behaviorally relevant for WM. However, MTL involvement during the WM task did change across learning, as hippocampal activity increased over time in the WM probe period. Altogether, the results suggest that cortico-hippocampal interactions may emerge across learning to facilitate the comparison of information from WM and LTM. This work also shows that human lPFC develops stimulus-selective WM responses with learning and establishes novel evidence for long-term memory influences on WM maintenance. This influence of LTM over WM representations of stimuli in sequences likely reflects an integration process in lPFC that may emerge as a function of memory consolidation.

Disclosures: J. Miller: None. **A. Tambini:** None. **A. Kiyonaga:** None. **M. D'Esposito:** None.

Poster

P807. Working Memory: Circuits

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Program #/Poster #: P807.06

Topic: H.05. Working Memory

Support: ONR MURI N00014-16-1-2832
The MIT Picower Institute Innovation Fund

Title: Dynamic stability underlies cortical computations during working memory

Authors: L. KOZACHKOV[1], M. LUNDQVIST[1,4], J.-J. SLOTINE[2,3], *E. K. MILLER[1];
[1]Picower Inst. Learning Memory, [2]Dept. of Mechanical Engin., [3]Dept. of Brain and Cognitive Sci., MIT, Cambridge, MA; [4]Dept. of Psychology, Stockholm Univ., Stockholm, Sweden

Abstract: How does cortex keep its computations precise yet flexible? Recent theory has suggested that a variety of biologically plausible synaptic mechanisms—including anti-Hebbian plasticity and sparsity—endow neural networks with Dynamic Stability. Dynamic Stability is the ability of network activity to "stay in its lanes": be dynamic yet consistent, performing the same computations regardless of initial state and in the face of being perturbed by other activity, noise, distractions. In short, to be precise yet flexible. Here we test this theory by recording prefrontal cortical activity from rhesus macaque monkeys performing a distracted delayed-match-to-sample task (DDMTS). In an ordinary DMTS task, the animal must remember a sample item across a delay period. In DDMTS, 50% of trials have a mid-delay distractor item which the animal must ignore. The distractor acts as a perturbation to the cortex, from which it must recover in order to perform the task correctly. Theory predicts that the recovery from this perturbation will be *exponential*. Our findings indicate that this is indeed the case. Moreover, we find that the time constant of this recovery is consistent with previous time constants measured from prefrontal cortex in the literature—between 200 and 300 milliseconds. Finally, we train artificial neural networks (ANNs) to perform DDMTS while measuring their stability, as well as their similarity to the neural data recorded from the brain. Preliminary analyses show a tradeoff between network stability, task performance, and similarity to the brain. Too stable or too unstable and the ANNs cannot perform the task and they are dissimilar to actual neural activity. We found that there is an inverted U-shaped curve of Dynamic Stability. In the optimal range (not too much or too little Dynamic Stability) ANNs perform the task optimally and are most similar to the brain.

Disclosures: L. Kozachkov: None. **M. Lundqvist:** None. **J. Slotine:** None. **E.K. Miller:** None.

Poster

P807. Working Memory: Circuits

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Program #/Poster #: P807.07

Topic: H.05. Working Memory

Support: NIMH R56MH116007
CUNY ASRC Seed Grant (Round 4)

Title: Blink-related oscillations during the working memory delay period show a larger early evoked component compared to rest

Authors: *J. ORTEGA[1], C. REICHERT PLASKA[2], B. A. GOMES[3], T. M. ELLMORE[1]; [1]Psychology, The City Col. of New York, New York, NY; [2]City Univ. of New York, New York, NY; [3]Neurosurg., Cedars-Sinai Med. Ctr., Los Angeles, CA

Abstract: Blink-related oscillations (BROs) are a neurophysiological response associated with spontaneous eye blinking and are believed to be reflective of environmental monitoring processes. BROs are characterized by an early negative peak around 100 ms followed by a positive peak occurring around 300 ms. Research on the modulation of these oscillations during visual and auditory sensory stimulation suggests that blink-related processing adapts to accommodate differences in sensory environments. These findings led us to investigate BROs during other cognitive processes such as working memory (WM). We investigated 17 adults (mean age 22.8 years, 7 males) while they completed a Sternberg scene WM task. High-density EEG data was collected using a Brain Products active electrode recording system. Independent component analysis (ICA) was performed, and artifact components were removed. Eye blinks were recorded using an electrooculogram. Blink epochs were evaluated post-ICA to confirm successful artifact removal. Denoised data was band-passed filtered to the 0-4 Hz frequency range, segmented into 3 s epochs, and separated into three conditions: a delay condition of blinks occurring during the delay period, a task condition of all blinks during the task excluding delay periods, and a rest condition of blinks occurring during rest. We found BRO waveforms during the delay period exhibited a greater negative peak (-3.173 μV ± 1.6 amplitude, p = .014) amplitude compared to the rest condition (-1.727 μV ± 1.6 amplitude) at approximately 75-120 ms latency (N1) in the POz electrode. This difference was not present during the task condition. We also found a smaller positive peak amplitude during the task condition (2.458 μV ± 1.3 amplitude, p = .004) compared to the rest condition (4.1 μV ± 2.4 amplitude) at approximately 225-300 ms latency (P3) in the POz electrode. We investigated BROs modulation during a working memory task and found a smaller positive P3 for the task condition compared to the rest. This expected reduction of the P3 amplitude is believed to reflect reduced resources for processing blink-related information. We also observed a novel finding: N1 amplitude during the delay condition exhibited a greater negative peak compared to the rest condition. BROs have been found to activate the parahippocampal gyri[1], a critical region involved in WM functions. Thus, our findings suggest that this early evoked component of BROs may play a role in WM.1.Liu, C. C. et al. Hum. Brain Mapp. 2019 Feb 1; 10.1002/hbm.24378

Disclosures: J. Ortega: None. **C. Reichert Plaska:** None. **B.A. Gomes:** None. **T.M. Ellmore:** None.

Poster

P807. Working Memory: Circuits

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Program #/Poster #: P807.08

Topic: H.05. Working Memory

Support: R01 MH121480
FBI Seed Funds

Title: Functional heterogeneity of prefrontal oscillations in a self-ordered working memory task

Authors: *F.-K. CHIANG, E. L. RICH;
Nash Dept. of Neuroscience, Friedman Brain Inst., Icahn Sch. of Med. at Mount Sinai, New York, NY

Abstract: Working memory (WM) is the ability to manipulate information temporarily held in mind, and is critical to cognitive processes such as planning and reasoning. The prefrontal cortex (PFC) is important to the processes of WM, but it remains unclear whether there is functional heterogeneity among different subregions of PFC. To assess this, we trained monkeys to perform a sequential self-ordered target selection task and recorded from arrays chronically implanted in the dorsolateral prefrontal cortex (DLPFC, 128 channels) and frontal eye field (FEF, 128 channels). On each trial, monkeys were presented with a target configuration consisting of eight identical filled circles. The task required monkeys to saccade to each target, one at a time in any order, returning their eyes to the center after each selection. The same target configuration was presented repeatedly for every selection in a block of 40 trials, and different target configurations were used in six blocks per recording session. Within a trial, reward was delivered only once for each target and target revisits were not rewarded. Therefore, monkeys had to use WM to track which targets had been visited and prepare for the next target selection. Local field potentials (LFPs) were aligned to fixation and reward epochs across correct saccades, and preliminary data revealed specific modulations as a function of saccade order. Increased alpha/beta (10-25 Hz) activity was commonly observed in both DLPFC and FEF during the fixation epoch prior to the first target selection of a trial, when WM was not yet required, suggesting alpha/beta activity is involved in attending to a configuration at the beginning of the new trial. Theta (4-8 Hz), but not alpha/beta, in DLPFC, but not FEF, increased during the reward epochs and following fixation epochs after correct target selections, suggesting that DLPFC theta is important for updating or retrieving target-reward contingencies held in mind. Lastly, alpha/beta activity in FEF was observed across saccades during fixation epoch, suggesting this signal represents attention or saccadic preparation. These results indicate that LFPs in DLPFC and FEF are distinct and dynamically modulated during information updating and response selection in a self-ordered WM task.

Disclosures: F. Chiang: None. **E.L. Rich:** None.

Poster

P807. Working Memory: Circuits

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Program #/Poster #: P807.09

Topic: H.05. Working Memory

Support: NIH grants R01 MH117996
NIH grants R01 MH116675

Title: Prefrontal Neural changes in Early Adolescence related to working memory

Authors: *J. ZHU[1], C. M. GARIN[1], X. QI[2], A. W. LODISH[2], L. SILENZI[2], E. A. KATTNER[2], G. D. MYATT[2], T. R. STANFORD[2], E. SALINAS[2], C. T. WHITLOW[3], B. LUNA[4], C. CONSTANTINIDIS[1];
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Abstract: Humans and nonhuman primates experience a protracted period of cognitive development that extends through adolescence and parallels the maturation of the prefrontal cortex (PFC). Little is known about changes in prefrontal neurons activities over this time period. To address this question, we tracked developmental measures, behavioral performance, neurophysiological activity in the PFC, and neuroanatomical changes evaluated with structural MRI as monkeys transitioned from puberty into adulthood. Eight macaques (2F, 6M *Macaca mulatta*) were trained to perform variations of the Oculomotor Delayed Response (ODR) task, and we recorded neural activity at the ages of 3.4±0.2, 4.0±0.3, 4.4±0.2 and 4.9±0.3 yrs. Eight animals performed the ODR task with a 1.5 s delay, while four animals also performed a version of the task with a longer delay period, and the ODR + distractor task. The monkeys performed the ODR task robustly in adolescence, with an average performance in the ODR task for 8 animals at 83%. Between the time points, modest improvement in performance in both tasks was observed, with greatest improvement for the most difficult tasks and conditions. Single neuron activities were collected from dorsolateral PFC. Anatomical localization of the recording sites was based on MRI of each animal. An increase in the percentage of neurons that responded to the task (35%, 45%, 58%, 57% respectively) was observed in the 4 time points. Robust cue and delay period activities were present across time points, with relatively stronger activities in later stages. In parallel we performed MRI that measured structural volumetric changes (T1) and microstructural changes (Neurite Orientation Dispersion and Density Imaging (NODDI)). Substantial increase of gray matter volumes and more specifically in frontal, parietal and occipital lobe was detected until the age of ~3.4 yrs. before plateauing, and white matter

measures display a linear increase without plateau detected up to ~5.3 yrs., showing an extended volumetric and microstructural development. Our results provide insights on prefrontal neuronal activity during working memory at the earliest time points achieved. Tracking of the cohort is ongoing allowing us to characterize how behavior, prefrontal activity and cortical structure change in these animals as they enter adulthood so as to account for enhanced executive functions.

Disclosures: J. Zhu: None. **C.M. Garin:** None. **X. Qi:** None. **A.W. Lodish:** None. **L. Silenzi:** None. **E.A. Kattner:** None. **G.D. Myatt:** None. **T.R. Stanford:** None. **E. Salinas:** None. **C.T. Whitlow:** None. **B. Luna:** None. **C. Constantinidis:** None.

Poster

P807. Working Memory: Circuits

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P807.10

Topic: H.05. Working Memory

Title: Functional and microstructural connectivity patterns predict working memory performance in older adults

Authors: *E. GOZDAS, B. AVELAR-PEREIRA, H. HOSSEINI;
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Abstract: Introduction: Working memory enables the temporary storing of crucial information necessary for a wide range of cognitive tasks and is also a core component of age-related cognitive decline. Neuroimaging research over the past decades has improved our understanding of the neural substrates of working memory. However, the field lacks clarity on whether a specific neural architecture predicts item selection in working memory performance. Here, we integrated network neuroscience and predictive modeling with neurite imaging and functional MRI (fMRI) to identify microstructural and functional connectivity patterns that best predict working memory performance in older adults engaged in a classic working memory task.**Methods:** Seventy participants (age range 65-85 years, 37 female) were recruited for the study.The Sternberg working memory task fMRI (8 min.) and multi-shell diffusion MRI (dMRI) data were acquired for all participants. We preprocessed fMRI and dMRI data using standard procedures. Neurite orientation dispersion and density imaging (NODDI) were then computed using the preprocessed dMRI data to estimate neurite density (ICVF) and neurite complexity (ODI). The functional and neurite microstructural connectivity were computed for all pairs of nodes defined in the HCP brain atlas. Further, we adapted the connectome-based predictive modeling (CPM) method to identify functional and neurite microstructural connectivity patterns that predict working memory in older adults using leave-one-out cross-validation.**Results:** Significant correlations emerged between the predicted and observed working memory

performance (r=0.34, p=0.01) using functional connectivity. The functional connectivity pairs that contributed most to the prediction model were the lateral temporal cortex- posterior cingulate cortex, anterior cingulate and medial prefrontal cortex- posterior cingulate cortex, orbital and polar frontal cortex- posterior cingulate cortex. For the model trained on microstructural networks, stronger correlations were found between predicted and observed performance using ICVF (r=0.44, p=0.001) and ODI (r=0.59, p<0.001). The microstructural connectivity pairs with highest contribution to the model were the anterior cingulate and medial prefrontal cortex, and orbital and polar frontal cortex.**Conclusions:** We have demonstrated that specific functional and microstructural connectivity patterns predict decision in a working memory task in older adults. These results provide early evidence regarding the neural architecture predicting working memory performance in older adults above and beyond functional connectivity.

Disclosures: E. Gozdas: None. **B. Avelar-Pereira:** None. **H. Hosseini:** None.

Poster

P818. Prefrontal Networks: Dorsolateral Prefrontal Cortex

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P818.01

Topic: H.08. Learning and Memory

Support: NSF NCS 1631563

Title: Virtual dissection and comparative connectivity of the cortico-cerebellar network in chimpanzees

Authors: *A. N. PARKS[1], K. L. BRYANT[2], J. SMAERS[1], E. E. HECHT[3];
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Abstract: Chimpanzees (*Pan troglodytes*), along with bonobos, are the closest living relatives of humans. As such, chimpanzee brains offer critical insight into the evolution of the neural substrates that underlie human cognition. In recent years, it has been demonstrated that the lateral cerebellar hemispheres play an integral role in mediating cognitive function by virtue of their expansive connectivity network with the neocortex, the cortico-cerebellar system (CCS). Furthermore, the CCS contains topographically-organized connections between specific lobules of the posterior lobe of the cerebellum and heteromodal neocortical regions, including the prefrontal and posterior parietal cortex. However, while quantitative comparisons of the CCS between humans and chimpanzees would glean insight into the evolution of the neural correlates of human cognitive function, these bidirectional connections between cerebellar lobules and their

neocortical counterparts have yet to be quantified in chimpanzees. Here, we report quantitative connectivity of the chimpanzee cortico-cerebellar system by performing a virtual dissection of CCS tracts using diffusion tensor imaging (DTI). A DTI sequence was applied at 3 Tesla using diffusion-sensitizing gradients in 64 directions with 5 averages in each of 2 phase encoding directions for 67 chimpanzee individuals. White matter tracts were reconstructed by applying a multiple ROI (region of interest) tractography technique. Masks for lobules VI though IX of the posterior cerebellar lobe were generated in template space. Next, masks were registered to native DTI space using ANTS for each of the individual subjects. Tractography was performed in native space using bedpostX. Thresholded results were then binarized, registered to template space, summed, and then averaged to produce composite images where the intensity value at each voxel corresponds to the percent of subjects with above-threshold connectivity at a given voxel location. Similarities and differences with human cortico-cerebellar connectivity patterns are described. These results may help inform our understanding of brain-behavior evolution in the hominid lineage.

Disclosures: A.N. Parks: None. **K.L. Bryant:** None. **J. Smaers:** None. **E.E. Hecht:** None.

Poster

P818. Prefrontal Networks: Dorsolateral Prefrontal Cortex

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P818.02

Topic: H.08. Learning and Memory

Title: Characterization and manipulation of Corticothalamic neurons in associative cortices using Syt6 Cre transgenic mice

Authors: *L. VAASJO[1], X. HAN[1], A. TIEMROTH[2], A. N. THURMON[2], A. FIGUEROA[2], R. REYES[2], P. FELICIANO-RAMOS[4], M. GALAZO[3];
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Abstract: Cortico-thalamic interactions between associative cortices and higher-order thalamic nuclei are involved in high-cognitive functions, such as decision-making and working memory. Corticothalamic neurons (CTn) in the prefrontal cortex and other associative areas have been much less studied than their counterparts in the primary sensory areas. The availability of characterized transgenic tools to study CTn in associative areas will facilitate their study, and contribute to overcome the scarcity of data about their properties, network dynamics, and contribution to cognitive functions. Here, we characterized the Syt6-Cre (KI148Gsat/Mmud) transgenic mouse line. In this line, Cre is strongly expressed in the prefrontal, motor, cingulate, and retrosplenial cortices, as well as in other brain areas including the Cerebellum and the

Olfactory tuberculum. Cortical expression starts embryonically and reaches the adult expression pattern by postnatal day 15. In the cortex, Cre is expressed by layer 6 CTn, and by layer 5 CTn to a lesser extent. We quantified Syt6-Cre + CTn axon varicosities to estimate the distribution and density of putative corticothalamic driver and modulator inputs to thalamic nuclei in the medial, midline, intralaminar, anterior, and motor groups. Also, we characterized the effect of optogenetic stimulation of Syt6-cre-CTn in the activity of the prefrontal cortex. CTn stimulation in the prefrontal cortex induces an oscillatory activity in the local field potential that resembles the cortical down-states typically observed during slow-wave sleep or quiet wake.

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Poster

P818. Prefrontal Networks: Dorsolateral Prefrontal Cortex

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Program #/Poster #: P818.03

Topic: H.08. Learning and Memory

Support: National Multiple Sclerosis Society Research Grant (RG-1507-04951)
National Institute of Neurological Disorders and Stroke of the National Institutes of Health (F31NS115361)

Title: Dynamic Functional Connectivity Between the Cerebellum and Dorsolateral Prefrontal Cortex Explains Variability in Memory Ability in Multiple Sclerosis

Authors: *M. ZUPPICHINI[1], D. SIVAKOLUNDU[4], K. WEST[2], D. OKUDA[5], B. RYPMA[3]; [1]Behavioral and Brain Sci., Univ. of Texas At Dallas, DALLAS, TX; [2]Ctr. for BrainHealth, [3]Behavioral & Brain Sci., Univ. of Texas At Dallas, Dallas, TX; [4]Dept. of Neurol., Yale Sch. of Med., New Haven, CT; [5]Dept. of Neurol. & Neurotherapeutics, Univ. of Texas Southwestern Med. Ctr., Dallas, TX

Abstract: Dynamic Functional Connectivity Between the Cerebellum and Dorsolateral Prefrontal Cortex Explains Variability in Memory Ability in Multiple Sclerosis
Authors: Mark D. Zuppichini, Dinesh K. Sivakolundu, Kathryn L. West, Darin T. Okuda, and Bart Rypma
Background: Network connectivity is disrupted in multiple sclerosis (MS) and is related to cognitive function. Memory impairment is common in MS yet the underlying neuropathology is still unknown. Research suggests both dorsolateral prefrontal cortex (dlPFC) and cerebellar (Cb) involvement in mnemonic function, and the cerebellum is known to be adversely affected by MS. Dynamic resting-state functional connectivity, or the time-varying connectivity, is strongly related to cognition in network neuroscience. Thus, investigating the role that Cb dysfunction

plays in MS-related abnormal dynamic connectivity and memory impairment may aid in understanding MS-related memory impairment. **Objective:** To investigate the relationship between dlPFC-Cb dynamic connectivity and memory impairment in MS.**Methods:** 32 MS and 20 healthy controls (HC) participants completed the Digit Span Test – Forward as a measure of memory and underwent magnetic resonance imaging. Dynamic resting-state functional connectivity analysis was used to assess functional and structural connectivity. **Results:** The MS group (M=7.12, SD=1.29) performed comparably to the HC group (M=7.65, SD=1.23) on the DST-F (t=1.47, p=.147). Analysis of dlPFC-Cb connectivity revealed differences in the relationship between dynamic connectivity and digit-span ability across groups. In the MS group, connectivity between the left dlPFC and Cb explained 15% of the variance in digit-span variability after controlling for age and education (F(1,28)=6.63, p=.016, ΔR^2=.15), and connectivity between the right dlPFC and Cb explained 13% of the variance in digit-span variability after controlling for age and education (F(1,28)=5.17, p=.031, ΔR^2=.13). In the HC, connectivity between the left (F(1,14)=2.43, p=.142, ΔR^2=.12) and right (F(1,14)=0.00, p=.949, ΔR^2=.00) dlPFC and Cb did not significantly explain any variance in the digit-span.**Conclusion:** Dynamic resting-state connectivity between the cerebellum and dlPFC is critical for the maintenance of memory function observed in MS. **Keywords:** Network connectivity; cerebellum; memory; resting-state connectivity; prefrontal cortex.

Disclosures: M. Zuppichini: None. **D. Sivakolundu:** None. **K. West:** None. **D. Okuda:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Biogen. F. Consulting Fees (e.g., advisory boards); Celgene, EMD Serono, Genentech, Genzyme, Novartis. **B. Rypma:** None.

Poster

P818. Prefrontal Networks: Dorsolateral Prefrontal Cortex

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Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P818.04

Topic: H.08. Learning and Memory

Title: The role of mPFC projecting populations in recent and remote expression of memories

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Abstract: As memories age, they undergo reorganization at the systems level. In particular, memories are known to initially depend on the hippocampus for expression, but become more dependent on the medial prefrontal cortex (mPFC) at remote time-points. Although retrieval-induced activity of mPFC neurons increases with time, it is unclear whether distinct mPFC projecting populations contribute to retrieval at different time points. Here, we aimed to delineate

how these distinct projection-specific populations of mPFC neurons contribute to memory expression at recent vs. remote time-points. To examine this, we virally labeled projection-specific populations in the mPFC and examined the extent to which these distinct mPFC populations are activated during contextual fear learning (i.e. encoding), and subsequently reactivated during recent vs. remote recall. Then, we used an activity-dependent tagging method to express excitatory or inhibitory opsins in these projection-specific mPFC populations during learning, and asked whether reactivating them is sufficient for memory expression at recent and remote time-points in a novel context. Conversely, we asked if inhibition of these populations can impair recall at recent or remote time-points. Our results provide evidence that different mPFC projection-specific populations differentially contribute to recent vs remote recall. In particular, basolateral amygdala (BLA) projecting neurons are reactivated at both recent and remote time-points, while Nucleus Reuniens (NRe) projecting population exhibited increased reactivation at the remote compared to recent time-point. Consistently, the manipulation of BLA projecting sub-population affected memory expression at both time-points while manipulation of NRe projecting sub-population affected only remote recall. Furthermore, the excitation of only the mPFC-NRe circuit decreased hippocampal activity in CA1 region at the remote time point, whereas inactivation of this circuit increased of CA1 activity at the recent time point. Together, our results suggest a model for temporal changes in brain circuits that express the memory. During expression at recent time-point, a limited engagement of mPFC is observed (e.g. mPFC to BLA projecting population) while at remote time-point the mPFC's engagement is enhanced by involvement of other projecting populations (e.g mPFC to NRe projecting population). This enhanced engagement is associated with decreased activity in CA1 region at remote time-point which suggests a top-down regulatory mechanism by mPFC on hippocampus during remote recall of memories.

Disclosures: A. Golbabaei: None. **S.A. Josselyn:** None. **P.W. Frankland:** None.

Poster

P818. Prefrontal Networks: Dorsolateral Prefrontal Cortex

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Title: Medial prefrontal cortex flexibly links stimuli and conditioned responses through behavior-dependent sensory encoding

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Abstract: To survive in challenging environments, animals need to learn to perform adaptive behaviors in the presence of sensory information that predicts threats or rewards. The medial prefrontal cortex (mPFC) has been suggested to link sensory and motor aspects of such associations during learning and behavior execution. During learning, sensory-driven prefrontal responses emerge as stimuli gain behavioral relevance, and it has been shown that manipulation of mPFC activity can drive and inhibit the execution of learned behaviors. Yet, how prefrontal circuits map relevant sensory signals onto specific behaviors is still unclear. Here we investigate the involvement of mPFC in mediating such mappings using aversive auditory conditioning paradigms with dynamic stimulus-response associations. In addition to using a classic fear conditioning paradigm, we developed novel active avoidance experiments in which we vary the predictive stimuli and the instrumental behaviors induced by those stimuli by manipulating task contingencies. During these paradigms, we used miniaturized microscopes for calcium imaging in freely behaving mice to track the activity of mPFC neurons throughout learning. To isolate the relation between individual task variables and the recorded neural activity, we employed a model-based analysis approach, which allowed us to quantify coding changes over the course of an experiment. We find that neural correlates of sensory and behavioral signals are encoded by independent subpopulations. While tone-evoked responses are tightly linked to behavioral relevance, they do not directly correlate with behavior execution. Yet, how the identified tone signals are encoded depends on the associated tone-induced behavior. Specifically, changing the behavior that is induced by a given tone modulates the tone's representation through coordinated changes of the tone responses of individual cells. Conversely, the representations of different tones show increased similarity when the tones are associated with the same behavior. These results suggest that mPFC implements abstract stimulus-response mappings through behavior-dependent sensory representations, providing a flexible link between sensory information and conditioned responses. Our findings are thus consistent with a model in which mPFC provides contextualized information to downstream areas, rather than directly driving specific behaviors.

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Poster

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Title: The geometry of category and choice representations in human ventromedial prefrontal cortex.

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Abstract: The ventromedial Prefrontal cortex (vmPFC) has been studied extensively as one of the neurophysiological substrates that supports social and affective processing, and reward-based decision making. This prevailing sentiment naturally gives rise to the question: what, if anything, is then encoded and processed if the decisions being made are not socially relevant or value-based? To answer this question, we conducted single-unit recordings in human patients implanted for clinical purposes who were instructed to alternate between two tasks, one involving a sensory-perceptual decision (Category Task) and the other involving a memory-dependent decision (Memory Task). Transitioning between the Category and Memory tasks involved a screen-wide cue and question related to the presented stimulus: either membership to target category or novelty of the stimulus, to which the patient gave a yes/no response using either a button-press or a directed saccade. We recorded 320 well-isolated single neurons from the vmPFC of 13 patients performing this switching task. We found that both abstract categorical representations and choice representations exist in single vmPFC units, allowing for this information to be decoded with varying degrees of representational invariance across tasks during stimulus presentation. Furthermore, our task allowed for comparison of task-relevant signals during baseline periods lacking visual stimulation. We identified representations of contextual variables such as target category and response modality encoded in baseline vmPFC neuron firing rates that did not generalize across tasks, suggesting an orthogonalized representation like concurrently recorded neurons in medial temporal and frontal cortices (MTL/MFC respectively). In contrast to MFC, we found that switching between tasks led to an increase in contextual information at baseline. Neurons in the vmPFC also exhibited task-modulated correlations with MTL and MFC, supporting the emergence of task-relevant neuronal ensembles whose synchronization supports proper task execution. In summary, the vmPFC is highly active, encoding a mixture of task-relevant information which, in conjunction with other regions of cortex and subcortex, supports flexible behavior when switching between tasks.

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Poster

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Topic: H.08. Learning and Memory

Support: PRONEX (FAPEMIG: APQ-00476-14).

Title: The role of prefrontal neurons in memory and learning in the barnes maze apparatus

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Abstract: For over decades the means for how learning and memory is processed and consolidated within the brain has been discussed and studied. However, only in the past decades has science advanced to technologies that allow precise spatial and temporal observation of neuronal activation patterns to specific types and circuits, while also observing behavioural traces. Here we investigate the dynamics of excitatory and inhibitory neurons in the mPFC of mice while undergoing a learning and memory task (Barnes Maze (BM). Groups: a total of 11 mice (c57bl/6) were used. 6 mice expressed cre-protein in vGlut1 positive neurons, while 5 other mice presented cre-protein expression in Gad2 positive neurons. Surgery: all mice received a viral injection of a jGCamP6 construct and fiber stub implantation at the mPFC (M.L.: +0.35, A.P.: +2.4, D.V.: -1.8) and were then allowed to rest for 20 days. BM: All animals were submitted to the BM protocol: animals were placed in a round platform, with 1 true and 19 false escape holes, and were then allowed to explore the apparatus for 2-3 minutes, over 6 days, 3 trials/day. Daily activities were as followed: 1 day of habituation, 4 days of learning and 1 day of memory. BM protocol objective is a search and locate true escape hole task, in which training results in fewer mistakes (wrong hole investigation), traveling smaller distances, faster execution and strategy adapting. Fiber Photometry (FP): while performing the task, calcium surges were detected through FP at a 500-540 nm window for both 405 (10µW) and 465 (30µW) excitatory inputs. Data was acquired at 12kbps and then processed and analyzed with the z-score $\Delta f/f$ method. Behavioral events were set as a TTL input for calcium surges overlap. All animals presented improved performance, with smaller errors, distance traveled and latency to task compared to initial days. In addition, both groups showed a shift in search strategy, from random (low efficiency), to serial (medium efficiency) and direct (high efficiency). All behavioral events

above were analyzed and paired to calcium surges. Initial data found no specific surges correlated to specific events, although at some days we found significant differences for excitatory neurons in total number of calcium surges during daily tasks, and number of surges within correct hole investigations, compared to previous days, which may suggest a change in calcium dynamic based on learning. There are yet many experiments and analysis to be performed, in addition to tweaks to be done for both FP rig setup and BM.

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Poster

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Title: Neuronal activity in the lateral prefrontal cortex during cued object selection

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Abstract: To investigate object learning, retrieval and selection in the lateral prefrontal cortex (LPFC), two monkeys were trained in a cued selection task. A semi-chronic system allowed recording of neuronal activity across large areas of the dorsal and ventral LPFC. The task involved a total of eight objects, divided into two four-object sets associated with different color cues. Each session consisted of a series of problems, each consisting of four cycles of trials. In each problem, two objects were defined as targets, one from each set. On each trial, the animal was required to gaze at the red central fixation point (fp) followed by a cue in the centre with six black squares in a circular array (cue phase). Then the six black squares were replaced randomly with six objects consisting of three objects from each set (visual search phase). The red fp turned cyan, and the animal was required to select one object by making a saccade to it (selection phase). If the object selected on the current trial was a target, this was followed by its associated color cue and reward; if it was a nontarget, the response led to a negative feedback signal and no reward (feedback phase). In cycle 1 of each problem, the cue was dark grey and the animal had to work through a series of trials, sampling objects at random until the two targets were discovered. In subsequent cycles, the trial began with a color cue and the animal had to select its associated target. Trials continued in each cycle until each target had been selected once, after

which the next cycle began. After all four cycles, new target objects were defined for the next problem. In both animals, behavioral performance improved cycle by cycle to more than 90 % correct in cycle 4. In ventral LPFC, across cycles 2-4, population activity encoded the presented cue color, followed by gradual emergence of encoding for the associated target object (retrieval). In the search phase, location coding emerged with a similar time course in the two regions. Following feedback (learning), cue and object information reappeared in ventral LPFC, though with no cross-generalisation to the codes earlier in the trial. The results suggest the ventral LPFC integrates relevant information to control object learning and retrieval, coordinating with dorsal LPFC to guide search and selection.

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Poster

P874. Cognitive Natural Aging: Behavior

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Topic: H.12. Aging and Development

Support: NIH/NIA Grant 5R01AG055500

Title: Global functional connectivity of the cognitive control network predicts task-switching performance in older adults

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Abstract: Aging populations show declining performance in tasks that require recall and switching between multiple rule sets, commonly referred to as task-switching. Evidence suggests that age-related neurodegeneration disrupts white matter connectivity contributing to poor connections in large-scale networks such as the Cognitive Control network (CCN), which is believed to be critical for executive functions involved in task-switching. However, no research has directly examined the relationship between task-switching performance and CCN connectivity. Aging research using variants of switching tasks has shown an increased cost (i.e., Mixing Cost) when adding rule sets, but it is unknown how the underlying neural mechanisms are taxed (Verhaeghen 2011; Wasylyshyn et. al., 2011; Nashiro et. al., 2018). Studies using Global Functional Connectivity (GFC) from resting-state functional magnetic resonance imaging (rs-fMRI) have shown a positive relationship between GFC and fluid intelligence. These data suggest GFC in regions and networks supporting cognitive control is sensitive to individual differences in fluid cognitive ability in adults (Cole et al., 2012; Franzmeier et al., 2017). The present study bridges these two fields by examining whether GFC in the CCN predicts individual

differences in Mixing Costs in older adults. The sample included behavioral and rs-fMRI data from 47 neurotypical older adults who performed a modified task-switching paradigm consisting of switching conditions that reflect low (separate) and high (overlap) response rule overlap to magnify individual differences in Mixing Cost. We predicted that overlap in response rules would increase the difficulty to recall and manage multiple task rules, resulting in larger Mixing Costs (Mayr 2001). As in previous work with older adults (Franzmeier et al., 2017), the CCN was operationalized as the frontoparietal control network in the Yeo7 network parcellation. Consistent with predictions, older adults have greater Mixing Cost in the overlap compared to the separate condition. We also show that greater GFC in the CCN predicts lower Mixing Cost, an effect amplified by the overlap condition. Therefore, the more functionally integrated the CCN is with the rest of the brain, the better older individuals were able to maintain and switch between multiple, overlapping rule sets. This unique relationship to Mixing Costs and the CCN suggest a specialized mechanistic role for CCN integration in the aspects of task-switching that are affected most with cognitive aging.

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Poster

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Topic: H.12. Aging and Development

Support: Pomona College research grant

Title: Neurocognitive differences between older professional and amateur orchestral musicians

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Abstract: Musical training can contribute to plastic changes in the brain. While there is evidence of increased cognitive functioning in musicians compared to non-musicians, differences in cognition between professional and amateur musicians have not been explored. The cognitive and neurological benefits of music may be more evident in amateur musicians compared to professional musicians due to certain risk factors. The current study examined cognitive functioning of older professional orchestral musicians compared to older amateur orchestral musicians. Musicians (at least 65 years old) were recruited from various professional and community-based orchestras. Participants' (20 professional and 21 amateur musicians) demographics, health status, and musical background were collected using Qualtrics. The CNS Vital Signs (CNSVS) Remote Test was used to assess cognitive functions. Using the CNSVS

Remote Test, domain scores from several neuropsychological tests were compared between professional and amateur musicians. For processing speed, professional musicians showed significantly lower scores than amateur musicians ($p < 0.05$). When comparing musicians age 65-69, professional musicians had significantly lower processing speed, verbal memory, and lower composite memory compared to amateur musicians ($p < 0.05$). When looking at highest level of education, professional musicians with professional degrees had a significantly greater psychomotor speed compared to professional musicians with 4 year degrees ($p < 0.05$). Additional results show that female professional musicians had significantly lower processing speed compared to female amateur musicians. Male professional musicians had significantly higher motor speed compared to male amateur musicians ($p < 0.05$). Our results show that the CNSVS test reveals differences in the cognitive abilities between older professional and amateur orchestral musicians. Age, sex, and education level differences also attribute to differences in test performance within and between the musician groups, while ethnicity does not. This information can be utilized to understand how musical training can affect the development of neurodegenerative diseases such as Alzheimer's disease.

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Poster

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Topic: H.12. Aging and Development

Support: National Institutes of Health Grant P01- AG055365

Title: Pupil diameter indexes time-course shifts in older adults' sustained listening effort

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Abstract: Understanding speech in noisy environments is a key component of everyday life and requires effortful listening. Older adults' listening effort may be especially affected by noise, arising in part from a reduced ability to effectively track a target speaker among background speakers across prolonged time periods. The present study used 60-second audiobook segments to evaluate how different listening situations influence older adults' sustained effort when listening to meaningful speech. This experiment investigated the effect of 1) different signal-to-noise ratios (SNRs: quiet, 0dB, or -6dB) and 2) number of passage exposures (three times each in

the two noisy conditions) on listening effort, for 13 older adults with clinically normal hearing. We hypothesized that poorer SNRs would result in increased listening effort and that repeated exposures would result in decreased listening effort. While pupillary measures were recorded, participants listened to the audiobook segments, with larger pupil dilations indicating greater listening effort. Generalized additive mixed model (GAMM) results revealed that listeners showed evidence of increased listening effort in both noisy conditions relative to quiet. In the noisy conditions, participants exhibited decreased listening effort in the second, compared to the first, presentation of a passage. Furthermore, listening effort varied nonlinearly with listeners' subjective intelligibility ratings across SNR blocks. In comparison to the quiet condition, listening effort in the 0dB condition was associated with higher self-reported intelligibility ratings, suggesting that engaging in effortful listening only benefited perceived intelligibility when the speech was at least moderately challenging. The more difficult -6dB condition revealed dynamic changes in listening effort over time; higher intelligibility ratings were associated with increased listening effort at the beginning of the passage that decreased over time. The opposite effect was found at lower intelligibility ratings; initially participants showed low listening effort that increased over time. Taken together, our findings illustrate how SNR and repeated exposure influence listening effort when listening to sustained, continuous speech, and how effort can deviate from intelligibility in some listening conditions. Acknowledgement: The views expressed in this abstract are those of the author and do not reflect the official policy of the Department of Army/ Navy/Air Force, Department of Defense, or U.S. Government.

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Poster

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Topic: H.12. Aging and Development

Support: NIA U01AG062371

Title: Cognitive Enhancement Training in Older Adults Improves Attention and Processing Speed

Authors: L. CHUKOSKIE[1], R. SPENCE[2], T. SIMMONS[1], A.-M. ENGLER[3], S. HACKER[3], L. HILL[4], Y. TAKARAE[6], *J. SNIDER[1], J. TOWNSEND[5];
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Abstract: In a 10-year follow-up NIH ACTIVE studies have shown that older adults trained to improve speed of processing and attention had a 29% decreased rate of dementia compared to a no-contact control group. We have completed a game-based training to enhance these cognitive functions in healthy older adults that resulted in improved cognitive function with some transfer to practical function. Methods: Previous ACTIVE studies required training in lab. Our games are gaze-driven which forces full engagement despite being played independently at home. Gaze is an observable indicator of attention focus and a powerful tool for training attention since eye movement and spatial attention recruit highly overlapping neural circuitry. We supplied participants with the intervention laptop and eye tracker. Training games were designed with embedded principles to improve processing speed and attentional control, including resistance to distraction and inhibitory control. Participants played 20-30 minutes a day five times per week for 12 weeks. We monitored compliance over a secure remote connection. Primary outcome measures were processing speed and selective attention. Additional measures were of general cognitive and practical function. Results: Attention distractibility improved significantly with training as did processing speed. Additional evidence for reduced attention distractibility was improved inhibition of eye movement in an anti-saccade task. A wait-list control showed no change in any of the assessed outcome measures following a wait period of 12 weeks but showed improved attention and processing speed following the subsequent 12-weeks of cognitive training. A 12-18-month follow-up assessment of 14 early participants found that gains in selective attention were stable after a year, but improvements in processing speed were reduced. After 2-weeks of booster training, however, improvements in processing speed were increased to previous post-training levels. We saw no change in general cognitive function, but performance on a driving simulator task that required processing of peripheral visual cues improved. Conclusions: We have developed an approach that successfully trains foundational attention and processing skills in older adults with some transfer to practical function. If successful in larger trials with appropriate controls, this approach would provide an inexpensive and broadly available tool that would improve cognitive function and that may have the potential to decrease risk of dementia.

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Poster

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Translational Medicine Institute Translational Acceleration Program, Colorado State University

Title: Plasticity in the adult human white matter: the effects and modifiers of response to aerobic exercise interventions

Authors: *A. MENDEZ COLMENARES[1], M. VOSS[3], J. FANNING[4], E. SALERNO[5], N. GOTHE[6], M. L. THOMAS[1], E. MCAULEY[6], A. F. KRAMER[7], A. BURZYNSKA[2]; [1]Psychology/Molecular, Cell. and Integrative Neurosciences, [2]Dept. of Human Develop. and Family Studies/Molecular, Cell. and Integrative Neurosciences, Colorado State Univ., Fort Collins, CO; [3]Dept. of Psychological and Brain Sci., Univ. of Iowa, Iowa City, IA; [4]Dept. of Hlth. and Exercise Sci., Wake Forest Univ., Winston-Salem, NC; [5]Div. of Publ. Hlth. Sciences, Dept. of Surgery, Washington Univ. Sch. of Med. in St. Louis, St. Louis, MO; [6]Dept. of Kinesiology and Community Hlth., Univ. of Illinois at Urbana-Champaign, Urbana, IL; [7]Dept. of Psychology, Northeastern Univ., Boston, MA

Abstract: White matter provides a scaffold for brain function and cognition. Animal studies suggest that white matter plays an active role in brain plasticity and learning. However, evidence for experience-induced plasticity in the adult human white matter remains scarce and inconsistent, especially in old age. Here, we report the effects of 6-month aerobic exercise interventions (walking and dance, NCT01472744) on white matter integrity in healthy older adults (n = 180, 60-79 years) measured by changes (Δ) in the ratio of calibrated T1- to T2-weighted signal (T1w/T2w). We observed significant time-by-group interactions, with positive changes in the T1w/T2w signal in late-myelinating regions in the aerobic exercise groups and declines in the active control. The increases in T1w/T2w signal in the walking group correlated positively with Δ in memory performance, as reported in our recent publication (Mendez Colmenares et al., 2021). We further explored the possible modifiers of intervention-induced change in the T1w/T2w signal. Among participants randomized to the walking intervention, those who increased their participation in total daily light physical activity showed greater increases in T1w/T2w in the total white matter, cingulum, splenium of the corpus callosum, and superior longitudinal fasciculus. These effects persisted after controlling for Δ in moderate-to-vigorous physical activity, systolic blood pressure, age, and sex. Our findings suggest that increasing engagement in moderate-to-vigorous physical activity (i.e., walking and dance) results in group-level increases in white matter T1w/T2w signal, and these increases were the largest among those who additionally increased their daily light physical activity engagement. Our results show evidence that white matter regions that are vulnerable to aging retain some degree of plasticity that can be induced by aerobic exercise training and moderated by light daily activity.

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Poster

P874. Cognitive Natural Aging: Behavior

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Program #/Poster #: P874.06

Topic: H.12. Aging and Development

Title: Spatial navigation declines with age in clinically normal participants

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Abstract: Spatial navigation performance declines with age. We studied a virtual navigation task, the "Apple Game" (Bierbrauer et al. 2020) focusing on path integration. Here, we expanded these results by investigating how navigation performance and the employment of different navigational strategies differ across the lifespan (median age 58.2, 21-81).Participants navigate in a virtual field and have to remember their initial location to return to it (path integration). Difficulty varies between trials according to two parameters: (1) the number of interim locations participants have to visit (0, 1, or 2) before they return to their initial location; and (2) the presence or absence of a landmark. This paradigm is a simplified version of the original task, suitable for older participants. We recruited 155 clinically normal participants (Mini Mental State Exam > 24/30). We used linear mixed models to predict the drop error, i.e., the Euclidean distance between the initial and final locations. We also analyzed the employment of landmark-based navigational strategies. As previously shown, the drop error increased with the number of interim locations and decreased with the presence of the landmark. Importantly, the drop error strongly increased with age. This age effect was present in all subtasks except one, which included two distractors and no landmark, suggesting this subtask was too difficult for all participants, including the younger. In contrast, in the easiest subtask, with a landmark and no distractor, the drop error increased with age, indicating it is appropriate for investigating age effects.
Interestingly, the distance between the landmark and the final position decreased with age, suggesting that older participants were attracted by the landmark instead of using the landmark to navigate correctly. Indeed, the positive effect of the landmark on spatial navigation decreased with age. The decrease in the positive effect of the landmark with age was only present with one or two distractors highlighting the importance of having at least one distractor to evaluate how the landmark affects spatial navigation in older participants.
This simplified version of the Apple Game is appropriate to test how path integration changes with age. Without distractors, older participants benefit from the presence of the landmark as

much as younger participants do. With distractors, the landmark's positive effect decreases in older participants, suggesting that older participants have difficulties to integrate their position with the landmark position. As a future direction, we aim to investigate the interplay between age effects and preclinical neurodegenerative disease.

Disclosures: L. Colmant: None. **Y. Bellaali:** None. **A. Bierbrauer:** None. **N. Axmacher:** None. **P. Lefevre:** None. **B. Hanseeuw:** None.

Poster

P874. Cognitive Natural Aging: Behavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P874.07

Topic: H.12. Aging and Development

Support: NSF Grant 1460682
NIH Grant 5T32AG000175
Fulton Country Elder Health Scholarship

Title: Choice-relevant bias influences decision-making measure differently as a function of age

Authors: *J. WALKER[1], M. E. WHEELER[2];
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Abstract: During time-dependent binary perceptual decision-making, evidence accumulation is defined as the timeframe when individuals are gradually integrating choice-relevant information.[1] There are many factors that influence this process, including the presence of prior knowledge that pushes individuals towards/away from given options and, thereby, may manipulate the amount of evidence required to reach decisions.[2] This effect is likewise reflected in blood oxygen level-dependent (BOLD) activity in the inferior temporal cortex (ITC). For example, a 2018 functional magnetic resonance imaging (fMRI) study by Dunovan & Wheeler used a task where participants were presented with visually noisy images and asked to classify them as either faces or places. Prior to each trial, they viewed probabilistic cues (80% chance face, 80% chance house, or unbiased 50/50 control). They found that bias toward the correct answer (valid trials) was associated with higher accuracy, shorter reaction times (RTs), and decreased ITC BOLD activity. Bias away (invalid trials) was associated with poorer accuracy, slower RTs, and increased ITC activity. The unbiased 50/50 control condition (neutral trials) was in the middle. Such findings are crucial for understanding brain-behavior relationships during decision-making. However, this 2018 work only included younger adults and age is another important factor to consider going forward. This is because some literature suggests that older adults may approach decision-making from a relatively more inflexible perspective, disregarding prior knowledge in favor of taking their time to consider all choices more equally.[3] It is unclear if

or how that is reflected in ITC activity. Therefore, the current fMRI study replicates the design in the aforementioned 2018 work and extends it to include a cross-sectional age comparison of younger (21-35 years old, n=16) and older adults (60-75 years old, n=18). Results indicate that only behavioral outcomes were consistent across age groups. Both younger and older adults were faster and more accurate for valid trials, followed by neutral, followed by invalid. However, while younger adults replicated the finding that lower BOLD signals are associated with valid trials and higher BOLD signals are associated with invalid trials in a selection of ITC regions, older adults' BOLD activity was consistent across conditions. These findings suggest, that as people age, the patterns of activity underlying their bias-relevant decision-making processes may change to maintain performance.
1. Ploran, et al., JNeurosci 27, 144 (2007) 2. Dunovan & Wheeler, Sci Rep 8, 1 (2018) 3. Ratcliff, et al., Psychol Aging 19, 2 (2004)

Disclosures: J. Walker: None. **M.E. Wheeler:** None.

Poster

P878. Spatial and Relational Learning

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P878.01

Topic: H.10. Human Learning and Cognition

Support: Max Planck Society

Title: Integrating knowledge about structure and reward contingencies for generalization and inference

Authors: *F. F. DEILMANN[1], M. M. GARVERT[1], C. F. DOELLER[1,2];
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Abstract: The hippocampal-entorhinal system is remarkably efficient in organizing relations between sensory stimuli such as state transition probabilities in a cognitive map. Such a representation of knowledge is assumed to enable fast learning of novel relations and the generalization of reward, which likely facilitates goal-directed behavior. However, in addition to experienced transition probabilities, objects may simultaneously share other types of relational information, such as reward contingencies. Here we investigate how the representation of relational knowledge is influenced by a subsequently learned latent reward structure. Are structural relationships between objects still represented veridically, or do reward contingencies influence the representation of relational knowledge? In our task, participants initially acquire knowledge about the relationships between objects based on object transitions that follow a hidden graph structure. Behavioral data suggest that participants successfully acquire this

knowledge and can utilize it to find shortcuts when navigating around the graph. During a subsequent decision-making task, each object gets associated with fluctuating reward values. Critically, two parts of the graph structure share the same reward contingencies. Model-based analysis reveals that participants can extract this underlying latent reward structure and use it to their advantage by generalizing over states which share the same reward contingencies. Furthermore, participants can apply their acquired structural knowledge to correctly infer the current reward values of objects whose values they never directly experienced. This suggests that participants represent both the experienced transitions and the shared reward contingencies between objects; moreover, they can combine both types of information for generalization and inference. Using functional magnetic resonance imaging, we now investigate how object relationships and shared reward contingencies are represented in the hippocampal-entorhinal system and interact with other brain regions to guide choice behavior.

Disclosures: F.F. Deilmann: None. **M.M. Garvert:** None. **C.F. Doeller:** None.

Poster

P878. Spatial and Relational Learning

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P878.02

Topic: H.10. Human Learning and Cognition

Support: Max Planck Society

Title: Effects of prototype abstraction on pattern completion and inference in concept space

Authors: *T. A. J. SCHÄFER[1], E. SCHULZ[2], S. THEVES[1], C. F. DOELLER[1,3];
[1]MPI for Human Cognitive and Brain Sci., Leipzig, Germany; [2]MPI for Biol. Cybernetics, Tübingen, Germany; [3]Kavli Inst. for Systems Neuroscience, NTNU, Trondheim, Norway

Abstract: Processing of complex environments is greatly facilitated by the formation and use of concepts. Concepts represent combinations of features shared by similar entities and allow generalisation from limited experience to novel situations. Recent research suggests that concept formation is supported by cognitive maps within the hippocampal-entorhinal system that encode multiple relationships between experiences in a common representational format for flexible access. In a behavioral and functional magnetic resonance imaging experiment, we investigate if this map-like representation of concepts supports the retrieval of abstracted information to guide inference. In a novel behavioral paradigm, participants are first trained to categorize a set of exemplars based on the ratio of their two features. Subsequently, they encounter exemplars that exhibit only one of the features and are instructed to complete the missing feature according to their category label. Here we find that feature inferences are attracted more towards the category center (i.e. prototype) than to the nearest experienced exemplars, suggesting the retrieval of an

abstract representation. In an ongoing functional magnetic resonance imaging study, we examine to what degree the hippocampal system, in concert with visual areas, represents prototypes for feature inference. By decoding the inferred features and feature-combinations from brain patterns, we will test if each category's unseen prototype is formed during categorization and allows feature inference. This project can help us to understand the relationship between abstract representations and cognitive maps and the role of pattern completion in concept formation.

Disclosures: T.A.J. Schäfer: None. **E. Schulz:** None. **S. Theves:** None. **C.F. Doeller:** None.

Poster

P878. Spatial and Relational Learning

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P878.03

Topic: H.10. Human Learning and Cognition

Support: ERC-CoG GEOCOG 724836
223262/F50
197467/F50

Title: Deepmreye: mr-based camera-less eye tracking using deep neural networks

Authors: *M. NAU[1], M. FREY[2], C. F. DOELLER[1];
[1]Max Planck Inst. for Human Cognitive and Brain Sci., Leipzig, Germany; [2]Kavli Inst. for Systems Neuroscience, NTNU, Trondheim, Norway

Abstract: Viewing behavior provides a window into many central aspects of human cognition and health, and it is an important variable of interest or confound in many fMRI studies. To make eye tracking freely and widely available for MRI research, we developed DeepMReye: a convolutional neural network that decodes gaze position from the MR-signal of the eyeballs. It performs camera-less eye tracking at sub-imaging temporal resolution in held-out participants with little training data and across a broad range of scanning protocols. Critically, it works even in existing datasets and when the eyes are closed. Decoded eye movements explain network-wide brain activity also in regions not associated with oculomotor function. This work emphasizes the importance of eye tracking for the interpretation of fMRI results and provides an open-source software solution that is widely applicable in research and clinical settings.

Disclosures: M. Nau: None. **M. Frey:** None. **C.F. Doeller:** None.

Poster

P878. Spatial and Relational Learning

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P878.04

Topic: H.10. Human Learning and Cognition

Support: Norwegian University of Science and Technology
Max Planck Society

Title: Structural connectivity-based segmentation of the human entorhinal cortex

Authors: *I. F. SYVERSEN[1], M. P. WITTER[1], A. KOBRO-FLATMOEN[1], P. E. GOA[2], T. NAVARRO SCHRÖDER[1], C. F. DOELLER[1,3];
[1]Kavli Inst. for Systems Neuroscience, NTNU, Trondheim, Norway; [2]Dept. of Physics, NTNU, Trondheim, Norway; [3]MPI for Human Cognitive and Brain Sci., Leipzig, Germany

Abstract: The medial (MEC) and lateral entorhinal cortex (LEC), widely studied in rodents, are well defined and characterized. In humans, however, the exact locations of their homologues remain uncertain. Previous functional magnetic resonance imaging (fMRI) studies have subdivided the human EC into posterior-medial (pmEC) and anterior-lateral (alEC) parts, but uncertainty remains about the choice of imaging modality and seed regions, in particular in light of a substantial revision of the classical model of the EC based on novel insights from rodent anatomy. Here, we used structural, not functional imaging, namely diffusion tensor imaging (DTI) and probabilistic tractography to segment the human EC based on differential connectivity to other brain regions known to project selectively to MEC or LEC. We defined MEC as more strongly connected to presubiculum and retrosplenial cortex (RSC), and LEC as more strongly connected to distal CA1 and proximal subiculum (dCA1pSub) and orbitofrontal cortex (OFC). Although our DTI segmentation had a larger medial-lateral component than in previous fMRI studies, our results show that the human MEC and LEC homologues have a border oriented both towards the posterior-anterior and medial-lateral axes, supporting the differentiation between pmEC and alEC. In the future, we plan on testing the reproducibility of these results in another DTI dataset, as well as comparing results from DTI with fMRI.

Disclosures: I.F. Syversen: None. **M.P. Witter:** None. **A. Kobro-Flatmoen:** None. **P.E. Goa:** None. **T. Navarro Schröder:** None. **C.F. Doeller:** None.

Poster

P878. Spatial and Relational Learning

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P878.05

Topic: H.10. Human Learning and Cognition

Support: Max Planck Society

Title: Investigating geometric determinants of semantic memory

Authors: *S. THEVES[1], T. A. J. SCHÄFER[1], W. J. DE COTHI[2], C. BARRY[3], C. F. DOELLER[1];
[1]MPI for Human Cognitive and Brain Sci., Leipzig, Germany; [2]CoMPLEX, Univ. Col. London, London, United Kingdom; [3]UCL, London, United Kingdom

Abstract: Recent work suggests that hippocampal codes, originally considered to underlie spatial memory, support the formation of conceptual knowledge by organizing information in cognitive maps that represent multiple relations in a common space. Yet evidence for the involvement of spatially tuned cells in the formation of such concept spaces in humans is elusive. As a window into the respective mechanisms, we leverage insights about the sensitivity of place cell firing to deformations of the environmental geometry. It has been shown that shifting the boundaries of a rodent's enclosure along one dimension causes firing fields of hippocampal place cells to stretch in the direction of environmental expansion. This 'stretchy box' effect has also been observed at the level of human spatial memory where recall of object locations mimicked place field distortions in rodents, indicative of a place cell code for spatial memory. Under the hypothesis that place cells might encode positions in concept space, we here ask whether relocation of categorical boundaries likewise distorts semantic memory. To this end, we first train participants to acquire a concept of two categories, defined as a square enclosure (category A) and its surrounding (category B) in a two-dimensional feature space. Further, participants learn to associate certain category A members with objects, thus assigning the objects a position in concept space. Subsequently, we relocate the boundaries of category A along one feature dimension, resulting in a rectangular shape of category A. Participants encounter this change and update their concept via a recategorization task. To probe whether deformation of the concept's geometry affects memory, we test participants on the associations between objects and category A members immediately before and after the recategorization. We find a significantly higher increase in memory errors along the stretched as compared to the non-stretched dimension, thus demonstrating a 'stretchy box' effect in concept space. We will further evaluate whether such distortions can be explained by a boundary vector cell model of place cell population activity. This project can provide insight in spatial coding mechanisms underlying the representation of conceptual knowledge.

Disclosures: S. Theves: None. **T.A.J. Schäfer:** None. **W.J. De Cothi:** None. **C. Barry:** None. **C.F. Doeller:** None.

Poster

P878. Spatial and Relational Learning

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Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P878.06

Topic: H.10. Human Learning and Cognition

Support: Max Planck Society
Kavli Foundation
European Research Council (ERC-CoG GEOCOG 724836)
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The Egil and Pauline Braathen and Fred Kavli Centre for Cortical Microcircuits
National Infrastructure scheme of the Research Council of Norway – NORBRAIN (197467/F50)

Title: Hippocampus and striatum encode distinct task regularities that guide human timing behavior

Authors: *I. POLTI[1], M. NAU[2], R. KAPLAN[1], V. VAN WASSENHOVE[3], C. F. DOELLER[4,1];
[1]Kavli Inst. for Systems Neuroscience, Ctr. for Neural Computation, The Egil and Pauline Braathen and Fred Kavli Ctr. for Cortical Microcircuits, Jebsen Ctr. for Alzheimer's Disease, Norwegian Univ. of Sci. and Technol., Trondheim, Norway; [2]Natl. Inst. of Mental Hlth. (NIMH), Bethesda, WA; [3]CEA DRF/Joliot, NeuroSpin; INSERM, Cognitive Neuroimaging Unit; CNRS, Univ. Paris-Saclay, Gif Sur Yvette, France; [4]Max Planck Inst. for Brain, Cognition & Behavior, Leipzig, Germany

Abstract: The brain encodes the statistical regularities of the environment in a task-specific yet flexible and generalizable format. How it does so remains poorly understood. Here, we seek to understand this by converging two parallel lines of research, one centered on striatal-dependent sensorimotor timing, and the other on hippocampal-dependent cognitive mapping. We combined functional magnetic resonance imaging (fMRI) with a visual-tracking and time-to-contact (TTC) estimation task, revealing the widespread brain network supporting sensorimotor learning in real-time. Hippocampal and caudate activity signaled the behavioral feedback within trials and the improvements in performance across trials, suggesting that both structures encode behavior-dependent information rapidly. Critically, hippocampal learning signals generalized across tested intervals, while striatal ones did not, and together they explained both the trial-wise performance and the regression-to-the-mean biases in TTC estimation. Our results suggest that a fundamental function of hippocampal-striatal interactions may be to solve a trade-off between specificity vs. generalization, enabling the flexible and domain-general expression of human timing behavior broadly.

Disclosures: I. Polti: None. **M. Nau:** None. **R. Kaplan:** None. **V. van Wassenhove:** None. **C.F. Doeller:** None.

Poster

P878. Spatial and Relational Learning

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P878.07

Topic: H.10. Human Learning and Cognition

Support: Max Planck Society

Title: Representing values for prospective decision making

Authors: *A. NITSCH[1], M. M. GARVERT[1], N. W. SCHUCK[2], C. F. DOELLER[1,3];
[1]MPI for Human Cognitive and Brain Sci., Leipzig, Germany; [2]Max Planck Inst. For Human Develop., Berlin, Germany; [3]Kavli Inst. for Systems Neuroscience, NTNU, Trondheim, Norway

Abstract: Everyday decisions require us to plan ahead and predict how valuable different choice options will be in the future. Efficient prediction of such future values is facilitated by an internal model representing state transitions and associated reward contingencies in a task. Recently, it has been suggested that the hippocampal system represents relationships between states in a map-like format akin to its representation of relationships between landmarks and places in physical space. Such a map-like representation allows for efficient prediction of future states because it facilitates the computation of distances and directions. It is unclear, however, whether and how these spatial coding principles and geometric computations might contribute to mapping abstract dimensions such as values. Here, we aim to test the idea that the hippocampal system supports prospective decision making by representing values of choice options in a relational map. To this end, we used a novel decision task requiring participants to monitor and predict changing values of two options over a sequence of time points. Crucially and unknown to participants, such a sequence represents a trajectory in an abstract two-dimensional value space characterized by the reward magnitudes associated with the two options. Recognizing the nature of the particular trajectory allows for prediction of future values of the two options. Behavioral results show that participants are able to integrate changes along the two value dimensions to guide decision making, suggesting they form a map of the relationships between options. Preliminary fMRI results indicate prefrontal regions tracking the values of the options characterized by the trajectories in the two-dimensional value space. Ultimately, this study can help us elucidate whether the hippocampal system adopts spatial coding principles to support efficient prospective value-based decision making.

Disclosures: A. Nitsch: None. **M.M. Garvert:** None. **N.W. Schuck:** None. **C.F. Doeller:** None.

Poster

P878. Spatial and Relational Learning

Location: Virtual Only

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Program #/Poster #: P878.08

Topic: H.10. Human Learning and Cognition

Support: Max Planck Society
ERC-CoG GEOCOG 724836
Kavli Foundation
Jebsen Foundation

Title: Adaptive cognitive maps for curved surfaces in the 3D world

Authors: *M. KIM, C. F. DOELLER;
MPI for Human Cognitive and Brain Sci., Leipzig, Germany

Abstract: Terrains in a 3D world can be undulating but most of previous research exclusively investigated spatial representations on a flat surface, leaving a 2D cognitive map as a dominant model in the field. Here we investigated whether humans represent a curved surface by building a dimension-reduced flattened 2D map or a full 3D map. In the online experiments, participants learned the location of objects lying on a flat square and curved surface (similar to a half-pipe in a skate park) in the virtual environment by driving on the surface (Experiment 1, n=87), driving and looking vertically (Experiment 2, n=88), or flying (Experiment 3, n=41). Later they were asked to retrieve either the path distance on the surface or the 3D Euclidean distance between the objects. Path distance estimation performance was overall good, but we found a significant underestimation bias for the path distance on the curve, suggesting the influence of potential 3D shortcuts even though participants were only driving on the surface (Experiment 1). Euclidean distance estimation performance was better when participants were more exposed to the global 3D structure of the environment by looking and flying (Experiment 2,3). These results suggest that the representation of the 2D manifold embedded in a 3D world is neither purely 2D nor 3D, rather it is flexible and dependent on the behavioral experience and demand.

Disclosures: M. Kim: None. **C.F. Doeller:** None.

Poster

P878. Spatial and Relational Learning

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ERC-CoG GEOCOG 724836
The Centre of Excellence scheme of the Research Council of Norway—Centre for Neural Computation (223262/F50)
NORBRAIN (197467/F50)
Kavli Foundation

Title: Structuring time: The hippocampus constructs sequence memories that generalize temporal relations across experiences

Authors: *J. L. S. BELLMUND[1], L. DEUKER[2], N. D. MONTIJN[3], C. F. DOELLER[1];
[1]MPI for Human Cognitive and Brain Sci., Leipzig, Germany; [2]Donders Institute, Radboud Univ., Nijmegen, Netherlands; [3]Utrecht Univ., Utrecht, Netherlands

Abstract: The hippocampal-entorhinal region supports memory for episodic details, such as temporal relations of sequential events, and mnemonic constructions combining experiences for inferential reasoning. However, it is unclear whether hippocampal event memories reflect temporal relations derived from mnemonic constructions, event order, or elapsing time, and whether these sequence representations generalize temporal relations across similar sequences. Here, participants mnemonically constructed times of events from multiple sequences using infrequent cues and their experience of passing time. After learning, event representations in the anterior hippocampus reflected temporal relations based on constructed times. Temporal relations were generalized across sequences, revealing distinct representational formats for events from the same or different sequences. Structural knowledge about time patterns, abstracted from different sequences, biased the construction of specific event times. These findings demonstrate that mnemonic construction and the generalization of relational knowledge combine in the hippocampus, consistent with the simulation of scenarios from episodic details and structural knowledge.

Disclosures: J.L.S. Bellmund: None. **L. Deuker:** None. **N.D. Montijn:** None. **C.F. Doeller:** None.

Poster

P878. Spatial and Relational Learning

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Program #/Poster #: P878.10

Topic: H.10. Human Learning and Cognition

Support: Wellcome Grant 212281/Z/18/Z
ERC-CoG GEOCOG 724836

Title: Probing neural representations of scene perception in a hippocampally dependent task using artificial neural networks

Authors: *M. FREY[1], C. F. DOELLER[2], C. BARRY[3];
[1]Kavli Inst. for Systems Neuroscience, NTNU, Trondheim, Norway; [2]Max Planck Inst. for Human Cognitive and Brain Sci., Leipzig, Germany; [3]UCL, London, United Kingdom

Abstract: Deep artificial neural networks (DNNs) trained through backpropagation provide effective models of the mammalian visual system, accurately capturing the hierarchy of neural responses through primary visual cortex to inferior temporal cortex (IT) (Yamins et al. 2016, Zhuang et al. 2021). However, the ability of these networks to explain representations in higher cortical areas is relatively lacking and considerably less well researched. For example, DNNs have been less successful as a model of the egocentric to allocentric transformation embodied by circuits in retrosplenial and posterior parietal cortex. These brain regions play a central role in spatial memory and perception being part of the 'pipeline' that transforms perceptual information to the spatial representations found in entorhinal cortex and hippocampus (Byrne et al. 2007, Bicanski et al. 2019). In humans, the Four Mountains task (Hartley et al. 2007) provides a test of egocentric to allocentric topographical processing. Subjects are required to match spatial scenes presented from different perspectives with performance being considerably impaired by damage or degradation to the ego-allocentric pipeline (Bird et al. 2010, Chan et al. 2016). We describe a novel virtual environment, inspired by the Four Mountains task, designed to probe the ability of DNNs to transform scenes viewed from different egocentric perspectives. The 'participant' is required to match scenes that correspond to the same configuration of distinct objects viewed from a different perspective - lures consist of the same or similar objects arranged in other configurations. Using a network architecture inspired by the connectivity between temporal lobe structures and the hippocampus, we demonstrate that DNNs trained using a triplet loss can learn this task. Analysis of the representations learnt in these artificial networks revealed late layers accurately encode the allocentric Euclidean distance between objects in the scene - the clearest representation of this quantity being in layers analogous to hippocampus region CA1.

Disclosures: M. Frey: None. **C.F. Doeller:** None. **C. Barry:** None.

Poster

P923. Light and Electron Microscopy

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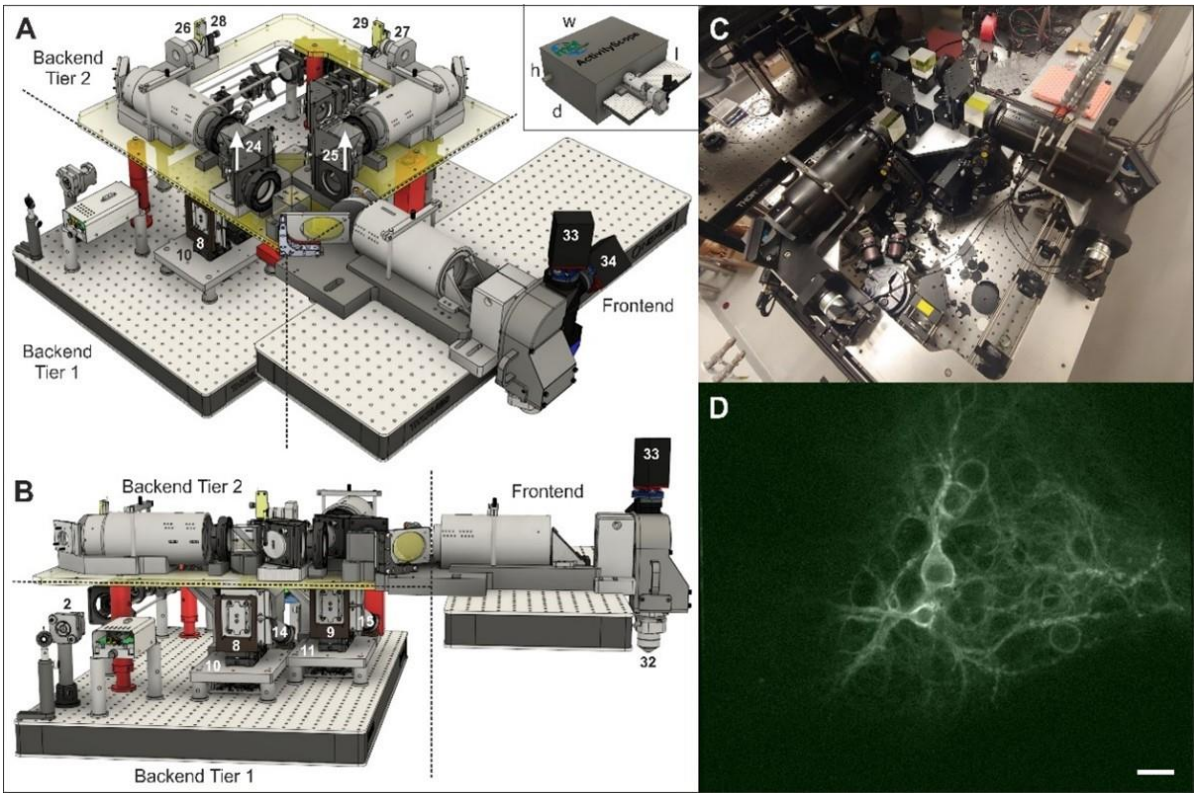
Program #/Poster #: P923.01

Topic: I.03. Anatomical Methods

Title: Activityscope: second-generation scanned line angular projection(slap2) two photon laser scanning microscopy for high speed live imaging at subcellular resolution

Authors: G. E. JAINDL[1], B. E. KIMMEL[1], J. KING[1], K. PODGORSKI[2], D. A. FLICKINGER[3], J. ARNOLD[2], N. ROUSSEL[1], P. ANGSTMAN[1], **S. TAPPAN**[1], *J. GLASER[1];
[1]MBF Biosci., Williston, VT; [2]Janelia Res. Campus, HHMI, Ashburn, VA; [3]Inst. Design & Fab, Howard Hughes Med. Inst., Ashburn, VA

Abstract: Optical imaging of in vivo neuronal populations using fluorescent voltage and neurotransmitter indicators is an emerging frontier that requires microscopes capable of kilohertz-rate volumetric imaging in scattering brain tissue. The ActivityScope is a collaboration between Vidrio Technologies, MBF Bioscience the Podgorski lab, inventors of Scanned Line Angular Projection (SLAP) 2-photon laser scanning microscopy. ActivityScope uses a novel and unique scan technology. A vertical laser line is horizontally swept nearly 10,000 times per second across the surface of a Digital Micromirror Device (DMD) from where the excitation light is projected to the specimen. This approach enables scanning of multiple cell-sized ROIs at kilohertz rates. The DMD can switch “on" and “off" individual pixels within the field of view (FOV) of the ActivityScope, allowing highly flexible random-access imaging without the access time costs associated with previous random-access technologies.
We have developed software that improves the reliability and performance of the system and achieves user-friendly control of the specialized hardware for rapid imaging of multiple ROIs. Key features include automated calibration tools for tuning timing parameters, and real-time display of acquired data, which can be saved to disk. We are developing tools to allow experimentalists and technicians to easily configure complex imaging experiments comprised of both structural and functional activity imaging of multiple neurons, without requiring computer programming expertise.
ActivityScope is capable of imaging at subcellular spatial resolution and temporal resolution in the millisecond range (i.e., at 1000 Hz and higher) in the brain of mice in vivo and in brain slices in vitro. We believe that the ActivityScope microscope will be an asset to many experimental paradigms focused on live imaging of neuronal circuits.



Disclosures: **G.E. Jaindl:** A. Employment/Salary (full or part-time):; MBF Bioscience. **B.E. Kimmel:** A. Employment/Salary (full or part-time):; MBF Bioscience. **J. King:** A. Employment/Salary (full or part-time):; MBF Bioscience. **K. Podgorski:** None. **D.A. Flickinger:** None. **J. Arnold:** None. **N. Roussel:** A. Employment/Salary (full or part-time):; MBF Bioscience. **P. Angstman:** A. Employment/Salary (full or part-time):; MBF Bioscience. **S. Tappan:** A. Employment/Salary (full or part-time):; MBF Bioscience. **J. Glaser:** A. Employment/Salary (full or part-time):; MBF Bioscience.

Poster

P923. Light and Electron Microscopy

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Topic: I.03. Anatomical Methods

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"Fondazione CR Firenze" (private foundation)

Title: 3d molecular phenotyping of the human brain with light-sheet fluorescence microscopy

Authors: *I. COSTANTINI[1], M. SCARDIGLI[4], L. PESCE[4], J. RAMAZZOTTI[4], N. BRANDY[4], M. BAGHDAD[4], V. GAVRYUSEV[4], G. MAZZAMUTO[5], F. CASTELLI[4], C. CHECCUCCI[4], M. ROFFILLI[6], L. SILVESTRI[2], P. FRASCONI[3], F. PAVONE[2];
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Abstract: Studying the 3D architecture of the human brain at subcellular resolution is one of the current outstanding challenges. Having a robust method to map the anatomy, without introducing any visual artifacts or sampling bias, is becoming essential to obtain reliable quantitative information. Here, we report an innovative pipeline that allows 3D mapping of neurons in cm^3 of the human brain at high resolution using Light-Sheet Fluorescence Microscopy (LSFM). At first, we optimized SHORT, a new tissue clearing method to clear and stain mesoscopic volumes of human brain tissue. Then, to perform fast 3D imaging with subcellular resolution over cm-sized samples, we built a custom-made inverted light-sheet fluorescence microscope able to acquire up to four channels. Finally, we implemented a data management system able to deal with the large data volume produced during the acquisition and a machine learning technique to automatically count the neurons in 3D with high precision and sensitivity. We demonstrated the reliability of our approach by analyzing various human brain areas, and in particular, reconstructing a whole human Broca's Area of 4 x 4 x 2 cm^3. In conclusion, the proposed combination of a specialized tissue preparation protocol, an advanced LSFM and big data analysis allows mapping human brain architecture at the cellular level, paving the way to perform routinary analysis of 3D reconstructions of different tissue blocks up to the entire organ.

Disclosures: I. Costantini: None. **M. Scardigli:** None. **L. Pesce:** None. **J. Ramazzotti:** None. **N. Brandy:** None. **M. Baghdad:** None. **V. Gavryusev:** None. **G. Mazzamuto:** None. **F. Castelli:** None. **C. Checcucci:** None. **M. Roffilli:** A. Employment/Salary (full or part-time):; Bioretics Srl. **L. Silvestri:** None. **P. Frasconi:** None. **F. Pavone:** None.

Poster

P923. Light and Electron Microscopy

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P923.03

Topic: I.03. Anatomical Methods

Support: U01MH114824
U01MH105971

Title: Whole brain architecture and anatomical cell-type organization of the rodent brain

Authors: *R. MUÑOZ CASTAÑEDA[1], R. PALANISWAMY[1], J. PALMER[1], R. DREWES[1], M. GUSTISON[3], C. ELOWSKY[1], K. HIROKAWA[5], N. H. CAIN[5], K. UMADEVI VENKATARAJU[2], S. M. PHELPS[4], J. A. HARRIS[5], P. OSTEN[1];
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Abstract: The distribution of cell types in anatomical regions is fundamental to understand brain functions, yet classical stereological methods have been too laborious for comprehensive analyses across animals. New imaging techniques, such as serial two-photon tomography (STPT) and light-sheet fluorescence microscopy (LSFM), allow us to generate whole-brain datasets at a cellular resolution. To acquire and understand this enormous amount of information, however, it is also essential to automate acquisition and analysis of such data. In order to analyze and understand the organization of the brain, we developed an automated pipeline for the acquisition and analysis of cellular resolution histological brain data in rodents. First, we used NeuroTrace fluorescent Nissl-staining as a standard method to define anatomical brain structures to perform 3D brain imaging with STPT and LSFM. We use genetic labels such as Cre-based reporter mouse lines (eg ChAT-IRES-Cre) and whole mount immunolabeling of specific cell-types that will help to provide deeper understanding of neuroanatomical organization. We finally apply the developed pipeline to understand specie-specific neuroanatomical organization and cell-types patterns distribution.

Disclosures: R. Muñoz Castañeda: None. **R. Palaniswamy:** None. **J. Palmer:** None. **R. Drewes:** None. **M. Gustison:** None. **C. Elowsky:** None. **K. Hirokawa:** None. **N.H. Cain:** None. **K. Umadevi Venkataraju:** None. **S.M. Phelps:** None. **J.A. Harris:** None. **P. Osten:** None.

Poster

P923. Light and Electron Microscopy

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Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P923.04

Topic: I.03. Anatomical Methods

Support: NIMH Grant MH116827

Title: Clearscope: theta light sheet imaging for large intact tissues

Authors: P. J. LANG[1], D. S. DENU[1], S. J. TAPPAN[1], B. W. HAYDOCK[1], C. THOMAS[1], *N. J. O'CONNOR[1], B. S. EASTWOOD[1], I. C. MACFARLANE[1], R. TOMER[2], P. J. ANGSTMAN[1], J. R. GLASER[1];
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Abstract: Advances in tissue clearing methods and improved Light Sheet Microscopy (LSM) techniques have opened new avenues of research into brain-wide exploration of connectivity patterns and molecular indicators of behavior and pathology. Working with the Tomer laboratory at Columbia University we have developed ClearScope, a revolutionary light-sheet microscope capable of imaging large, intact specimens at high resolutions. ClearScope uses advanced technology known as Light Sheet Theta Microscopy (LSTM), which utilizes a unique arrangement of two light sheets oblique to the specimen and detector axes. This arrangement allows the LSTM to image specimens of unprecedented size. ClearScope is a robust commercial technology that can be deployed in most neuroscience laboratories for use with virtually all tissue clearing techniques. It can be configured with up to seven laser excitation wavelengths for multi-channel imaging of most fluorescent reporters. ClearScope also provides a wide-field fluorescence imaging mode for quickly evaluating a specimen to select regions of interest for very high-resolution scans in light-sheet mode. ClearScope provides efficient imaging of cleared tissues at cellular resolution without a limitation on lateral tissue size. We present our most recent ClearScope developments including Streamlined workflows for high-speed image acquisition and hardware control; automated light-sheet optimization which maximizes axial resolution and overall image quality while minimizing training time required for system operation; and image volume stitching automatically performed as images are acquired and resulting in complete 3D images of tissue volumes viewable immediately after acquisition. A variety of file formats can be saved from ClearScope for easy analysis and collaboration.

Disclosures: P.J. Lang: A. Employment/Salary (full or part-time):; MBF Bioscience. **D.S. Denu:** A. Employment/Salary (full or part-time):; MBF Bioscience. **S.J. Tappan:** A. Employment/Salary (full or part-time):; MBF Bioscience. **B.W. Haydock:** A. Employment/Salary (full or part-time):; MBF Bioscience. **C. Thomas:** A. Employment/Salary (full or part-time):; MBF Bioscience. **N.J. O'Connor:** A. Employment/Salary (full or part-time):; MBF Bioscience. **B.S. Eastwood:** A. Employment/Salary (full or part-time):; MBF Bioscience. **I.C. MacFarlane:** A. Employment/Salary (full or part-time):; MBF Bioscience. **R. Tomer:** None. **P.J. Angstman:** A. Employment/Salary (full or part-time):; MBF Bioscience. **J.R. Glaser:** A. Employment/Salary (full or part-time):; MBF Bioscience.

Poster

P923. Light and Electron Microscopy

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P923.05

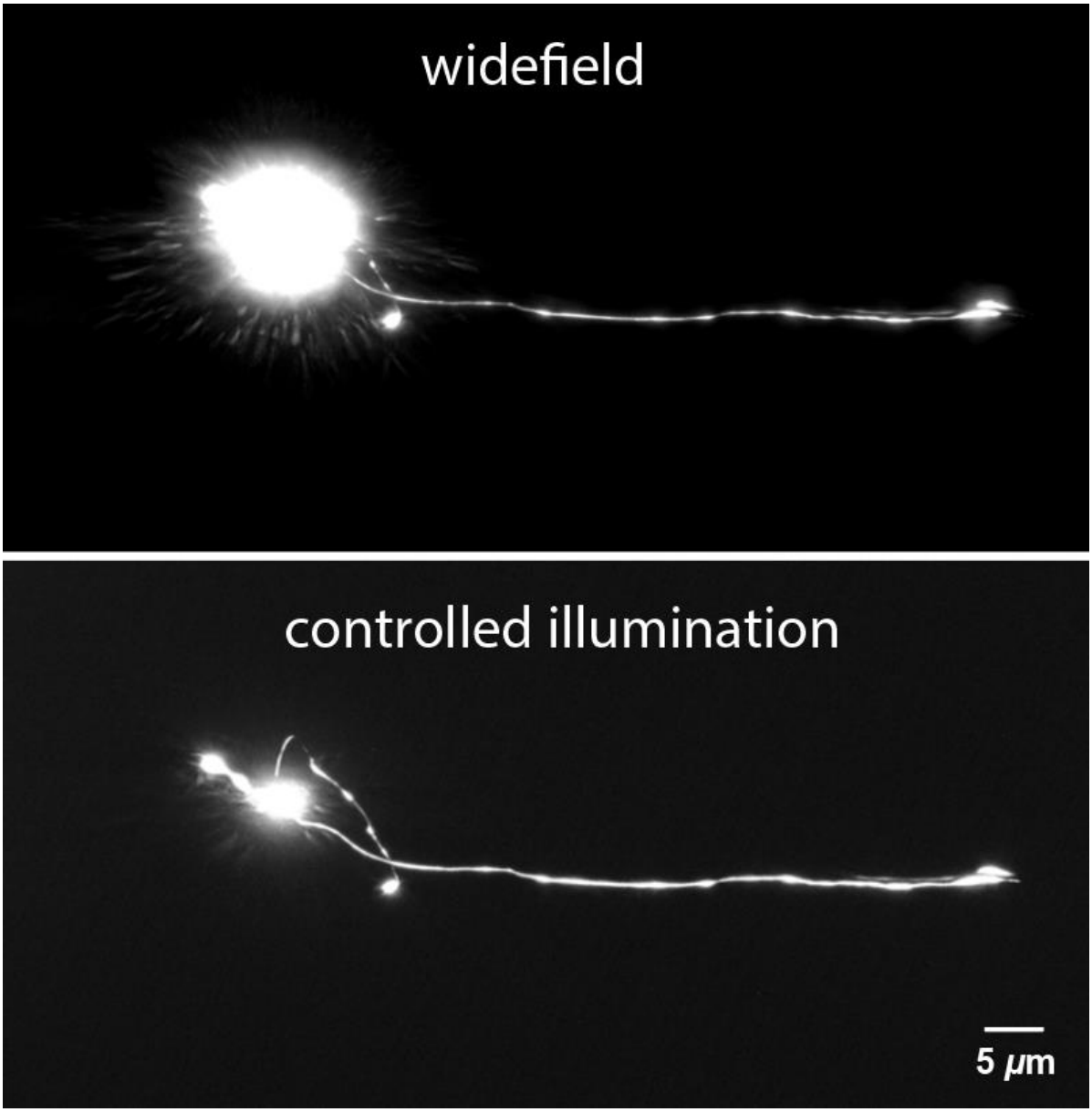
Topic: I.03. Anatomical Methods

Support: Northeastern University TIER 1 grant

Title: Improve widefield imaging contrast of neurons using actively controlled illumination

Authors: *Y. WANG, J. FAN, S. CHUNG;
Bioengineering, Northeastern Univerisity, Boston, MA

Abstract: In widefield neuron imaging, out-of-focus and scattered light often obscures dim fluorescent fibers. The scattering of light coming from bright cell bodies can also degrade the signal-to-background ratio (SBR) of nearby fibers. This phenomenon makes it difficult for widefield neuron imaging to visualize axons and dendrites near to their cell bodies. Scanning techniques, such as confocal and two-photon microscopy, can solve this problem but are limited by reduced imaging speed and increased cost, making them less accessible than widefield microscopes in many biological labs. We solve this obstacle by illuminating different structures with different light intensities in a common widefield microscope. With a simple spatial light modulator add-on to the microscope and real-time image processing, we illuminate bright structures, such as neuron cell bodies, with low light intensity. We illuminate dim, fiber-like structures, such as axons and dendrites, with high light intensity. Illuminating bright structures with dim light reduces the scattering surrounding them, exposing dim nearby fibers. In the actively controlled illumination setup, we illuminate only the in-focus portion of the targeted neuron. Moreover, we illuminate those parts with different light intensity levels to highlight fibers. Thus, we minimize background and enhance the visibility of fibers in the final image. Active illumination significantly improves fiber contrast quality while maintaining a fast imaging speed and low cost. Using a widefield microscope with our active illumination setup, we demonstrate improved neuron image quality in complex neurons of *C.elegans.*



Disclosures: Y. Wang: None. **J. Fan:** None. **S. Chung:** None.

Poster

P923. Light and Electron Microscopy

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Program #/Poster #: P923.06

Topic: I.03. Anatomical Methods

Support: NIH R01NS111028

NIH RF1NS113287
NIH R21NS112666
University of Minnesota MnDrive Graduate Assistant Fellowship

Title: Miniaturized head-mounted microscope for whole cortex mesoscale imaging in freely behaving mice

Authors: *M. RYNES[1], D. A. SURINACH[2], S. LINN[1], M. LAROQUE[3], V. RAJENDRAN[2], J. DOMINQUEZ[2], O. HADJISTAMOULOU[2], Z. S. NAVABI[2], L. GHANBARI[2], G. JOHNSON[2], M. NAZARI[4], M. H. MOHAJERANI[5], S. B. KODANDARAMAIAH[2];
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Abstract: The advent of genetically-encoded calcium indicators, along with surgical preparations such as thinned skulls (Drew et al Nature Methods 2010) or refractive index matched skulls (Silasi et al Nature Methods 2016), have enabled mesoscale cortical activity imaging in headfixed mice. Such imaging studies have revealed complex patterns of coordinated activity across the cortical surface during spontaneous behaviors (Mohajerani et al Nature Neurosience 2013), goal-directed behavior (Allen et al Neuron 2017), locomotion (Musall et al Biorxiv 2018), motor learning (Makino et al Neuron 2017) and perceptual decision making (Orsolic et al BiorXiv 2019). However, studies have shown that neural activity during free, unrestrained behavior significantly differs from that recorded in headfixed animals. Furthermore, freely moving animals exhibit a repertoire of behaviors that is vastly increased from headfixed animals. The ability to perform mesoscale imaging of the cortex in freely behaving mice will potentially open up new avenues of scientific enquiry.Here we present the 'mini-mScope', a miniature, head-mountable imaging device compatible with transparent polymer skulls recently developed by our group ('See-Shells', Ghanbari et al Nature Communications 2019). With a 8x10 mm field of view, the mini-mScope can image most of the mouse dorsal cortex with resolution ranging from 39.37 to 55.68 micrometers. The current prototype weighs 3.8 g and incorporates a magnetic interlocking mechanism that allows quick fixation (<1s of temporary restraint) to a See-Shell implanted on an awake mouse. Open Field behavior tests indicate neither See-Shell implantation nor addition of mini-mScope significantly inhibits locomotion, even during trial durations > 30 min. The mini-mScope employs an array of blue LEDs to illuminate the cortical surface uniformly, with up to 8mW of light power. An additional green LED provides illumination for reflectance imaging of intrinsic optical signals arising from changes in cortical blood volume and oxygenation. We have used the mini-mScope to successfully record mesoscale cortical activity of the whole cortex in freely behaving mice, revealing intriguing coordinated activity patterns across cortical regions during a wide variety of behaviors which would be difficult to study in headfixed animals. Additionally, the mini-mScope is capable of simultaneously imaging mesoscale calcium dynamics and LFP electrode measurements during unrestrained behaviors.

Disclosures: M. Rynes: None. **D.A. Surinach:** None. **S. Linn:** None. **M. Laroque:** None. **V. Rajendran:** None. **J. Dominquez:** None. **O. Hadjistamoulou:** None. **Z.S. Navabi:** None. **L.**

Ghanbari: None. **G. Johnson:** None. **M. Nazari:** None. **M.H. Mohajerani:** None. **S.B. Kodandaramaiah:** None.

Poster

P923. Light and Electron Microscopy

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Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P923.07

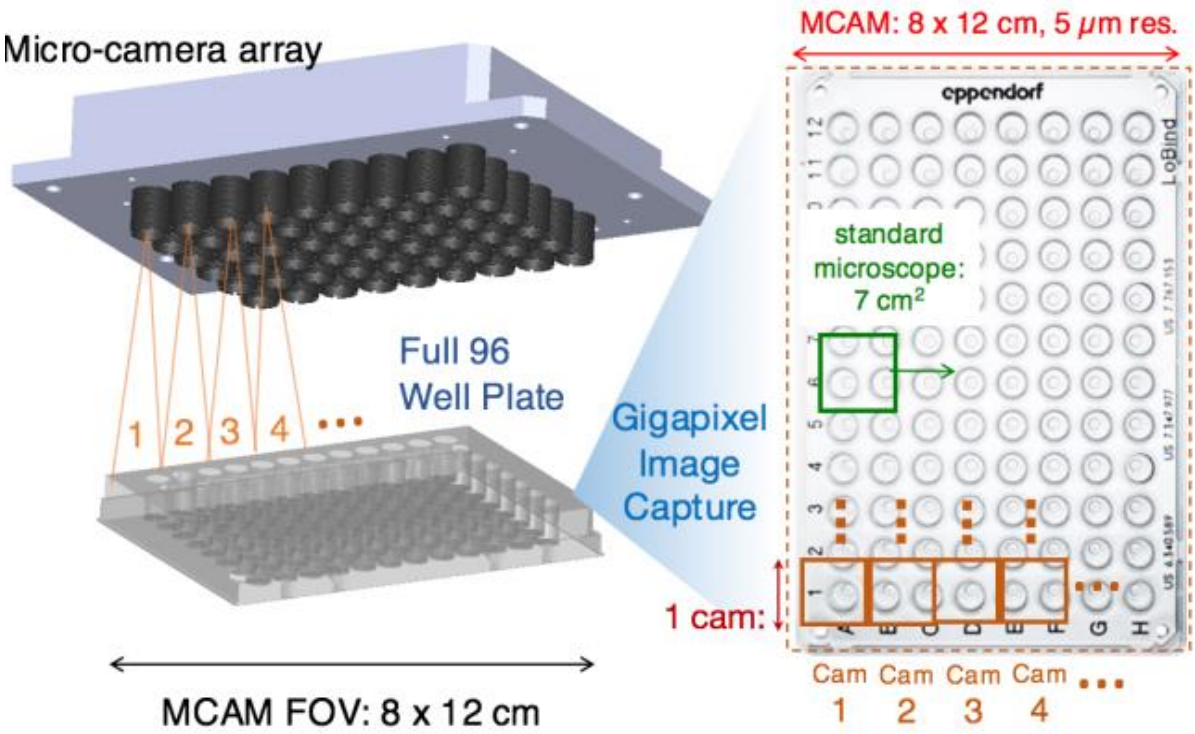
Topic: I.03. Anatomical Methods

Support: NIH Grant R44OD024879

Title: The Micro-Camera Array Microscope - A Gigapixel Imaging and Video Platform for Freely Moving Small Organism Imaging

Authors: K. KIM[1], P. C. KONDA[1], E. E. THOMSON[2], S. SHARMA[3], C. SEITZ[2], K. ZHOU[1], C. COOKE[1,3], S. XU[1], X. YANG[1], X. YAO[1], V. PATHAK[1], J. PARK[3], R. APPEL[3], V. SALIU[3], M. ZHANG[3], C. B. COOK[3], T. J. J. DOMAN[3], P. REAMEY[3], J. EFROMSON[3], A. BEGUE[3], G. HORSTMEYER[3], R. HORSTMEYER[1,3], E. A. NAUMANN[2], ***M. HARFOUCHE**[3];
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Abstract: We present a novel microscopy platform that enables video-rate gigapixel-scale imaging of small model organisms. The optical system consists of a collection of 54 densely packed “micro-cameras” that each view a unique area of an 8 x 12 cm arena. Image data from all 54 sensors is computationally combined into large composite frames, containing many hundreds of megapixels, to reveal model organism behavior at high resolution (5 um/pixel) over an area large enough to enable natural movement and interaction. The demonstrated imaging system’s 54 micro cameras, each taking a volume of 7 cm^3, are tiled together in a compact rectangular configuration (1.35 cm between micro- cameras) to jointly stream data at high rates (~4 GB/sec) to a single computer, thus also enabling video capture. We demonstrate the performance of our new micro-camera array microscope (MCAM) in a variety of unique imaging and video experiments using model organisms including zebrafish, Drosophila and C. Elegans. For example, we simultaneously monitor the behavior of one hundred zebrafish larvae during free swim, and also record the fluorescent neural activity of freely swimming larvae at 5 um per pixel resolution over a 8x12 cm swim arena. We additionally highlight the ability of our new platform with recording 3D video by measuring the 3D movement of dozens of naturally interacting Drosophila. We additionally discuss future improvements for our new imaging method, which include higher video frame rates, automated organism detection and tracking, and the ability to acquire multi-channel fluorescence images.



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Poster

P923. Light and Electron Microscopy

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Program #/Poster #: P923.08

Topic: I.03. Anatomical Methods

Support: Francis Crick Institute Grant FC001153
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Wellcome Trust Investigator Grant 110174/Z/15/Z
Diamond Light Source, beamline I13-2 proposal 20274
Swiss Light Source, beamline TOMCAT proposal 20190417
European Research Council, European Union's Horizon 2020 Research and Innovation Programme grant 852455

Title: Sample preparation and warping accuracy for correlative multimodal imaging in the mouse olfactory bulb using two-photon, synchrotron X-ray and volume electron microscopy

Authors: *Y. ZHANG[1,2], T. ACKELS[1,2], A. PACUREANU[4,2,1], M.-C. ZDORA[5,3,6,7], A. BONNIN[5], A. T. SCHAEFER[1,2], C. BOSCH[1];
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Abstract: *In vivo* physiology and circuit structure complement each other in understanding brain function as they respectively reveal the computations a neural circuit performs and their biological implementation. We aim to apply this function-structure approach to mechanistically understand how odour representations are transformed in the mouse olfactory bulb (OB).
To this end, we aim to image a genetically targeted portion of mouse OB of $(\sim 0.5mm)^3$ containing tens of input structures (glomeruli) and hundreds of output projection neurons (PNs) with *in vivo* two-photon (2P) calcium imaging (using GCaMP6f). In order to link PN activity to corresponding inputs, we will then trace apical dendrites of identified PNs to their parent glomeruli in subsequently acquired synchrotron X-ray tomography (SXRT) and/or volume electron microscopy (vEM) datasets.
A key limiting factor of this multimodal workflow is the success rate of heavy metal staining necessary for generating X-ray or electron contrast (41% without 2P, n = 143; 33% with 2P, n = 24). To improve this, we tested several *en bloc* staining protocols on 0.6mm-thick dorsal mouse OB slabs[1,2], and examined staining homogeneity by X-ray microtomography and ultrastructure preservation by vEM. Without prior 2P, a perfusion based, two-step reduced osmium protocol stained all 8 samples homogeneously but with varying ultrastructural quality. Replacing perfusion with immersion dropped the yield to 50% (n = 4) but obtained consistently good ultrastructure. Improving tissue handling during surgery, imaging and dissection raised it to 70% (n = 10).
Another challenge of this workflow is to accurately locate structures of interest in each dataset. After functional 2P imaging, we imaged, in the same volume, fluorescently labelled blood vessels alongside GCaMP signal, which provides ample features for registering the 2P and SXRT/vEM fields of view. To assess warping accuracy, blood vessels in each dataset were independently traced, paired and aligned to a common space. We found that 2P to SXRT warping is as accurate as 2P to vEM warping. With an average error of ~10 µm, both glomeruli and PNs imaged *in vivo* by 2P can be located in the structural SXRT/vEM datasets.
With improved staining and reliable warping, future developments may focus on incorporating more complex physiology and targeted synaptic resolution post-SXRT vEM acquisitions to the workflow, which will give a richer, fully correlated function-structure dataset, driving a more complete understanding of mammalian olfactory processing across length scales and time.
1. Y. Hua, P. Laserstein, M. Helmstaedter, *Nat. Commun.* **6**, 1-7 (2015).
2. M. Pallotto *et al.*, *Elife* **4**, e08206 (2015).

Disclosures: Y. Zhang: None. **T. Ackels:** None. **A. Pacureanu:** None. **M. Zdora:** None. **A. Bonnin:** None. **A.T. Schaefer:** None. **C. Bosch:** None.

Poster

P923. Light and Electron Microscopy

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Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P923.09

Topic: I.03. Anatomical Methods

Support: NSF 2014862
NIH MH119517

Title: Preservation of Ultrastructure and Antigenicity after Rehydration of Cryofixed Brain Tissue

Authors: *J. PEREZ-GARZA, E. PARRISH-MULLIKEN, Z. DEANE, L. OSTROFF; Univ. of Connecticut Physiol. & Neurobio., Storrs, CT

Abstract: Immunohistochemistry at the EM level (immuno-EM) can reveal subcellular localization of molecules with full subcellular structural details, but compromises between sensitivity and morphological preservation are often required. Cryofixation followed by freeze-substitution can achieve an especially high level of structural preservation because tissue is dehydrated at low temperature. Freeze-substituted tissue is normally either embedded in resin at low temperature or fixed with osmium tetroxide before warming so that membranes are not extracted by the dehydration solvent, and both of these options preclude pre-embedding immunolabeling, which must be performed on hydrated samples near room temperature. Pre-embedding labeling is typically more sensitive than post-embedding and allows the use of higher-contrast stains for easier serial reconstruction, while cryofixation has the additional advantage of allowing long-term storage of tissue samples, making collection of large and complex sets of samples feasible. Here, we have developed a cryosubstitution approach that allows rehydration and sensitive immunolabeling of cryofixed tissue without loss of ultrastructure. Once rehydrated according to our method, tissue can be labeled with gold probes or chromogenic immunohistochemistry followed by standard EM staining and embedding. To assess the effect of our protocol on labeling and ultrastructure, we compared tissue freshly labeled and embedded without cryofixation to rehydrated cryofixed from the same animals. We choose to target the localization of three native antigens in the rat lateral amygdala (LA); gamma-aminobutyric acid (GABA), and two calcium-binding proteins, calbindin (CalB), and parvalbumin (PV). We found that rehydrated cryofixed tissue has better preservation of some subcellular structure contents while membrane ultrastructure was not affected by rehydration, showing smooth membrane continuity. For immunolabeling, differences in labeling patterns

between the two samples were not visible. This rehydration approach offers a new alternative to flexibly integrate sample storage, molecular labeling, and high-quality EM ultrastructure.

Disclosures: J. Perez-Garza: None. **E. Parrish-Mulliken:** None. **Z. Deane:** None. **L. Ostroff:** None.

Poster

P923. Light and Electron Microscopy

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Program #/Poster #: P923.10

Topic: I.03. Anatomical Methods

Support: NIH Grant 5P50MH094271-09

Title: Immersion fixation and staining of large volumes of human brain tissue for large scale connectomics

Authors: *N. KARLUPIA[1], R. SCHALEK[1], Y. WU[1], Y. MEIROVITCH[1], A. W. CHARNEY[2], B. KOPELL[3], J. W. LICHTMAN[1];
[1]Mol. and Cell. Biol., Harvard Univ., Cambridge, MA; [2]Icahn Sch. of Med. Mount Sinai, New York, NY; [3]Dept. of Neurosurg., Mount Sinai Hlth. Syst., New York, NY

Abstract: Despite its obvious importance, the cellular and subcellular structure of healthy and diseased human brain is understudied. Recent advances in the field of connectomics has allowed mapping of cells and their circuits at the nanometer scale in volumes of about 1 mm^3. However, given that the human cerebral cortex can be 3 mm in thickness, substantially larger volumes are required. Larger volume electron microscopy (EM) studies in humans have been limited by three major challenges: a) availability of fresh brain biopsy samples instead of poorly preserved postmortem tissue; b) the requirement for excellently preserved large volume tissue blocks using immersion fixation instead of trans-cardiac perfusion fixation routinely used in animal studies; and c) the need for preservation of extracellular space (ECS) and heavy metal (osmium) stain penetration throughout the tissue block. Human cerebral cortical samples from neurosurgical patients are now accessible owing to interventions for epilepsy or lead placement for deep brain stimulation that involve removal of overlying tissue. We devised an immersion fixation and heavy metal staining method that provides excellent ultrastructure of both rodent and human surgical brain samples up to volumes of 8-27 cubic millimeters. Punched or otherwise excised biopsies were obtained from freshly harvested adult mice brains and also collected from the dorsolateral prefrontal cortex of anonymized patients undergoing lead placement for deep brain stimulation. For immersion fixation we optimized the composition and temperature of the fixative solution and minimized the time so as to allow excellent preservation of ultrastructure. We also tested whether anion channel regulators, anti-edematous drugs and higher

concentrations of buffer would benefit maintenance of ECS. To achieve homogenous heavy metal staining (Osmium-Reduced osmium-Thiocarbohydrazide-Osmium or ORTO) all the way to the core of 8 mm^3 tissue blocks without cracking we did variations at each step including the concentration of reagents, duration and incubation temperature of staining, washing, dehydration, infiltration and also tested thiocarbohydrazide alternative. We conclude that rapid immersion fixation in cold fixative containing mannitol and an optimized ORTO method that includes mannitol containing normal saline gives well preserved, homogenously stained, crack-less and non-brittle large volume blocks for EM.

Disclosures: N. Karlupia: None. **R. Schalek:** None. **Y. Wu:** None. **Y. Meirovitch:** None. **A.W. Charney:** None. **B. Kopell:** None. **J.W. Lichtman:** None.

Poster

P971. Data Analysis and Statistics: Human Data III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P971.01

Topic: I.07. Data Analysis and Statistics

Support: MOST, Taiwan Grant 110-2628-B-A49A-509
MOST, Taiwan Grant110-2321-B-A49A-502
Mt. Jade Young Scholarship Award from Ministry of Education, Taiwan
National Yang Ming Chiao Tung University and the Ministry of Education (Aim for the Top University Plan), Taipei,Taiwan

Title: Use scalp electroencephalography to evaluate the severity of depression

Authors: *C. CHEN[1], A.-C. YANG[1,2,3];
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Abstract: Use scalp electroencephalography to evaluate the severity of depression
Authors*Ching Hao Chen , Albert C. Yang
Abstract
Background: The World Health Organization predicts that depression will be the most costly disease on average in the world by 2030(N Bains et al., 2020), with more than 300 million people suffering from depression and the number of patients continuing to rise. However, the current clinical diagnosis of depression relies on consultations, scales, and tests to assess the severity of depression, which is a time-consuming and costly process, and still relies on the judgment of the medical team in the assessment process, which can easily lead to subjective differences in the definition of depression severity. Early detection of depression requires a

patient-friendly inexpensive method based on easily measurable objective indicators. (M Bachmann et al., 2018) To solve the problem of subjectively assessing the severity of patients, we hypothesized that the severity of symptoms in patients with depression is associated with the features of electroencephalography (EEG), so that the features can be used as an indicator to assess the severity of depression by using scalp EEG analysis as a basis for diagnostic markers using artificial intelligence approach. **Objectives** (1) To create a highly accurate prediction model to assess the severity of depression in patients. (2) To identify indicators of depression severity in scalp EEG. **Methods:** The data were collected from 103 depression patients at Kaohsiung Kai-Shuan Hospital, and their 2-minute EEG were collected at resting state. 80 females and 23 males, with a mean age of 45 years, standard deviation of ±11, and a mean age at diagnosis of 39 years, standard deviation of ±12. The educational stages were divided into 7 stages and categorized as 1-7, with a mean educational stage of 3.8 and standard deviation of ±1.3. To avoid the error caused by the machine, we only took the middle 100 seconds and cut it into 10 segments. And used the previous study (Zimmerman et al., 2013) as the segments for the HAMD-17 severity classification. More than 4,000 pieces of data were compiled. Then, we created a prediction model to achieve the most accurate severity prediction, and used statistical analysis to identify the most significant features. **Results**: We created up to 90% - 80% of the depression severity evaluation models and found features in the spectrum transformation method that had a significant effect on depression severity. **Conclusions:** By using this depression severity prediction model, we can assist clinical care in assessing the severity of depression and improving the treatment process for depressed patients.

Disclosures: C. Chen: None. **A. Yang:** None.

Poster

P971. Data Analysis and Statistics: Human Data III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P971.02

Topic: I.07. Data Analysis and Statistics

Title: Head motion in functional MRI scans does not drive results of brain-wide association study: an ABCD study multivariate analysis

Authors: *G. GRIMSRUD[1], N. BYINGTON[1], M. MOONEY[2], M. CORDOVA[2], O. DOYLE[2], R. HERMOSILLO[1], E. EARL[2], A. PERRONE[2], L. MOORE[2], A. GRAHAM[2], J. NIGG[2], W. THOMPSON[3], E. FECZKO[1], O. MIRANDA DOMINGUEZ[1], D. A. FAIR[1];
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Abstract: Motion artifacts reduce both structural and functional image quality and are a major confound in studies of brain development and individual differences. Studies have highlighted

that results obtained with an original sample before quality control (QC) compared with a quality-controlled subset of the data results in significantly different findings, including differences in effect sizes and functional neuroanatomy. Moreover, neurodevelopmental studies often involve participants predisposed to high motion (e.g. ADHD, Autism), and may have increased motion relative to typically developing samples - exacerbating these misleading artifacts. Determining the optimal QC parameters of motion for such neuroimaging data could allow for more comparable and efficient analysis practices. The Adolescent Brain Cognitive Development study (ABCD; N=11,877, ages 9-10) has implemented several conservative methods to minimize motion artifacts, including new 'scrubbing' techniques. Utilizing the ABCD BIDS Community Collection (ABCC, Collection-3165) and the ABCD Reproducible Matched Samples, we built and validated a multivariate ABCD motion method for an additive model, the PolyNeuro Risk Score (PNRS) model of executive function. We conducted our analysis with varying QC thresholds to 1) determine the optimal parameters to conserve the largest participant pool and 2) confirm any result was not driven by head motion. Connectivity matrices were generated for varying combinations of framewise displacement (FD) thresholds (0.08, 0.1, 0.2, 0.4 mm) and amounts of data (5, 8, 10 min). All analyses included an additional 'outlier detection step' to scrub any additional frames with poor QC. Each QC threshold combination produced a different sample size for analysis ranging from N=1,775 to N=8,519. 3,076 participants, who survived at an FD $\leq$ 0.1 mm and 8 minutes of data, became our test sample. The analysis was run several times for this test group using varying amounts of their lower quality data. Overall, our model's predictive power increased when a larger sample size was used, and such power did not vary greatly using iterations of FD thresholds in the test group. These findings indicate: 1) that our procedures, that include outlier detection, are particularly adept at controlling for motion in PNRS studies, and 2) the increase in predictive power for PNRS studies is a function of sample size and not of head motion (for the thresholds tested). Overall, findings serve to better inform future analyses using the PNRS model in large-scale datasets and could help preserve more high-motion participants to better understand both typical and atypical brain-behavior relationships.

Disclosures: G. Grimsrud: None. **N. Byington:** A. Employment/Salary (full or part-time):; University of Minnesota. **M. Mooney:** A. Employment/Salary (full or part-time):; Oregon Science and Health University. **M. Cordova:** None. **O. Doyle:** None. **R. Hermosillo:** A. Employment/Salary (full or part-time):; University of Minnesota. **E. Earl:** A. Employment/Salary (full or part-time):; Oregon Health and Science University. **A. Perrone:** A. Employment/Salary (full or part-time):; University of Minnesota. **L. Moore:** A. Employment/Salary (full or part-time):; Oregon Health and Science University. **A. Graham:** A. Employment/Salary (full or part-time):; Oregon Health and Science University. **J. Nigg:** A. Employment/Salary (full or part-time):; Oregon Science and Health University. **W. Thompson:** A. Employment/Salary (full or part-time):; University of California San Diego. **E. Feczko:** A. Employment/Salary (full or part-time):; University of Minnesota. **O. Miranda Dominguez:** A. Employment/Salary (full or part-time):; University of Minnesota. **D.A. Fair:** A. Employment/Salary (full or part-time):; University of Minnesota. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); University of Minnesota.

Poster

P971. Data Analysis and Statistics: Human Data III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 9:45 AM - 10:45 AM

Program #/Poster #: P971.03

Topic: I.07. Data Analysis and Statistics

Support: R01 (1 477922 299000 191100)

Title: Scale-invariant connectome dynamics across slow and fast timescales

Authors: *T. ALDERSON[1], J. WIRSICH[2], S. KOYEJO[1], S. SADAGHIANI[1];
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Abstract: Connectome dynamics arise from the moment-to-moment co-activation patterns among brain regions. To date, these interactions have principally been constrained to a single timescale of observation, namely, fluctuations in the hemo-dynamic properties of brain. A promising future direction lies in extending this approach to fast dynamics of electro-physiological interactions and characterizing the spatio-temporal rules governing dynamics across both slow and fast timescales. Accordingly, we analyzed concurrently recorded fMRI and source localized EEG from 26 young adults during resting state. Signals were averaged for each of 68 parcels of the Desikan atlas. To permit a frame-by-frame comparison, simultaneously recorded fMRI time series (<0.1 Hz) and bandpassed EEG amplitude envelopes (delta, theta, alpha, beta, gamma) were resampled to the same number of data points. Recurrent co-activation patterns-or states-corresponding to all possible interactions among canonical intrinsic connectivity network were detected and characterized in terms of their dynamic properties, including probability of occurrence, mean lifetime, switching frequency, and transition probability. Connectomes were expressed at all frequency scales (infraslow to gamma) and displayed dynamics defined by several highly correlated properties, including probability of occurrence, mean lifetime, switching frequency, and transition probability. Of special note was the correspondence in the transition probabilities of connectomes across modalities, which were scaled to express transitions at different rates (infraslow to gamma). Given that the observed self-similarity of connectome state sequences at all timescales was consistent with a scale-free phenomenon, we deployed wavelet-based fractal analysis and determined scale-free behavior in the connectomes of all frequency bands (infraslow to gamma). Finally, self-similarity of connectomes across modalities was confirmed by compressing slow fMRI sequences into fast EEG sequences. This simple transformation brought scaling exponents associated with long-range temporal correlations into register across “simulated” and observed EEG In conclusion, we find strong evidence that connectome sequences in concurrent fMRI-EEG recordings are scale-free over several orders of magnitude including infraslow (<0.1 Hz) fluctuations of the hemo-dynamic signal and considerably faster neural oscillations (~1-60 Hz). Our findings suggest that

fMRI- and EEG-derived connectomes are composed of the same spatial ingredients traversing the same state space, but independently, at different rates.

Disclosures: T. alderson: None. **J. Wirsich:** None. **S. Koyejo:** None. **S. Sadaghiani:** None.

Poster

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Program #/Poster #: P971.04

Topic: I.07. Data Analysis and Statistics

Support: NIH grant R01MH118370

Title: High Amplitude BOLD Co-fluctuations: Discrete Events or Continuous Signals?

Authors: *Z. LADWIG[1], B. A. SEITZMAN[2], Y. YU[3], D. M. SMITH[3], A. DWORETSKY[3], B. ADEYEMO[2], S. E. PETERSEN[2], C. GRATTON[3];
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Abstract: Resting state functional connectivity (rsFC) in fMRI has been an important tool for studying functional brain organization in humans. In order to make reliable individual estimates of rsFC, it is necessary to collect large amounts of data per subject. Recently, researchers identified single time points ("events") during which many areas of the brain fluctuate together. Remarkably, rsFC network organization calculated from concatenating just a few of these time points resembles the rsFC network organization estimated from the entire data set. This observation offers at least two interpretations — (1) events are discrete points at which network-defining rsFC is inordinately strong or (2) rsFC is present over time, but is embedded in a background of varying noise. Here, we examined the discreteness of events in real and simulated resting state fMRI data. We find that events are not discrete - they show continuous properties with timepoints of intermediate fluctuation in real data. Second, we show that events are present in simulated data with a static rsFC structure, and that their presence is associated with the modularity of the static correlation structure. These results suggest that variable co-fluctuations in rsFC are a byproduct of a static network organization in a noisy measure, and moments of high cofluctuation may identify timepoints that are less affected by noise.

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Poster

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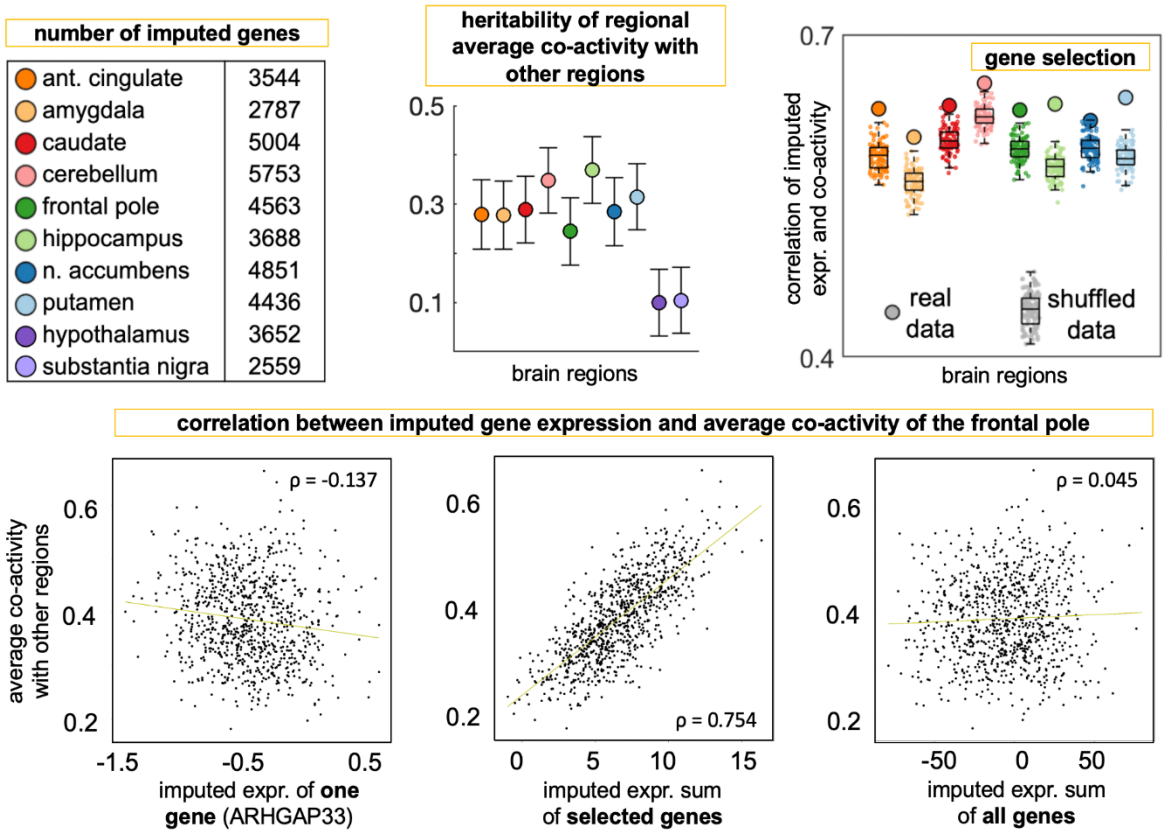
Program #/Poster #: P971.05

Topic: I.07. Data Analysis and Statistics

Title: Transcriptomic correlates of individual variation in brain activity

Authors: *N. HOANG[1], J. PARK[1], Y. CHEN[1], N. SARDARIPOUR[1], M. BENTON[2], J. CAPRA[3], M. RUBINOV[1];
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Abstract: A lack of simultaneous genotype, gene expression, and imaging data for large human populations constrains the search for genetic correlates of regional brain activity. Existing studies use data from a small number of individuals, imaged by consortia such as the Allen Institute Human Brain Atlas (AHBA). These data provide a useful snapshot of interregional transcriptomic relationships but cannot reveal relationships between gene expression and activity across individuals. Here, we integrated multiple neuroimaging and genomic datasets to impute gene expression profiles of ten brain regions in over 800 people. We used this method to probe the relationship between regional brain activity and gene expression levels across individuals. We modeled the relationship between individual-level genotypes and gene expression levels in the brain using PrediXcan, a machine learning method that finds combinations of regulatory alleles reflecting heritable gene expression and estimates the genetically regulated component of this expression (Gamazon, et al. 2015). We applied PrediXcan models for ten brain regions to 888 people from the Human Connectome Project (28.7 ± 3.7 mean age, 484 females), including twins and non-twin siblings. This produced imputed expression across individuals for an average of 4,000 genes per brain region. We found that our method accurately captures existing knowledge of genetic structures in families and populations, as well as gene co-expression on data from the AHBA. We then validated the utility of this method by linking the expression of several gene groups to commonly studied neural phenotypes. We found a substantial overlap between selected genes in relation to mean and variance of interregional coactivity. We thus report strong correlations between individual variation in gene expression and in brain activity and coactivity patterns. Our analyses open the way towards future large-population studies of relationships between heritable gene expression and large-scale brain organization.



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Poster

P971. Data Analysis and Statistics: Human Data III

Location: Virtual Only

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Program #/Poster #: P971.06

Topic: I.07. Data Analysis and Statistics

Support: NIH Grant T32 HD101395

Title: Influence of mechanical stress on peripheral nerve shear wave velocity in a tension-controlled setting

Authors: *C. L. RUGEL[1], S. S. LEE[1], C. K. FRANZ[2];
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Abstract: Clinical studies using shear wave ultrasound elastography to evaluate changes in the mechanical properties of pathologic nerve, consistently report greater nerve shear wave velocity in both diabetic and entrapment neuropathies. This finding is often interpreted as pathologic nerve having greater stiffness than unimpaired nerves, as the relationship between stiffness and shear wave velocity has been validated in various soft tissues. However, this relationship has yet to be evaluated in peripheral nerve. Furthermore, findings from in vivo studies that examine nerve shear wave velocity across limb positions suggest that shear wave velocity could be more indicative of nerve tension than stiffness, as nerve tension varies at different joint angles. Thus, it is the purpose of this study to determine the relationships between peripheral nerve shear wave velocity, tension, and stiffness. To do this, we performed traditional mechanical testing on feline sciatic nerves ex vivo (N=2), elongating the nerve by 0.25mm after one-minute relaxation periods while simultaneously recording the resulting nerve tensile force. Shear wave velocity of the nerve was measured both instantaneously after elongation and at the end of each relaxation period. To avoid potential bias of nerve morphometry, we performed data analysis in terms of nerve stress (tension normalized to nerve cross sectional area) and percent strain (nerve elongation normalized to initial nerve length). From stress and strain values, we were able to calculate elastic modulus, stiffness normalized to nerve size. Our results indicate that nerve shear wave velocity is highly influenced by nerve stress (R^2=0.75, p<0.001), but the relationship is not as strong as that of nerve stress and elastic modulus (R^2=0.97, p<0.001). These findings are not surprising considering stress is used to calculate elastic modulus, but the disparity between the coefficients of determination suggest that there are intrinsic properties of nerve that contribute to its shear wave velocity. This work highlights the importance of controlling nerve tension in clinical evaluations of shear wave velocity, such as by maintaining standard limb positioning. However, additional studies are needed to elucidate other potential factors, such as nerve structure, composition, and fiber directionality, that also affect nerve shear wave velocity.

Disclosures: C.L. Rugel: None. **S.S. Lee:** None. **C.K. Franz:** None.

Poster

P971. Data Analysis and Statistics: Human Data III

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Program #/Poster #: P971.07

Topic: I.07. Data Analysis and Statistics

Support: CONACyT México, PhD Fellowship, DA-B

Title: The importance of racial identity in neuroimaging research: Differences in brain morphometry between White and African Americans

Authors: D. ATILANO-BARBOSA, *F. A. BARRIOS;
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Abstract: There is a marked underrepresentation of ethnic and racial minorities in biomedical research, causing disparities in the understanding of genetic and acquired diseases, as well as in clinical treatments efficacy. These disparities are also present in neuroscience, emphasizing the negative consequences of extrapolating findings from small and non-representative samples of the population in the neurobiological and neuro-functional characterization of the human brain. Racial identity, understood as the sense of collective identity based on a perceived common heritage with a racial group, has generally been used as a demographic variable and nor as a variable of interest in neuroimaging research. Few brain morphometric studies between Whites and African Americans have reported differences in orbitofrontal volumetry and insula cortical thickness. Nevertheless, these studies are mainly implemented in small samples and in population with cognitive impairment. In this sense, we hypothesized that, in a representative sample of neurotypical young adults, there may be differences in brain morphometry between *Whites* and *African Americans*. To test this hypothesis, we analyzed the Human Connectome Project (HCP) brain morphometric data of a group of *Whites* (n = 266, $M_{age(y)}$ = 28.92, SD = ±3.59, $M_{education(y)}$ = 15.35, SD = ±1.60) and *African Americans* (n = 47, $M_{age(y)}$ = 29.11, SD = ±3.65, $M_{education(y)}$ = 14.55, SD = ±1.98). Permutations analysis between groups adjusting for *age*, *education* and *economic income* were implemented. Results indicated significant differences between Whites and African Americans in cortical thickness and surface area involving distinct brain regions in frontal, parietal, temporal and occipital lobes. In this regard, racial identity and inclusion of sub-representative minorities in neuroimaging research, such as African Americans, are fundamental for the comprehension of human brain morphometric variability.
Data were provided by the Human Connectome Project, WU-Minn Consortium (Principal Investigators: David Van Essen and Kamil Ugurbil; 1U54MH091657) funded by the 16 NIH Institutes and Centers that support the NIH Blueprint for Neuroscience Research; and by the McDonnell Center for Systems Neuroscience at Washington University.

Disclosures: D. Atilano-Barbosa: None. **F.A. Barrios:** None.

Poster

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Title: Establish a machine learning approach to predict sleep stages based on ECG

Authors: *P.-C. CHIU[1], A.-C. YANG[1,2,3];
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Abstract: Sleep study with polysomnography (PSG) contains multiple physiological signals.These signals have shown different conditions in sleep status of human body especially sleep stage of electroencephalography (EEG). However, PSG used for sleep staging may interrupt the subject's sleep quality and cannot easily be used for following the trend of sleep. In order to estimate sleep stage in long-term record and less sleep state interference using electrocardiography (ECG) through measuring autonomic system can quantify sleep stages by less than PSG signal outside the sleep lab environment. We used a dataset of a community cohort study from the American National Heart Lung & Blood Institute, which focused on cardiovascular disease and sleep breathing disorders. The dataset included 8,444 PSG records which were used for developing a machine learning model for sleep stages prediction. We identified heart rate rhythms between R peak waveform (R-R interval) and respiratory patterns based on ECG signal. Extracting the features of time domain, frequency domain and nonlinear method of heart rate variability (HRV) are to establish a machine learning model to predict EEG sleep stages based on physiological measures. For using a segment of every 120 seconds of most recorded epoch of sleep stage like continuing sleep stage 2 of ECG data to extract the features from heart rate including time domain, frequency domain, nonlinear methods have the performance of 83.8 percent accuracy. The significant features of machine learning are entropy of nonlinear method, total power of frequency domain, standard deviation of R-R intervals(SDNN) of time domain. Using machine learning approach, our results suggested that specific HRV features of continuing sleep stage pattern are associated with EEG sleep stages. We can use these approach to record sleep stage on a subject in any condition of household and hospital environment outside sleep lab than PSG.

Disclosures: P. Chiu: None. **A. Yang:** None.

Poster

P001. Nervous System Patterning and Developmental Cell Death

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P001.01

Topic: A.01. Neurogenesis and Gliogenesis

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NIH grant R01NS97846 to Dr. Michael Selzer
NIH grant R01NS097846-02 to Dr. Michael Selzer
NIH grant R01NS092876 to Dr. Michael Selzer
USA Pennsylvania State Department grant Project 10: 420491-04400-02 to Dr. Nune Darbinian

Title: Prenatal alcohol exposure disrupts fetal eye and brain development in humans and rats

Authors: *N. DARBINIAN[1], A. DARBINYAN[6], G. TATEVOSIAN[1], N. MERABOVA[1,7], F. D'AMICO[1], T. K. KHADER[1], A. BAJWA[1], D. MARTIROSYAN[1], A. K. GAWLINSKY[1], R. PURSNANI[1], H. ZHAO[2], S. AMINI[3], M. MORRISON[4], L. GOETZL[8], M. E. SELZER[1,5]; [1]Ctr. for Neural Repair and Rehabil. (Shriners Hosp. Pediatric Res. Center) LKSOM, [2]Dept. of Biomed. Educ. and Data Science, Lewis Katz Sch. of Med., [3]Dept. of Biology, Col. of Sci. and Technol., [4]5Department of Psychiatry Lewis Katz Sch. of Med., [5]Dept. of Neurology, Lewis Katz Sch. of Med., Temple Univ., Philadelphia, PA; [6]Dept. of Pathology, Yale Univ. Sch. of Med., New, CT; [7]Med. Col. of Wisconsin-Prevea Hlth., Green Bay, WI; [8]Dept. of Obstetrics & Gynecology, Univ. of Texas, Houston, TX

Abstract: Introduction: Prenatal ethanol exposure (EtOH) can result in a complex developmental abnormality called fetal alcohol syndrome (FAS). Diagnostic criteria include ocular, periocular and other craniofacial abnormalities, deficient somatic and brain growth with abnormal morphogenesis, and neurobehavioral impairment. Although the ocular abnormalities are considered diagnostic of FAS, their pathogenesis is poorly understood. **Methods**: Fetal brain and eye tissues were collected at 11 to 21 wks gestation. The EtOH-exposed and control groups were matched for gestational age (GA), fetal gender, race, and maternal BMI, and were analyzed using ELISA, flow cytometry, immunohistochemistry, western-blot assay and the Caspase-3/7 assay. RNA expression for biomarkers was assessed using Real-Time PCR (qPCR) and Droplet Digital PCR (ddPCR), using a Pearson correlation for p-values. Eye morphometric assays were performed using a sliding digital caliper. *In vivo* rat FAS model was established for studies of toxic effects of EtOH on rat fetal brain and eye development. **Results**: Ten human EtOH-exposed cases (1st or 2nd trimester) were compared with GA-matched controls. EtOH exposure was associated with a consistent pattern of reduced eye and iris diameter. Markers of synaptogenesis were reduced, and caspase-3 activity was increased in human fetal brains. The reduction in eye diameter correlated with levels of EtOH intake ($p=0.001$). EtOH-associated caspase-3 activation correlated with decreased eye size ($p=0.0003$). Studies in the rat FAS model showed similarly increased levels of activated caspase-3 in the FAS pup brains compared to control. Body and brain weight were significantly lower in the EtOH groups at P8 and P15. The levels of Myelin Basic Protein (MBP) and Peripheral Myelin Protein (PMP22), which play important roles in oligodendrocyte development and myelin formation were decreased in EtOH-exposed human fetal brains and eyes. The levels of β-catenin were increased, and BDNF decreased in the EtOH groups of both human and animal brains. Expressions of these molecules play essential roles in neural development. There was a negative correlation between prenatal EtOH intake and brain and eye development. **Conclusion**: Prenatal EtOH exposure alters the

development of the eye in a dose-dependent manner, resulting in microphthalmia, which can be detected in human fetuses as early as 11 wks GA. Thus, recognition of reduced eye size, in combination with specific molecular markers of EtOH toxicity in the fetus may be predictive of FAS postnatally and be useful in the future as biomarkers to identify patients for *in-utero* treatments with targeted neuroprotective therapies.

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Poster

P001. Nervous System Patterning and Developmental Cell Death

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P001.02

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH/NIMHD- Grant Number: S21MD001830
NIH/NIAAA- Grant Number: 1R01AA027808-01A1

Title: Effects of lead exposure (Pb) on cell survival and expression of BDNF/TrkB, in cells: BJ (Fibroblasts), HEK 293 (Embryonic kidney cells) and Hippocampal Neurons

Authors: *J. GONZÁLEZ[1,3], R. GONZALEZ[2,3], K. CORDERO[4], E. CARTAGENA[3], B. MADERA[5], D. BRACHO[6], C. VELAZQUEZ-MARRERO[7];
[2]Natural Sci., [1]Univ. of Puerto Rico, Cayey, PR; [3]Med. Sci. Campus, [4]Anatomia y Neurobiologia, Inst. of Neurobiology-RCMI, San Juan, PR; [5]Med. Sci. Campus, Mol. Sci. Res. Ctr., San Juan, PR; [6]NIEF, Inst. de Neurobiologia-RCMI, San Juan, PR; [7]Inst. of Neurobio., Univ. of Puerto Rico Med. Sci. Campus/ Inst. of Neurobio., San Juan, PR

Abstract: The purpose of this study is to examine the effect of different concentrations of lead on both cell survival and BDNF/TrkB expression. This research arises from the absence of studies that directly address the effect of lead contamination at the relevant concentrations measured in different regions in Puerto Rico. Physiologically relevant concentrations have been determined and used to treat human postnatal fibroblast (BJ) and embryonic kidney (HEK293) cell lines characterizing the effects of lead on both survival and BDNF/TrkB expression using immunocytochemistry. Results indicate significant changes in cell morphology associated with significant changes in both BDNF and TrkB expression. Interestingly, expression of BDNF and TrkB in BJ cells, showed an opposite trend to that observed in HEK293 cells. These results potentially highlight the importance of developmental stage as a key factor in determining the effects of which these cell models are found, one embryonic and the other postnatal. We further

examined the effects of lead in isolated hippocampal neuronal preparations indicating significant effects in both survival and protein expression. Our data thus far, once again highlights the importance of implementing and enforcing regulations on lead contamination in the Island.

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Poster

P001. Nervous System Patterning and Developmental Cell Death

Location: Virtual Only

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Program #/Poster #: P001.03

Topic: A.01. Neurogenesis and Gliogenesis

Support: Knut and Alice Wallenberg Foundation (2015.0041, 2018.0220, 2018.0172)
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Silicon Valley Community Foundation (2018-191929)
Swiss National Science Foundation (CRSK-3_190495 and PZ00P3_193445)

Title: Molecular architecture of the developing mouse brain

Authors: G. LA MANNO[1], ***K. SILETTI**[2], A. FURLAN[3], D. GYLLBORG[4], E. VINSLAND[2], A. MOSSI ALBIACH[2], C. MATTSSON LANGSETH[4], I. KHVEN[1], A. R. LEDERER[1], L. M. DRATVA[1], A. JOHNSSON[2], M. NILSSON[4], P. LÖNNERBERG[2], S. LINNARSSON[2]; [1]École Polytechnique Fédérale de Lausanne, Lausanne, Switzerland; [2]Karolinska Inst., Stockholm, Sweden; [3]Cold Spring Harbor Lab., Cold Spring Harbor, NY; [4]Stockholm Univ., Solna, Sweden

Abstract: The developing nervous system is highly complex, generating an enormous diversity of cellular states over a lengthy gestational window. Although single-cell RNA sequencing is increasingly used to study the molecular architecture underlying this process, most studies to date have focused on particular regions, cell types, or developmental stages. To gain a comprehensive picture of brain development, we performed single-cell RNA sequencing on nearly 300,000 cells isolated from the embryonic mouse forebrain, midbrain, and hindbrain each day between neurulation and birth (E7 to E18.5). An analysis based on our Cytograph pipeline resulted in 798 clusters, which we manually annotated. In addition to a highly diverse set of radial glia, neuroblasts, and neurons, we characterized developing non-neuronal cells, particularly glial progenitors, immune cells, and meningeal fibroblasts. The dataset revealed the striking regional heterogeneity of neural progenitors in the brain, including the brain's

specialized secondary organizers that pattern the developing neural tube. To further investigate the spatial organizations of these cellular states in our dataset, we performed *in situ* mRNA sequencing of 119 genes across 24 sagittal sections in the embryonic day 10.5 mouse brain. Together the single-cell and spatial datasets provide a rich resource for exploring the genes and cellular lineages that contribute to mammalian nervous-system development and disease.

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Poster

P001. Nervous System Patterning and Developmental Cell Death

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Program #/Poster #: P001.04

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH R01 NS105748
Start-up funds, The University of Chicago

Title: Regulation of neural lineages by Hox genes to specify post-mitotic neuronal characteristics during circuit development

Authors: *D. VASUDEVAN[1], H. CARR[1], Y.-W. WANG[2], J. ZUNIGA[1], S. CORCORAN[1], E. HECKSCHER[2];
[2]Mol. Genet. and Cell Biol., [1]Univ. of Chicago, Chicago, IL

Abstract: During development, neural stem cells (NSCs) differentiate into neurons that wire together, forming circuits that drive behaviors. NSCs have enormous therapeutic potential in treating neurodevelopmental disorders and injury. But, this potential is untapped because how NSCs give rise to neural circuits is not fully understood. In the vertebrate spinal cord and invertebrate nerve cord, one set of NSCs are repeatedly found at all segmental levels along the anterior-posterior (A-P) axis. In different A-P locations there are different circuits that control specialized A-P functions. Thus, there must be naturally occurring variations in how stem cells give rise to circuits along the A-P axis. In general, Hox genes underlie A-P variation in multiple tissue types, but it is unknown how Hox genes generate A-P circuit diversity in the CNS. Here, I am developing the Drosophila larval nerve cord as a genetically tractable and numerically simple model to address this question. I am testing which post-mitotic characteristics of neurons is modulated by the posterior Hox gene, Abdominal-B (AbdB). The objectives of this study are first, to test whether the levels of Hox gene expression in NSC sets the levels of Hox gene expression in post-mitotic neuronal progeny. The second objective is to determine how changing

levels of Hox in post-mitotic neurons modulates their morphology, as well as the formation of anatomical and functional input synapses. Specifically, I will focus on NB3-3 neuronal stem cells and the lineage of even-skipped lateral (EL) interneurons that it produces, and investigate the role of the Hox gene, AbdB. This will provide insight into the Hox-based genetic mechanisms that regulate how stem cells give rise to neural circuits.

Disclosures: D. Vasudevan: None. **H. Carr:** None. **Y. Wang:** None. **J. Zuniga:** None. **S. Corcoran:** None. **E. Heckscher:** None.

Poster

P001. Nervous System Patterning and Developmental Cell Death

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P001.05

Topic: A.01. Neurogenesis and Gliogenesis

Support: NSF Grant 1754340
NIH-NINDS Grant R01NS105886
NIH-NINDS Grant R01NS113859
Shriners Hospital 86700-NCA
Shriners Hospital 85111-NCA
NSF Supplement 1754340

Title: Trpm8 activity is necessary for preventing spinal cord motor neuron apoptosis in Xenopus laevis larvae reared at cold temperature

Authors: *A. NAVARRETE VARGAS, O. VISINA, K. SPENCER, L. BORODINSKY; Univ. of California, Davis, Sacramento, CA

Abstract: During nervous system development neurons are produced in excess and as much as 50% of them are pruned by undergoing programmed cell death, in what is known as developmental apoptosis. The mechanisms underlying this process are unclear. Previous studies in the lab demonstrated that *Xenopus laevis* embryos and larvae grown at cold temperature exhibit a higher number of motor neurons than siblings grown at warm temperature and that this cold temperature-dependent increase in motor neuron number correlates with a decrease in motor neuron apoptosis and enhanced motor performance when cold temperature-grown larvae are tested at cold temperature. The mechanisms that contribute to the survival of motor neurons at cold temperatures remain unknown. Here we assessed whether the cold-sensitive transient receptor potential cation channel subfamily M member 8 (TRPM8) is necessary and sufficient in preventing apoptosis of spinal cord neurons, specifically motor neurons, in animals reared at cold temperature. To assess necessity, embryos were injected bilaterally at the two-cell stage with either control or TRPM8-morpholino and reared at cold (16°C) temperature until reaching larval

stage 40, when they were processed for immunostaining for the motor neuron transcription factor, HB9, and for Terminal deoxynucleotidyl transferase dUTP Nick End Labeling (TUNEL) assay to detect apoptotic cells. We observed that TRPM8 knockdown in animals reared at cold temperature leads to a 65-fold increase in number of apoptotic spinal cord cells compared to control and 48% of spinal cord motor neurons are apoptotic in TRPM8-deficient animals compared to only 1% in control counterparts. In contrast, TRPM8 knockdown in animals reared at warm (22.5°C) temperature did not change the number of apoptotic spinal cord cells or the proportion of apoptotic motor neurons. Current efforts are focused on assessing the sufficiency of TRPM8 activation in preventing motor neuron apoptosis in embryos grown at warm temperature and to rescue the deficient motor performance these larvae exhibit when tested at cold temperature. The results so far suggest that TRPM8 is necessary for preventing motor neuron apoptosis in cold temperature-reared animals and highlight the role of the environment in nervous system development. This study may contribute to the understanding of the mechanisms underlying the pathological loss of motor neurons in disease, in which a developmental origin has been considered.

Disclosures: A. Navarrete Vargas: None. **O. Visina:** None. **K. Spencer:** None. **L. Borodinsky:** None.

Poster

P001. Nervous System Patterning and Developmental Cell Death

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P001.06

Topic: A.01. Neurogenesis and Gliogenesis

Support: NSERC grant

Title: Brief exposure to delta^9 (-) THC affects embryonic and adult locomotion; the effect persisted in the offspring

Authors: *M. AMIN[1], K. AHMED[1], J. SZASZKIEWICZ[2], T. HAMILTON[3], D. ALI[1]; [1]Univ. of Alberta, Edmonton, AB, Canada; [3]ASSOCIATE PROFESSOR, PSYCHOLOGY, [2]MACEWAN UNIVERSITY, Edmonton, AB, Canada

Abstract: The stereoselectivity of a compound plays an essential role in it's pharmacological activity. δ^9 (-) THC is a stereoisomer of THC and is the predominant form present in the cannabis plant, *Cannabis sativa*. The goal of this study was to determine if brief exposure of zebrafish embryos to δ^9 (-) THC during the gastrulation stage impacted development. Our results show that embryos treated with concentrations of THC of 1 mg/L and higher did not survive past 15 dpf. Treatment with δ^9 (-) THC resulted in morphological defects, defective synaptic activity and altered motor neuron development in young zebrafish. Additionally, δ^9 (-) THC treatment

altered development of the swimbladder. To investigate the mechanism of action of THC, we stimulated hedgehog and Wnt signaling pathways prior to THC treatment since these pathways are involved in swimbladder development. Our findings showed that overactivation of the Shh pathway using purmorphamine prevented the THC-mediated abnormal swimbladder development, whereas activation of Wnt signaling (using BIO) did not. RNAseq analysis showed that more than 2750 genes were differentially expressed in embryos following treatment with 3 mg/L and 0.5 mg/L of δ^9 (-) THC. δ^9 (-) THC treatment resulted in the downregulation of sonic hedgehog-related genes such as ptch2, smo, gli1, and gli2b. Together, these findings suggest that THC treatment induces changes in the activity of the sonic hedgehog pathway, as suggested by previous work. To determine the long-term effects of exposure to δ^9 (-) THC, we reared THC treated animals to adults and investigated their behavior. Our results showed that there were changes in locomotor activity and swimming behaviors in adults and even into the next generation of animals (F1 embryos). Briefly, adult zebrafish (treated with 0.5 mg/L THC during gastrulation) spent more time near the walls of the tank and less time in the center zone compared with age matched controls. In addition, the F1 embryos from the THC treated parents exhibited decreased spontaneous activity and travelled smaller distances following mechanical, touch stimuli. These
findings suggest that a brief exposure to δ^9 (-) THC, of only 5 hr during gastrulation, may have persistent and long term effects on zebrafish mand even into the next generastion of animals.

Disclosures: M. Amin: None. **K. Ahmed:** None. **J. Szaszkiewicz:** None. **T. Hamilton:** None. **D. Ali:** None.

Poster

P001. Nervous System Patterning and Developmental Cell Death

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P001.07

Topic: A.01. Neurogenesis and Gliogenesis

Support: BBSRC (BB/G007632/1)
Royal Society

Title: Nell2/nel promotes neuronal survival in the developing and aging brain

Authors: C. NAKAMOTO, E. BAKER, A. JARABEK, M. DUNSON, E. FRANGOPOULOS, ***M. NAKAMOTO**;
Dept. of Biol., Valparaiso Univ., Valparaiso, IN

Abstract: Correct functioning of the nervous system is critically dependent on the generation and maintenance of the proper numbers of neurons. During development, a significant portion of the newly generated neurons die through programmed cell death, and neuronal survival is

maintained only if they encounter appropriate environmental cues. Once neurons are selected for survival during the developmental cell death period, the proper numbers of neurons are normally maintained for the lifetime of the organism. Whereas previous studies have revealed factors that regulate developmental neuronal survival, the molecular mechanisms that control the generation of proper numbers of neurons during development and support the long-term neuronal survival in the adult brain are not fully understood. Nell2 (also known as Nel) is a multimodular extracellular glycoprotein that is predominantly expressed in the nervous system. Nell2 exerts diverse functions in neural development, including neuronal proliferation and differentiation, and axon guidance. Our previous study has shown that Nell2 acts as an inhibitory guidance cue for retinal axons in the eye-specific retinogeniculate projection. Nell2 has been shown to promote survival of cerebral and hippocampal neurons *in vitro* and protects retinal ganglion cells (RGCs) from cell death after optic nerve injury. We previously showed that overexpression of Nell2 in the developing chick retina increases the number of RGCs, whereas knockdown of Nell2 expression results in decrease in RGC numbers. Our study revealed that Nell2 positively regulates the RGC numbers by promoting their differentiation and survival during development. In the current study, we investigated the survival-promoting functions of Nell2 in neural development and aging by using Nell2 knockout mice. Nell2 is strongly expressed in RGCs and hippocampal neurons in developing and adult mice. We have found that the numbers of RGCs and hippocampal neurons are significantly decreased in developing (embryonic day (E) 19.5 - postnatal day (P) 4) Nell2 knockout mice. Deletion of the Nell2 gene does not significantly affect neuronal proliferation. However, Nell2 knockout mice showed increased apoptosis both in developing RGCs and hippocampal neurons, suggesting that Nell2 promotes neuronal survival in those areas during development. Furthermore, compared to wild-type, apoptosis of RGCs and hippocampal neurons is significantly increased in the adult and aging (~ P480) Nell2 knockout mice. Taken together, those results suggest that Nells acts as a survival promoting factor that is essential for production and maintenance of proper numbers of neurons.

Disclosures: C. Nakamoto: None. **E. Baker:** None. **A. Jarabek:** None. **E. Frangopoulos:** None. **M. Nakamoto:** None.

Poster

P001. Nervous System Patterning and Developmental Cell Death

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P001.08

Topic: A.01. Neurogenesis and Gliogenesis

Support: Virginia Brain Institute Fellowship
UVA Raven Society Fellowship
Jefferson Scholars Foundation Fellowship

Title: Characterizing phagocytic neural crest cells in early zebrafish neural development.

Authors: *K. E. LIU[1], S. C. KUCENAS[2];
[1]Neurosci. Grad. Program, [2]Dept. of Biol., Univ. of Virginia, Charlottesville, VA

Abstract: During neural development, sculpting of the central and peripheral nervous systems (CNS and PNS, respectively) is necessary to generate a functional and adaptable organism. This sculpting involves the over-proliferation of neurons and subsequent "pruning back" of neuronal projections and synapses to homeostatic levels. During the assembly of activity-dependent circuits, this pruning is necessary for circuit function and plasticity throughout life. Several cell types prune by consuming extracellular debris, apoptotic neurons, and their synapses through a process called phagocytosis. Notably, aberrant phagocytosis is implicated in several devastating neurodevelopmental (e.g. Nasu-Hakola disease, Rett syndrome) and neurodegenerative (e.g. Parkinson's disease, multiple sclerosis) disorders. During development we know that debris is cleared during early neurogenesis, yet there are no professional phagocytes (microglia or macrophages) present in the CNS or PNS. In studies using zebrafish, our lab identified neural crest cells (NCCs) as novel phagocytes that actively consume dying cells during an early developmental period (~24 to 40 hours post-fertilization [hpf]). While we now know that NCCs aid in debris clearance during this window, the mechanisms, interactions, and effects of NCC phagocytosis on neural development are not known. Zebrafish provide an unparalleled advantage to examine NCCs in early neurodevelopment because of their accessibility at early developmental stages, genetic tractability, capacity for *in vivo* time-lapse imaging, and targetable early-formed circuits. The early zebrafish touch-response circuit consists of primary sensory neurons in the dorsal spinal cord (Rohon-Beard [RB] neurons) which synapse with local interneurons and primary motor neurons. By 24 hpf, these connections form a touch-response circuit that facilitates early recoil responses in developing embryos. This circuit changes drastically over development: RB neurons quickly degenerate and are replaced by dorsal root ganglion neurons in this circuit, producing cellular debris in need of phagocytic clearance. Using zebrafish transgenic lines that fluorescently mark phagocytic components in NCCs (Lamp1, PI(3)P), we are characterizing the effects of NCC phagocytosis on the touch-response circuit *in vivo*. Understanding the molecular basis behind phagocytes in neural development contributes vital knowledge to their aberrant function in neurodevelopmental and neurodegenerative disorders and could lead the way to novel therapeutic strategies for these conditions.

Disclosures: K.E. Liu: None. **S.C. Kucenas:** None.

Poster

P001. Nervous System Patterning and Developmental Cell Death

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P001.09

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH R01HD092593
3R01HD092593-S1
Research Foundation of Cerebral Palsy Alliance #3720

Title: Preterm birth alters the maturation of the GABAergic system in the human prefrontal cortex

Authors: *H. LACAILLE, C.-M. VACHER, J. SALZBANK, J. O'REILLY, A. A. PENN; Pediatrics, Columbia Univ. Med. Ctr., New York, NY

Abstract: Alterations in neurotransmitter systems, particularly changes in GABAergic and glutamatergic systems, during frontal lobe development have been hypothesized to play a key role in neurodevelopmental disorders seen in children born very preterm (<32 weeks gestation) and at very low birth weight. Prematurity frequently results in neurodevelopmental disorders such as autism, attention deficit disorders or epilepsy in the absence of radiologically visible lesions, suggesting that long-term physiological alterations might underlie these pathologies. Recent magnetic resonance spectroscopy imaging studies have suggested that GABA concentrations may be lower in preterm than term infants, but cannot assess the components of the GABAergic system that may have altered development. Here we studied the molecular development of the GABAergic system specifically in the dorsolateral prefrontal cortex (Brodmann area 10-46), a region that plays a role in attention and working memory and that has been implicated in neurodevelopmental and psychiatric disorders. The maturation state of the GABAergic system in this region was assessed in human post-mortem brain samples of 17 male and 9 female term infants, ranging in age from 0 to 8 months obtained from the NIH NeuroBiobank. Gene expression levels were measured for 47 genes related to GABAergic development and this gene expression data was used to calculate a maturation index. The index correlated with the age of the donor and was more dynamic in male than in female infants, suggesting a link to the increased male infant susceptibility to impaired neurodevelopment . To evaluate the impact of premature birth on the GABAergic system development, 9 male and 4 female one-month-old term and 8 male and 6 female one-month corrected-age very preterm infants, were compared using the same gene list and methodology. The maturation index for the GABAergic system was significantly lower (-50% $p<0.05$) in male preterm infants, with major alterations in genes linked to GABAergic function in astrocytes. These preliminary findings support further investigations of astrocytic GABAergic development as a key contributor to abnormal neurodevelopment in preterm infants.

Disclosures: H. Lacaille: None. **C. Vacher:** None. **J. Salzbank:** None. **J. O'Reilly:** None. **A.A. Penn:** None.

Poster

P001. Nervous System Patterning and Developmental Cell Death

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P001.10

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH Grant R01MH122478
NIH Grant R01EY025174
NIH Grant R01NS116598

Title: Activity of Transplanted MGE-derived Interneurons over Their Period of Programmed Cell Death

Authors: *B. RAKELA[1], W. R. MANCIA LEON[2], M. HOSEINI[4], A. R. HASENSTAUB[5], A. ALVAREZ-BUYLLA[3], M. P. STRYKER[6];
[1]Physiol., [3]Eli and Edythe Broad Ctr. for Regeneration Med. and Stem Cell Res., [2]Univ. of California San Francisco, San Francisco, CA; [4]Physiol., Univ. of California, San Francisco, CA; [5]Otolaryngology / Ctr. for Integrative Neurosci., UCSF, San Francisco, CA; [6]Ctr. for Integrative Neurosci, Dept Physiol, Univ. California Med. Sch., San Francisco, CA

Abstract: Two major classes of inhibitory cortical interneurons (cINs) derived in embryonic development from the medial ganglionic eminence (MGE), one expressing parvalbumin (PV) and the other somatostatin (SST), are required for the induction of critical period plasticity within primary visual cortex (V1) in mouse. After their migration from MGE into the developing cortex, they undergo a period of selection via programmed cell death around postnatal day 7, ~60% survive to integrate into the local circuit. Why do some cINs survive and others die? cIN precursors transplanted into postnatal hosts develop according to the intrinsic cIN timeline making it possible to study genetic mutations that would render the donor animal non-viable and offering access *in vivo* to early developmental time points where the same cells can be tracked across maturation. Earlier studies have shown that programmed cell death takes place at the same time after neurogenesis in both endogenous and transplanted cINs (Southwell et al., 2012), and suggest that activity of cINs at the onset of programmed cell death predicts their survival (Wong et al., 2018). Here, we have studied mutant cINs lacking the cell surface homophilic-binding proteins in the protocadherin (Pcdh)-γ gene cluster, in which programmed cell death of PV and SST cINs is significantly increased (Mancia et al, 2020), to probe the role of correlated activity in the selection of cINs for survival. We transplanted wildtype and mutant cINs expressing GCaMP7f in order to monitor their activity *in vivo* in relation to that of host neurons during the period of programmed cIN death. These data show that transplanted cINs are active before the time of programmed cell death, activity was more highly correlated among host neurons than among transplanted cINs, and activity among mutant cINs was even less correlated. These findings on correlated activity are consistent with the notion that the increased cell death of Pcdh-γ mutant cINs may be due to impairment of their connectivity with surrounding cells or to signaling events that depend on particular patterns of activity.

Disclosures: B. Rakela: None. **W.R. Mancia Leon:** None. **M. Hoseini:** None. **A.R. Hasenstaub:** None. **A. Alvarez-Buylla:** E. Ownership Interest (stock, stock options, royalty,

receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Neurona Therapeutics. F. Consulting Fees (e.g., advisory boards); Neurona Therapeutics. **M.P. Stryker:** None.

Poster

P010. Postnatal Neurogenesis: Environmental and Pharmacological Regulation I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P010.01

Topic: A.02. Postnatal Neurogenesis

Support: NIH Grant U54 MD007601

Title: The critical role of selenium in perineuronal net and GABAergic inhibitory circuit development

Authors: *A. SASUCLARK, M. PITTS;
Cell and Mol. Biol., Univ. of Hawaii, Honolulu, HI

Abstract: Parvalbumin interneurons (PVIs) are an important class of highly metabolic GABAergic interneurons known for their role in modulating neuronal synchrony and activity. During development, PVIs are selectively surrounded by perineuronal nets (PNNs), extracellular matrix structures that assist in stabilizing synaptic input to PVIs and serve to protect against oxidative damage. Conversely, redox dysregulation disrupts maturation of both PVIs and PNNs, and is hypothesized to be a key contributing factor to PVI/PNN abnormalities in neuropsychiatric disorders, most prominently schizophrenia. Oxidative stress is countered by a host of antioxidant enzymes, including multiple selenoproteins, a unique clade of proteins characterized by the co-translational incorporation of selenium as the amino acid selenocysteine. Moreover, selenoproteins are required for PVI development. Yet, despite the known indispensable role of selenoproteins in PVIs, their influence on the temporal dynamics of PNN maturation has yet to be investigated. Thus, primary neuron/glia mixed cultures were grown in serum-free media containing deficient (10 nM), optimal (100 nM), or excessive (1 uM) levels of selenium, and assessed at various time points for PNN maturation, antioxidant activity, and electrical activity using micro-electrode array (MEA) plates. At 28 days in vitro (DIV), the density of both PNNs and PNN-labeled PVIs were significantly increased in the 100 nM selenium condition relative to the other groups. In parallel, cellular GPx activity was significantly elevated in both the 100 nM and 1 uM groups relative to the 10 nM group at 28 DIV. Finally, we also observed significant differences in our MEA data at 28 DIV, with our 100 nM selenium condition exhibiting significantly lower mean firing rates and burst frequencies than the 10 nM condition. In conclusion, our results show that selenium is an important modulator of PNN maturation and suggest that selenium-deficiency impedes proper development of GABAergic inhibitory circuits.

Disclosures: A. Sasuclark: None. **M. Pitts:** None.

Poster

P010. Postnatal Neurogenesis: Environmental and Pharmacological Regulation I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P010.02

Topic: A.02. Postnatal Neurogenesis

Support: NIH Grant R00NS089013 and R56AG064077
Whittier Foundation
Baxter Foundation
Broad Foundation
USC-CONACYT Postdoctoral Fellowship
USC Provost's Postdoctoral Research Grant

Title: Neural regeneration in adult Drosophila

Authors: *I. FERNANDEZ-HERNANDEZ, E. MARSH, E. HU, M. A. BONAGUIDI;
Stem Cells Biol. and Regenerative Med., USC, Los Angeles, CA

Abstract: Adult neurogenesis occurs at different extents in specific locations of the vertebrate nervous system throughout life, and its deregulation has been linked to diverse pathologies. Therefore, interventions aimed at restoring functional neurogenesis upon injury and ageing represent a promising strategy to rejuvenate the adult nervous system. With an unparalleled genetic and functional toolbox, *Drosophila* represents a compelling model to accelerate the investigation of mechanisms promoting neural regeneration. Yet, generation of new neurons in the adult fly has been elusive thus far. We generated a modified lineage tracing system which allows for the sustained identification of adult-born neurons in the fly and their genetic manipulation in a cell type-specific manner. Here I present evidence for the continuous generation of new neurons in the optic lobes, the auditory/vestibular, as well as the olfactory system of adult flies in physiologic conditions. Furthermore, acute injury and oral administration of small molecules enhance neuronal regeneration. These results unveil the hidden proliferative capacity of the adult *Drosophila* nervous system, and underscore its potential to expedite the identification of conserved mechanisms and compounds promoting nervous system regeneration at the single-cell, circuitry, and functional levels.
-Fernández-Hernández, I., Rhiner, C., Moreno E. 2013; *Adult neurogenesis in Drosophila*. Cell reports. Volume 3, Issue 6: 1857-1865
-Fernández-Hernández, I.*, Marsh, E., Bonaguidi, M.*, 2020 *Mechanosensory neuron regeneration in adult Drosophila*. Development. 2021 Mar 10;148(5)
-Fernández-Hernández, I.*, Hu, E., Bonaguidi, M.* *Olfactory neuron turnover in adult Drosophila*. (bioRxiv: doi.org/10.1101/2020.11.08.371096)

Disclosures: I. Fernandez-hernandez: None. **E. Marsh:** None. **E. Hu:** None. **M.A. Bonaguidi:** None.

Poster

P010. Postnatal Neurogenesis: Environmental and Pharmacological Regulation I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P010.03

Topic: A.02. Postnatal Neurogenesis

Support: New Jersey Governor's Council for the Medical Research and Treatment of Autism CAUT20AFP004
NIEHS P30 ES005022

Title: Caesarean delivery stimulates neural stem cell markers in the postnatal mouse brain.

Authors: *J. K. LESSING[1], X. ZHOU[2], H. SUN[1], J. WANG[1], M. G. DOMINGUEZ-BELLO[1], E. DICICCO-BLOOM[2];
[1]Dept. of Biochem. and Microbiology, Rutgers Univ., New Brunswick, NJ; [2]Dept. of Neurosci. and Cell Biol., Rutgers Univ., Piscataway, NJ

Abstract: Neurodevelopmental diseases - including autism spectrum disorder, attention deficit hyperactivity disorder (ADHD), and learning disability - are thought to be caused by a combination of genetic susceptibility and environmental triggers. Caesarean-section delivery (CS) may be an environmental trigger of developmental disorders. Human cohort studies correlate CS to a ~20% increased risk of developing autism, a ~17% increased risk of ADHD, and increases in metabolic diseases like obesity and type-1-diabetes. Though CS is a medical necessity, the World Health Organization estimates CS rates above 10% do not improve maternal or neonatal health, in healthy mothers. While the global CS rate in 2015 reached 21.1%, it was 31.7% in US in 2019. Disorders associated with CS may be caused by many factors, including perinatal distress, maternal anesthesia, altered perinatal hormonal environment, and disrupted vertical transmission of microbiome from mother to neonate. To study effects of CS on neurodevelopment we designed a model system in outbred mice. Previous data from this model indicates CS alters gut microbiome composition by weaning at postnatal day 21 (P21). Further, CS induces changes in ileal transcriptome at P7. We now extend the paradigm to consider how CS affects neurodevelopment. Timed-pregnant dams were allowed to give birth vaginally (control) or underwent terminal CS at gestational day 19. Pups from both groups were cross-fostered by dams giving birth in the previous 24-hours. Following sacrifice on P8, we isolated cerebellum, hippocampus, and medial prefrontal cortex based on associations with neurodevelopmental diseases and periods of neurogenesis. Cerebellar external germinal layer proliferation peaks from P5-P10 and continues to P18; hippocampal dentate gyrus precursors proliferate throughout life; medial prefrontal cortex serves as negative control because neuronal

precursor proliferation is largely complete by E17.5, though gliogenesis is ongoing. Brain region protein extracts were assessed by immunoblotting for neurodevelopmental markers. Preliminary data in hippocampus shows that male mice that underwent CS had higher levels of proliferation marker cyclin E ($p = 0.001$; $n = 7$/group) and potentially increased doublecortin ($p = 0.08$). No significant differences were detected in females. In sum, CS was found to increase levels of neural stem cell markers in the hippocampus, and other regions are being analyzed. Interestingly, this CS effect was observed only in males. Future studies will analyze additional brain regions and time points, and investigate transcriptional and microbiome-based mechanisms underlying these effects.

Disclosures: J.K. Lessing: None. **X. Zhou:** None. **H. Sun:** None. **J. Wang:** None. **M.G. Dominguez-Bello:** None. **E. DiCicco-Bloom:** None.

Poster

P010. Postnatal Neurogenesis: Environmental and Pharmacological Regulation I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P010.04

Topic: A.02. Postnatal Neurogenesis

Support: NSF grant IOS-1923094 to EDK
Second-year Transformational Experience Program from OSU to ND

Title: Tamoxifen administered via intraperitoneal injections leads to long-term suppression of adult hippocampal neural progenitor cell proliferation

Authors: *N. DEVASTHALI, B. M. SMITH, D. L. EINSTEIN, P. SARCHET, E. D. KIRBY; Dept. of Psychology, The Ohio State Univ., Columbus, OH

Abstract: Research on neurogenesis in the adult rodent hippocampus frequently uses the Cre-Lox system as a method for manipulating gene expression in the adult neurogenic cell lineage. Cre recombinase catalyzes recombination of LoxP-flanked genetic sequences, which can be used to knock down gene expression by excising LoxP-flanked target genes, or to express a novel transgene, via removal of a LoxP-flanked stop sequence. To specifically target such recombination to adult neural stem and progenitor cells, cell specific promoters, such as Nestin or GLAST, are often used to drive a TAM-inducible version of Cre: $CreER^{T2}$. In this system, TAM is required to drive Cre nuclear localization and thus LoxP recombination, but there are conflicting studies about the effects of TAM itself on neurogenesis. Thus, we investigated the effects of TAM on adult hippocampal neurogenesis. Male and female adult C57/Bl6J mice were treated with 5 daily IP injections of either TAM (n=11) or oil (n=12). On the last 3 days of TAM treatment, all mice also received daily IP injections of the mitotic marker 5-bromo-2'-deoxyuridine (BrdU). 21 days later, all mice received an IP injection of 5-ethynyl-2´-

deoxyuridine (EdU) 2h before perfusion and brain harvest. We found no significant differences in body weight between groups during TAM injections, but 21 days after injections, female TAM-treated mice weighed significantly more than oil-treated female mice. TAM had no effect on body weight in males. Neither sex showed any differences between groups in the number of surviving BrdU+ cells or in BrdU co-labeling with neuronal phenotype markers 3 weeks after TAM. In contrast, cell proliferation was significantly suppressed 3 weeks after TAM in both sexes: TAM treated mice had fewer total EdU+ proliferating cells, fewer EdU+/Sox2+ proliferating intermediate progenitors, and fewer Sox2+ total intermediate progenitors than oil treated mice. Our findings suggest that TAM suppresses the proliferation of progenitor cells 3 weeks after TAM is administered, while neurogenesis that begins during the period of TAM administration is not affected 3 weeks later. Similarly, body weight is unaffected by TAM during administration, but in female mice, previous TAM exposure leads to greater weight gain in the weeks following TAM. These results indicate that when studying genetic models that rely on TAM-dependent Cre recombination, oil injection is not an appropriate control for TAM injections even if evaluation time points are weeks after TAM. Rather, all mice should receive TAM and genotype comparisons should be used to avoid the confound of TAM effects of neurogenesis processes.

Disclosures: N. Devasthali: None. **B.M. Smith:** None. **D.L. Einstein:** None. **P. Sarchet:** None. **E.D. Kirby:** None.

Poster

P010. Postnatal Neurogenesis: Environmental and Pharmacological Regulation I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P010.05

Topic: A.02. Postnatal Neurogenesis

Support: NIH NIA

Title: Activity of Ciliated Ependymal Cells Controls Directional Flow and Polarity of the Ciliated Cells

Authors: *K. MELLANSON[1], N. PEUNOVA[2];
[1]State Univ. of New York, Stony Brook, EAST SETAUKET, NY; [2]Stony Brook Univ., Stony Brook, NY

Abstract: ACTIVITY OF CILIATED EPENDYMAL CELLS CONTROLS DIRECTIONAL FLOW AND POLARITY OF THE CILIATED CELLS Kennelia Mellanson[1,2,3], Tatyana V. Michurina[2,3], Anatoli Mikhailik[2,3], Ken-ichi Takemaru[4], Grigori Enikolopov[2,3], and Natalia Peunova[2,3] [1]Molecular and Cellular Pharmacology, SBU, NY 11794[2]Center for Developmental Genetics, SBU, NY 11794[3]Dept. of Anesthesiology,

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Efficient directional flow of the cerebrospinal fluid along of the walls of the brain ventricles requires coordinated and robust activity of the ciliated cells in the ependymal layer of the ventricles. Reciprocally, flow is recognized as a signal by multiciliated ependymal cells and by primary cilia of neural stem cells. Compromised flow has a negative feedback effect on the activity of the multiciliated cells and on the division of neural stem and migration of progenitor cells in the subventricular zone (SVZ) of the lateral ventricles. We investigated the effect of impaired ciliated cell activity in mouse mutant lines on directional flow, cilia beat frequency (CBF), ciliated cell polarity, and proliferation and migration of progenitor cells We studied the explants of the brain ventricles using live imaging, assessing the function of the ciliated cells in the experimental setup that we have developed to measure in parallel the ciliary beat and directional flow and to quantify the responses to various types of treatment within the same group of individual cells before and after a particular treatment, (rather than applying conventional averaging of the responses across different regions of a tissue), thereby significantly increasing the accuracy of the ciliary function analysis. We applied super-resolution fluorescent microscopy to determine the effects of mutations on the ciliated cell structure. We found that conditional mutation of CEP164, a component of the distal appendage of the cilia, when targeted specifically to the ciliary cells, compromises directional flow and CBF. Specifically, it affects the overall arrangement of the ciliated cells, their shape, and their rotational, translational, and tissue-level polarity. Ependymal cells of the CEP164fl/fl mutant are smaller and are more densely packed; their basal bodies cover a greater fraction of the apical surface of the ciliated cell and show greater displacement. We also show that decreased number of cilia generated in CEP164 mutant due to the defects in the docking of the basal bodies to the apical membrane of the ciliated cells. We also studied how proliferation and migration of neural progenitors in the CEP164$^{-/-}$ SVZ is dependent on the fluid flow.

Disclosures: K. Mellanson: None. **N. Peunova:** None.

Poster

P010. Postnatal Neurogenesis: Environmental and Pharmacological Regulation I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P010.06

Topic: A.02. Postnatal Neurogenesis

Support: R01AA016959
T32AA007471

Title: Similar reactive proliferation and neurogenesis in male and female rat dentate gyrus following alcohol dependence.

Authors: *K. R. THOMPSON, S. P. GUERIN, N. N. NAWARAWONG, K. NIXON; Pharmacol. & Toxicology, The Univ. of Texas at Austin, Austin, TX

Abstract: Neurodegeneration occurs with excessive alcohol consumption, the hallmark of an alcohol use disorder (AUD). However, in abstinence, cognitive and structural recovery is observed. Adult hippocampal neurogenesis is a mechanism by which recovery may occur in the hippocampus following alcohol dependence. Reactive neural stem / progenitor proliferation and subsequent neurogenesis has been reported in male rats following dependence, but it is unknown if reactive neurogenesis occurs similarly in females. To study this, we quantified the effects of alcohol dependence on proliferating cells in the dentate gyrus using 5'-bromo-2'-deoxurdidine (BrdU) and immature newborn neurons using Neuronal Differentiation 1 (NeuroD1) in the dentate gyrus. Ethanol was given to adult Sprague-Dawley rats (PND 65-70) by intragastric gavage (25% w/v ethanol in vanilla Ensure Plus®) every 8 hours for 4 days. Control animals received an isocaloric caloric diet. Average intoxication behavior was 1.4 ± 0.1 for females (0-5 scale; $p<0.05$ vs. males) corresponding to hypoactive activity and 1.8 ± 0.1 for males corresponding to ataxic with an elevated abdomen, resulting in animals receiving 10.9 ± 0.2 g/kg/day (F) and 9.8 ± 0.2 (M) ethanol and blood ethanol concentrations of 317.2 ± 19.8 (F) and 358.4 ± 15.7 (M). Withdrawal severity was scored 10 - 28 hours after the last dose of ethanol. Female rats averaged 0.3 ± 0.1 and males 0.6 ± 0.1 (0-4 scale; hyperactivity) with peak withdrawal scores of 2.1 ± 0.3 (F; arched back) and 2.7 ± 0.2 (M; general tremor). Females had significantly lower intoxication behavior, but no other differences were found between groups. Rats were injected with BrdU (300 mg/kg, i.p.) and sacrificed 2 hours later at either 7 days (T7) or 14 days (T14) after the last dose of ethanol. At T7, BrdU+ cells were significantly higher in ethanol-exposed animals (main effect of diet, $p<0.05$), but no differences were found for sex. As previously shown, at T14 NeuroD+ cells were significantly higher in the inner 1/3 of the granule cell layer of ethanol-exposed animals and in males (main effects of diet and sex, $p<0.05$), but no significant diet by sex interaction. Potentially ectopic NeuroD+ cells were also quantified (hilus, molecular layer), but no significant differences were found between either diet or sex groups. These results suggest that alcohol-induced reactive proliferation and neurogenesis are similar in male and female rats, but that basal rates of neurogenesis differ. Further, the lack of ectopic newborn neurons could indicate that reactive neurogenesis is also beneficial in adult female rats following alcohol dependence and may contribute to recovery of the hippocampus.

Disclosures: K.R. Thompson: None. **S.P. Guerin:** None. **N.N. Nawarawong:** None. **K. Nixon:** None.

Poster

P010. Postnatal Neurogenesis: Environmental and Pharmacological Regulation I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P010.07

Topic: A.02. Postnatal Neurogenesis

Support: NIDCD F31DC018469

Title: Microglia-neuroblast interactions in the developing rostral migratory stream

Authors: *S. MELLER[1], L. HERNANDEZ[2], E. MARTÍN LÓPEZ[4], T. LIBERIA VAYÁ[3], C. A. GREER[5];
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Abstract: This work examines the interactions between microglia, neuroblasts and radial glia in the developing rostral migratory stream (RMS) and investigates if microglia ablation disrupts neuroblast migration. CX3CR1-GFP mice, which express EGFP under the Cx3cr1 locus in microglia, were electroporated with a tdTomato plasmid in the cerebral lateral ventricles at postnatal day 0 (P0) to label proliferating neuroblasts and sacrificed at P4, P7, and P14. Additional pups were sacrificed at P8 and P21 for electron microscopy analysis. To selectively deplete microglia in vivo, CX3CR1-GFP/DCX-DsRed mice, which express Ds-Red under the dcx promoter, were injected with liposomal clodronate in the lateral ventricles bilaterally at P0. Mice were injected with 25 mg/kg BrdU two hours later to label dividing cells and sacrificed at P3. To assess microglia and neuroblast density, cells were segmented and quantified using Fiji (ImageJ) software. Differences in cell density were evaluated with a two-way ANOVA and Bonferroni's multiple comparisons posthoc test. Microglia closely associate with migrating neuroblasts in the RMS core and with radial glia processes at the dorsal border during early postnatal development. There was a significant decrease in RMS microglia density three days after liposomal clodronate injection. However, this was not accompanied by a corresponding decrease in neuroblast density at either the start of the RMS or along its descent. Thus, we found that microglia interact with migrating neuroblasts and radial glia during early postnatal development of the RMS. However, microglia ablation did not impact the density of migrating neuroblasts in the RMS. In ongoing work we are using a conditional genetic model to examine the effects of microglia knockdown on neuroblast migration and RMS architecture at later developmental ages.

Disclosures: S. Meller: None. **L. Hernandez:** None. **E. Martín López:** None. **T. Liberia Vayá:** None. **C.A. Greer:** None.

Poster

P010. Postnatal Neurogenesis: Environmental and Pharmacological Regulation I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P010.08

Topic: A.02. Postnatal Neurogenesis

Support: R01 AA016959
R01 AA025591
F31 AA023459
T32 AA007471

Title: Similar Morris Water Maze-induced neuronal activation in the dentate gyrus following alcohol dependence: relationship to adult neurogenesis

Authors: *N. N. NAWARAWONG[1], C. G. NICKELL[2], J. R. PAULY[2], K. NIXON[1,2]; [1]Pharmacol. & Toxicology, The Univ. of Texas at Austin, Austin, TX; [2]Pharmaceut. Sci., Univ. of Kentucky, Lexington, KY

Abstract: Abstinence from alcohol leads to structural and functional recovery in a number of brain regions, especially the hippocampus. Striking increases in neural stem cell (NSC) proliferation and subsequent "reactive neurogenesis" coincides with the structural recovery of the hippocampal dentate gyrus (DG) in a model of an alcohol use disorder (AUD). However, whether these reactively born neurons are functionally integrated into hippocampal circuits is not known. Utilizing a triple label immunohistochemical approach, we previously showed newborn neuronal c-Fos activation in the DG of ethanol-treated rats following Morris Water Maze (MWM) training / testing. Despite the specificity this method provided, we were limited by minimal DG c-Fos expression, which is known to decline with repeated trainings. Unlike c-Fos, the immediate early gene (IEG) Arc remains more consistent with repeated training experiences. Thus, we utilized Arc to examine whether neurons born during alcohol-induced reactive neurogenesis are incorporated into a learned MWM-induced ensemble. Male Sprague-Dawley rats were gavaged with either a 25% (w/v) ethanol diet or an isocaloric control diet every 8 h for 4 days in a Majchrowicz model. Rats received a mean dose of 9.7 ± 0.2 g/kg/day resulting in peak blood ethanol concentrations of 401 ± 14 mg/dL. Typical withdrawal severity was observed with scores averaging 1.4 ± 0.2 out of four (tail tremors) and peaking at 3.6 ± 0.1 (chattering teeth). On day 7 of abstinence (peak reactive NSC proliferation), rats were injected with bromodeoxyuridine (BrdU) to label dividing cells and left undisturbed for 6 wks. In the 7th week, all rats underwent the MWM for 4 days, then were sacrificed and tissue was collected 90 min after the final MWM session. We previously reported significantly more newborn neurons (BrdU+/NeuN+) in the dentate gyrus of the ethanol-treated rats compared to controls ($p<0.05$). Despite this increase, there were no observed group differences in the number of c-Fos+ cells or MWM-activated newborn neurons (BrdU+/c-Fos+/NeuN+). Here, we found 3.4x more Arc+ cells in the DG following MWM training / testing compared to our previous report of c-Fos+ cells ($p<0.05$). Despite the greater quantity of Arc+ cells overall, there was no group difference in the number of Arc+ cells in the DG of ethanol rats compared to controls, similar to our prior report with c-Fos. Thus, MWM training induces the IEG Arc similarly between control and ethanol rats with reactive neurogenesis. These data contribute to our growing understanding that ethanol-induced reactive neurogenesis may contribute to recovery of functional hippocampal circuitry.

Disclosures: N.N. Nawarawong: None. **C.G. Nickell:** None. **J.R. Pauly:** None. **K. Nixon:** None.

Poster

P054. Fragile X: Behavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P054.01

Topic: A.07. Developmental Disorders

Support: ZonMV MKMD 114024154

Title: Testing time and light as environmental factors regulating behavioral phenotype expression in FMR1-KO mice

Authors: *M. ARROYO ARAUJO, R. KAT, R. HAVEKES, P. MEERLO, M. J. H. KAS; Groningen Inst. for Evolutionary Life Sci., Univ. of Groningen, Groningen, Netherlands

Abstract: The value of animal models in biomedical research relies on their capacity to partly represent a human disorder and to predict an outcome given specific baseline conditions. However, the translation of animal models into clinical trials has proven to be challenging given the high rate of conflicting results found in preclinical literature. One of the most informative approaches to investigate the consistency of results in big pools of scientific literature is by performing a systematic search and meta-analysis of published studies. The inconsistency in phenotype expression of the Fragile-X syndrome mouse model (FMR1-KO) drove us to run a systematic search on the behavioral phenotype of this line; 3194 articles showed as hits for the search terms used. Out of this set of publications, 336 articles were fully screened to extract the behavioral outcome data reports. Additionally, study characteristics of the studies' methodology were collected to inform us whether environmental conditions could explain the inconsistency seen in the phenotype expression. Given the large variability of results between studies and the incomplete methodological reporting, we conducted an experiment to investigate whether environmental factors such as the time of testing relative to the light-dark cycle and the light conditions during the test could influence the phenotype differences between FMR1 KO mice and their wild-type (WT) littermates. To this end, four of the most used behavioral tests according to our meta-analysis results were performed; these tests measure some of the typical Fragile-X/autism-like symptoms. Elevated plus maze, Open field test (OF), Acoustic startle response, and Pre-pulse inhibition (PPI) were carried out at the beginning of the light or dark phase. Animals were tested under lighting conditions matching those of their housing room at the time of testing or the opposite. With this study, we aimed to separate the influence of the lighting conditions *per se* from the possible circadian influence. Results suggest that the tests we performed are differentially sensitive to light and time of day. For example, while in the OF there was a robust hyperactivity phenotype in all conditions, in the PPI test there was a larger variation

in terms of the direction of the genotype effect depending on the environmental conditions. Altogether, this suggests that reporting of pre-clinical studies should become more transparent, and that environmental factor's influence should be further explored given the suggested impact on the interpretation and use of animal models and their potential translation to clinical trials.

Disclosures: M. Arroyo Araujo: None. **R. Kat:** None. **R. Havekes:** None. **P. Meerlo:** None. **M.J.H. Kas:** None.

Poster

P054. Fragile X: Behavior

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Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P054.02

Topic: A.07. Developmental Disorders

Support: NIH Grant HD097067
USDA Grant 2018-67001-28266

Title: Parental Reports on Early Autism Behaviors in Their Children with Fragile X Syndrome as a Function of Infant Feeding

Authors: *C. J. WESTMARK;
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Abstract: This study evaluates the prevalence of autistic behaviors in fragile X syndrome as a function of infant diet. Retrospective survey data from the *Fragile X Syndrome Nutrition Study*, which included data on infant feeding and caregiver-reported developmental milestones for 190 children with fragile X syndrome enrolled in the Fragile X Online Registry with Accessible Database (FORWARD), were analyzed. Exploratory, sex-specific associations were found linking the use of soy-based infant formula with worse autistic behaviors related to language in females and self-injurious behavior in males. These findings prompt prospective evaluation of the effects of soy-based infant formula on disease comorbidities in fragile X syndrome, a rare disorder for which newborn screening could be implemented if there was an intervention. Gastrointestinal problems were the most common reason cited for switching to soy-based infant formula. Thus, these findings also support the study of early gastrointestinal problems in fragile X syndrome, which may underly the development and severity of disease comorbidities. In conjunction with comorbidity data from the previous analyses of the *Fragile X Syndrome Nutrition Study*, the findings indicate that premutation fragile X mothers should be encouraged to breastfeed.

Disclosures: C.J. Westmark: None.

Poster

P054. Fragile X: Behavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P054.03

Topic: A.07. Developmental Disorders

Support: NIH Grant F32DC015160
NARSAD Grant 27206
NIH Grant K01DC018310

Title: Auditory Hypersensitivity and Perceptual Disruptions in a Rat Model of Fragile X Syndrome

Authors: *B. AUERBACH[1,2], X. LIU[1,2], K. RADZIWON[2], R. SALVI[2];
[1]Mol. & Integrative Physiol., Univ. of Illinois at Urbana-Champaign, URBANA, IL; [2]Ctr. for Hearing and Deafness, State Univ. of New York at Buffalo, Buffalo, NY

Abstract: Abnormal sensory processing is a hallmark of autism spectrum disorders (ASD) and related neurodevelopmental disorders like Fragile X syndrome (FX), most notably manifesting as extreme sensitivity to sound (i.e. hyperacusis). Auditory hypersensitivity is not only a pressing clinical problem in FX and ASD, but it also likely reflects fundamental circuit defects that extend to more complex but less accessible features of the disorders, such as impaired communication and language development. We have developed a model of auditory hypersensitivity in a *Fmr1* knockout (KO) rat model of FX using an operant conditioning task to assess auditory reaction time-intensity (RT-I) functions, a reliable psychoacoustic measure of loudness growth, using a variety of stimulus frequencies, bandwidths and durations. *Fmr1* KO and littermate wildtype (WT) rats learn the task at the same rate and reach the same level of peak performance but *Fmr1* KO animals exhibited faster auditory RTs over a broad range of intensities and steeper RT-I slopes compared to WT controls, consistent with perceptual evidence of excessive loudness growth. *Fmr1* KO animals also displayed altered perceptual integration of sound duration and bandwidth compared to WT controls, suggesting that faster RTs in these animals is evidence of aberrant auditory processing rather than generalized hyperactivity or altered motor responses. This novel phenotype has the potential to uncover fundamental deficits at the core of FX and ASD pathophysiology while also having direct clinical implications for one of the most disruptive features of these disorders.

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Poster

P054. Fragile X: Behavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P054.04

Topic: A.07. Developmental Disorders

Support: Smith Foundation ODYSSEY

Title: Altered Developmental Trajectory in Fragile X Knockout Rats Revealed By Ultrasonic Vocalizations

Authors: *J. H. BLADON[1], A. HOOKER[1], Z. PRANSKE[1], N. DIAZ-RODRIGUEZ[1], S. P. JADHAV[2];
[2]Psychology, [1]Brandeis Univ., Waltham, MA

Abstract: Fragile X is the single most prevalent monogenetic cause of intellectual disability. Those with Fragile X are more likely to exhibit behavioral disabilities consistent with Autism Spectrum Disorder (ASD), that include delayed developmental milestones, poor communication skills, and cognitive inflexibility. One common early indicator Autism and Fragile X is delayed development of social skills. To more closely examine the altered developmental trajectory as it relates to the pathology of Fragile X, we examined the vocal development of young Fragile X knockout rats (FMRP null) using a maternal separation model. To do this, we recorded ultrasonic vocalizations (USV's) for three minute every two days in P4 to P18 rats during social isolation in a soundproof chamber. We then used image processing software in Matlab (DeepSqueak & USVseg) to isolate and analyze vocalizations. We found significant sex, age, and genotype effects, as well as interactions between the three on the number, principal frequency, duration, and tonal variety of the pups' USVs. Across all sexes and genotypes, call rate and length began low, peaked around P8, and fell again as the animals grew older. Interestingly, while call rate was not impacted by genotype, call length peaked at an older age in Fragile X males than it did in controls. Overall, calls became significantly shorter, higher in frequency, and more complex in their structure as animals aged (ancova on age, all $p<.0001$). These changes occurred abruptly within an animal, such that consecutive recordings in the same animal could be completely different. After considering age, we found significant effects of genotype on call duration, call frequency, and the variance of frequency within calls. Fragile X pups began with a higher call frequency but that fell to below wildtype rats with age (ancova, genotype*age $f(1)=7.64$, $p<.01$). Fragile X rat pups also uniformly had lower variance in the shape of their calls (ancova, tonality*age $f(1)=5.21$, $p<0.05$ and duration variance*age $f(1)=4.1$, $p<0.05$). These results first provide a normative description of rat vocal development during the initial two weeks of development. Then, by describing alterations in this trajectory in Fragile X null pups, they provide validity to the rat knockout model of Fragile X for studying altered communication skills during development.

Disclosures: J.H. Bladon: None. **A. Hooker:** None. **Z. Pranske:** None. **N. Diaz-Rodriguez:** None. **S.P. Jadhav:** None.

Poster

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Program #/Poster #: P054.05

Topic: A.07. Developmental Disorders

Support: NIH grant MH120498
NIH grant MH120569
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NIH grant MH123742

Title: Fmr1 knockout rats show social interaction deficits but enhancement in observational learned cued fear conditioning

Authors: *S. NAVABPOUR[1], M. MUSAUS[2], T. J. JAROME[3];
[1]Translational Biology, Medicine, and Hlth., [2]Sch. of Neurosci., [3]Deparment of Animal and Poultry science, Virginia Tech., Blacksburg, VA

Abstract: Autism spectrum disorder (ASD) is one of the most complex and severe neurodevelopmental psychiatric disorders, characterized by impaired social interaction and communication with multiple etiologies. Fragile X syndrome (FXS), the most common monogenetic, inherited disorder, is a leading genetic cause of ASD. FXS results from silencing the Fragile X Mental Retardation gene (*Fmr1*) and the loss of its protein product FMRP, an inhibitor of protein synthesis. Since the initial description of *Fmr1* knockout rats, there have been few studies on the behavioral characterization of this model. However, it is essential to define its phenotypes before using them in translational studies. Here, using the elevated plus maze (EPM), an open field task (OFT), and observational cued fear conditioning, we investigated anxiety responses and social interaction behavior and learning and memory in male *Fmr1* knockout (KO) Sprague Dawley rats. Consistent with the literature but unlike FXS patients, *Fmr1* KO rats did not show elevated anxiety levels. During the OFT, the rats (8-9 weeks, n=8/group) could freely choose to interact with a novel object (NO) or a novel rat (NR) matched by their sex and age. We found that the *Fmr1* KO rats interacted with the NR less frequently than wild-type (WT) rats. Also, the ratio of the time spent with the NR to the time spent with the NO was less for *Fmr1* KO rats than that of the WTs, suggesting a social deficit similar to the social phobia FXS patients present. Next, we placed the *Fmr1* KO or WT rats (observer) in an auditory fear conditioning chamber with a mesh divider separating it from a NR on the other side (demonstrator). There was a black plexiglass panel covering the grid floor on the observer rats' side so they could neither see nor feel the shocks during the training. However, they could see, hear, and smell the demonstrator rat through the mesh divider during the conditioning session in which the demonstrator was presented with several pairings of an auditory cue and foot shock. During this session, the observer rats could also hear the auditory cue that was presented to the demonstrator rat. During testing to the auditory cue in a novel context, we found that while both *Fmr1* KO and

WT observer rats acquired fear to the auditory cue, which occurred without ever receiving any foot shock presentations, *Fmr1* KO rats freeze significantly more than the WT rats. Taken together, these data suggest that *Fmr1* KO rats are impaired in social behavior but have improvements in observationally learned cued fear memory, which is not likely due to differences in anxiety. These results have important implications for translational studies on the FXS rat model.

Disclosures: S. Navabpour: None. **M. Musaus:** None. **T.J. Jarome:** None.

Poster

P054. Fragile X: Behavior

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Topic: A.07. Developmental Disorders

Support: This work was funded by Ulysses Neuroscience Ltd. and Angelini Pharma S.p.A.

Title: Chronic treatment with the selective GSK-3 inhibitor SB216763 reverses behavioral impairments in the Fmr1 knock out mouse model of Fragile X Syndrome

Authors: *J. KEALY[1], A. M. THORNTON[2], A. FREEBURN[1], C. GREENE[1], B. GARRONE[3], C. MILANESE[3], F. P. DI GIORGIO[3], C. K. CALLAGHAN[1], M. BIANCHI[1];
[1]Ulysses Neurosci. Ltd., Dublin, Ireland; [2]Trinity Col. Dublin, Dublin, Ireland; [3]Angelini Pharma S.p.A., Rome, Italy

Abstract: Fragile X Syndrome is a complex neurodevelopmental disorder resulting from an inactivation of the *FMR1* gene on the X chromosome, leading to a loss of FMRP protein expression. Individuals with Fragile X Syndrome can be mildly or severely affected, with symptoms including intellectual disability, developmental delay, autism-like symptoms, and anxiety, amongst others. In mice, knocking out the *Fmr1* gene disrupts a wide range of signaling pathways, including the glycogen synthase kinase-3 (GSK-3) pathway, and results in a behavioral phenotype analogous to the human condition including cognitive impairment, disruption of social behavior, and an increase in stereotypic behaviors. For this study, male *Fmr1* knock out (*Fmr1*$^{-/y}$) mice and c57BL6/J wild type (WT) controls underwent behavioral testing following chronic treatment with SB216763 (30 mg/kg, one intraperitoneal dose per day over 10 days), an inhibitor of both GSK-3α and GSK-3β. Marble burying was used as a measure of stereotypy/repetitive behavior, and sociability was measured using the 3-chamber sociability task. Locomotor activity was assessed across the three trials of the 3-chamber sociability task. SB216763 significantly reduced marble burying in *Fmr1*$^{-/y}$ mice. SB216763 also reversed the social discrimination deficit observed in *Fmr1*$^{-/y}$ mice. There was no social recognition deficit in vehicle-treated *Fmr1*$^{-/y}$ mice and GSK-3 inhibition had no effect in *Fmr1*$^{-/y}$ mice. In 3-chamber

sociability trials where hyperlocomotion was observed, SB216763 had no effect on attenuating this behavior. These data show that GSK-3 inhibition is a robust strategy for ameliorating some of the behavioral deficits seen in *Fmr1*$^{-/y}$ mice, highlighting a signaling mechanism for novel therapeutics for treating some symptoms of Fragile X Syndrome. All animal experiments were approved by the Trinity College Dublin Animal Research Ethics Committee and the Health Products Regulatory Authority under the European Union directive 2010/63/EU.
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Poster

P054. Fragile X: Behavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P054.07

Topic: A.07. Developmental Disorders

Support: NIH: NS112706

Title: Mitochondrial inefficiency: from abnormal metabolism to behavioral deficits in Fragile X

Authors: *P. LICZNERSKI[1], S. NAIR[2], E. A. JONAS[1];
[1]Yale Univ. Sch. of Med., New Haven, CT; [2]Yale Univ., New Haven, CT

Abstract: Loss of function of the gene (Fmr1) encoding Fragile X mental retardation protein (FMRP) results in unregulated, elevated mRNA translation and aberrant synaptic morphology. We have recently discovered a completely novel function of a “leak channel” termed ATP synthase c-subunit leak channel (ACLC), formed by the C-subunit octamer within the ATP-synthase in the mitochondrial inner membrane. We showed in Fragile X syndrome that pharmacological inhibition of ACLC by the ATP synthase modulator Dexpramipexole (Dex) normalizes the elevated protein translation rates, induces synaptic maturation, and attenuates autistic behaviors, which raises the spectrum of new possibilities for therapeutic intervention in FX. We have also found that Fragile X neurons have a mitochondrial leak-dependent metabolic phenotype that is normalized by pharmacological inhibition of ACLC. We here further

investigate the effects of ACLC activity on mRNA translation. We find preliminarily that Dex alters the proximity of specific ATP synthase mRNAs to mitochondria, which could contribute to changes in ATP synthase stoichiometry by inducing assembly of ATP synthase and eliminating free c-subunit in the inner mitochondrial membrane. This stoichiometric alteration provides for a long-term change in the efficiency of ATP synthesis that may be required for normalization of behavioral abnormalities in Fragile X mice.

Disclosures: P. Licznerski: None. **S. Nair:** None. **E.A. Jonas:** None.

Poster

P054. Fragile X: Behavior

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Program #/Poster #: P054.08

Topic: A.07. Developmental Disorders

Support: NIH Grant U54HD082008

Title: Bottom-up temporal cortical generators drive sensory-related neural oscillatory deficits in Fragile X Syndrome

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Abstract: Neural and sensory hyperexcitability are prominent in many individuals with Fragile X Syndrome (FXS). Previous work from our group and others has established translational electrophysiological biomarkers for sensory processing abnormalities in FXS, primarily at the EEG scalp/sensor level. While EEG provides robust equivalency across FXS patients and Fmr1 KO mice, the neural sources of these abnormalities remain poorly characterized. Transformation of scalp electrical data into source space allows for the separation of temporal and frontal cortical sources of neural activity, and thus can better characterize the pathways in which functional alterations exist. Participants (36 adolescents and adults with FXS and 39 age- and sex-matched typically developing controls (TDC) listened passively to an auditory chirp stimulus during continuous recording of dense-array EEG. We constructed a depth-weighted minimum norm estimate. Current source density was computed across a resultant triangular mesh (15,002 vertices) which represented the cortical envelope. Vertices were parcellated into 68 cortical nodes including sources of interest in left and right temporal and frontal cortices and a control node in cuneus. Morlet wavelets were calculated on source waveforms to reconstruct phase and

amplitude characteristics of neural responses to auditory chirps. In the temporal source, individuals with FXS showed increased gamma power, t(73)=-2.56, p=.012, theta power t(73)=-3.11, p=.003, and theta/alpha inter-trial phase coherence (ITPC) to stimulus onset t(73)=-2.37, p=.02, and decreased gamma phase synchrony to the auditory stimulus at 40 Hz, t(73)=2.18,p=.032, compared to TDC. For frontal and cuneus sources the only difference was during stimulus onset. Increased gamma power in temporal cortex was significantly correlated with increased autism-like behaviors and negative cognitive/clinical scores, whereas increased gamma phase synchrony was associated with positive clinical/cognitive scores. Disentangling early sensory (temporal) sources of group differences from top-down (frontal) sources is an important due to the orientation of Heschl's gyrus, which confounds temporal and frontal signals at the scalp. Our findings confirm expectations of a primarily early sensory deficit in temporal cortex, while suggesting that multiple sources may contribute variance in scalp signal power. Early sensory processing in auditory sensory cortex is thus the likely primary driver of FXS-related auditory processing deficits in neocortex in the chirp task, rather than top-down modulation via frontal cortices.

Disclosures: L.E. Ethridge: None. **E.V. Pedapati:** None. **L.A. De Stefano:** None. **R. Liu:** None. **L.M. Schmitt:** None. **J.A. Sweeney:** None. **C.A. Erickson:** None.

Poster

P054. Fragile X: Behavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P054.09

Topic: A.07. Developmental Disorders

Support: UT Rising STARs
Voelcker Biomedical Research Academy
Northside Health Careers High School

Title: Contagious behavior deficits in the mouse model of fragile X syndrome

Authors: *R. GONZALES-ROJAS, A.-N. RANA, P. MASON, C. RENFRO, V. ANNALURU, H. LEE;
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Abstract: Imitative deficits in patients with autism spectrum disorders (ASDs) foundationally impact their abilities to learn and socially interact. However, the exploration of these deficits in preclinical autism mouse models is limited. Fragile X syndrome (FXS) is a common form of inherited intellectual disabilities with a high risk of co-morbid ASDs, and the mouse model of FXS—*Fmr1* KO mice—recapitulates many autistic behaviors. Wild-type (WT) mice exhibit a rudimentary form of imitation through socially contagious itch behavior. When presented with a

video of a scratching demonstrator mouse, WT mice will look towards the screen and copy the observed behavior, as opposed to WT mice watching an ambulating demonstrator mouse as a control stimulus. We recently demonstrated that adolescent (P45-P59) and adult (P60-P130) male *Fmr1* KO mice fail to imitate scratching behavior, relative to age-matched WT controls (Gonzales-Rojas & Rana *et al*. 2020, *Scientific Reports*). Furthermore, WT and *Fmr1* KO mice showed no differences in the total number of looks towards scratching or ambulating demonstrator stimuli. This look behavior phenotype is notable because patients with ASDs show well-documented eye contact deficits, and FXS patients also show impaired vision. However, our findings illustrate that contagious itch deficits in *Fmr1* KO mice are independent of visual impairments. We are currently exploring if our autism mouse model shows similar deficits in other forms of contagious behavior. Collectively, we hope our novel behavioral assessments will enable the preclinical exploration of targeted therapeutic treatments for learning impairments in ASDs.

Disclosures: R. Gonzales-Rojas: None. **A. Rana:** None. **P. Mason:** None. **C. Renfro:** None. **V. Annaluru:** None. **H. Lee:** None.

Poster

P111. LTP: Modulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P111.01

Topic: B.07. Synaptic Plasticity

Support: NIH Grant R01-NS113600
NIH Grant R01-MH125772
2020 Junior Investigator Neuroscience Research Award
Fondation pour la Recherche Médicale
Fondation Bettencourt Schueller

Title: BDNF-induced BDNF release mediates presynaptic LTP in the hippocampus

Authors: *C. BERTHOUX[1], K. NASRALLAH[1], P. E. CASTILLO[1,2];
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Abstract: The nervous system has evolved to adjust behavior in response to new experiences, ultimately ensuring animal survival. This crucial process mainly relies on activity-induced modifications of synaptic contacts, such as long-term potentiation (LTP) and depression (LTD). The mechanisms underlying these forms of plasticity are remarkably diverse and not fully understood. One key mediator of synaptic plasticity is the brain-derived neurotrophic factor (BDNF) and its receptor, tyrosine kinase B (TrkB). Despite many years of research, how exactly

BDNF/TrkB signaling mediates plasticity remains unclear. In the dentate gyrus, the main input region of the hippocampus proper, we have recently identified a presynaptic form of LTP at the mossy cell (MC) to dentate granule cell (GC) synapse. While MC-GC LTP requires postsynaptic BDNF/TrkB signaling, the precise source of BDNF and the downstream signaling pathway supporting synaptic plasticity is unknown. Here, we report that selective deletion of *Bdnf* in either the presynaptic (MC) or the postsynaptic (GC) neuron impaired MC-GC LTP in acute mouse hippocampal slices, indicating that both pre- and postsynaptic BDNF are required for this form of plasticity. Using two-photon live imaging of a BDNF sensor (BDNF-pHluorin) in combination with selective pharmacology and conditional knockout, we found that BDNF released from the presynaptic compartment can elicit postsynaptic BDNF release in a TrkB- and calcium-dependent manner. In addition, activation of type-1 cannabinoid (CB_1) receptors, which are highly expressed in MC axon terminals, negatively regulated presynaptic BDNF release and LTP, likely through inhibition of voltage-gated calcium channels. Finally, we found that BDNF is required for the induction of MC-GC LTP following exposure to an enriched environment. Altogether, these results strongly suggest that BDNF-induced BDNF release mediates presynaptic LTP and may contribute to dentate gyrus-dependent learning, as well as neuropsychiatric disorders such as epilepsy and anxiety.

Disclosures: C. Berthoux: None. **K. Nasrallah:** None. **P.E. Castillo:** None.

Poster

P111. LTP: Modulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P111.02

Topic: B.07. Synaptic Plasticity

Support: NIH R01-NS092716
NIH T32GM136503

Title: Heterosynaptic endocannabinoid-mediated long-term potentiation of non-nociceptive synapses

Authors: *A. D. FRANZEN, R. T. PAULSEN, B. D. BURRELL;
Univ. of South Dakota, Vermillion, SD

Abstract: Chronic pain is pain lasting longer than three months and while prevalent, its mechanism is poorly understood. One element of this condition is enhanced non-nociceptive afferent input onto nociceptive circuitry. Our laboratory studies modulation of nociceptive vs. non-nociceptive synapses via dual intracellular recordings in the CNS of the medicinal leech (*Hirudo verbana*). Previous results from our lab show that high-frequency stimulation (HFS) of a nociceptive neuron results in heterosynaptic potentiation of a non-nociceptive synapse. In this

study we examine how endocannabinoids, which are normally associated with synaptic depression, mediate LTP. HFS of the nociceptor elicits a depression of tonic inhibition onto the non-nociceptive afferent and this depression is 2-AG and TRPV-dependent. This resulting disinhibition leads to potentiation of the non-nociceptive synapses that requires NMDA receptor activation. We have also observed that this potentiation is PKMζ-dependent. As PKMζ is a neuron-specific aPKC isoform believed to be a memory engram, further studies are underway to determine PKMζ's role in eCB-mediated plasticity. These findings are relevant to understanding how the endocannabinoid system modulates pain and the utility of cannabinoid-based treatments to treat persistent pain.

Disclosures: A.D. Franzen: None. **R.T. Paulsen:** None. **B.D. Burrell:** None.

Poster

P111. LTP: Modulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P111.03

Topic: B.07. Synaptic Plasticity

Support: NIH Grant NS111986

Title: Endocannabinoid-mediated enhancement of hippocampal long-term potentiation

Authors: *F. LEMTIRI-CHLIEH, E. S. LEVINE;
Univ. of Connecticut Sch. of Med., Farmington, CT

Abstract: It is widely accepted that exogenous cannabinoids can impair short-term memory and cognition in humans and other animals. This is likely related to the inhibition of long-term potentiation (LTP), a form of synaptic plasticity, by the global and sustained activation of CB1 cannabinoid receptors in the presence of exogenous agonists. Conversely, the temporally and spatially-restricted release of endogenous cannabinoid ligands may enhance synaptic plasticity in a synapse-specific manner. The functional roles of endocannabinoids (eCBs) are complex because they modulate synaptic transmission via suppression of GABA and glutamate release, with opposing effects on postsynaptic excitability. We examined the role of eCB signaling in LTP by recording field excitatory postsynaptic potentials (fEPSPs) in the CA1 stratum radiatum in hippocampal slices from juvenile mice. Significant LTP (~50% increase from baseline) was induced by either 1 or 3 trains of TBS. The magnitude of this potentiation was significantly reduced by preventing cannabinoid receptor activation with the selective CB1 receptor antagonist NESS-0327 (~25% increase from baseline). LTP was inhibited to a similar extent by preventing the synthesis of the endogenous ligand 2-AG using the DAG lipase inhibitor DO34. A comparable effect of CB1 blockade was also observed with pharmacologically-induced LTP. These results suggest that activation of CB1 receptors by endogenous 2-AG normally enhances

the magnitude of LTP, leading to the prediction that the predominant eCB effect under these conditions is to suppress inhibition. We therefore examined TBS-induced LTP while blocking inhibitory synapses with the GABA-A receptor blocker picrotoxin (PTX). The addition of PTX alone caused a slight, but not significant, increase in LTP. Consistent with our hypothesis, PTX completely prevented the effect of blocking CB1 receptors, indicating that intact GABAergic transmission is required for the eCB effect. The presence of PTX did not unmask any enhancing effect of CB1 receptor blockade, suggesting minimal eCB-mediated suppression of glutamate release under these conditions. These results indicate that 2-AG activation of CB1 receptors enhances TBS-induced and pharmacologically-induced LTP, and this effect is caused by the actions of 2-AG at inhibitory synapses.

Disclosures: F. Lemtiri-Chlieh: None. **E.S. Levine:** None.

Poster

P111. LTP: Modulation

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Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P111.04

Topic: B.07. Synaptic Plasticity

Support: Wellcome Trust/DBT India Alliance Grant IA/I/12/1/500529
DST/INSPIRE Fellowship2016/IF60611

Title: Cholinergic signals optimize information transmission and learning in the hippocampus

Authors: *R. SHARMA, S. NADKARNI;
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Abstract: Neuromodulators regulate and fine-tune nonlinear membrane and synaptic properties. This provides neural circuits with flexibility in function and across behavioral states. We focus, in particular, on acetylcholine which is released during active waking in the cortex and hippocampus. In the hippocampus, acetylcholine modulates neuronal activity predominantly via its action on M1 and M4 metabotropic receptors. Their distinct localizations and biophysical properties produce rich dynamics across multiple timescales. We have developed a detailed biophysical model for the presynaptic activation of M4 and postsynaptic activation of M1 receptors in the hippocampus. Our model describes activation of M1 receptors leading to IP3 production from membrane PIP2 molecules. The binding of IP3 to IP3 receptors ultimately causes calcium release from the endoplasmic reticulum (ER). The additional calcium release from the ER enhances the action of potassium channels like the calcium-activated SK channels and modifies other downstream synaptic signaling. Interestingly, an opposing effect of M1 receptors is to suppress SK channels and the voltage-activated KCNQ2/3 channels directly. We quantify the effect of enhanced calcium signaling from the ER and antagonistic effects on SK

channels via M1 receptor activation on long-term plasticity at Schaffer-collateral synapses in the hippocampus. Our model also describes the presynaptic reduction of the voltage sensitivity of voltage-gated calcium channels (VGCCs) and the resulting suppression of the neurotransmitter release via M4 receptors. We characterize the influence of the apparent dichotomy of suppression of synaptic transmission by M4 and enhanced neuronal excitability by M1 on synaptic signaling and memory formation in a range of physiologically relevant activity regimes.

Disclosures: R. Sharma: None. **S. Nadkarni:** None.

Poster

P111. LTP: Modulation

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Program #/Poster #: P111.05

Topic: B.07. Synaptic Plasticity

Support: NIA Grant AG028271
NIA Grant AG067061

Title: Short-term high-fat diet consumption impairs long-term potentiation in the aged hippocampus

Authors: *B. M. GONZALEZ OLMO, F. ZHAO, C. ASKWITH, R. M. BARRIENTOS; The Ohio State Univ., Columbus, OH

Abstract: More Americans are consuming diets higher in saturated fats and refined sugars than ever before, and obesity rates among older adults alone have doubled since 1980. These trends could have serious consequences for the older population because in addition to the gradual memory declines observed with normal aging, high-fat diet (HFD) consumption, known to induce neuroinflammation, has been shown to aggravate and accelerate memory declines. Because obesity is a complex disease with many comorbidities, making the study of underlying mechanisms difficult and confounded, we employ a short-term diet manipulation protocol. We have previously demonstrated that short-term consumption (3 days) of a HFD among aged rats produced profound impairments to contextual and emotional/fear memories, which depend on an intact hippocampus and amygdala. These impairments were precipitated by increases in proinflammatory cytokines, primarily interleukin-1 beta (IL-1b), in both brain regions. Here, we explored the extent to which HFD consumption amongst aged rats disrupts hippocampal long-term potentiation (LTP), the form of synaptic plasticity thought to underlie long-term memory consolidation in mammals. Young adult (3 months old) and aged (24 months old) F344xBNF1 rats were assigned to either chow or HFD (containing 60% saturated fat) for three days. Using transverse hippocampal slices, we examined the individual and combined effects of age and diet on several forms of synaptic activity. Specifically, excitatory post-synaptic potentials were

induced in the stratum radiatum of CA1 and LTP expression was triggered with a theta-burst stimulation protocol. Our preliminary data demonstrate that late-phase LTP was particularly compromised by the combination of aging and HFD while LTP maintenance was robust in chow-fed young and aged rats. These findings suggest that the previously observed neuroinflammation-mediated hippocampal memory impairments in aged HFD-fed rats occurs, at least in part, through deterioration of synaptic plasticity, as measured by LTP, in the hippocampus. Future studies will examine LTP in the amygdala.

Disclosures: B.M. Gonzalez Olmo: None. **F. Zhao:** None. **C. Askwith:** None. **R.M. Barrientos:** None.

Poster

P111. LTP: Modulation

Location: Virtual Only

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Program #/Poster #: P111.06

Topic: B.07. Synaptic Plasticity

Support: NIH Grant R01 NS10742101
NIH Grant P01 HL144454

Title: Postnatal age influences how Intermittent Hypoxia impacts hippocampal synaptic plasticity and NMDA receptor subunit expression.

Authors: *A. ARIAS-CAVIERES, A. J. GARCIA, III;
Univ. of Chicago, Chicago, IL

Abstract: The ability to scale synaptic strength supports the robustness and flexibility of learning and memory throughout a lifetime. Intermittent Hypoxia (IH) is a hallmark of several clinical conditions such as sleep apnea and apneas of prematurity. These conditions are associated with neurocognitive deficits. In adult mice, IH-dependent oxidative stress downregulates hippocampal GluN1, the obligatory subunit of the NMDA receptor, and eliminates NMDA receptor-dependent synaptic plasticity. However, how IH impacts NMDA receptor subunit expression and synaptic plasticity during early postnatal life is not well-understood. This ongoing study characterizes how IH impacts NMDA receptor-dependent synaptic plasticity during early neonatal life. Electrophysiological studies and protein analyses were performed using hippocampal brain slices taken from mice (P14-P15) exposed to ten days of neonatal IH (nIH, starting at P4-P5). In contrast to the effects of IH on the adult hippocampus, nIH neither suppressed GluN1 expression nor eliminated NMDAr-dependent synaptic plasticity. Additionally, nIH caused a downregulation of GluN2A while upregulating GluN2B. These findings suggest that postnatal age is a significant factor influencing the consequences of IH in the hippocampus.

Disclosures: A. Arias-Cavieres: None. **A.J. Garcia:** None.

Poster

P111. LTP: Modulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P111.07

Topic: B.07. Synaptic Plasticity

Support: European Union Horizon 2020 Research and Innovation Programme under Grant Agreement number GrapheneCore3 (881603).

Title: Graphene oxide nanoflakes prevent potentiation at amygdala synapses: a novel tool to block dysfunctional plasticity in anxiety disorders

Authors: E. PATI[1], G. CELLOT[1], A. FRANCESCHI BIAGIONI[1], K. KOSTARELOS[2,3], L. BALLERINI[1];
[1]Intl. Sch. of Advanced Studies, Trieste, Italy; [2]Catalan Inst. of Nanoscience and Nanotechnology (ICN2), CSIC and BIST, Campus UAB, Bellaterra, 08193, Barcelona, Spain; [3]Nanomedicine Lab, Natl. Graphene Inst. and Fac. of Biology, Med. & Health, The Univ. of Manchester, AV Hill Building, Oxford Rd, Manchester, M13 9PL, United Kingdom

Abstract: In the last decade, graphene-based nanomaterials have been proposed as innovative therapeutic tools in the field of precision medicine for neuroscience applications. In detail, graphene oxide nanoflakes with small lateral dimension (sGO) were found to target specifically and transiently glutamatergic synapses of the hippocampus *in vivo*, by reducing the release of neurotransmitter from the presynaptic terminal[1].Recently, this ability was used to rescue the aberrant glutamatergic transmission characterizing post-traumatic stress disorders (PTSD). Since PTSD is related to a hyper-function of the amygdala resulting from the long term potentiation (LTP) of glutamatergic synapses, our group reported that, in a rat model of PTSD, a single intra-amygdala injection of sGO administered after the stressful event triggering the disease and the related altered plasticity of the amygdala, could revert the anxiety related behaviours[2]. In order to elucidate how sGO could interfere at synaptic level with the pathological LTP underlying PTSD, we developed an *in vitro* model of amygdala potentiated glutamatergic synapses. To this aim, in rat dissociated amygdala cultures we induced chemical LTP through the brief application of glutamate (50 µM for 30 seconds), that was monitored through patch clamping as a 30-minute-long lasting increase in the amplitude of postsynaptic currents (PSCs). The co-application of sGO (20 µgr/mL) with glutamate abolished the expression of the LTP. By combining the recording of miniature postsynaptic currents (mPSC), informative for modifications in synaptic structure, with confocal analysis of pre- (VGlut1) and postsynaptic (PSD95) markers, we detected that LTP relied on functional and structural changes of the synaptic terminals, that were both impaired by the treatment with sGO. We recorded simultaneously mono-synaptically

connected neuron pairs and we observed that sGO decreased the probability of glutamate release from the presynaptic site, measured as a pair pulse ratio (PPR) increase between two consecutive evoked unitary PSCs. Such changes were concurrent with a lack of LTP induction.In our work, we explored from a mechanistic perspective how sGO, by targeting precisely the release of glutamate from presynaptic site of excitatory synapses, could prevent a form of pathological plasticity associated with PTSD. These findings might find application for the treatment for a broad spectrum of neuro-pathologies.
[1]Rauti et al. 2019, *https://doi.org/10.1021/acs.nanolett.8b04903*[2]Franceschi Biagioni et al. 2021, *https://doi.org/10.1016/j.biomaterials.2021.120749*

Disclosures: E. Pati: None. **G. Cellot:** None. **A. Franceschi Biagioni:** None. **K. Kostarelos:** None. **L. Ballerini:** None.

Poster

P111. LTP: Modulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P111.08

Topic: B.07. Synaptic Plasticity

Support: National Research Foundation of Ukraine Grant 2020.01/0266

Title: Impact of virus sialidase blockage on synaptic plasticity in rat brain

Authors: A. SAVOTCHENKO, T. DREVYTSKA, Y. TKACHENKO, R. MORHACHOV, M. STEFANENKO, S. DEMCHENKO, D. ISAEV;
Bogomoletz Inst. of Physiol., Kyiv, Ukraine

Abstract: Impact of virus sialidase blockage on synaptic plasticity in rat brain
Sialidase controls the level of sialylation and can alter cell–cell or cell–virus recognition processes. Sialidase neuraminidase (NEU) play a pivotal role during last stages of virus development. The lack of NEU in viruses makes them inactive viral particles which unable to infect host cells. NEU is a key enzyme that cleaves negatively charged sialic acid residues from membrane proteins and lipids. Polysialic acids (PSA) are widely distributed in neuronal tissue. Due to their position on the outer cell membranes and anionic nature, PSA are involved in a multiple cell signaling events. Using the specific blocker of NEU – N-Acetyl-2,3-dehydro-2-deoxyneuraminic acid (NADNA) we show that downregulation of NEU activity causes a significant increase in the level of hippocampal tissue sialylation. In the present work we investigated effect of blockage of endogenous NEU on synaptic plasticity in different regions of hippocampus and on EPSP-pop-spike (E-S) potentiation.Temporal lobe slices were prepared from Wistar rats aged postnatal day 19-21. Evoked synaptic responses were elicited through a bipolar electrode placed in the Schaffer collateral-commissural pathway. Extracellular recordings

were obtained from pyramidal cell layer and stratum radiatum CA1 regions. We examined the effect of bath application of NADNA on synaptic plasticity in cellular and dendritic layers of hippocampus. Analyzing mean and peak values of fPSP in control and NADNA-treated slices, we observed significant increase of both values in stratum pyramidale and radiatum. Data obtained shows that blockade of NEU leads to increase of different types of short-term plasticity (STP) in pyramidal cell layer without changes in STP of radiatum layer. We observed decrease of level and LTP-probability in NADNA-treated slices in stratum radiatum. Our findings indicate that origination of LTP lead to E-S potentiation in control slices, while in NADNA-treated slices its occurrence induced E-S depression. Also we show the increase of short-term depression level due to polysialilation. Our findings demonstrate that neuraminidase blockade primes long-term depression and suppresses long-term potentiation of synaptic transmission in the rat hippocampus. We propose that in NADNA-treated slices originate enhancement of transmitter release probability following synaptic activation.

Disclosures: A. Savotchenko: None. **T. Drevytska:** None. **Y. Tkachenko:** None. **R. Morhachov:** None. **M. Stefanenko:** None. **S. Demchenko:** None. **D. Isaev:** None.

Poster

P111. LTP: Modulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P111.09

Topic: B.07. Synaptic Plasticity

Support: NIH Grant R15NS078645

Title: Effects of prophylactic treatment on hippocampal and amygdalar synaptic plasticity and gene expression in a rodent model of PTSD

Authors: *A. C. EVERETT, R. MILLER, E. SAITO, E. WINZENRIED, C. SMITH, Z. BOYCE, J. G. EDWARDS;
Brigham Young Univ., Provo, UT

Abstract: Post-traumatic stress disorder (PTSD) is a complex anxiety and depression disorder induced by exposure to traumatic stress and is characterized by symptoms of re-experiencing (flashbacks, nightmares, etc.), avoidance, and hyper-arousal. While it is widely accepted that brain regions of the limbic system - the amygdala, ventral hippocampus, and prefrontal cortex - are dysregulated in PTSD, the pathophysiology of the disorder is not well defined and therefore, pharmacological interventions are severely limited. Because stress hormones norepinephrine and cortisol (corticosterone in rats) are heavily implicated in the disorder, we explored whether antagonizing beta-adrenergic and glucocorticoid receptors via systemic propranolol and mifepristone administration is sufficient to mitigate pathological changes in LTP, gene

expression, and behavior induced by a novel social defeat (SD) protocol. Young adult, male Sprague Dawley rats were exposed to continuous light and social isolation with acute SD to induce PTSD-like symptoms. Drug-treated rats were given propranolol and mifepristone injections beginning prior to and continuing throughout SD stress. Using competitive ELISAs on plasma, field electrophysiology at CA1 of the ventral hippocampus (VH) and the basolateral amygdala (BLA), quantitative RT-PCR, and behavioral assays, we demonstrate that SD stress increased plasma corticosterone and behavioral anxiety, elevated long-term potentiation in the VH (~25% greater than control) and BLA (~35% greater than control), and altered the expression of mineralocorticoid, glucocorticoid, beta-3 adrenergic receptors, AMPA, and NMDA receptors. These measures largely reverted to control levels with the administration of propranolol and mifepristone, with the exception of plasma corticosterone, which remained elevated with drug administration likely due to corticosterone negative feedback inhibition by mifepristone or stress-inducing injections. Together these data validate our novel SD stress protocol as means of inducing PTSD-like symptoms, and suggest therapeutic potential for propranolol and mifepristone as a prophylactic treatment for preventing PTSD before stress onset.

Disclosures: A.C. Everett: None. **R. Miller:** None. **E. Saito:** None. **E. Winzenried:** None. **C. Smith:** None. **Z. Boyce:** None. **J.G. Edwards:** A. Employment/Salary (full or part-time):; Brigham Young University.

Poster

P123. Transcription in Plasticity

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P123.01

Topic: B.07. Synaptic Plasticity

Support: NSERC Discovery Grant

Title: Expression of the immediate early gene, Arc, identifies a neural network supporting classical conditioning to time-of-day.

Authors: *R. R. MCHUGH[1], R. J. SUTHERLAND[2];
[1]Neurosci., Canadian Ctr. For Behavioral Neurosci. (unive, Lethbridge, AB, Canada; [2]Univ. Lethbridge, Lethbridge Alberta, AB, Canada

Abstract: The activity of the hippocampus is necessary for memory acquisition, storage, and retrieval. Much of the research focus has been on its role in spatial navigation and contextual memory. However, the encoding of temporal cues is not well understood in long-term memory. Here, we explore the expression of the *Arc* immediate early gene (IEG) after condition to a time-of-day cue in a novel conditioning procedure in rats. For 14 days, we video-recorded locomotor

activity and delivered a clocktime-specific, multimodal alarm signal to 12 cage-paired rats. The quantification of IEG expression in neurons is a useful method for mapping temporally-specific neuronal activity in the brain. Specifically, the expression of *Arc* mRNA participates in synaptic changes associated with long-term plasticity. We predicted the neural activity associated with a temporally-conditioned response would include expression of *Arc* genes in the hippocampus. The role of components of the observed network is under experimental examination.

Disclosures: R.R. McHugh: None. **R.J. Sutherland:** None.

Poster

P123. Transcription in Plasticity

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P123.02

Topic: B.07. Synaptic Plasticity

Support: NIH Grant NS083085

Title: Activity-dependent localization of Camk2a mRNAs in dendritic spines of hippocampal neurons

Authors: *D.-W. HWANG[1], S. DAS[1], R. H. SINGER[2];
[2]Anat. & Structural Biol., [1]Albert Einstein Col. of Med., Bronx, NY

Abstract: It has been demonstrated that calcium-mediated activation of Calcium/calmodulin-dependent protein kinase type II (CaMKII) is sufficient to induce spine-specific structural long-term plasticity (sLTP). The synaptic tagging and capture hypothesis postulates that the induction of CaMKII kinase activity serves as a molecular tag, which further potentiates downstream molecular events such as the recruitment of protein components that are needed for long-lasting synaptic changes. The timing with respect to the onset of stimulation and spine-specific subcellular localization of this activation of CaMKII is extensively studied, whereas the preceding molecular events have not been clearly delineated. In particular, it has yet to be known how inactive CaMKII becomes available for calcium-mediated activation in stimulated spines. To answer this question, we combined single-molecule fluorescent imaging approaches with a novel bacteriophage stem loop-based genetic tagging system (i.e. MS2/PP7 hybrid) to assess the kinetics and subcellular localization of endogenous *Camk2a* mRNAs in response to neuronal stimulation. Our preliminary data indicate that upon global and local stimulation, *Camk2a* mRNAs are recruited to the spines on a time scale of seconds and minutes. Importantly, the spine-localization of *Camk2a* mRNAs coincides with the activation of spines, suggesting that the localization is functionally connected to calcium-mediated activation of CaMKII in dendritic spines. We are currently investigating the spatiotemporal aspects of this

activity-dependent *Camk2a* localization in dendritic spines and subsequent molecular events following the localization as they relate to the establishment of long-lasting synaptic changes.

Disclosures: D. Hwang: None. **S. Das:** None. **R.H. Singer:** None.

Poster

P123. Transcription in Plasticity

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P123.03

Topic: B.07. Synaptic Plasticity

Support: R01MH105592

Title: Bdnf-dt: a novel divergent rna in the bdnf locus

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Abstract: Divergent transcription from bidirectional promoters is frequently seen among transcriptionally active protein-coding genes, where divergent RNAs (divRNAs) fine-tune the expression of mRNAs. Here we characterize a novel divergent transcript in the brain-derived neurotrophic factor (*BDNF-DT*) locus that is conserved across both mouse and human genomes. The *BDNF* gene structure is complex, containing nine short non-coding exons that are alternatively spliced to one common-coding exon to generate multiple unique transcripts, all of which encode the same mature BDNF protein. In addition to *BDNF* mRNA transcripts, a *BDNF* antisense (*BDNF-AS*) long non-coding RNA has been described. We used RNA isolated from human postmortem dorsolateral prefrontal cortex (DLPFC) to conduct RACE-PCR with primers specific to novel antisense exons and detect the 5' start and the 3' end of multiple novel *BDNF-DTs*. We found two alternative 3' ends and three alternative 5' starts for a total of five novel divergent transcripts (*BDNF-DT1, 2, 3* and *BDNFAS-DTa, b*), three of which are evolutionary conserved and two of which have protein-coding potential. Using mouse primary cortical cultures we characterized the kinetics of *Bdnf-DT* expression following 6 hours of depolarization (100 mM KCl). While expression of *Bdnf* exon *1, 2* and *4* transcripts peaked 3 hours after KCl wash off, the *Bdnf-DT* transcript showed a delayed peak expression at 6 hours upon KCl wash off. Using a human neural progenitor cell (NPC) line stably transduced with CRISPR activation constructs (VPR - ReNcell VM), we showed that selective upregulation of *BDNF* exon *1* and *4* transcripts drive the expression of *BDNF-DT* 3 and 4-fold, respectively. Further, we found that

the expression of *BDNF-DT* is significantly higher in postmortem human dorsal prefrontal cortical tissue from brain donors diagnosed with schizophrenia compared to neurotypical controls (N=378, t=3.805, p= 0.000167). Finally, we found that the schizophrenia-associated C allele (Val-BDNF) of the rs6265 SNP is highly-significantly (p<1e-10) associated with the lower expression of the *BDNFAS-DT* read-through fusion transcript, as compared to the T allele (Met-BDNF). In summary, we describe a novel divRNA in the *BDNF* locus, define the structure of its isoforms, characterize its activity-dependent expression in mouse primary cortical cultures, and drive its expression with CRISPR activation constructs in human NPCs. Our functional studies suggest that *BDNF-DT* expression may regulate *BDNF* mRNA, which could explain the observation of reduced *BDNF* levels in patients with schizophrenia. Our study sheds light and poses new questions on the complex biology of *BDNF*.

Disclosures: S.V. Bach: None. **G. Punzi:** None. **N.E. Smith:** None. **J. Shin:** None. **L. Collado-Torres:** None. **K.R. Maynard:** None. **S.C. Page:** None. **A. Straub:** None. **D.R. Weinberger:** None. **K. Martinowich:** None. **G. Ursini:** None.

Poster

P123. Transcription in Plasticity

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P123.04

Topic: B.07. Synaptic Plasticity

Support: Sumitomo Dainippon Pharma

Title: Acute stress differentially regulates BDNF expression in prefrontal excitatory and inhibitory neurons of chronically stressed rats. Modulation by lurasidone treatment.

Authors: *F. MARCHISELLA[1], I. PISANO[1], F. DE ROSA[1], K. C. CREUTZBERG[1], V. BEGNI[1], M. PAPP[2], M. A. RIVA[1];
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Abstract: One key trait in psychiatric patients is represented by disturbances in emotional control. While, emotional dysregulation is a human feature, exposure to acute challenges in rodents can be used to investigate key pathways and systems that may be relevant in emotional control. Here, we used the chronic mild stress (CMS) paradigm to identify changes in the responsiveness to an acute stress and we investigated the potential impact of a chronic treatment with the antipsychotic drug lurasidone (Lur).80 Male Wistar rats were exposed to either 7-week CMS paradigm or left undisturbed in home-cages. Lur (3.0 mg/kg, per os) or vehicle (1% w/v) were administered for 5 consecutive weeks, starting 2 wks after CMS. 1% sucrose intake was measured at weekly intervals to assess anhedonia. 30 min-restraint stress was performed before killing on half the animals per group. Analyses include ELISA, qRT-PCR and in situ

hybridization. Two-way ANOVA followed by Tukey's multiple comparison test was performed with significance set at p<0.05. CMS significantly lowered sucrose intake, which was progressively ameliorated by lurasidone (p<0.001). Following exposure to the acute restraint stress (ARS), peripheral CORT levels were significantly increased in control rats (p<0.05), an effect that was blunted by Lur treatment (p<0.01). We next investigated the mRNA levels of 4 markers of neuronal activity (*C-fos; Zif268, Arc/Arg3.1, Npas4*) and the data were combined in an overall Z-score. Exposure to ARS significantly increases PFC activity both in control and CMS-exposed animals (p<0.0001), an effect that was not affected by Lur treatment. We next analyzed *Bdnf* expression, a key trophic factor linking neuronal activity with functional stress responsiveness. *Bdnf* mRNA levels in the PFC were increased upon ARS exposure in control group but not in CMS-exposed rats, while Lur treatment was able to restore its modulation in rats subjected to CMS in comparison to chronically stressed animals treated with vehicle (p < .05). We also used RNAscope to map *Bdnf* expression. We found that the effect of the ARS in control animals is driven by parvalbumin- positive neurons (p < 0.05), whereas lurasidone increased *Bdnf* mRNA levels selectively in CAMKII- positive neurons (p < 0.01). In summary, these data indicate that lur may promote adaptive mechanisms that are crucial for the ability to improve specific psychopathological domains, such as anhedonia, associated with CMS exposure. Lur is also able to potentiate specific mechanisms under acute stress exposure to promote resilience.

Disclosures: F. marchisella: None. **I. Pisano:** None. **F. De rosa:** None. **K.C. Creutzberg:** None. **V. Begni:** None. **M. Papp:** None. **M.A. Riva:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Sumitomo Dainippon Pharma.

Poster

P123. Transcription in Plasticity

Location: Virtual Only

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Program #/Poster #: P123.05

Topic: B.07. Synaptic Plasticity

Title: Rna-seq analysis reveals plasticity changes that co-occur with therapeutic electrical brain stimulation

Authors: *K. SANDERS[1], T. H. SANDERS[2];
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Abstract: Deep brain stimulation (DBS) and vagus nerve stimulation (VNS) are known to ameliorate pathological brain signaling in conditions such as Parkinson's disease and epilepsy. However, the mechanisms of action remain largely unknown. Previously, we investigated gene

expression profiles in the cortex, subthalamic nucleus, striatum and hippocampus in stimulated and control rodents to show that chronic (4 days or more) stimulation modulates the transcriptome both locally and remotely. We used behavioral correlation and principal component subset analysis (PCSA) to identify transcriptome signatures of effective stimulation [1,2]. In the current study, we compare how stimulation of DBS and VNS impacts the transcriptome and report previously unpublished evidence indicating antiviral effects of VNS. DBS animals received stimulation at both high (100 Hz, 130 Hz) and low (30 Hz) frequencies. VNS rats were implanted with stimulation cuffs over the left vagus nerve and received a train of fifteen 100 ms pulses delivered at 30Hz every 17.5 s for 30 minutes each day over 4 days. Stimulated rats were compared to sham stimulated controls. Brains were removed, dissected, and frozen after the last session of stimulation. RNA was extracted, then quantified using RNA-seq and RT-qPCR. Differential mRNA and miRNA expression were performed using MATLAB and R. **We found that** DBS modulated a set of genes involved in plasticity, proliferation, and calcium signaling. Genes differentially transcribed in stimulated animals compared to controls included striatal nestin (Nes), cortical doublecortin (Dcx), and hippocampal S100a4. PCSA analysis identified region-specific optimal sets of features associated with effective DBS. Widespread, but different, transcriptome changes were identified in the hippocampus and cortex of VNS stimulated rats [2]. These rats also had greatly increased cortical expression of miRNAs associated with antiviral activity compared to age-matched sham stimulated rats from the same population.

1) Sanders, Katherine N., and Teresa H. Sanders. "Local and Remote Signatures of Effective Deep Brain Stimulation." *Brain Stimulation: Basic, Translational, and Clinical Research in Neuromodulation* 12.2 (2019): e8-e9.
2) Sanders, Teresa H., et al. "Cognition-enhancing vagus nerve stimulation alters the epigenetic landscape." *Journal of Neuroscience* 39.18 (2019): 3454-3469.

Disclosures: K. Sanders: None. **T.H. Sanders:** None.

Poster

P123. Transcription in Plasticity

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Program #/Poster #: P123.06

Topic: B.07. Synaptic Plasticity

Support: NIH Grant R15 NS on "Investigating How Chromatin Remodeling Affects Endocytosis and Synaptic Organization"

Title: Determining the interaction model between Kismet and Amyloid Precursor Protein-like using the Drosophila neuromuscular junction

Authors: *B. PRATES DE OLIVEIRA ALBANEZ, E. HENDRICKS, F. L. LIEBL; Southern Illinois Univ. Edwardsville, Edwardsville, IL

Abstract: Determining the interaction model between Kismet and Amyloid Precursor Protein-like using the *Drosophila* neuromuscular junctionSynaptic structure and function rely on the temporally appropriate expression of genes. Kismet (Kis) is a *Drosophila* CHD protein that is most similar to CHD7 and CHD8, which are implicated in CHARGE Syndrome and autism spectrum disorders, respectively. Kis regulates synaptic structure, the localization and clustering of postsynaptic glutamate receptors (GluRs), synaptic transmission, and endocytosis. *Kis* loss-of-function mutants phenocopy animal models of Alzheimer's Disease (AD) by displaying impaired endocytosis. *Kis* loss-of-function mutants also exhibit impaired locomotion and increased amyloid precursor protein like (*appl)* mRNA transcripts. Similar to *kis* mutants, expressing human APP and β-site APP cleaving enzyme (BACE) in *Drosophila* larval neurons decreases endocytosis. Further, overexpressing APPL in neurons decreases GluRIIC and expressing a secretion defective APPL in neurons decreases FasII. Based on these findings, Kis could interact with APPL in three ways, by regulating *appl* expression, in cooperation with AICD (a fragment generated during APP cleavage), or by acting as a cofactor to promote AICD transcriptional activity. I will examine these possibilities first by knocking down APPL in *kis* mutant neurons. These experimental results will contribute to better understand how the chromatin remodeling protein Kismet influences APPL at the synapse.Authors: Bruna Prates, Emily Hendricks, and Faith Liebl, PhD.

Disclosures: B. Prates De Oliveira Albanez: None. **E. Hendricks:** None. **F.L. Liebl:** None.

Poster

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Program #/Poster #: P123.07

Topic: B.07. Synaptic Plasticity

Support: NIH Grant R01MH105592

Title: Novel ReNcell VM neural progenitor line for CRISPR activation

Authors: *N. E. SMITH[1], S. V. BACH[2], E. A. PATTIE[2], S. C. PAGE[3], K. MARTINOWICH[3]; [1]Johns Hopkins Univ., Baltimore, MD; [2]Lieber Inst. for Brain Develop., Baltimore, MD; [3]Lieber Inst. For Brain Develop., Baltimore, MD

Abstract: The ReNcell VM neural progenitor line, generated by overexpression of the MYC family transcription factors in human cells derived from the ventral mesencephalon, has been used for developing stem cell therapies and validating therapeutic interventions. Here we

describe generation of a ReNcell VM line that constitutively expresses a CRISPR activation (CRISPRa) system to facilitate convenient upregulation of desired target genes. ReNcell VM cultures were transduced with high-titer lentivirus carrying deactivated Cas9 (dCas9) endonuclease fused to a strong transcriptional activator, VPR (VP64-p65-Rta). Successfully-transduced cells were sub-cultured and passaged to create a stably-transduced VPR - ReNcell VM line. As with the original ReNcell VM, VPR - ReNcell VM cultures can be differentiated into neurons by the removal of growth factors (EGF and FGF) from the culture media and maintenance for 15 days in vitro (DIV). The expression of neuronal markers after differentiation was confirmed by qRT-PCR (NES, S100B, SLC6A1, RELN, GRIA1, GRIA2, GRIN1) and immunocytochemistry (TUJ1, MAP2, NEUN) analyses. Differentiated VPR - ReNcell VM cultures are responsive to depolarization with 100 mM KCl as assessed by robust induction of immediate early genes, such as ARC and FOS. Transduction with a single guide RNA targeting the promoter regions of BDNF exon 1 or exon 4, selectively upregulated the expression of corresponding BDNF transcripts 23-fold and 830-fold, respectively. These data describe development of a convenient cell culture-based model to rapidly test human guide RNA specificity for CRISPRa applications. The ability to efficiently manipulate target genes of interest in this line may facilitate novel methods for in vitro human disease modeling.

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Poster

P154. Epilepsy: From Molecular Mechanisms to Seizure Generation

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Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P154.01

Topic: B.10. Epilepsy

Support: Bio-CARe (RGO).BT/AB/08/01/2008 (III) Grant, funded by Department of Biotechnology, Ministry of Science & Technology, Govt. of India [BT/MED/122/SP24580/2018], Grant, funded by Department of Biotechnology, Ministry of Science & Technology, Govt. of India

Title: Potential role of deregulated Cdk5 in the altered synaptic transmission associated with Mesial temporal lobe epilepsy with hippocamapal sclerosis (MTLE-HS) and Focal cortical dysplasia (FCD).

Authors: *A. DIXIT[1], J. BANERJEE[2], A. SRIVASTAVA[3], D. SHARMA[3], S. DEY[3], M. TRIPATHI[4], M. SHARMA[5], P. S. CHANDRA[3];
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Abstract: Rationale: Mesial temporal lobe epilepsy with hippocampal sclerosis (MTLE-HS) and Focal cortical dysplasia (FCD) are the most common forms of drug resistant epilepsy (DRE) pathologies. Identifying factors involved in the abnormal synaptic transmission associated with MTLE-HS and FCD remains a challenge. Cyclin-dependent kinase-5 (Cdk5) regulates synaptic transmission by modulating synaptic composition of various synaptic proteins. Altered Cdk5 activity has been implicated in animal model of epilepsy and in patients with MTLE-HS, FCD and other neurodegenerative diseases. However, its role in abnormal synaptic transmission in DRE is not clearly understood. The present study is designed to delineate the contribution of Cdk5 signalling to the altered synaptic transmission in MTLE-HS and FCD patients. **Methods**: Brain tissue samples resected during surgery from the hippocampal and the anterior temporal lobe (ATL) regions of patients with MTLE (n=8), frontal or temporal lobe regions of patients with FCD (n=23) and non-epileptic controls (n=15), were analysed for the protein expression levels of Cdk5, p-Cdk5, p35/p25, NR2A, pNR2A in MTLE-HS and Cdk5, p-Cdk5, gephyrin, p-gephyrin, γ2 containing $GABA_A$ receptor in FCD by western blotting. Cdk5 activity was analysed by kinase assay in the samples obtained from patients with MTLE-HS. NMDAR mediated glutamatergic activity was measured with and without treatment of Cdk5-specific inhibitor roscovitine, using patch clamp in ATL and hippocampal samples of patients with MTLE-HS. Co expression analysis of Gephyrin and γ2 containing $GABA_A$ receptor was carried out by immunofluorescence in patients with FCD. **Results:** We found significant upregulation of protein expression of Cdk5, p-Cdk5, p35/p25, NR2A, pNR2A in ATL region as compared to hippocampal region in samples resected from patients with MTLE-HS. Similarly, Cdk5 activity was significantly higher in ATL region as compared to hippocampal region. Magnitude of reduction in the frequency of EPSCs by roscovitine in the ATL samples was higher than that in the hippocampal samples. Upregulation of Cdk5 and p-Cdk5 and co-localization of gephyrin with γ2 containing GABAA receptor was observed to be significantly high in FCD patients. **Conclusion:** Our results suggest that Cdk5 might be contributing to the pathophysiology of MTLE-HS and FCD by different mechanisms. Cdk5 might differentially regulate excitatory synaptic activity in the hippocampal and ATL region of patients with MTLE-HS by phosphorylating NR2A subunits. Whereas it might have a potential role in increasing the stability of gephyrin-dependent γ2 subunit containing GABAA receptor clusters in FCD.

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Poster

P154. Epilepsy: From Molecular Mechanisms to Seizure Generation

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Topic: B.10. Epilepsy

Support: Leon Levy Foundation postdoctoral fellowship
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American Epilepsy Foundation

Title: Investigating cellular and circuit mechanism of epilepsy in acute slices from resected human brain tissue

Authors: *T. BUTOLA[1], V. ROBERT[1], L. PENG[1], C. GONG[2], W. DOYLE[1], M. DUSTER[1], S. DEVORE[1], S. LIVINGSTON[1], M. SPINO[1], R. TOMER[2], O. DEVINSKY[1], J. BASU[1];
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Abstract: Nearly one-third of epilepsy patients suffer ongoing seizures despite multiple medication trials. There is an urgent need for therapies and studies that recognize better neural targets for therapeutic intervention. In humans, the hippocampus and the entorhinal cortex are central to the pathophysiology of temporal lobe epilepsy, the most common type of drug-resistant epilepsy. However, most of what we know about cortico-hippocampal circuitry is from rodent studies. Little is known about the cellular organization and synaptic function of the cortico-hippocampal circuit in the human brain. In our study, we aim to fill this gap by characterizing cellular intrinsic properties, circuit connections and integration of inputs in human cortex and hippocampus. We have leveraged the resolution and high-throughput of intracellular patch clamp electrophysiology, and light sheet theta microscopy to parse the functional properties, and structural morphology of human entorhinal cortex and hippocampal neurons. Further, using electrical stimulation we have established the strength, excitation-inhibition coupling and short-term plasticity dynamics of the long-range and local synaptic inputs that these human neurons receive. By systematically comparing these properties between human and mouse neurons derived from equivalent brain areas, we shed light on fundamental cross-species differences and similarities in the cortico-hippocampal circuit. Importantly, capitalizing on our close collaboration with clinicians at the NYU Comprehensive Epilepsy Center, we corroborate our single neuron and circuit level physiology with patient specific pathology of seizures vis a vis developmental timeline, focal area, spread and severity. Elucidating the cellular and circuit pathophysiology governing aberrant neural dynamics in the epileptogenic human brain is crucial to identifying potential targets and strategies for therapeutic intervention at the neuronal level.

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Poster

P154. Epilepsy: From Molecular Mechanisms to Seizure Generation

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Support: Fondation pour la Recherche Médicale (FDM20170839111)
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Title: Metabolomic signatures of status epilepticus: a human case-control study.

Authors: *A. HANIN[1,2], C. CHOLLET[4], S. DEMERET[3], F. CASTELLI[4], V. NAVARRO[1,2,5]; [1]Paris Brain Inst., Paris, France; [2]Epileptology Unit, [3]Neuro-Intensive Care Unit, Pitié-Salpêtrière Hosp., Paris, France; [4]Médicaments et Technologies pour la Santé (MTS), Univ. Paris Saclay, CEA, INRAE, Saclay, France; [5]DMU Neurosciences 6, Sorbonne Univ., Paris, France

Abstract: Objective: Status epilepticus is a life-threatening prolonged epileptic seizure. Seizure recurrence may lead to cellular alterations and homeostasis disturbances responsible for neuronal loss, secondary cerebral atrophy and irreversible neurologic sequels. Understanding the metabolomic processes associated with status epilepticus excitotoxicity is of major importance in order to propose neuroprotective therapies for status epilepticus patients. **Methods**: A comprehensive untargeted metabolomic analysis was conducted on samples from status epilepticus patients (blood n=78; CSF n=29) and control patients (blood n=107; CSF n=29). In order to identify metabolomic signatures from status epilepticus, we used ultra-performance liquid chromatography coupled with an Exactive mass spectrometer. The features were annotated accordingly to biochemical and metabolomic databases. The chromatographic peak areas were compared between status epilepticus and control patients. We manually selected metabolites for which differences could not be explained by the treatment used or by the alimentation provided in intensive care unit, and we performed a metabolic pathway analysis on these features. **Results**: Significant differences between status epilepticus and control patients were identified for 1214 metabolites in the plasma and 358 metabolites in the CSF. We identified 188 metabolites in the plasma and 57 in the CSF for which differences can only be explained by status epilepticus. The pathway analysis identified that metabolic deregulations in status epilepticus were significantly associated with amino acid metabolism (pyruvate, arginine, tryptophan, cysteine and methionine), pyrimidine metabolism, lipid homeostasis and carbohydrate metabolism. We especially found, in SE patients' samples, higher levels of pyruvate, lactate, methionine, serine and ketobutyric acid, and lower levels of arginine, tryptophan, uracil and uridine. **Significance**: Together, our data indicate that status epilepticus is associated with impaired amino acid metabolism. Thus, modulation of amino acid metabolism might be a possible therapeutic approach to prevent status epilepticus excitotoxicity.

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Poster

P154. Epilepsy: From Molecular Mechanisms to Seizure Generation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P154.04

Topic: B.10. Epilepsy

Support: NINDS Grant R01-NS110669

Title: The source of fast traveling waves during human seizures: resolving a controversy

Authors: *E. SCHLAFLY[1], F. A. MARSHALL[1], E. M. MERRICKS[4], U. EDEN[2], S. S. CASH[5], C. A. SCHEVON[6], M. A. KRAMER[3];
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Abstract: Two seemingly contradictory theories describe the source of ictal traveling waves. The first posits that the source arises from a slowly advancing ictal wavefront characterized by collapsing inhibitory restraint (*Smith et al., Nature Communications, 2016*). The second is that the source of traveling waves arises from a fixed location (*Martinet et al., Nature Communications, 2017*). Because these theories were developed using independent microelectrode array recordings and methods from different institutions, the different theories may reflect patient variability or method sensitivity. In this work, we combine the methods and microelectrode array recordings (n=11 patients, n=31 seizures) from the previous studies to analyze the source of ictal traveling waves and understand the factors that led to the different observations. We validate both existing methods on an *in silico* dataset before applying each method to the full cohort of seizures. We show that both of the previously established methods to characterize ictal traveling waves perform similarly, and that seizures show evidence of both theoretical paradigms. When an ictal wavefront is present (17/31 seizures), application of a novel third method shows the pattern of ictal wavefront propagation is correlated with that of traveling waves immediately before ictal wavefront passage, and anticorrelated after ictal wavefront passage. However, both correlations appear only transiently (n=17 seizures, t in [-10, 20] s surrounding ictal wavefront passage). Moreover, we identify multiple shifts in traveling wave directions, suggesting that neither a fixed network nor an ictal wavefront acts alone to produce traveling waves. Using a computational model, we illustrate how an ictal wavefront interacts with a fixed source, and show that this model reproduces features of the traveling wave dynamics observed *in vivo*. We conclude that the existing controversy around the source of ictal traveling waves does not result from differences in methods or patient populations. Instead, we find that ictal traveling waves are generated by interactions between both types of sources - ictal wavefront and fixed location - proposed in previous theories.

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Poster

P154. Epilepsy: From Molecular Mechanisms to Seizure Generation

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Program #/Poster #: P154.05

Topic: B.10. Epilepsy

Support: Hamot Health Foundation. "The Role of Basal Ganglia in Language and Motor Control"

Title: Peri-ictal thalamocortical synchronization in focal epilepsy

Authors: *W. J. LIPSKI[1], S. W. COLLINS[2], M. CAPOGROSSO[2], J. GONZALEZ-MARTINEZ[2];
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Abstract: Medically refractory epilepsy is highly debilitating and lethal. Surgical treatment is often the only option; however, the conventional surgical interventions are often ineffective, costly, and lead to compilations. One of the major impediments to improving the diagnosis and treatment of refractory epilepsy is a basic lack of knowledge about how seizures are organized, how they start, spread and terminate. Intracranial monitoring of brain activity via stereo-electroencephalography (SEEG) has been employed to study electrophysiological signatures of seizures, in order to identify the epileptogenic zone (EZ) for possible resection or neuromodulation. However, despite considerable evidence for subcortical involvement in epilepsy collected over the past century, such studies now focus mainly on examining seizures in cortical brain regions, leaving largely unexplored the vast thalamo-cortical networks that are known to underlie many aspects of cortical synchronization. To characterize the patterns of ictal functional connectivity between thalamic nuclei and related cortical structures that are characteristic of medically refractory focal epilepsy, we studied 11 patients with MRE who were implanted with SEEG depth electrodes that simultaneously explore thalamic nuclei and cortical regions. Specific patterns of synchronized activity in these networks were quantified in the pre-ictal, ictal and post-ictal periods. We hypothesized that peri-ictal synchronization occurs between the cortical EZ and the associated thalamic nuclei. In support of this hypothesis, preliminary analyses showed a selective slow peri-ictal potential shift in thalamic electrodes accompanied by high-frequency (75-300 Hz) phase synchrony between the pulvinar nucleus and EZ identified in occipital cortex; whereas the anterior nucleus showed a similar synchronization with EZ located in frontal cortical areas in another patient. Furthermore, in a patient with posterior quadrant epilepsy, high-frequency pulvinar stimulation (30 seconds train duration, 100 Hz) produced a suppression of epileptiform activity in the EZ during the stimulation epoch. The effect appeared

to be frequency and location dependent, with optimal responses at 100 Hz stimulation applied in the posterior lateral pulvinar nucleus. These results support the hypothesis that thalamo-cortical networks critically participate in seizure initiation and termination via their reciprocal connections. This knowledge will advance the development of treatment options by defining optimal neuromodulation targets and parameters of therapeutic electrical stimulation intended to modify these networks.

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Poster

P154. Epilepsy: From Molecular Mechanisms to Seizure Generation

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Program #/Poster #: P154.06

Topic: B.10. Epilepsy

Support: Tiny Blue Dot

Title: Linking altered sleep homeostasis to ictal recruitment using human intracranial recordings

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Abstract: Rationale: In neurotypical brains, synaptic strengthening during wake leads to an increase in slow wave activity (SWA, i.e. 1-4 Hz) during sleep, while, sleep dependent downscaling leads to SWA power an exponential decline overnight. Here we analyzed ictal and interictal sleep recordings from patients implanted with intracranial electrodes for clinical purposes to investigate the correlation between sleep SWA and ictal recruitment at the level of local cortical networks. We investigated the link between ictal increases in phase-locked high gamma (PLHG) - a proxy for neuronal firing - and interictal increases in sleep SWA.**Methods:**6 human epileptic patients were included in the analysis: 3 patients with depth electrodes (16, 24, and 59 electrodes in cingulate or hippocampal areas) and 3 patients with a combination of grid and strip subdural electrodes (55, 94, and 80 electrodes; 1 with poorly localized seizures, and 1 without seizures). For each patient, a whole night of recordings without seizures was selected and visually sleep scored using slow waves and spindles. Bad channels, bad epochs and 1-second epochs around interictal spikes identified by an epileptologist were removed from the analysis.

We computed the power for SWA (delta (δ)) and beta (β) bands, and compared the average δ power and δ/β ratio between the first and last hour of sleep in each electrode. δ power and δ/β ratio values within the first and last hour and their difference overnight were then Spearman-correlated to ictal PLHG. **Results:** Visual inspection of individual patient data suggested that highest δ power co-localized with the clinically identified SOZ. The average correlation between δ power during sleep and ictal PLHG was positive in each of the 5 patients with seizures (significant at $p<0.05$ at the individual level in the 3 patients with the most frequent seizures). δ power and δ/β ratio showed an average decline across all channels in 4/5 patients. In the 4 patients where seizure focus could be localized, a paradoxical positive correlation was observed between ictal PLHG and an increase (rather than a decrease) in δ/β ratio overnight. Interestingly, in contrast, overnight change in δ/β ratio was negatively correlated with ictal PLHG in the patient with unidentified focus. **Conclusions:** These preliminary results from intracranial recordings in 6 patients with focal epilepsy suggest that power adaptively decreases across the whole cortex during a night of sleep, while sleep homeostasis is selectively altered in the epileptic focus. If findings are confirmed in larger studies, interictal sleep SWA patterns may provide useful localization information about epileptogenic regions in patients.

Disclosures: B. Sevak: None. **D. Scott:** None. **C. Denis:** None. **V. Kremen:** None. **A. Struck:** None. **E.M. Merricks:** None. **G. McKhann:** None. **S.A. Sheth:** None. **L.M. Bateman:** None. **G. Tononi:** None. **C.A. Schevon:** None. **M. Boly:** None.

Poster

P154. Epilepsy: From Molecular Mechanisms to Seizure Generation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P154.07

Topic: B.10. Epilepsy

Support: The Children's Foundation r1-2021-31

Title: Impact of cannabidiol (CBD) on anxiety symptoms in pediatric epilepsy patients: A preliminary analysis

Authors: *S. SINGH, B. BORG, A. MORALES, J. EVANSKI, J. K. MACDOUGALL, A. R. WEBER, A. LUAT, **H. MARUSAK**;
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Abstract: Title: Impact of cannabidiol (CBD) on anxiety symptoms in pediatric epilepsy patients: A preliminary analysis**Authors:** Saurav Singh, Breanna Borg, Austin Morales, Julia Evanski, Jamie MacDougall, Amanda Weber, Aimee Luat, Hilary Marusak
Funding: The Children’s Foundation
Cannabidiol (CBD) is used as a medication for treating seizures in children. Recent research

suggests that CBD can also reduce anxiety in adults. Importantly, anxiety disorders are the most common childhood mental illness, affecting up to one in three before age 18. Further, anxiety affects up to 50% of children with epilepsy which may negatively affect their quality of life. However, little is known about the effects of CBD on anxiety in children. Our ongoing study examines CBD as a potential treatment for childhood anxiety, by leveraging a group of patients undergoing clinically indicated treatments. We conducted a preliminary analysis (n = 4; ages 2-20; 3 female) of our ongoing 4-6-week observational study of pediatric epilepsy patients at the Children's Hospital of Michigan. Participants were followed as they started on CBD (Epidiolex, Jazz Pharmaceuticals) for clinically indicated seizures. Parents/guardians completed surveys about their child before starting on CBD, and again after achieving the maintenance dose (4-6 weeks; ~25 mg/kg). Surveys were about seizure frequency, medication side effects, and anxiety symptoms. At the end of the study, three out of the four parents reported that they would recommend Epidolex to other families (one parent was undecided). Three out of the four parents also reported a reduction in their child's seizure frequency from baseline ($M \pm SD$ = 6.05 ± 7.85 seizures/month) to the end of the study (2 ± 4 seizures/month), and three out of four reported fewer average negative side effects (e.g., speech difficulties, unstable walking) after starting the medication (baseline: 49 ± 29.13, end: 35.25 ± 17.58). All four parents reported more frequent positive side effects (e.g., improved mood, better sleep) at the end of the study (15 ± 6.92) as compared to baseline (6.75 ± 3.2). Three out of four parents reported a reduction in their child's anxiety symptoms from baseline (12 ± 11.6) to the end of the study (2.25 ± 1.71); one patient showed no change in anxiety symptoms. Taken together, these data are consistent with prior studies demonstrating the effectiveness of CBD for reducing seizures in pediatric populations, and that CBD may have fewer side effects than other anti-seizure medications. Although preliminary, these data also indicate that CBD may be a promising approach for reducing anxiety symptoms in pediatric samples. This notion will be explored in future studies.

Disclosures: S. Singh: None. **B. Borg:** None. **A. Morales:** None. **J. Evanski:** None. **J.K. MacDougall:** None. **A.R. Weber:** None. **A. Luat:** None. **H. Marusak:** None.

Poster

P226. Apoe and Associated Pathways I

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Program #/Poster #: P226.01

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: R01DC014447
RF1AG069196

Title: Cellular and Circuit Vulnerability to Apolipoprotein E4 in the Olfactory System

Authors: *D. GILHOOLY[1], R. BUDHATHOKI[1], E. STARR[1], *S. LIU[2];
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Abstract: Accumulating evidence supports a causative relationship between the allele ε4 of apolipoprotein E (APOE4), the strongest genetic risk factor for Alzheimer's disease (AD), and olfactory dysfunction (OD), which is the prodromal AD symptom. Consistently, animal studies have showed olfactory behavior deficits and network dysfunction in the olfactory bulb (OB) and piriform cortex in transgenic (APOE4) mice carrying the human APOE4 gene at the age of 6 and 12 months, respectively, indicating initiation of APOE4 pathogenesis in the OB. However, the cellular and circuit substrates of these age-dependent APOE4 actions in the olfactory system remain unclear. We focused on APOE4 actions in the OB and its adjacent downstream structure the anterior olfactory nucleus (AON) at the cellular and circuit levels in 6-month-old APOE4 mice and had the following main findings: (1) APOE4 elevates excitability of OB mitral cells (MCs) and AON neurons in awake head-fixed and anesthetized mice, respectively; (2) APOE4 significantly prolongs MC responses to the stimulation of olfactory nerve, spontaneous excitatory (sEPSCs) and inhibitory postsynaptic currents (sIPSCs) in MCs, indicating neurotransmitter uptake deficit in the OB of APOE4 mice; (3) consistently, APOE4 mice display atrophic alteration in OB astrocytes, which play key roles in neurotransmitter uptake; (4) APOE4 mice exhibit deficit in olfactory habituation and dishabituation, which is rescued by chemogenetic activation of OB astrocytes. Ongoing work will expand to target the OB-AON synaptic pathways. Taken together, our study suggests that APOE4 acts on OB output neurons to dysregulate AON functions leading to OD at the early age.

Disclosures: D. Gilhooly: None. **R. Budhathoki:** None. **E. Starr:** None. **S. Liu:** None.

Poster

P226. Apoe and Associated Pathways I

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Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P226.02

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH R01AG061114

Title: An autocrine role for apolipoprotein E in brain endothelial cell function

Authors: *F. M. MAROTTOLI[1], L. M. TAI[2];
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Abstract: The association of the apolipoprotein E4 genotype (*APOE4*) and neurovascular dysfunction with age and neurodegeneration underscores the importance of identifying the underlying mechanisms. Specialized brain endothelial cells (BECs) are the core component of

the neurovasculature yet changes in their function are often viewed solely as a consequence of altered astrocyte and pericyte signaling. Given that BECs are directly responsible for neurovascular function, this perspective critically overlooks their dynamic and complex functions. To better understand the extent *APOE* genotype of BECs modulates their phenotype, we isolated primary BECs from 2-month-old male and female *APOE*-targeted replacement mice and performed experiments without subculture. Initial biochemical and RNA-sequencing characterization revealed that compared to *APOE3*, *APOE4* BECs produce less apolipoprotein E (apoE) and have altered metabolic and inflammatory gene expression profiles. Further, through specific functional assays, we found that *APOE4* BECs demonstrate a greater preference for oxidative phosphorylation over glycolysis and, accordingly, higher markers of mitochondrial activity. Complementarily, *APOE4* BECs had higher mitochondrial superoxide levels, lower levels of antioxidants related to heme and glutathione, and higher markers/outcomes of oxidative damage to proteins and lipids. Either in parallel or through sequential processes, markers of inflammation were also higher in *APOE4* BEC cultures under both basal conditions and following a low dose of lipopolysaccharide (LPS). Further, a high dose of LPS resulted in greater paracellular permeability in *APOE4* BECs when assessed by electric cell-substrate impedance sensing. Importantly, guided by our transcriptomic data, we found that the nuclear receptor Rev-Erb agonist SR9009 improved functional metabolic markers, lowered inflammation, and modulated paracellular permeability at baseline and following LPS treatment in *APOE4* BECs. Collectively, our data identify a role for autocrine apoE signaling in BECs and represents a novel cellular mechanism for how *APOE* regulates neurovascular function.

Disclosures: F.M. Marottoli: None. **L.M. Tai:** None.

Poster

P226. Apoe and Associated Pathways I

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Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P226.03

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: T32AG20506

Title: The effects of apolipoprotein E isoforms on spread of phosphorylated tau

Authors: *A. AFFANEH, A. K. LINDEN, J. A. KESSLER;
Neurol., Northwestern Univ., Chicago, IL

Abstract: The development and progression of Alzheimer’s disease (AD) are strongly correlated with the human apolipoprotein E (*hAPOE)* genotype. Tau pathology consisting of hyperphosphorylated neurofibrillary tangles is a hallmark of AD. The spread of phosphorylated tau (p-tau) is known to spread in a stereotyped manner starting in the entorhinal cortex

progressing lastly to the hippocampus and neocortex. While both *APOE*4 genotype and prion-like hyperphosphorylated tau propagation are linked to advanced disease progression, it remains unclear how heterologous expression of *APOE* effects p-tau spread. Here we sought to address this question by differentiating *APOE* E4/E3 and CRISPR/Cas9 corrected isogenic *APOE* E3/E3 into excitatory forebrain neurons while using microfluidic devices. This, in conjunction with the development plasmonic sensors sensitive to multiple species of tau, facilitates the ability to track release of pathological tau in real time. Using this nanotechnology, we evaluated the levels of p-tau spread between *APOE* genotypes. Specifically, *APOE*4 neurons have higher levels of intracellular p-tau/tau, as well as secreted p-tau/tau. Under biological stressors, levels of apoE increases in parallel with increases of p-tau. Future studies will investigate specific mechanisms in which apoE interactions effect the spread of neurotoxic proteins.

Disclosures: A. Affaneh: None. **A.K. Linden:** None. **J.A. Kessler:** A. Employment/Salary (full or part-time):; Northwestern University. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Northwestern University. Other; Editor in Chief of Annals of Clinical and Translational Neurology.

Poster

P226. Apoe and Associated Pathways I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P226.04

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH grant AG061776 (DMH)
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Title: APOE-ε4 but not APOE-ε3 genotype synergizes with chronic sleep disruption to accelerate Aβ plaque deposition in APPPS1-21 mice

Authors: *C. WANG, C. LEE, S. PARHIZKAR, C. E. WALLACE, A. C. MOORE, J. R. CIRRITO, D. M. HOLTZMAN;
Dept. of Neurol., Washington Univ. In St. Louis, Saint Louis, MO

Abstract: Alzheimer’s Disease (AD) is the most common cause of dementia and a major public health problem. Currently, around 47 million people worldwide are living with dementia. Thus, identifying modifiable risk factors for AD is critical. The apolipoprotein E *(APOE)* gene is the strongest genetic risk factor for late-onset AD. *APOE*-ε4 genotype is strongly associated with increased AD risk and amyloid-β (Aβ) plaque accumulation in humans, as compared to the other common *APOE* genotypes such as ε3. In relation to sleep, recent human studies suggest that

APOE genotype modulates the effect of sleep disruption on AD risk, suggesting a possible link between apoE and sleep in AD pathogenesis which is relatively unexplored. To test our hypothesis that *APOE* genotype modifies Aβ deposition in response to chronic sleep deprivation (SD), we used APPPS1-21 mice with targeted replacement of the murine Apoe locus with human *APOE*-ε3, or -ε4. At four months of age, experimental mice were placed in automated SD chambers set at a 30 sec interval for 6 hours per day (8 am to 2 pm) for 8 weeks or in normal cages with normal sleep (NS) conditions. After treating mice with chronic SD or normal sleep, we quantified total or fibrillar Aβ plaques. Interestingly, chronic SD in *APPPS1-21* mice significantly increased Aβ deposition in the presence of apoE4 but not apoE3. To have a better understanding why *APPPS1-21:ε4/4* mice are more significantly affected by SD, we are currently using several approaches, including assessing whether SD influences interstitial fluid (ISF) Aβ clearance in an isoform-dependent fashion.
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Disclosures: C. Wang: None. **C. Lee:** None. **S. Parhizkar:** None. **C.E. Wallace:** None. **A.C. Moore:** None. **J.R. Cirrito:** None. **D.M. Holtzman:** A. Employment/Salary (full or part-time):; Co-founder, C2N Diagnostics LLC.. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; National Institutes of Health, Cure Alzheimer's Fund, Tau Consortium, the JPB Foundation, Good Ventures, C2N Diagnostics, Novartis, NextCure. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); a patent licensed by Washington University to C2N Diagnostics on the therapeutic use of anti-tau antibodies. This anti-tau antibody program has now been licensed to Abbvie, a patent licensed by Washington University to Eli Lilly on a humanized anti-Aβ antibody. F. Consulting Fees (e.g., advisory boards); Genentech, Denali, C2N Diagnostics, Cajal Neurosciences, Takeda, Casma, Eli Lilly.

Poster

P226. Apoe and Associated Pathways I

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Program #/Poster #: P226.05

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH/NIA R01AG060195
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Harold and Ronna Cooper Family

The Consolidated Anti-Aging Foundation

Title: Loss of apolipoprotein E exacerbates microglial and astrocyte reactivity to sustained systemic glucocerebrosidase inhibition.

Authors: *K. J. CONNOLLY, O. R. BREKK, S. N. ROSENTHAL, C. N. CLOUSE, *P. J. HALLETT, O. ISACSON;
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Abstract: Elevated lipid storage is frequently observed in late-onset neurodegenerative diseases such as Alzheimer's disease (AD) and Parkinson's disease (PD). Genetic risk factors for AD and PD also include factors involved in lipid distribution and metabolism, such as *APOE* and *GBA*. Mechanisms for lipid transport between neurons and glia in response to mitochondrial stress and NMDA-induced excitotoxicity have been shown to depend on glial apolipoprotein E (apoE) expression *in vitro*. The promotion of lipid transport between neurons and glia during stress suggests that apoE, as the major transcellular lipid carrier in the brain, is a key factor in the cellular adaptation to stress. This in turn suggests that insufficient apoE activity may underpin the elevated lipid storage associated with neurodegenerative diseases. To determine whether apoE function is required for cellular adaptation to increased lipid load, 3-month-old WT and apoE-KO mice received daily intraperitoneal injections of the glucocerebrosidase inhibitor, CBE, for 9 or 18 d. Animals were perfused 1 d after the final dose for histological and biochemical analysis. Microglial and astrocyte reactivity to CBE were assessed by Iba1 and GFAP immunohistochemistry. Microglial reactivity was greater in the dentate gyrus, substantia nigra and layer V of the motor cortex in apoE-KO mice after 9 and 18 d CBE, relative to WT. Loss of apoE also elevated astrocyte reactivity in the dentate gyrus, substantia nigra and layer V of the motor cortex after 18 d CBE. Ongoing analyses into the molecular and cellular consequences of prolonged CBE include changes in lipid transport, α-synuclein aggregation, tau phosphorylation, lysosome biogenesis, autophagy and proteostasis, to determine how loss of apoE function may impact cellular adaptation to intracellular lipid formation resulting from sustained glucocerebrosidase inhibition. The exacerbated glial activation in apoE-KO animals receiving CBE suggests that apoE expression and function may, in part, determine the capacity of neuron-glia networks to overcome damaging stimuli. When apoE function is insufficient, failed resolution of lipids may then lead to persistent damage and degeneration. Therefore, characterizing the consequences of limited lipid clearance on the adaptation to rising lipid load is highly relevant to understanding how failure to overcome increased lipid storage contributes to neurodegenerative processes in AD, PD and related disorders.

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Poster

P226. Apoe and Associated Pathways I

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Program #/Poster #: P226.06

Topic: C.02. Alzheimer's Disease and Other Dementias

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Title: Human iPSC-derived astrocytes as a model for studying the impact of APOE polymorphism in Alzheimer´s disease

Authors: E. DÍAZ-GUERRA, R. VECINO, E. ARRIBAS-GONZÁLEZ, I. SERRA-HUETO, E. MORENO-JIMÉNEZ, M. NAVARRETE, R. MORATALLA, *C. VICARIO;
Cajal Institute, CSIC, Madrid, Spain

Abstract: Human iPSC-derived astrocytes as a model for studying the impact of APOE polymorphism in Alzheimer´s disease

Authors E. DÍAZ-GUERRA[1,2], R. VECINO[1,2], E. ARRIBAS-GONZÁLEZ[1,2], I. SERRA-HUETO[1], E. MORENO-JIMÉNEZ[1,2], M. NAVARRETE[1], R. MORATALLA[1,2], *C. VICARIO[1,2]. [1]Instituto Cajal-CSIC, Madrid, Spain; [2]CIBERNED-ISCIII, Madrid, Spain

Disclosures E. Díaz-Guerra: None. **R. Vecino:** None. **E. Arribas-González:** None. **I. Serra-Hueto:** None. **E. Moreno-Jiménez:** None. **M. Navarrete:** None. **R. Moratalla:** None. **C. Vicario:** None.

Abstract The epsilon4 (ε4) allele of the APOE gene, which encodes the apolipoprotein E4 (ApoE4), is a strong genetic risk factor for developing Alzheimer´s disease (AD), the most common form of dementia. Astrocytes play a role in AD pathology but the impact of the disease and APOE alleles on human astrocyte maturation and function remains to be fully elucidated. To address this question, we have obtained induced pluripotent stem cells (hiPSCs) from fibroblasts of AD patients carrying the ε4 and ε3 alleles (in homozygosis), from healthy controls and from isogenic iPSCs lines carrying the different APOE alleles. Astrocyte differentiation was achieved by sequential addition of small molecules and growth factors. The hiPSC-derived astrocytes expressed GFAP, GLT1, AQP4 and S100ß. These cells showed Aβ and glutamate uptake capacity, produced Ca^{2+} waves and responded to inflammatory stimuli in both AD- and APOE allele-dependent manners. Our findings support the use of hiPSC-derived astrocytes as a model to study mechanisms of cell dysfunction in AD and reveal a tight connexion between APOE, astrocyte homeostasis and AD pathology.

Disclosures: E. Díaz-Guerra: None. **R. Vecino:** None. **E. Arribas-González:** None. **I. Serra-Hueto:** None. **E. Moreno-Jiménez:** None. **M. Navarrete:** None. **R. Moratalla:** None. **C. Vicario:** None.

Poster

P226. Apoe and Associated Pathways I

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Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant DP2NS087943
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AG047644

Title: Regulation of neuronal beta-amyloid production by astrocytic cholesterol and mechanical force

Authors: *H. WANG[1], J. A. KULAS[2], C. WANG[3], D. A. KOZLOWSKI[4], D. M. HOLTZMAN[5], H. A. FERRIS[2], S. B. HANSEN[1];
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Abstract: Alzheimer's Disease (AD) is characterized by the presence of β-Amyloid (Aβ) plaques, tau tangles, and loss of cognitive function. ApoE4 isoform and traumatic brain injury (TBI) are both risk factors for AD, but it is unclear if there are common underlying mechanisms. In this study we show that Aβ generation in neurons is regulated by apoE delivery of cholesterol and the nanoscale clustering of the amyloid precursor protein (APP) processing proteins in neurons while mechanical force works opposite to cholesterol. Using super-resolution imaging in mouse brain, we show that astrocyte-derived cholesterol is carried by apoE to specifically traffic neuronal APP in and out ganglioside (GM1) lipid clusters where it interacts with β- and γ-secretases to generate Aβ-peptide. We find that targeted deletion of astrocyte cholesterol synthesis robustly reduces amyloid and tau burden in a mouse model of AD. Treatment with cholesterol-free apoE or knockdown of cholesterol synthesis in astrocytes decreases cholesterol levels in cultured neurons and causes APP to traffic out of GM1 clusters where it interacts with α-secretase, however γ-secretase does not respond to cholesterol regulation and remains in the GM1 clusters. APOE4 expressing mouse brains have more cholesterol and greater association of APP with GM1 domains than APOE3 brains. In cell culture, we show that both APP and γ-secretase are mechanosensitive and they sense different thresholds of mechanical shear (normal and noxious respectively). In a controlled cortical impact rat model of TBI, high intensity (noxious) mechanical force causes both APP and γ-secretase to move from thicker GM1 clusters to the thinner disordered regions. Translocation of APP's γ-cleavage from thick to thin membrane results in a shift from Aβ40 to Aβ42 production. As animals recovered from the

injury, APP moves back into the GM1 clusters regulated by increased membrane cholesterol while γ-secretase does not respond to cholesterol change. We conclude that astrocytes regulate Aβ production through the delivery of cholesterol to neurons by apoE. This suggests that the cholesterol is used as a signaling molecule, and the concentration is kept low in neurons to allow for astrocyte signaling. In contrast, mechanical force translocates γ-secretase to the thinner membrane, leading to an increased Ab42/40 ratio.

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Poster

P226. Apoe and Associated Pathways I

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Program #/Poster #: P226.08

Topic: C.02. Alzheimer's Disease and Other Dementias

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Foundation for Barnes-Jewish Hospital 3770

Title: Structure of Human Lipidated Apolipoprotein E

Authors: *M. STRICKLAND[1], Y. CHEN[2], M. RAU[3], B. SUMMERS[3], R. KOSSINA[3], H. JIANG[1], J. ULRICH[1], J. FITZPATRICK[3], D. M. HOLTZMAN[1];
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Abstract: The Apolipoprotein E (*APOE*) gene is the most important genetic risk factor for late-onset Alzheimer's Disease (AD). ApoE is a lipid associated protein that plays a vital role in intercellular cholesterol and lipid transport, especially in the central nervous system (CNS) where it is the primary apolipoprotein. ApoE has three major isoforms (ApoE2, ApoE3, and ApoE4) of which ApoE3 is the most common. A single copy of the ApoE4 allele increases AD risk approximately 4-fold and two copies increase disease risk approximately 12-fold relative to the ApoE3 allele. The ApoE2 allele decreases risk for AD relative to the ApoE3 allele. Despite resulting in a dramatic increase in disease risk, ApoE4 only differs from ApoE3 by a single amino acid. To better understand the significance of this change, we aim to study the structure of lipidated ApoE; its physiological confirmation. Previous studies investigating the structure of

ApoE have been hampered by both the lipid environment of the physiological forms of ApoE and the tendency of non-lipidated to rapidly aggregate. We aim to address these issues by utilizing single particle cryo-electron microscopy (cryoEM) in order to determine the structure of lipidated ApoE. Advances in direct electron detectors and automated data acquisition strategies now routinely allow the determination of protein structures to near-atomic resolution. Using this method, we plan to determine the structure of lipidated ApoE2, ApoE3, and ApoE4 in order to understand conformational differences between the isoforms to reveal mechanistic insights into the role of ApoE for AD risk. In this study, we utilize both astrocyte-produced ApoE particles and artificially-lipidated recombinant ApoE. We have characterized these particles using size-exclusion chromatography and nondenaturing gradient gel electrophoresis. We have shown in our study that the artificial particles recapitulate the features of astrocyte produced ApoE particles and are a good initial model for understanding ApoE structure. In this study, we have also utilized monoclonal Fab fragments to further characterize the ApoE particles and assist in alignment of the ApoE particles during 2D classification. Using this method, we show two ApoE proteins are present per lipoparticle and that ApoE adopts at least two major conformations on lipid containing particles. Currently, we have obtained low resulotion 3D electron density maps of the ApoE lipoparticles in the two major conformations. Future research will pursue obtaining a high-resolution structure of the three isoforms to better understand any conformational differences that exist between the ApoE isoforms.

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Poster

P226. Apoe and Associated Pathways I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P226.09

Topic: B.11. Glial Mechanisms

Support: COLCIENCIAS

Title: Different gliovascular alterations characterize the frontal cortex of sporadic and familial Alzheimer's disease: APOEch protection

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Abstract: In response to brain insults astrocytes become reactive. This process modifies the structural and functional properties of these cells giving rise to highly context-specific effects that frequently become maladaptive. In fact, astroglial reactivity is characteristic of brain pathologies such as Alzheimer's disease (AD). Interestingly, Colombia has the largest human group known with familial Alzheimer's disease (FAD). This kindred carry the autosomal dominant mutation E280A in PSEN1, which causes early-onset dementia. Recently, our group has identified an E280A carrier who did not develop FAD. The individual was homozygous for the Christchurch mutation in APOE3 (APOEch). Remarkably, most of apoE (the APOE gene product) -which is the main genetic risk factor for developing sporadic Alzheimer's disease (SAD)- is produced by the astroglia. Since astrocytes are central in highly delicate brain processes -as neuroinflammation, the maintenance of glutamate homeostasis, and the preservation of the gliovascular unit (GVU)-, in this study we characterized some of the cellular aspects of the astrocytes that could influence the mechanisms of pathogenesis or protection of the three different biological backgrounds of SAD, FAD and APOEch. Interestingly, we show that the frontal cortex of APOEch displays the mildest histological alterations despite presenting the highest amyloid burden. Furthermore, while FAD is characterized by a hyperreactive-like microenvironment and SAD exhibits astrodegeneration, the astroglia of APOEch resembles that of control. Also, we show that glutamate-regulatory astrocytic proteins are altered in the three conditions, but to a lesser extent in APOEch. Lastly, we demonstrate that gliovascular deterioration is characteristic of SAD and FAD but not of APOEch. Our results suggest a preponderant role of astrocytes in the maintenance of the GVU under the context of AD and, therefore, in the mechanisms of brain health and disease.

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Poster

P253. Dopamine and Non-Dopamine Pathways I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P253.01

Topic: C.03. Parkinson's Disease

Support: University of Florida Colledge of Medicine

Title: Lrrk2 kinase activity and dopamine neurotransmission is differentially affected by vps35 p.d620n or knockdown

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Abstract: VPS35 is an essential component of the retromer complex required for endosomal sorting for lipid and protein recycling. A mutation in VPS35 (p.D620N) leads to autosomal dominant Parkinson's disease, whereas a reduction in VPS35 expression is associated with Alzheimer's dementia. Mutations in LRRK2 that constitutively increase its kinase activity and downstream Rab phosphorylation are also a leading genetic cause of Parkinson's disease, and LRRK2 and VPS35 may directly interact. Modulating this endosomal-vesicular trafficking system is an important therapeutic opportunity to slow or halt neurodegeneration. To better understand the relationship between retromer and Lrrk2, and to assess gain- versus loss-of-function, we have examined striatal dopamine neurotransmission by ex-vivo slice voltammetry in Vps35 p.D620N knock-in (VKI) and haploinsufficient Vps35 mice (hVKO, in which retromer is ~50% reduced). Evoked striatal dopamine release is significantly enhanced in 3-month-old VKI mice but declines to a level comparable to wild type (WT) in 12-month-old animals. Lrrk2 kinase activity is also constitutively elevated in 3-month-old VKI mice. As a corollary, in vivo treatment of VKI animals with the Lrrk2 kinase inhibitor, Mli2, normalizes both dopamine transporter (Dat) expression and striatal dopamine release. Here we demonstrate hVKO mice also exhibit a significant increase in evoked dopamine release compared to age-matched WT mice that is maintained at 12 months. Together, our results suggest Vps35 p.D620N and Vps35 haploinsufficiency may result in a similar loss-of-function with respect to Dat recycling and striatal dopaminergic neurotransmission. However, in contrast to VKI animals, LRRK2 kinase activity in hVKO mice is reduced compared to WT littermates. Thus hyper- and hypoactivation of LRRK2 kinase is associated with retromer induced dysregulation of dopamine neurotransmission, and these observations should be reconciled before LRRK2 kinase inhibitors are recommended for therapeutic use. Defects in vesicular trafficking from retromer and/or LRRK2 dysfunction may unproportionally affect dopamine specific cargos and dopaminergic neurons due to their extensively arborized, poorly myelinated axons and requirement for nigrostriatal axonal trafficking. Cellular context is important and is to be more fully explored.

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Poster

P253. Dopamine and Non-Dopamine Pathways I

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Topic: C.03. Parkinson's Disease

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NIH Grant 1S10OD020026-01
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Title: Dopaminergic network properties: implications in behavior

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Abstract: Dopaminergic neurons of the substantia nigra (SNC) and ventral tegmental area (VTA) exhibit spontaneous firing activity. The dopaminergic neurons in these regions have been shown to exhibit differential sensitivity to neuronal loss and psychostimulants targeting dopamine transporter. However, it remains unclear whether these regional differences scale beyond individual neuronal activity to regional neuronal networks. Here we utilized live-cell calcium imaging to show that network connectivity greatly differs between SNC and VTA regions with higher incidence of hub-like neurons in the VTA. Specifically, the frequency of hub-like neurons was significantly lower in SNC than in the adjacent VTA, consistent with the interpretation of a lower network resilience to SNC neuronal loss. We tested this hypothesis when activity of an individual dopaminergic neuron is suppressed, through whole-cell patch clamp electrophysiology, in either SNC, or VTA networks. Neuronal loss in the SNC increased network clustering, whereas the larger number of hub-neurons in the VTA overcompensated by decreasing network clustering in the VTA. We further show that network properties are regulatable via a dopamine transporter dependent mechanism. Our ongoing work expands our findings to an in vivo model system where we stimulate dopaminergic networks with simultaneous real time calcium imaging. The behavioral responses and dopaminergic network are tracked across experimental time and session instances. Thereby changes in dopaminergic networks are matched to behavioral changes beyond single neurons. Our results demonstrate novel regulatory mechanisms of functional network topology in dopaminergic brain regions and how they relate to behavior.

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Poster

P253. Dopamine and Non-Dopamine Pathways I

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DA043895
The Karen Toffler Charitable Trust

Title: In drug naive Parkinson's disease patients, Dopamine transporter and Tyrosine hydroxylase expression are increased in peripheral immune cells

Authors: *A. GOPINATH[1], P. M. MACKIE[1], A. SMITH[1], T. RIAZ[1], C. HANSEN[1], A. H. GITTIS[4], R. BOUCHARD[5], J. SCHWENK[5], P. HASHEMI[6], A. BUCHANAN[6], P. J. GASKILL[7], R. NOLAN[7], S. MATT[7], A. DOTY[2], A. RAMIREZ-ZAMORA[1], M. OKUN[1], W. J. STREIT[3], H. KHOSHBOUEI[3];
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Abstract: Introduction: While the scientific debate about the origin of sporadic Parkinson's Disease (PD) i.e. body-first vs. brain-first hypothesis, has not been settled, it is well-established that PD pathology affects both CNS and peripheral dopamine transmission. The vast majority of PD research has focused on neuronal and glial dysfunction, but there is strong evidence demonstrating the dysregulation of innate immunity in PD at the systemic level. In PD, there is a loss of dopamine neurons as measured by decreased markers of dopaminergic neurons, the dopamine transporter (DAT) and tyrosine hydroxylase (TH). Objective: Since peripheral immune cells express dopaminergic proteins, including DAT and TH, we tested the hypothesis that in drug naïve PD patients there is a similar reduction in DAT+/TH+ peripheral blood mononuclear cells (PBMCs). Methods/Results: We first established a highly sensitive and reproducible flow cytometry assay to assess DAT and TH expression in PBMCs of 12 drug naïve PD patients (newly diagnosed), 84 PD patients receiving treatments and 67 aged/sex matched healthy subjects. We found there is no change in total PBMCs ($p>0.91$, healthy vs. PD, vs. drug naïve, One-way ANOVA), but contrary to our hypothesis we found that percentages of DAT+/TH+, but not NET+, PBMCs are significantly higher in drug naïve PD patients (n=12) compared to healthy controls (n=67) ($P<0.001$, One-way ANOVA). Importantly, in PD patients receiving treatments (n=84), irrespective of the treatment modality, the percentages of

DAT+/TH+ monocytes declined significantly compared to drug naïve patients, but remain significantly higher than healthy controls (P<0.001, one-way ANOVA), suggesting that treatment for PD can modulate peripheral immune cells expressing DAT/TH. Similar to human PD, the percentages of DAT+/TH+ myocytes are increased in two animal models of PD and reduced following L-dopa+Carbidopa, but not L-dopa alone. To test whether changes in DAT+/TH+ PBMCs are increased in other neurodegenerative diseases, we recruited a patient cohort with Alzheimer's disease (n=6) and found increased DAT+/TH+ PBMCs to be specific for PD. Alzheimer's patients did not exhibit changes in the DAT+/TH+ PBMCs. We also discovered that PD patients exhibit increased DAT+/TH+ myeloid derived suppressor cells (MDSCs) suggesting a compensatory immune response in PD. Ongoing studies are focused on investigating the mechanism underlying increased DAT+/TH+ PBMCs in PD. Collectively, these data suggest a system-wide dysregulation of the dopamine signaling on peripheral immune cells in PD, providing a novel diagnostic and therapeutic perspective for PD.

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Poster

P253. Dopamine and Non-Dopamine Pathways I

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Title: In Parkinson's disease patients Tyrosine Hydroxylase expression is increased in CD14+ monocytes and linked to TNF

Authors: *A. DAGRA[1], A. GOPINATH[1], M. G. TANSEY[3], G. SHAW, Dr[4], D. MILLER[1], C. HANSEN[1], P. M. MACKIE[1], A. RAMIREZ-ZAMORA[1], M. OKUN[1], W. J. STREIT[1], H. KHOSHBOUEI[2];
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Abstract: Introduction: Peripheral immune cells (human and mouse) express tyrosine hydroxylase (TH), the rate limiting enzyme in catecholamine synthesis. TH is classically studied in the context of brain catecholamine signaling, but little is known about TH production and function in peripheral immune cells. This knowledge gap is due, in part, to the lack of an adequately sensitive assay to measure TH in immune cells expressing lower TH levels compared to cells with high TH expression. **Objective/Methods**: We developed the TH-Bio-ELISA that can quantify TH picogram levels of TH protein. We then applied the TH-Bio-ELISA, to measure TH in the $CD14^{+}$ monocytes of 11 Parkinson's disease (PD) patients, and 11 aged matched healthy control subjects from both sexes. We hypothesized that the loss of TH+ neurons in the brains of PD patients is accompanied by lower levels of TH in the peripheral monocytes of PD patients. **Results**: We found that TH-Bio-Elisa is highly sensitive and reproducibly quantifies picogram levels of TH in multiple TH expressing cells such as primary human macrophages, murine dopamine neurons, PC12 cells and primary human monocytes. We applied this assay to monocytes isolated from venous blood of PD patients (n=11) and healthy age-matched control subjects (n=11). Contrary to our hypothesis, we found that monocytes from PD patients express significantly higher levels of TH protein than those form healthy controls ($P<0.01$, two-tailed t-test). Next, we focused on understanding the mechanism of increased monocytic TH in PD. It is known that tumor necrosis factor (TNF), a pro-inflammatory cytokine, is increased in the brains and peripheral circulation of PD patients and animal models of PD. Therefore, we investigated a possible connection between higher levels of TH protein and increased TNF in PD. Monocytes isolated from healthy donors were treated with TNFα or with TNF in the presence of an inhibitor (XPro1595). Tissue plasminogen activator (TPA) was used as a positive control. We observed that TNF stimulation increased both the number of TH+ monocytes and the quantity of TH per monocyte (N=3-6 independent biological replicates per group, $P<0.01$), without increasing the total number of monocytes. **Conclusions**: Our results show that TNF modifies monocytic TH production and serves a regulatory role in peripheral immune function. In addition, the TH-Bio-ELISA provides a novel and sensitive tool to investigate the immune regulatory pathways between brain and peripheral immune system in PD.

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Poster

P253. Dopamine and Non-Dopamine Pathways I

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DA043895

Title: Loss of function in Parkinson's patient-derived dopamine neurons precedes neuronal degeneration

Authors: *H. KHOSHBOUEI;
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Abstract: The pathophysiological mechanisms for degenerative changes in dopamine neurons during early stages of Parkinson's disease (PD) remain unknown. The inaccessibility of living diseased tissue has been a limitation in studying progression of PD pathophysiology prior to degeneration of dopamine neurons. To address these issues, we differentiated induced pluripotent stem cells (iPSCs) from a PD patient carrying the α-synuclein triplication mutation and an unaffected first-degree relative into spontaneously firing dopaminergic neurons. Around 96% of iPSC-derived dopamine neurons expressed DAT, TH, D2, GIRK2 and indicators for neuronal maturation such as Nurr1, FOXA2, PRK8 and MAP2 (n=6 from three independent rounds of differentiation). In 157 iPSC-derived dopamine neurons from three independent rounds of differentiation, we found that human-like dopamine neurons overexpressing α-synuclein exhibited a unique up-state (depolarized plateau) and high frequency of spontaneous firing activity with a pause between subsequent broadbrimmed bursts. Investigating the underlying mechanism revealed that α-synuclein overexpression diminished D2 receptors and GIRK channels autoinhibition. Back-translating these findings into primary mouse neurons overexpressing α-synuclein revealed a similar phenotype and mechanism, supporting the causal role for α-synuclein reduction of autoinhibition in dopamine neurons. Live-cell GCaMP6 calcium imaging revealed α-synuclein overexpression perturbed intracellular calcium homeostasis, increased TH and extracellular dopamine levels along with reducing arborization of dopaminergic neurons and neuronal survival. Importantly, a GIRK channel activator or application of D2 receptor agonist, quinpirole, prevented neuronal loss, restored the firing activity, TH levels and the morphology of PD patient-derived dopaminergic neurons to the quantity measured in dopaminergic neurons from healthy subjects. These results provide novel insights into the pre-degenerative pathophysiology induced by α-synuclein overexpression and introduce a potential mechanism for the long-established clinical efficacy of D2 receptor agonists in the treatment of PD.

Disclosures: H. Khoshbouei: None.

Poster

P253. Dopamine and Non-Dopamine Pathways I

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Topic: C.03. Parkinson's Disease

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DA043895

Title: Dopamine transporter regulation and dysregulation of the macrophage immune response in Parkinson's disease

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Abstract: Nolan, Kaitlyn Runner, Stephanie Matt, Peter J. Gaskill, and Habibeh Khoshbouei Monocyte-derived macrophages are key players in tissue homeostasis and disease regulated by a variety of signaling molecules. Recent literature has highlighted the ability for the biogenic amines dopamine and norepinephrine to regulate macrophage functions, but the mechanisms governing dopamine and norepinephrine signaling on and around immune cells remains nebulous. In the central nervous system, biogenic amine transporters are regarded as the master regulators of neurotransmitter signaling. While we and others have shown macrophages express these transporters, relatively little is known of their function on these cells. To address these knowledge gaps, we interrogated the function of norepinephrine (NET) and dopamine (DAT) transporters on human monocyte-derived macrophages. Using multiple complementary approaches, we found that both NET and DAT are present and can uptake substrate from the extracellular space at baseline. Not only was DAT expressed in cultured macrophages, but it was also detected in a subset of human intestinal macrophages in situ. Surprisingly, we discovered a NET-independent, DAT-mediated immuno-modulatory mechanism in response to lipopolysaccharide (LPS). LPS induced reverse transport of dopamine through DAT, engaging autocrine/paracrine signaling loop that regulated macrophage cytokine release and phagocytic capacity. Pharmacologically inhibiting this signaling loop enhanced the pro-inflammatory response to LPS. Finally, we found that this DAT-immune axis was disrupted in Parkinson's disease macrophages with macrophages from a cohort of PD patients exhibiting dysregulated DAT activity and subsequent DAT de-regulation of the LPS-induced immune response. Collectively, our data introduce a novel role for DAT in the regulation of innate immunity during health and disease.

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Poster

P253. Dopamine and Non-Dopamine Pathways I

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Topic: C.03. Parkinson's Disease

Support: CONACyT. México (scholarship 893764, to JRGP)

Title: Multiunitary activity characterization of Inferior Olive, Dentate Nucleus and Crus II in an acute model of parkinsonism

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Abstract: Ventrolateral striatum injury modifies the activation pattern of the cerebellum, supporting the idea that both structures function as an integrated system (Vásquez, 2019). In contrast, Herrera Meza, 2014 described an expression pattern of IR-Fos, which shows an increase in the hemispheres, specifically in sim B, crus II and dentate nucleus (with emphasis on the last 2 structures) that was interpreted as an increase in multiunit activity (MUA). For this reason, the MUA was recorded in Crus II, Dentate Nucleus and Inferior Olive (IO) was added to complete the cerebellar circuit. The aim of this study was to analyze and characterize the effect on the MUA of cerebellar neurons after damage caused by the insertion of an electrode in the ventrolateral striatum (VLS). Male rats (N=21) of the Wistar strain weighing (250-350 g) were used. Animals were divided into control (n=10) (without VLS lesion) and experimental (n=11) groups (with VLS lesion). Each subject was implanted with 1 to 3 recording electrodes in Crus II, dentate nucleus, and inferior olive. Finally, 10 traces were randomly selected to characterize and compare basal activity and jaw tremor, in this way, jaw tremor traces were analyzed and compared between groups and structures for 4 weeks. Recordings showed an increase in the maximal amplitude in the experimental group compared with the control group. On the other hand, the IO increases its maximal amplitude during the tremor with respect to dentate nucleus and Crus II. During the 4 weeks of recording, the maximal amplitude shows a downward trend.

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Poster

P253. Dopamine and Non-Dopamine Pathways I

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Topic: C.03. Parkinson's Disease

Support: CAPES
CNPq (Grant No: 309567/2019-0)

Title: Involvement of raphe nuclei in depressive-like behavior and short-term memory in an animal model of rotenone-induced Parkinson's disease

Authors: *D. C. RAMOS[1], J. C. F. VIEIRA[1], L. C. SOUZA[1], M. BORTOLANZA[2], R. ANDREATINI[1], M. A. B. F. VITAL[1];
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Abstract: Parkinson's disease (PD) is a neurodegenerative disease characterized by motor symptoms and non-motor disturbances such as fatigue, cognitive impairment, constipation, anxiety and depression. The dopaminergic neurodegeneration is a hallmark of PD, however the noradrenergic and serotonergic systems also present deficiencies. Studies have shown that an impaired serotonergic neurotransmission plays a role in the development of cognition deficits and mood disorders. In the central nervous system, serotonin-containing neurons are located, mainly, in the raphe nuclei, and send projections to several encephalic regions, such as limbic structures, and basal ganglia. In this context, the tryptophan hydroxylase (TPH), the rate-limiting enzyme in serotonin biosynthesis, has the potential to affect several brain functions through the regulation of encephalic serotonin levels in those regions. Thus, we aimed to investigate the involvement of serotonergic neurons in the raphe nuclei in depressive-like behavior and short-term memory. In this study, male Wistar rats were intraperitoneally treated for 10 days with rotenone (2.5 mg/kg) or its vehicle (sunflower oil). Both groups underwent locomotor and behavioral assessments, and we performed immunohistochemical and neurochemical analysis. One day after the last rotenone injection, we observed hypolocomotion in the rotenone-lesioned animals tested in the open field. Conversely, animals recovered the motor function when tested 7, 14, and 21 days afterwards. Furthermore, the systemic administration of rotenone was able to impair the short-term memory assessed in the social recognition test performed 22 days after the last rotenone injection. In the modified swim test, conducted on day 29, we observed an increase in immobility time in the rotenone group, compared to the control. Subsequently the behavioral tests, we performed the immunohistochemical analysis for TPH positive neurons in the raphe nuclei. Our results did not reveal significant differences between the groups regarding the number of TPH-immunoreactive neurons, neither in the dorsal raphe nucleus (rostral, mid, and caudal parts), nor in the median raphe nucleus. In contrast, we observed a significant reduction in serotonin levels in striatum, hippocampus and prefrontal cortex of rotenone-lesioned rats. These data indicate that the nigrostriatal lesion induced by rotenone - revealed by reductions of tyrosine

hydroxylase-positive neurons in substantia nigra compacta and dopamine levels in striatum - did not affect the number of TPH-immunoreactive neurons in raphe nuclei but led to lower serotonin levels and behavioral disturbances.

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Poster

P253. Dopamine and Non-Dopamine Pathways I

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Topic: C.03. Parkinson's Disease

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Swedish Brain Foundation
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Swedish Foundation for Strategic Research
Science for Life Laboratory

Title: Drd1 induced Nr4a2, controls locomotor sensitization to L-DOPA

Authors: *I. MANTAS[1], X. ZHANG[1], E. FRIDJONSDOTTIR[2], M. SAARINEN[1], K. TIKLOVA[1], T. LI[1], P. ANDRÉN[2], T. PERLMANN[1], P. SVENNINGSSON[1];
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Abstract: Nr4a2 is a transcription factor, which is associated with disorders of the dopaminergic system such as Parkinson's disease. However, Nr4a2's function as an immediate early gene is largely unknown. In the present study, we revealed that chronic L-dihydroxyphenylalanine (L-DOPA) administration causes a robust Nr4a2 upregulation in the Drd1 expressing neurons of the dopamine depleted striatum and deep cortical layers. Mice lacking Nr4a2 in Drd1 cells (Drd1-cKO) exhibit profound less locomotor sensitization and slightly worsened dyskinesia scores. The behavioural changes observed in Drd1-cKO mice were accompanied by alterations in dynorphin and enkephalin neuropeptides at both mRNA and protein level. Even though Nr4a2 is well known to control dopamine related genes, we showed that both wild type and Drd1-cKO mice display L-DOPA induced striatal tyrosine hydroxylase mRNA upregulation. Accordingly, we unveiled that striatal Nr4a2 cells compose a distinct group, which is not overlapping with the tyrosine hydroxylase population. Nevertheless, mass spectrometry imaging of metabolites revealed an L-DOPA associated increase of α-glycerophosphocholine in control striata, which was absent in Drd1-cKO mice. Since Nr4a2 has been proposed to control mitochondrial

integrity, these results indicate a possible role of L-DOPA induced Nr4a2 in regulating lipid oxidation. The current study unravels unexplored functions of Nr4a2 in Parkinson's disease

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Poster

P283. Cellular and Molecular Mechanisms I

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Program #/Poster #: P283.01

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

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Title: Charge neutralization of lysines by carbamylation and acetylation creates multiple aggregation hot spots in tau sequence

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Abstract: The impact of various post-translational modifications (PTMs) on tau protein aggregation has been well documented. However, whether these PTMs such as phosphorylation or glycosylation are drivers of aggregation or mere functional modulators is still an open question. As many PTMs are very dynamic in nature and endless combinations exist, perhaps a site-specific context-dependent model makes the most sense. Recent cryo-EM structural analysis of tau fibrils extracted from patients suffering from various tauopathies, including Alzheimer's Disease (AD), Picks Disease, Progressive supranuclear palsy (PSP), and Corticobasal degeneration (CBD) has revealed the presence of unidentified charge densities near lysine residues. As there are 44 lysine residues in full-length tau protein and charge neutralization on any of them can alter the nature of lysine from anti-aggregation to pro-aggregation, lysine modifying charge neutralizing PTMs are back in focus. In this context, carbamylation is one such charge neutralizing non-enzymatic age-dependent PTM, which is very similar to acetylation, an enzymatic dynamic PTM. We looked beyond the core fibril and assessed the aggregation potential of "lysine-rich" regions. Thus formed amyloid fibrillar assemblies were characterized by various biophysical methods, including ThT fluorescence enhancement, CD spectroscopy, congo red staining, fluorescence microscopy, and SEM. We observed that lysine carbamylation could convert previously unsuspecting sites in the flanking region to aggregation hotspots. We also demonstrate that this stickiness is strong enough to overcome the resistance from neighboring amino acids such as Proline and arginine. We further compared the aggregation propensity of carbamylated sequences with acetylated ones and observed that carbamylation is a

much stronger driver for aggregation. Atomic-level MD simulation studies further supported this. We have also looked at the impact of carbamylation and acetylation on the stability of AD core-derived peptides and determined that the β8 fragment that aligns opposite to the notorious β1 (VQIVYK) fragment aggregates if acetylated or carbamylated. When we ran the MD simulation, core fibrils unfold unless lysines on β8 are carbamylated or acetylated. Other core fragments were also impacted by modification but didn't seem to affect the stability much. Overall our results suggest that while carbamylation and acetylation could be significant drivers of tau aggregation in general, carbamylation on the non-core fibril region could further accelerate the aggregation process by generating additional aggregation hotspots.

Disclosures: S. Gupta: None. **J. Gadhavi:** None. **S. Shah:** None. **A. Jain:** None.

Poster

P283. Cellular and Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P283.02

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Title: The Potential Impact of Zero Gravity on Amyloid Aggregation

Authors: S. AMSELEM[1], N. SZENKIER[2], A. NAVON[2], A. GROSS[2], L. STEINMAN[3], J. ROTHBARD[4], ***M. V. KARPUJ**[5];
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Abstract: Neurodegenerative diseases, ranging from Alzheimer's dementia to Parkinson's Disease pose a formidable challenge to medical science, as well as a burden on those who are afflicted and their families. The societal costs are enormous. Protein aggregation is a hallmark of these diseases. Various environmental factors such as temperature, and pH affect amyloid formation. However very little is known regarding the impact of gravity on these aggregates. Here, we demonstrate the effect of microgravity on amyloid formation in the presence and absence of some potential anti-amyloidogenic drugs. The microgravity disruptive environment allows for additional biophysical screening. The opportunity to study drugs and cells at microgravity permits exploration of chemical and biological properties that might not be present at usual conditions. Such explorations under extreme conditions is an elegant tool for drug discovery.

Disclosures: S. amselem: None. **N. Szenkier:** None. **A. Navon:** None. **A. Gross:** None. **L. Steinman:** None. **J. Rothbard:** None. **M.V. Karpuj:** None.

Poster

P283. Cellular and Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P283.03

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Title: Somatic mutations in PRNP identified in patients with prion diseases using Nanopore sequencing

Authors: *A. DIMITRIADIS, F. KROLL, T. CAMPBELL, L. DARWENT, J. COLLINGE, S. MEAD, E. VIRE;
MRC Prion Unit at UCL, London, United Kingdom

Abstract: Accurate and rapid sequencing of the prion gene PRNP is paramount to human prion disease diagnosis and for animal surveillance programmes. Prion diseases are fatal neurodegenerative conditions that affect humans and animals. Current methods for PRNP sequencing are based on Sanger or next-generation sequencing of relatively small fragments within the coding region of the gene. Yet, the contribution of variants in the non-coding regions of PRNP is poorly understood. Here we combined long-range PCR and nanopore sequencing to sequence the full-length PRNP in human blood (n=21) and brain (n=5). In genetic cases of prion diseases, we show that variants in PRNP are picked up as efficiently by nanopore as they are by Sanger. In sporadic cases of prion disease, nanopore also identified novel variants but none of them with highly pathogenic potential. Finally, we used our method to explore somatic mosaicism of the PRNP octapeptide repeat region in inherited cases of prion diseases. Here we uncover, for the first time, somatic events in the prion gene detected a rate ranging from 0 to 0.28% of the reads per individual. These somatic mutations are more frequent in inherited prion diseases, suggesting that genomic instability of PRNP provide fertile grounds for more genetic mutations. Our study highlights the power of long-read sequencing for prion disease diagnosis and monitoring.

Disclosures: A. Dimitriadis: None. **F. Kroll:** None. **T. Campbell:** None. **L. Darwent:** None. **J. Collinge:** None. **S. Mead:** None. **E. Vire:** None.

Poster

P283. Cellular and Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P283.04

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Support: 1 RF1 Ag058778

Title: "Investigating the mechanisms responsible for turnover of the ER stress sensing protein PERK"

Authors: *H. GUDLAVALLETI[1], B. J. STOVEKEN[2], R. VISWANTHAN[1], S. POLUSANI[3], J. D. LECHLEITER[1];
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Abstract: PKR like endoplasmic reticulum (ER) Kinase (PERK) is an early ER stress sensor. It is a transmembrane protein that activates the unfolded protein response (UPR), first by luminal-dependent dimerization, followed by autophosphorylation of the cytosol domains and subsequent phosphorylation eIF2α. This early stress response reduces elF2α-mediated translation in cells when there is an accumulation of unfolded proteins to help restore homeostasis. PERK variants associated with tauopathies have been identified that show increased turnover rates and decreased kinase activity. However, the underlying mechanisms of PERK turnover have not been clearly resolved, which could help explain how PERK variants increase the risk of developing neuropathology. Preliminary studies indicate that pharmacological inhibition of autophagy with the drug chloroquine increases detectable levels of PERK using western blot analysis. Early estimates suggest chloroquine treatments (50uM) of cultured mouse embryonic fibroblasts (MEFs) increase PERK levels by 8-fold over a 28-hour period. We are also testing the role of proteasomal inhibition on PERK turnover. Thus far, treatment of cultured MEFs with the proteasomal inhibitor MG-132 (25uM), does not appear to significantly affect total PERK levels at similar timepoints. Experiments are currently underway to use photoconvertible fluorescent proteins (PCFPs) fused to PERK. These studies will permit us to monitor protein turnover by imaging changes in converted fluorescent protein (green to red in response to near-UV irradiation). Following the total fluorescence of converted PCFPs is the experimental equivalent of measuring radioactivity in pulse chase methodology. The primary advantages of PCFP measurements are both total levels and the spatial location of PCFP-PERK can be monitored in live cells. Preliminary measurements indicated significant loss of PCFP-PERK, 24 hours after photoconversion. We plan to investigate the impact of various PERK mutations on turnover. Co-localization studies of PCFP-PERK with lysosomal markers are also planned to determine if PERK is degraded through a lysosomal or ER - Phagy pathway. Data generated from these studies will lead to a better understanding of PERK turnover, which in turn, may lead to new pharmacological targets for treating neurodegenerative disorders associated with PERK.

Disclosures: H. Gudlavalleti: None. **B.J. Stoveken:** None. **R. Viswanthan:** None. **S. Polusani:** None. **J.D. Lechleiter:** None.

Poster

P283. Cellular and Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P283.05

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Title: Postherpetic neuralgia - a new amyloid disease?

Authors: *A. BUBAK[1], M. NAGEL[2];
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Abstract: Postherpetic neuralgia (PHN) is defined as dermatomal-distribution pain that persists > 90 days after herpes zoster (HZ). The risk of PHN increases with age with up to 40% of HZ individuals over 65 years of age developing PHN. PHN is difficult to manage, significantly impacts activities of daily living and quality of life, and is the leading cause of suicide in older adults with chronic pain. The cause has been attributed to a chronic VZV ganglionitis and/or ganglionic damage leading to neuronal dysfunction but exact pathophysiological mechanisms are not well-characterized. Recent studies from our laboratory found that VZV infection of spinal astrocytes *in vitro* induced intracellular amylin and Ab42 expression, intracellular amyloid, and an amyloidogenic extracellular environment, likely in part through self-aggregating and catalytic VZV glycoprotein B peptides. Further support for VZV induction of an “amyloidogenic state” is provided by studies of: (1) plasma from acute herpes zoster individuals that contain increased amyloid and promotes aggregation of cellular amyloidogenic peptides; and (2) cerebrospinal fluid from VZV vasculopathy patients that similarly contain increased amyloid and an amyloidogenic environment. Because amyloid deposition on neurites tags them for complement-mediated microglial pruning, we hypothesized that VZV infection of sensory ganglionic neurons leads to amyloid deposition on neurites leading to neuronal degeneration. To test this hypothesis, we infected human sensory ganglionic neuronal cells (HD10.6s) with VZV, conducted targeted RNA sequencing, then confirmed findings by immunocytochemistry (IHC). Compared to mock-infected cells, VZV-infected cells had significant enrichment of gene sets associated with amyloidosis and had predicted upstream regulators associated with the amyloid-associated disease, Alzheimer's disease (APP, PSEN1, and Tau); upregulation of complement pathways and synaptic pruning were also seen. IHC revealed striking amylin aggregates (amyloid) along VZV-infected neurites that also co-localized with the post-synaptic density marker PSD93. Taken together, VZV-infection of sensory neurons produces amylin aggregates on neurites that can potentially induce microglial pruning and neurodegeneration. In combination with host and genetic factors, this may explain the development of PHN in a subset of HZ patients.

Disclosures: A. Bubak: None. **M. Nagel:** None.

Poster

P283. Cellular and Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

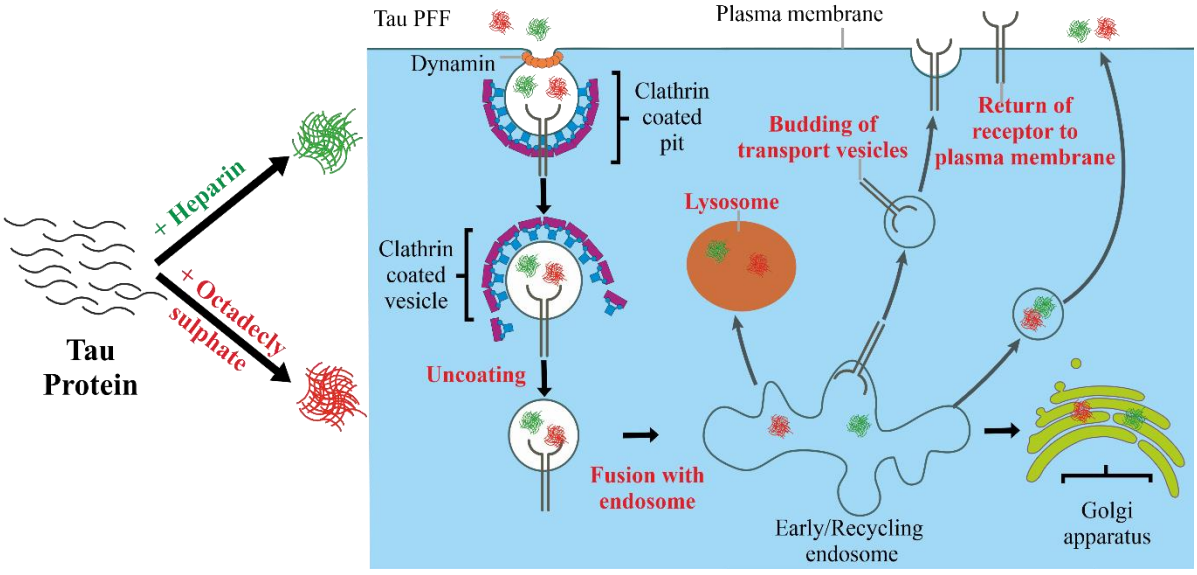
Program #/Poster #: P283.06

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Title: Heparin and octadecyl sulphate induced tau fibrils follow clathrin-mediated endocytosis in SH-SY5Y-derived differentiated neurons

Authors: *P. HIVARE, G. SWARUP, A. BANSAL, D. BHATIA, S. GUPTA;
Indian Inst. of Technol. Gandhinagar, Gandhinagar, India

Abstract: Tau is a microtubule-associated protein that is intrinsically disordered in nature and associated with more than 20 tauopathies in progressive neuropathological disorders (NDs). The intracellular proteinaceous aggregates of misfolded tau protein form the paired helical filaments (PHFs) and neurofibrillary tangles (NFTs). The aggregated tau species spread across neurons via transcellular propagation, and the mechanism of spreading remains unclear. The heparin-induced tau filaments differ structurally and do not reproduce the key structural features as found in Alzheimer's disease (AD) filaments raising questions about the use of *in vitro* tau assembly. Here in this study, we have prepared tau fibrils using heparin and octadecyl sulphate and studied the cellular internalization pathway in SH-SY5Y neuroblastoma cells and differentiated neurons. We studied the tau fibrils internalization using multiple cell-based approaches such as cellular uptake and recycling kinetics, standard endocytic probes, and protein colocalization. Transferrin is used to mark Clathrin-mediated endocytosis (CME), while Galectin-3 mark Clathrin-independent endocytosis (CIE). We further blocked the specific endocytosis pathway with inhibitors and found that tau fibrils follow the CMEpathway. The internalization pathway of tau fibrils formed by heparin and octadecyl sulphate will help understand the spread of tauopathies and Alzheimer's disease.1



Disclosures: P. Hivare: None. **G. Swarup:** None. **A. Bansal:** None. **D. Bhatia:** None. **S. Gupta:** None.

Poster

P283. Cellular and Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P283.07

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Support: NIH K99/R00 AG068359

Title: Creutzfeldt-Jakob disease with distinct PrP plaque phenotypes and PrP type 1 variants

Authors: R. BAYAZID, R. ASLAM, *I. CALI;
Pathology, Case Western Reserve Univ., Cleveland, OH

Abstract: Sporadic Creutzfeldt-Jakob disease (sCJD) comprises five distinct subtypes, which are identified by two major molecular modifiers of disease phenotype: (i) gel mobility of the pathogenic prion protein (PrP^D), commonly identified as PrP^D type 1 (T1) and type 2 (T2), and (ii) the polymorphic genotype (methionine (M)/valine (V)) at codon 129 of the PrP gene (129MM, -MV or -VV). The five sCJD subtypes are: MM(MV)1, VV1, MM2, MV2 and VV2 (Parchi et al., Ann Neurol 1999). Sporadic CJDMM1, the most common human prion disease, is characterized by T1 protease-resistant PrP^D and disease phenotype with fine spongiform degeneration (SD). The sCJDMM2 subtype harbors T2 PrP^D and large vacuoles SD whereas -MM1-2 shows mixed -MM1 and -MM2 histopathological features that correlate typically with the proportion of co-existing T1 and T2 PrP^D (Cali et al, Brain 2009). Furthermore, all sCJD cases with 129MM genotype (sCJDMM) are free of PrP amyloid plaques. Recently, sCJDMM1 with PrP^D kuru-plaque pathology has been described (Rossi et al., Acta Neuropathol Communic 2017). Here, we retrospectively examined 523 sCJDMM cases – referred to the National Prion Disease Pathology Surveillance Center (NPDPSC) – comprising 366 sCJDMM1, 48 -MM2 and 109 -MM1-2 cases. PrP^D plaques (p) were detected in 14 cases (~3%) populating either the white matter (p^{WM}-sCJD, n=7) of virtually all brain regions examined or the grey matter (p^{GM}-sCJD, n=7) of the cerebellar cortex. The two phenotypes correlated with distinct T1 gel mobilities: T1 migrated to ~20 kDa ($T1^{20}$) in p^{GM}-sCJD or to ~21 kDa ($T1^{21}$) in p^{WM}-sCJD. Furthermore, $T1^{20}$ and $T1^{21}$ showed distinct molecular features and brain distribution, indicating that T1 variants may be different human prion strains. These findings highlight the presence of PrP^D amyloid plaques in subjects with sCJDMM and suggest that $T1^{20}$ and $T1^{21}$ PrP^D are modifiers of histopathological phenotype.

Disclosures: R. Bayazid: None. **R. Aslam:** None. **I. Cali:** None.

Poster

P283. Cellular and Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P283.08

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Title: The outer brain

Authors: *R. A. NORMAN;
Robert A. Norman, D.O., P.A., Tampa, FL

Abstract: Much of my interest and research over the last decade has been on the connection between the skin and the brain, including searching for skin biomarkers to detect Alzheimer's disease (AD) and the connection between Parkinson's disease (PD) and melanoma. Parkinson's disease is a neurodegenerative disorder that triggers death of dopaminergic neurons within the brain's substantia nigra. Melanocytes are the pigmented cells that give rise to skin, hair and eye color, and melanoma is a potentially deadly cancer arising from melanocytes. As noted in a recent article by Dexter N. Dean, PhD, and Jennifer C. Lee, PhD, "Although these two diseases fundamentally differ, with PD leading to cell degeneration and melanoma leading to cell proliferation, epidemiological evidence has revealed a reciprocal relationship where patients with PD are more susceptible to melanoma and patients with melanoma are more susceptible to PD." Where is the hypothesis here? Let's look into the key pathology of each disease. Intracellular inclusions primary composed of proteinaceous α-synuclein (α-syn) amyloid fibrils, is the hallmark of PD. The α-Syn also has been detected in cultured melanoma cells and tissues derived from patients with melanoma. As Dean and Lee point out, an inverse correlation exists between α-syn expression and pigmentation and this has led to the prevailing hypothesis that α-syn inhibits enzymes involved in melanin biosynthesis. The two researchers recently reported an alternative hypothesis in which α-syn interacts with and modulates the aggregation of Pmel17, a functional amyloid that serves as a scaffold for melanin biosynthesis. In addition, α-syn may disrupt melanogenesis in melanoma cells by a disruption of TH and TYR enzymes involved in melanin biosynthesis. The hypothesis here is that uncovering the behavior of α-syn in melanoma could both provide novel approaches for treating melanoma and insights into the role of α-syn in PD.Although Tau protein has previously been assumed to be exclusive to the central nervous system, it is also found in peripheral tissues. My colleagues and I reported our research to determine whether there is a differential Tau expression in oral mucosa cells according to cognitive impairment.RT-QuIC and PMCA assays for formalin-fixed and paraffin-embedded skin tissues have also been utilized.The poster will provide a clear overview of the data in the abstract.

Disclosures: R.A. Norman: None.

Poster

P283. Cellular and Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P283.09

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Support: Department of Defense W81XWH2110189
National institute of aging R21AG068597
Lisa Dean Moseley foundation
NIH R01NS115977

Title: Accelerated accumulation of mature tau isoforms in Tau P301S-hiPSC-derived cerebral organoids instigates microtubule defects and early phases of axonal degeneration

Authors: *X. SUN[1], P. CHANG[1], C. M. KARCH[2], P. W. BAAS[1], L. QIANG[1];
[1]Drexel Univ. Col. of Med., Philadelphia, PA; [2]Psychiatry, Washington Univ. In St Louis, Saint Louis, MO

Abstract: Tauopathies are hallmarked by accumulation of pathological tau species derived from either MAPT mutations or other disease pathways. Tau, which is encoded by MAPT, is one of the most abundant microtubule (MT) associated proteins in the axon, but surprisingly, little is known about how tau impacts the properties of axonal MTs or how axonal MTs are affected by pathological changes in tau. Because pathological tau binds less avidly to them, conventional wisdom has been that MTs respond by becoming less rich in tau, which in turn results in a reduction in their levels and stability. However, recent studies have called into question the dogma of tau as a MT stabilizer, suggesting that tau's true role in this regard is to maintain portions of labile MTs. In addition, tau pathology is complicated by the existence of both mature and immature isoforms. To elucidate changes in axonal MTs resulting from pathological tau (separate from the contribution of non-tau confounders in diseases such as Alzheimer's), we used forebrain organoids derived from human induced pluripotent stem cells (hiPSCs) bearing the tauP301S mutation found in patients with Frontotemporal Dementia. A significant increase in the levels of 4R-tau and 1N-tau was observed in 2.5-month tauP301S-organoids, but not 3R and 0N tau isoforms, compared to the isogenic controls. Such phenotypes, further characterized by greater phosphorylation at the isotopes recognized by AT8 and TNT2 but not TOC1, were not identified in 2-dimensional differentiated glutamatergic neurons cultured for the same period. An early axonal degeneration phenotype in the tauP301S-organoids was documented via multifaceted analyses using amino-cupric silver stain, immunohistochemistry via SMI32, axonal swelling revealed by bIII tubulin and neuronal activity recording via multielectrode array. Other studies revealed that the increase in tau levels is accompanied by a decrease in MT stability as well as a decrease in the levels of MAP6, which is a genuine stabilizer of MTs. These results are

consistent with the view that tau keeps MTs from being stabilized (rather than the inverse) such that a significant rise in tau levels early in the pathology overcomes the reduced binding affinity of the tau to the microtubules. As for a toxic gain-of-function effect of the pathological tau, an uptick in mal-oriented MTs in the axons of the tauP301S-organoids was corrected by a drug that increases MT mobility, and this in turn rescued the aberrant axonal swelling in these neurons. Overall, our findings suggest that accelerated accumulation of mature tau isoforms instigates MT defects that accompany (and may underlie) early phases of axonal degeneration.

Disclosures: X. Sun: None. **P. Chang:** None. **C.M. Karch:** None. **P.W. Baas:** None. **L. Qiang:** None.

Poster

P283. Cellular and Molecular Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P283.10

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Support: K08 AG065426/AG/NIA NIH HHS/United States
RF1 AG055474/AG/NIA NIH HHS/United States
P30 AG066509/AG/NIA NIH HHS/United States
R01NS064131/NS/NINDS NIH HHS/United States

Title: Pathological tau drives the cytoplasmic aggregation of nuclear speckle scaffold protein SRRM2 in Alzheimer's disease neurons and models

Authors: *R. ECK[1], P. J. MCMILLAN[2], T. J. STROVAS[4], M. BAUM[4], B. K. MITCHELL[4], N. HENDRICKS[4], J. M. WHEELER[4], C. S. LATIMER[3], C. D. KEENE[4], B. C. KRAEMER[4]; [1]Univ. of Washington Grad. Program In Neurosci., Seattle, WA; [2]Dept Psychiatry & Behavioral Sci., Univ. of Washington, Seattle, WA; [3]Univ. of Washington, Department of Laboratory Medicine and Pathology, WA; [4]Geriatrics Res. Educ. and Clin. Ctr., Veterans Affairs Puget Sound Hlth. Care Syst., Seattle, WA

Abstract: Tau aggregation and toxicity requires several nuclear and well-conserved RNA binding proteins (*sut-1*, *sut-2*, *parn-2*, *sut-12*) in *C. elegans*, mice, and human cells. The mammalian homology of SUT-2 (MSUT2) normally resides in nuclear speckles (NS), membranelles organelles composed of phase-separated RNAs and RNA binding proteins that regulate transcription, mRNA processing, and splicing. Given the role of NS in RNA metabolism and the association of MSUT2 and other suppressors of tauopathy with NS, we used human pathological tissue and transgenic mice to identify any Alzheimer's disease-specific changes to NS. We observed that NS constituent scaffold protein SRRM2 is mislocalized and accumulates in cytoplasmic lesions in AD brain tissue. These SRRM2+ lesions significantly colocalize with

pathological tau. Furthermore, progression of tauopathy in transgenic mice is accompanied by increasing mislocalization of SRRM2 from the neuronal nucleus to the cytoplasm. In AD brain tissue, SRRM2 mislocalization is also associated with increased severity of pathological tau deposition. In *C. elegans,* we further explored the molecular mechanisms driving ectopic localization of RSR-2, the *C. elegans* homolog of SRRM2, and the resulting cellular consequences for neurodegeneration. Our findings suggest potential mechanisms by which pathological tau impacts nuclear speckle function in diverse organisms ranging from *C. elegans* to mice to humans. Future translational studies aimed at restoring nuclear speckle homeostasis may provide novel candidate therapeutic targets for pharmacological intervention.

Disclosures: R. Eck: None. **P.J. McMillan:** None. **T.J. Strovas:** None. **M. Baum:** None. **B.K. Mitchell:** None. **N. Hendricks:** None. **J.M. Wheeler:** None. **C.S. Latimer:** None. **C.D. Keene:** None. **B.C. Kraemer:** None.

Poster

P309. Neuroprotection and Therapy

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P309.01

Topic: B.01. Neurotransmitters, Transporters, and Signaling Molecules

Support: JSPS KAKENHI Grant 18K19720

Title: Pharmacological inhibition of histone deacetylases (HDACs) presents an enriched neuronal platform in the brain for neurorehabilitation

Authors: *H. MAEJIMA[1], M. KITAHARA[2], Y. TAKAMATSU[1], H. MANI[1,3], T. INOUE[2,4]; [1]Dept. of Rehabil. Science, Fac. of Hlth. Sci., [2]Grad. Sch. of Hlth. Sci., Hokkaido Univ., Sapporo, Japan; [3]Oita Univ., Oita, Japan; [4]JSPS, Tokyo, Japan

Abstract: Therepeutic exercse is a primary intervention to rehabilitate the motor impairment of patients with central nervous system (CNS) disorders. Specifically, aerobic exercise increases the expression of neurotrophins including brain-derived neurotrophic factor (BDNF) in the brain and contributes beneficially to the improvement of motor impairment. Histone deacetylases (HDACs) inhibitors acetylate histones and enhance gene transcriptions. In the present study, we examined the interactive effects of pharmacological treatment with an HDAC inhibitor and aerobic exercise on the modifications of epigenetic markers including HDAC activity and histone acetylation levels and neurotrophic gene expressions including neurotrophins and immediate-early genes (IEGs) in the motor cortex. The objective of this study was to find a more enriched neuronal platform in the brain for neuronal rehabilitation. ICR mice, 13-week-old, were divided into four groups based on two factors: HDAC inhibition and exercise. Intraperitoneal administration of an HDAC inhibitor (1.2 g/kg sodium butyrate, NaB) and moderate exercise at

10 m/min for 60 min were conducted five days a week for four weeks. NaB administration inhibited total HDAC activity and increased acetylation level of histone H4. NaB administration also increased the transcription levels of IEGs including c-fos and Arc, and neurotrophins including BDNF and NT-4 in the motor cortex. Moderate treadmill exercise enhanced HDAC activity and acetylated histone H4 and H3, whereas exercise did not affect transcription levels of IEGs and neurotrophins. In addition, there were no significant synergic effects between HDAC inhibition and the exercise regime on the gene expressions. Altogether, this study showed that daily HDAC inhibition could present more enriched condition for neuroplasticity to the motor cortex, suggesting that pharmacological treatment with HDAC inhibitor could precondition neuronal platform for neurorehabilitation in patients with CNS disorders.

Disclosures: H. Maejima: None. **M. Kitahara:** None. **Y. Takamatsu:** None. **H. Mani:** None. **T. Inoue:** None.

Poster

P309. Neuroprotection and Therapy

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P309.02

Topic: B.01. Neurotransmitters, Transporters, and Signaling Molecules

Title: The effects of exercise on GDNF and estrogen concentration in male and female rats

Authors: *J. M. VANGYSEGHEM, J. M. SPITSBERGEN;
Western Michigan Univ., Kalamazoo, MI

Abstract: Males and females differ hormonally and neurologically as they age yet are given the same treatment for many neurological diseases. As aging occurs there is a loss of motor neurons, which could be explained by the reduced neurotrophic factor concentration. A possible way to maintain neuroprotection would be the production and release of a target-derived neurotrophic factor, such as glial cell line-derived neurotrophic factor (GDNF). Previous studies have shown that GDNF concentration has increased in skeletal muscle after exercise. However, this study has only been done in male rats. While exercise has been shown to mitigate the rate of development of degenerative loss of skeletal muscle mass, quality and strength called sarcopenia it has additionally been shown to increase estrogen receptors on skeletal muscle and increase levels of GDNF in skeletal muscle. Furthermore, it has been shown that GDNF and estrogen signal through similar intracellular pathway, and through such pathways, estrogen can enhance intracellular GDNF signaling. Our hypothesis is that GDNF concentration will be lower in sedentary rats when compared to exercised. In order to gain a more complete picture of what is happening neurologically as we age, I will be looking at female rats and comparing them to male rats of the same age. Hindlimb skeletal muscle was taken from Sprague-Dawley rats, from both sedentary and voluntarily exercised males and females. Their ages ranged from 4 weeks to 18

months. Sedentary muscle was taken at 4 weeks, 6 weeks, 8 weeks, 12 weeks, 12 months and 18 months. Exercised muscle was harvested from 4 week old rats that had access to running wheels for 2 weeks, from, 8 week old rats exercised for 4 weeks, and from 12 month old rats who voluntarily exercised for 6 months. Western blot and ELISAs were used to measure GDNF concentration, while immunohistochemistry was used to visualize the motor neuron, end plates, and GDNF. There were differences in GDNF concentration between male and females as they aged and if they were sedentary vs. exercised. Motor end plates showed more complexity and larger surface area from skeletal muscle in exercised rats as compared to sedentary. Future studies will investigate if estrogen concentration changes in the same trend in these groups as GDNF concentrations. Understanding the role that neurotrophic factors play in neuroprotection as we age and exercise between different sexes, may help develop novel pharmacological treatments and could impact our healthcare system, as we can think about it in a more nuanced way.

Disclosures: J.M. Vangyseghem: None. **J.M. Spitsbergen:** None.

Poster

P309. Neuroprotection and Therapy

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P309.03

Topic: B.01. Neurotransmitters, Transporters, and Signaling Molecules

Support: NIMH 2T32MH067564
NIA 5R01AG054429

Title: Primary Cilia Regulate Hippocampal Bone Morphogenic Protein Signaling

Authors: *S. R. DUNLOP, C.-Y. PENG, J. A. KESSLER;
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Abstract: In the adult hippocampus, exposure to exercise or environmental enrichment (E/EE) attenuates bone morphogenetic protein (BMP) signaling and increases expression of the BMP inhibitor noggin. Additionally, E/EE has been shown to increase neurogenesis and enhance cognitive performance. Reducing BMP signaling in adult mice by ventricular infusion, transgenic overexpression, or viral overexpression of noggin reproduces many of the effects of exercise on both neurogenesis and behavior. Immuno-cytochemical and in-situ hybridization analyses indicate that noggin is expressed by dentate granule neurons, but the subcellular localization of the protein and the mechanisms regulating its synthesis and release are unknown. To address these questions, we generated primary hippocampal cultures from microdissected dentate gyri of postnatal day 3 (P3) C57B6 mice. Noggin protein expression was assessed by immunoblot, mRNA transcript expression via RT-qPCR, and extracellular release via sandwich

ELISA. Our results demonstrate that under normal conditions noggin is localized to primary cilia, immunolabeling in-vivo confirms this localization. However, under depolarizing conditions (20 mM KCl) noggin is redistributed in the cytoplasm and released extracellularly into the culture medium. Further, disruption of primary cilia leads to bulk release of noggin and depletion of intracellular stores of the protein. While TGF-beta signaling has been previously described in primary cilia, this is the first evidence of noggin localization in primary cilia and evidence of noggin protein regulation. Our observations indicate that ciliary integrity is required for noggin homeostasis and that primary cilia appear to be involved in noggin extracellular release.

Disclosures: S.R. Dunlop: None. **C. Peng:** A. Employment/Salary (full or part-time):; Northwestern University. **J.A. Kessler:** A. Employment/Salary (full or part-time):; Northwestern University. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Northwestern University. F. Consulting Fees (e.g., advisory boards); Northwestern University.

Poster

P309. Neuroprotection and Therapy

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P309.04

Topic: B.09. Network interactions

Support: NIH Grant NS093057

Title: Optogenetic re-activation of cortico-thalamic circuits for stroke recovery

Authors: *Z. CAO[1], T. C. CHIANG[2], A. KIM[1], M. Y. CHENG[2], G. K. STEINBERG[1]; [2]Neurosurg., [1]Stanford Univ., Stanford, CA

Abstract: Introduction: A stroke injury can severely disrupt neuronal activity in local and remotely connected regions, leading to network-wide deficits. In particular, cortical stroke disrupts neuronal activity in the cortico-thalamic circuit, resulting in the development of secondary degenerative injury in the remote thalamus. This secondary thalamic degeneration is associated with worsened functional outcomes. In this study, we use optogenetic strategy to selectively restore neuronal activity in the cortico-thalamic circuit and examine its effects on the thalamic injury and functional outcome after stroke. **Methods**: Optogenetic selective cortico-thalamic circuit re-activation was achieved by injecting AAV1-CaMKIIa-hChR2 virus in the ipsilesional somatosensory cortex (iS1) and implanting in the ipsilesional thalamus (iTH) (C57BL6 male mice, 6-8 weeks old). Cortical ischemic stroke was generated by permanent occlusion of the left middle cerebral artery. Stimulated stroke mice received daily stimulations between Post-stroke Day (PD) 4 to 14. Long-term functional outcome was evaluated by

ErasmusLadder tests at pre-stroke baseline, PD3, 7, 14 and 28. Mice were sacrificed at PD29 and immunostaining was used to detect primary cortical injury and secondary thalamic injury (NeuN), neuroinflammation (CD68), and oligodendrogenesis (Olig2 and CC1) in the thalamus. **Results**: Histological staining confirmed expression of ChR2 in the somatosensory cortico-thalamic tract. Stimulated group showed similar performance on the total H-H step and right side of the H-H step, but a trend of less total jump step compared to the non-stimulated group. We did not observe significant difference in the primary and secondary injuries, and neuroinflammation between the two groups. Interestingly, stimulated mice exhibited an increase in the number of mature oligodendrocyte ($Olig2^{+}CC1^{+}$) and oligodendrocyte progenitor cells ($Olig2^{+}$) in iTH. **Conclusion**: As the secondary thalamic injury allows an extended treatment window, targeting secondary injury is a promising approach to improve stroke recovery. Our preliminary results show that restoring neuronal activity in the cortical-thalamic circuits may promote functional recovery and oligodendrogenesis after stroke. Current studies are investigating the effects of cortico-thalamic circuit stimulation in oligodendrogenesis in the degenerative thalamus after stroke.

Disclosures: Z. Cao: None. **T.C. Chiang:** None. **A. Kim:** None. **M.Y. Cheng:** None. **G.K. Steinberg:** None.

Poster

P309. Neuroprotection and Therapy

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Program #/Poster #: P309.05

Topic: B.09. Network interactions

Support: K08NS114170

Title: Selective inhibitory circuit dysfunction after chronic frontal lobe contusion.

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Abstract: Traumatic brain injury (TBI) is a leading cause of neurologic disability and the most common associated cognitive deficits affect prefrontal cortex (PFC)-dependent functions such as: attention, working memory, social behavior, and mental flexibility. Despite this prevalence, little is known about the pathophysiology that develops in frontal cortical microcircuits after TBI. In a mouse model of frontal lobe contusion that recapitulates aberrant mental flexibility as measured by deficits in rule reversal learning, we investigated if alterations in subtype-specific inhibitory circuits are associated with cognitive inflexibility after TBI. Patch clamp recordings

and optogenetic stimulation were performed in layer V two months after injury in the orbitofrontal cortex (OFC). Selective vulnerability in the non-fast spiking, somatostatin-expressing subtype of inhibitory neurons was identified in chronic TBI. Specifically, these neurons exhibited a reduction in excitability and a decrease in their associated synaptic input onto pyramidal neurons. In contrast, fast spiking, parvalbumin-expressing neurons did not show changes in excitability or PV-specific synaptic input onto pyramidal neurons. SOM-mediated disinhibition with TBI was not simply action-potential dependent. Synapse specific properties including the amplitude and frequency of SOM-mediated quantal IPSCs were diminished with TBI as well. This selective vulnerability of SOM+ neuronal function identified in the OFC with chronic TBI was also associated with a hyperexcitable network in 0 mm Mg^{2+}/ 5mM K^{+}. These findings support targeting of subtype specific inhibition in therapeutic development for cognitive dysfunction in TBI.

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Poster

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Topic: B.09. Network interactions

Support: NSF Grant NSF-1854158
Alvin and Marion Birnschein Foundation

Title: Hidden Markov Modelling of altered cortical networks during reach in people with Multiple Sclerosis and intention tremor

Authors: *J. C. WAGNER[1,2], G. BALLARDINI[2], G. BOMMARITO[3], M. INGLESE[4,3], M. CASADIO[2], A. CANESSA[2], R. A. SCHEIDT[1], S. A. BEARDSLEY[1];
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Abstract: Up to 58% of Persons with Multiple Sclerosis (PwMS) live with tremor that impacts daily activities like reaching for items. These brain networks can be identified with Electroencephalography (EEG). Recently, Hidden Markov Modeling (HMM) has been used with EEG to identify brain network state changes during tasks. We used HMM to identify how cortical networks engaged in visually guided reaching are altered in MS.Ten Healthy Controls (HC) (7 male, 24-60 years) and 7 PwMS (5 male, 33-58 years) performed a reach and hold task using a 1-D passive wrist robot while 64-channel EEG was recorded. Subjects were to track a user-controlled cursor over a target that pseudorandomly moved to a new position (104 movements total). Kinematic measures such as Reaction Time (RT) and Target Acquisition Time

(TAT) were quantified from wrist position traces. After typical EEG preprocessing and source reconstruction, regions of interests were defined for each subject within visual-parietal-sensorimotor regions of both hemispheres. EEG signals ([-1 2] seconds about target movement) were used in the HMM to identify sequential brain states involved in the task. Timing of peak state probability and state transitions were estimated.On average, RTs were longer in PwMS than HC (PwMS: 485 ± 134 ms; HC: 387 ± 40 ms) as were TATs (PwMS: 327 ± 141 ms; HC: 193 ± 38 ms). Four brain states were identified for HC. State 1 ([-1 0.36] s), localized in the occipital lobe, coincided with attention to the trial. State 2 ([0.25 0.59] s), occurred in contralateral somatosensory, motor, and premotor cortices. Peak probability coincided with RT suggesting a role in movement planning and execution. State 3 ([0.51 1.24] s), occurred in visuomotor cortices, and captured the final stages of reach and stabilization. Cortical networks returned to baseline in State 4 ([0.97 2] s). Although the timing of states was similar between HC and PwMS, the brain networks present in states 2 and 3 were different. PwMS with tremor showed greater network variability and a shift in state dynamics towards bilateral brain activation. In contrast to HCs, occipital and parietal activity increased in state 2 associated with target tracking and motor planning. Importantly, PwMS RT occurred later, in the transition between states 2 and 3. Differences in brain network states in PwMS with tremor suggest disruptions in target detection and motor planning which delayed downstream processing of visually guided movement. Additionally, the shift to bilateral brain activation implies engagement of compensatory brain networks. Understanding how MS alters brain network states will facilitate development of targeted therapies to mitigate tremor.

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Poster

P309. Neuroprotection and Therapy

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Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P309.07

Topic: I.04. Physiological Methods

Title: Electrophysiological effects of taVNS on the basal cholinergic system, intracortical GABA-A-ergic system, and cerebellar inhibitory Purkinje-cell system: a TMS study

Authors: *V. M. VAN MIDDEN[1], Z. PIRTOSEK[1], M. ZIVIN[2], M. KOJOVIC[1]; [1]Neurol., Univ. Med. Centre, Ljubljana, Ljubljana, Slovenia; [2]Inst. of Pathophysiology, Ljubljana, Slovenia

Abstract: Introduction: Transcutaneous auricular vagus nerve stimulation (taVNS) is an emerging treatment option for various neurological and psychiatric diseases. Functional magnetic resonance imaging studies show increased activity of nucleus tractus solitarii, locus coeruleus, and raphe nuclei during taVNS, suggesting that taVNS may affect diffuse neuromodulatory networks. Yet, it remains unknown whether these changes translate to changes in downstream neuromodulator and neurotransmitter activity. We used different transcranial magnetic stimulation (TMS) paradigms to address the possible neurophysiological effects of taVNS: short latency inhibition (SAI) to assess the activation of the basal forebrain cholinergic system, short latency intracortical inhibition (SICI) to assess the activation of the intracortical inhibitory GABA-A system, and cerebello-cortical inhibition (CBI) to assess effect of inhibitory locus coeruleus projections on cerebellar Purkinje cells.**Methods:** 23 healthy participants (mean age 27 years, 57% females) completed 2 visits, separated by 5 days. In a randomized order they received 25min of taVNS at the left cymba conchae or sham stimulation (sVNS) at the left earlobe, using following stimulation parameters: square shaped 100Hz pseudobiphasic pulses, interphase interval 80 us, pulse duration 300ms, intensity 100uA above the sensory threshold level. For SAI we used 3 interstimulus intervals (ISI): 20, 22, and 24ms. For the SICI curve, we used 4 conditioning stimulus (CS) intensities: 70%, 80%, 90%, and 100% of the active motor threshold. For CBI ISI was 5ms and CS was at the maximum intensity tolerated by the participant. Each TMS paradigm was repeated before, during, and after taVNS/sVNS (b-d-a). For each TMS parameter we performed rmANOVA with within-subject factors: stimulation (real/sham), time (b-d-a), and additional factors ISI and CS intensity for SAI and SICI, respectively.**Results:** For SAI and SICI, rmANOVA revealed no significant 2-way or 3-way interactions, suggesting no difference in the effect of real and sham taVNS on these parameters. There was no significant difference between the effect of taVNS and sVNS on CBI. **Discussion and conclusions:** Our group analysis suggests that taVNS does not affect electrophysiological measures of intracortical cholinergic and GABAergic systems, nor does it affect cerebellar inhibition of the motor cortex. Our findings could be explained by a) a ceiling effect due to strong baseline inhibition in young healthy subjects that could not be further modified by intervention; b) high variability of TMS measures; c) suboptimal taVNS parameters; or d) a true absence of an effect of taVNS.

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Poster

P365. Spinal Cord Injury: Therapeutic Strategies: In Vivo: Pharmacological

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P365.01

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH R01 NS100531

Title: Exploiting polypharmacology: targeting multiple kinases to promote axon regeneration and behavioral recovery after CNS injury

Authors: *K. MAH[1], H. AL-ALI[1], W. WU[2], N. O'NEILL[1], J. LOWELL[1], M. MUNOZ[1], X.-M. XU[2], V. P. LEMMON[1], J. L. BIXBY[1];
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Abstract: Central nervous system (CNS) axons generally lack the ability to regenerate and reconnect, which hampers recovery after spinal cord injury. This can be attributed to both growth repressive factors in the CNS environment and a lack of intrinsic regenerative potential in adult CNS neurons. Kinases regulate many cellular processes including axon growth and regeneration; kinase inhibitors may therefore regulate multiple signaling networks to overcome regeneration inhibition. We developed a platform that combines phenotypic and target-based screening to identify a) kinase targets and b) kinase inhibitors of interest in neurite outgrowth. Using this approach on a library of kinase inhibitors, we identified “target” kinases whose inhibition promotes axon outgrowth and “anti-target” kinases whose inhibition limits this process (Al-Ali; PMID: 26056718). Simultaneously inhibiting each of two target kinases, Rho kinase (ROCK) and ribosomal protein S6 kinase 1 (S6K1) had a synergistic effect on promoting neurite outgrowth. Notably, we identified a single compound (RO48) that inhibits both ROCK and S6K1, as well as other target kinases, while exhibiting better overall kinome selectivity than several FDA approved kinase inhibitor drugs. RO48 promoted substantial axon outgrowth in a variety of cultured neurons, as well as corticospinal tract (CST) sprouting and regeneration in two mouse models of spinal cord injury, accompanied by improved recovery of sensorimotor function in rotarod, gridwalk, and pellet retrieval tasks. We subsequently identified RO48 derivatives with improved pharmacokinetic parameters that also produced robust neurite outgrowth in vitro. One of these derivatives was shown to promote CST axon growth in the pyramidotomy model. These results support the relevance of identified target kinases to regeneration, and are the first steps in generating candidate lead compounds for clinical development.

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Poster

P365. Spinal Cord Injury: Therapeutic Strategies: In Vivo: Pharmacological

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P365.02

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH Grant NS111037-02

Title: Rolipram delivered by PgP nanocarrier enhances motor function in a rat contusion SCI model

Authors: *Z. LIAO[1], J. GAO[2], M. KHANG[1], M. R. DETLOFF[3], J. LEE[2];
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Abstract: Spinal cord injury (SCI) is a major source of morbidity and mortality throughout the world. One of important pathological events after SCI is significant decrease of intracellular cAMP level in injured spinal cord. Phosphodiesterase 4 (PDE4) activation after injury causes excessive degradation of cAMP and this impairs cAMP signaling which affects cell metabolism, survival, and behavior. Rolipram (Rm), a PDE4 inhibitor, is used to effectively improve neuronal survival and reduce neuroinflammation after SCI. Previously, we reported that Rm was loaded in to an amphiphilic nanocarrier, poly(lactide-co-glycolide)-graft-polyethenimine (PgP) developed in my lab (US patent: US 10,232,050) and Rm- loaded PgP (Rm-PgP) nanoparticles injected at the SCI lesion can restore cAMP levels to sham level and reduce inflammation and apoptosis in rat compression SCI model. In this study, we evaluate the therapeutic efficacy of Rm-PgP treatment on secondary injuries, functional recovery, and neuropathic pain in a rat moderate contusion SCI model. Moderate contusion injury model was created at T9-T10 spinal cord of SD rats (200-250 g) using the impactor (IH-0400, PSI) with a force of 200 kdyn. Rm-PgP (10 μg Rm) was administered immediately after injury by intraspinal injection using microinjector (MICRO2T, WPI) with injection speed 2 μL/min. Rm-PgP effect on cAMP level was measured using cAMP ELISA kit at 1 and 2 days post treatment. Motor functional recovery of the hindlimb was evaluated using BBB locomotor rating scale and neuropathic pain was evaluated using von Frey filament up to 4 weeks after SCI. At 4 weeks post-injury, rats were sacrificed via cardiac perfusion and the spinal cord (1 cm) was retrieved, fixed, embedded in OCT block, longitudinally sectioned, and stained for inflammatory response, apoptosis, neuronal survival, and astrogliosis. We observed that cAMP level in Rm-PgP treated animal group was significantly different with that in untreated SCI group at 1 DPI and significantly fewer $ED1^+$ cells, $TUNEL^+$ apoptotic cells and $GFAP^+$ cells in the spinal cords from animals treated with Rm-PgP relative to the untreated animal group. For the motor function recovery, BBB scores of the treated animals were significantly higher than those of untreated SCI animals from 2 week upto 4 weeks. For the neuropathic pain, withlold threshold of the the treated animals were higher, but the threshold was not significantly different with untreated SCI animals at all time points. In conlussion, local delivery of Rm by PgP nanocarrier reduced secondary injury and improved motor function after SCI in a rat contusion SCI model.Funding was provided by NIH/NINDS grant 5R01 NS111037-02.

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Poster

P365. Spinal Cord Injury: Therapeutic Strategies: In Vivo: Pharmacological

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Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P365.03

Topic: C.11. Spinal Cord Injury and Plasticity

Title: The role of plasma derived purified exosome product for improved locomotor recovery in spinal cord injured rats

Authors: *F. MOINUDDIN, C. ONYEDIMMA, M. BYDON;
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Abstract: Study design: Animal Study **Introduction**: Preclinical studies indicate that mesenchymal stem cell-derived secretomes exert immunomodulatory and neuroprotective effects in various spinal cord injury models. Exosomes as one of the secretomes, have been shown to successfully aid wound healing for the skin and following myocardial infarction. However, the effects of human plasma-derived purified exosomal products (PEP) on spinal cord injury (SCI) have not been explored to this date. **Objective:** To determine the impact of PEP on locomotor recovery in the rat model of SCI followed by a comparison of the intrathecal and Intravenous (I.V) routes of administration for the delivery of PEP. **Methods**: Twenty-four female rats were randomly divided into five groups. These included 2 control groups for the I.V or I.T injected PEP groups and a sham group. Groups were subject to a T9 laminectomy, followed by a moderate contusion SCI. After 24 hours, lactated ringer solution or PEP was injected through I.V or IT routes. Rats were assessed weekly for a total period of 10 weeks by an open field Basso, Beattie, and Bresnahan (BBB) motor score and a postmortem quantification of external scaring, neuronal regeneration, cavity volume, and glial scarring was carried out at the end of the follow-up period. **Results**: Animals treated with I.V PEP or IT PEP had significant improvement within the first-week post-injury (1.05±1.2 vs. 2.16±3.15) and (1.08±3.5 vs. 4.5±0.083) respectively, with sustained improvement in BBB score at 10 weeks (11±1.16 vs. 13.8±1.90),(11.3±1.4 vs. 14±1.16). **Conclusion**: Our preliminary results suggest that I.V or IT PEP injection without immunosuppression might provide beneficial effects initially following traumatic SCI.

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Poster

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Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH Grant 5R21NS115094

Title: Critical role of mitochondrial aldehyde dehydrogenase -2 (ALDH2) in acrolein-mediated neurotoxicity in mouse spinal cord injury

Authors: *S. SUN[1], Z. ZHANG[2], S. HERR[2], R. SHI[3];
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Abstract: Oxidative stress-associated lipid peroxidation-derived aldehydes such as acrolein have emerged as key mediators in sustaining secondary injury that contributing significantly to the functional loss after spinal cord injury (SCI). Mitochondrial aldehyde dehydrogenase-2 (ALDH2), a key oxidoreductase and powerful endogenous anti-aldehyde machinery, is thought to play a critical role in acrolein-mediated pathology. Therefore, the gene mutant that encodes an inactive form isoenzyme (ALDH2*2), a genetic condition that exists in more than 600 million people worldwide, could potentially impair the individual's ability to remove toxic acrolein that leads to exacerbated injury following spinal trauma. Recently, a novel transgenic (TG) ALDH2*2 mouse that recapitulates the human condition was developed that offers an ideal model to study the clinically relevant scenario of heightened neurofunction impairment for individuals with this mutation. Further equipped by the ALDH2-selective agonist, Alda-1, such study could not only further demonstrate the critical role of ALDH2 in suppressing acrolein in SCI, but also explore ALDH2 as a potential target for anti-acrolein pharmaceutical intervention.The ALDH2*2 TG mice that received thoracic contusion SCI showed a more exaggerated ascension of the acrolein level when compared to the wildtype (WT) injury mice at 2-days post-injury. In addition, such acrolein buildup remained at a significantly higher level at 7 days when the WT mice had started to recover towards the baseline in the spinal cord. Moreover, the application of Alda-1 significantly rescued the ALDH2 enzymatic activity, mitigated the post-SCI acrolein elevation, and enhanced the neurofunctional recovery. These findings support our hypothesis that the activity of ALDH2 is an essential neuroprotective factor in acrolein sequestration and functional amelioration in SCI. A better outcome could be achieved by boosting its activity after SCI and possible other neurotrauma diseases, especially in individuals with such genetic deficits.

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Poster

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Program #/Poster #: P365.05

Topic: C.11. Spinal Cord Injury and Plasticity

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Title: Oxygen therapeutic to limit secondary spinal cord injury (SCI) in a rodent model

Authors: J. HOU[1,3], D. PLANT[3], S. TSUDA[1,3], B. RENGERT[3], J. ZHU[1], T. PARSONS[1], M. FRANKLIN[1], F. THOMPSON[3], C. GARVAN[1], B. SPIESS[1], P. BOSE[1,3,2];
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[3]North Florida/South Georgia Veterans Hlth. Syst., Gainesville, FL

Abstract: Acute SCI creates a spectrum of tissue damage. Damage results from both primary injury caused by immediate neural cell death and secondary injury resulting from ischemic and inflammatory cascades. Secondary injury contributes to lesion expansion and worsening neurological deficit. Injury and neuron death lead to swelling, reduced perfusion/O_2 delivery, and an expanding penumbra. This ischemic penumbra is an obvious target for therapeutic interventions because a rapid reduction of ischemia can show substantial benefits for neuron salvage and motor function. The objective of this study is to test the preclinical evaluation of the safety and efficacy of a novel patented IV emulsions of perfluorocarbon (PFC) nanoparticles, $NanO_2$ (NuVox Pharma LLC, Tucson, AZ), to limit the ischemic penumbra and improve motor disabilities following SCI. $NanO_2$ is a pentane-based, low molecular weight, nanodroplet (250-300 nm) PFC emulsion. Unlike other PFCs, $NanO_2$ has unique properties of higher levels of respiratory gas transfer and deeper O_2 penetration to areas not able to be perfused by red blood cells. NanO2 has a better tolerance and safety profile, and there have been no FDA hold safety concerns to date. Although $NanO_2$ is in Phase 1B and II clinical trials for stroke and cancer in the US, it has not been investigated for SCI. Randomized $NanO_2$ or normal saline (NS) were administered 20 minutes after SCI (0.6 ml/kg, 2 hrs apart, three doses total) via lateral tail vein. At post-injury week 5, the NS treated C-SCI animals showed significant increases in ankle torque and time-locked EMG burst amplitudes at 8 test velocities as compared to recordings in naïve animals, revealing an enduring velocity-dependent spasticity of the extensor muscles of the lower limb. In contrast, the $NanO_2$ treatment apparently blocked spasticity development. Moreover, $NanO_2$ treated animals exhibited rate-sensitive depression of the soleus H-reflex toward normal values especially in higher test frequency compared to significant loss of rate-depression exhibited in NS-treated SCI animals. Moreover, hindlimb step cycle knee-ankle coordination showed robust improvement following $NanO_2$ treatment compared to extensive loss of joint coordination in C-SCI animals. *Ex vivo* diffusion tensor imaging (DTI) MRI revealed robust preservation of the dorsal corticospinal and rubrospinal descending motor tracts following $NanO_2$ treatment compared to NS treated SCI animals. Our preliminary therapy-induced behavioral/physiological/MRI improvements data to date indicate that $NanO_2$ therapy in SCI may have prevented the ischemia-mediated neurological damage and improved motor disabilities.

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Poster

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Topic: C.11. Spinal Cord Injury and Plasticity

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Title: Lack of significant neuroprotection from either durotomy or methylprednisolone after cervical spinal cord injury in a rat model

Authors: *J. BERGQUIST, T. PRASSE, N. AL-KHAYAT, J. BUSHNELL, C. P. HOFSTETTER, Z. Z. KHAING;
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Abstract: Traumatic spinal cord injury (SCI) results in a wide range of outcomes from partial paralysis to complete tetraplegia. Importantly, there is a high density of motor neurons in the cervical region involved in important motor outputs such as breathing and hand function. Previous work in thoracic injury models has demonstrated surgical decompression (i.e., durotomy) is beneficial for tissue and neuron sparing, and for behavioral recovery; retrospective clinical studies suggest durotomy may improve outcomes in cervical injury as well, however prospective, randomized evidence is still needed to determine whether durotomy benefits this region. The present study aims to minimize the secondary damage after cervical SCI by addressing multiple interrelated contributors to neuron death via either surgical manipulation (durotomy), FDA-approved methylprednisolone sodium succinate (MPSS), or both surgical and pharmaceutical treatments. Clinically relevant 200 kD bilateral contusion-type injuries were introduced at the C5 spinal cord segment in a cohort of adult female Long Evans rats. Animals were randomly assigned to one of 5 groups: sham (n=1), SCI only (n=5), durotomy (n=5), MPSS (n=5), and durotomy + MPSS (n=5), and were allowed to survive for 14 days post injury (dpi). Treatment effectiveness was assessed by comparing neuron and tissue sparing, inflammatory response, and functional deficits between treatment groups. The durotomy + MPSS group showed statistically insignificant trends towards greater neuron sparing and tissue sparing 1mm caudal to injury epicenter compared to SCI-only controls (2162 +/- 250 vs. 1954 +/- 504 neurons), (83.6 +/-6 % vs. 73.5 +/- 10% spared). There were no differences in inflammatory (Iba1, GFAP) response between treatment and control groups, as measured by fluorescence intensity in immunohistochemistry ($p = 0.413$, 2-way ANOVA). Finally, all animals showed similar deficits in forelimb joints' range of motion and weight support at 4dpi and recovered most functions except forelimb-hindlimb coordination by 14dpi. The present study highlights a critical need for re-evaluation of the approved, yet still controversial use of MPSS. It is also likely that the 14 dpi survival time did not allow enough time for histological differences to manifest, therefore future studies will include longer survival times. Given the complex and

dynamic evolution of architecture and environment of the spinal cord after SCI, examining more chronic effects of durotomy and MPSS on different injury severities will be crucial.

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Poster

P365. Spinal Cord Injury: Therapeutic Strategies: In Vivo: Pharmacological

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P365.07

Topic: C.11. Spinal Cord Injury and Plasticity

Support: VA Merit Award (I01RX002969)
PVA Research Foundation
Taylor Foundation for Chronic Diseases

Title: Conditional Rac1 Knockout in Astrocytes Reduces Hyperreflexia Associated with Spasticity after Spinal Cord Injury

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Abstract: Spasticity is a hyperexcitability disorder resulting from damage to the spinal cord or brain (e.g., SCI, TBI, stroke). A clinical sign of spasticity is an amplified H-reflex response (aka knee-jerk spinal stretch reflex). Although conventional anti-spasticity drugs, such as baclofen, provide positive relief of spastic symptoms, they are not curative, only palliative, and may carry safety risks with long-term use. Our research has identified abnormal dendritic spines morphology on spinal alpha-motor neurons associated with spasticity after SCI. Specifically, we have shown that systemic pharmacological treatment with a Rac1-inhibitor restores close-to-normal dendritic spine morphology and reduces hyperreflexia after SCI. These studies, however, did not identify whether the beneficial effect of Rac1 inhibition was due a neuronal or non-neuronal mode of action. Here, our study aims to examine the relationship between Rac1 molecular signaling in astrocytes and spasticity after SCI. To address this aim, we investigated the effect of disrupting of Rac1 in GFAP+ astrocytes using a transgenic conditional knockout

(KO) mouse model, i.e., a cre-flox system, in a longitudinal study design. To induce spasticity, animals received a moderate contusion SCI at vertebral segment T10. Three weeks after SCI, injured wildtype (WT) animals showed a loss of H-reflex rate-dependent depression (RDD), a physiological sign of spasticity. In contrast, astrocytic Rac1 KO animals with SCI displayed a H-reflex response similar to measurements at baseline, before injury, and significantly lower than compared with WT controls. Importantly, the absence of significant hyperreflexia in astrocytic Rac1 KO animals was associated with near-normal dendritic spine profiles on alpha-motor neurons. As expected, control WT animals with SCI presented with abnormal dendritic spine morphology associated with hyperreflexia disorder. Finally, we observed that ventral spinal tissue in GFAP Rac1 KO expressed increased levels of the glial specific glutamate transporter, GLT-1, compared to WT controls. Collectively, structural plasticity along with changes in glutamatergic signaling appear to underlie the role of Rac1 activity in spasticity after SCI. Taken together, this study begins to deconstruct the contribution of Rac1 signaling in spinal motor neuron hyperexcitability after injury, and reveals a novel opportunity for developing targeted cell-specific therapies for alleviating SCI associated hyperexcitability disorders.

Disclosures: C. Benson: None. **K. Olson:** None. **S. Patwa:** None. **S.G. Waxman:** None. **A.M. Tan:** None.

Poster

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Program #/Poster #: P365.08

Topic: C.11. Spinal Cord Injury and Plasticity

Support: ICMR, GOVT OF INDIA
DST-SERB, GOVT OF INDIA

Title: Role of cyclosporine A in apoptosis after spinal cord injury

Authors: *P. P C[1], S. VENKATACHALAM[1], K. CHANDRAMOHAN[1], K. IYER[1], F. MICHAEL[1,2];
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Abstract: Role of Cyclosporine A in apoptosis after spinal cord injury Preeja Chandran P.C[1*], Khaviyaa Chandramohan[1], Krithika Iyer[1],Felicia Mary Michael[1,2], Sankar Venkatachalam[1]. [**]1-Department of Anatomy, Dr. ALM PGIBMS, University of Madras, Taramani Campus, Chennai 600 113. 2-University of Kentucky, Lexington, USA* Presenting Author; ** Corresponding Author
The cyclosporine A (CsA), an immunosuppressant is long known for its potential neuroprotective nature. The speculation was rooted in the finding that CsA improves

mitochondrial integrity and reduces the Cytochrome C release. Through its manipulative action on mitochondrial pores, it is believed to have an anti-apoptotic effect on the injured cells. Despite these facts, its usefulness is still controversial, especially after spinal cord injury. The present study was carried out to re-analyze its role in SCI treatment using a highly standardized rodent model of contusive SCI.
Contusion SCI model in rats was created following the MASCIS model guidelines. In Wister-albino rats of 70 to 90 days of age (Bodyweight: 150 to 250 g), moderate contusion injury (12.5 mm) was created using MASCIS impactor at T10-T11 spinal segment level. SCI animals were randomly divided into three groups as a) Lesion group b) SCI + 5mg/kg CsA i.p. 3 days following injury day and c). SCI + 5mg/kg CsA i.p., 3 days following injury day and again on sixth to the ninth day. received first three days and sixth day to the ninth day. All animals were sacrificed after 8 weeks of postoperative survival. Histology for assessing tissue sparing was carried out. Motor recovery during the postoperative period was objectively assessed through a series of behavioral tests such as BBB, Narrow Beam walking test, and ladder walking task. Numerical data were analyzed for statistical significance of the differences observed by GraphPad Prism Version 7.0.Results indicate improvement in the behavior scoring-based assessments. There was a gradual improvement of motor function - both gross locomotor ability and fine motor movements. Histological preparations showed evidence of tissue sparring at the injury site substantiating the anti-apoptotic effects of CsA. Further studies may be required to fine-tune the strategy to enhance the beneficial effects observed so that a statistically significant functional recovery may be achieved.Acknowledgment: Funding from SERB, Govt of India to SV, and ICMR-SRF to PC.

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Poster

P365. Spinal Cord Injury: Therapeutic Strategies: In Vivo: Pharmacological

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Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P365.09

Topic: C.11. Spinal Cord Injury and Plasticity

Support: DOD SC170121

Title: Bringing the clinic into the lab: An evaluation of how clinically common drugs and physical therapy muscle stretching influence the functional recovery of rodents after spinal cord injury

Authors: *M. SHARP[1], G. CESARZ[2], J. WILKERSON[3], A. SHUM-SIU[4], C. CASTILLO[4], J. C. PETRUSKA[5], D. S. MAGNUSON[4];
[1]Interdisciplinary Program in Translational Neurosci., [2]Dept. of Physiol., [3]Dept. of

Bioengineering, [4]Dept. of Neurolog. Surgery, [5]Dept. of Anatom. Sci. and Neurobio., Univ. of Louisville, Louisville, KY

Abstract: Stretch-based physical therapy remains a frontline intervention following spinal cord injury (SCI) utilized to combat secondary complications such as muscle atrophy, spasticity, and contractures. In addition to rehabilitation and physical therapy sessions, SCI patients receive a barrage of pharmacological treatments to ameliorate acute and/or chronic sequelae such as neuropathic pain and spasticity, among many others. However, the effects of many medications on motor/locomotor function after SCI remains unknown, and in some cases, has been shown to be detrimental to functional recovery. Furthermore, despite the clinical prevalence of stretching, numerous studies have shown that clinically modeled hindlimb stretching of rodents following SCI significantly impairs functional recovery and causes dramatic deficits in locomotor capacity. Thus, other interactions that may be occurring between physical therapy and pharmacological interventions after SCI remain unexplored in clinical and basic science research and may reveal novel cellular mechanisms mediating the detrimental effects of stretching. To address these fundamental questions, adult female Sprague-Dawley rats received a T10 spinal cord contusion (n=12). After 4 weeks of recovery, all animals received daily hindlimb muscle stretching for 2 weeks in combination with daily pharmacological intervention administered intraperitoneally at high functional doses, including acetaminophen, morphine, or control saline. After 2 weeks of stretching, animals recovered for 3 weeks without physical therapy or drug administration. Following recovery, animals underwent an additional 2 weeks of muscle stretching combined with daily administration of ibuprofen, baclofen, or control saline. Functional recovery was assessed weekly using the Basso, Beattie, and Bresnahan (BBB) Locomotor Scale and kinematic gait analysis. Von Frey, Hargreaves, and tail-flick sensory assays were performed weekly to assess mechanical and thermal cutaneous sensitivity. Locomotor analysis revealed that clinically common pharmacological interventions did not ameliorate the detrimental effects of stretching on functional recovery. Furthermore, BBB assessments suggested that hindlimb stretching combined with treatments such as opioids may exacerbate the deleterious effects of stretching on locomotion. These results emphasize the need to further model and evaluate commonly used drugs and therapies after clinical SCI and their combinatory effects on locomotor recovery in rodent models and the clinic.

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Poster

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Program #/Poster #: P365.10

Topic: C.11. Spinal Cord Injury and Plasticity

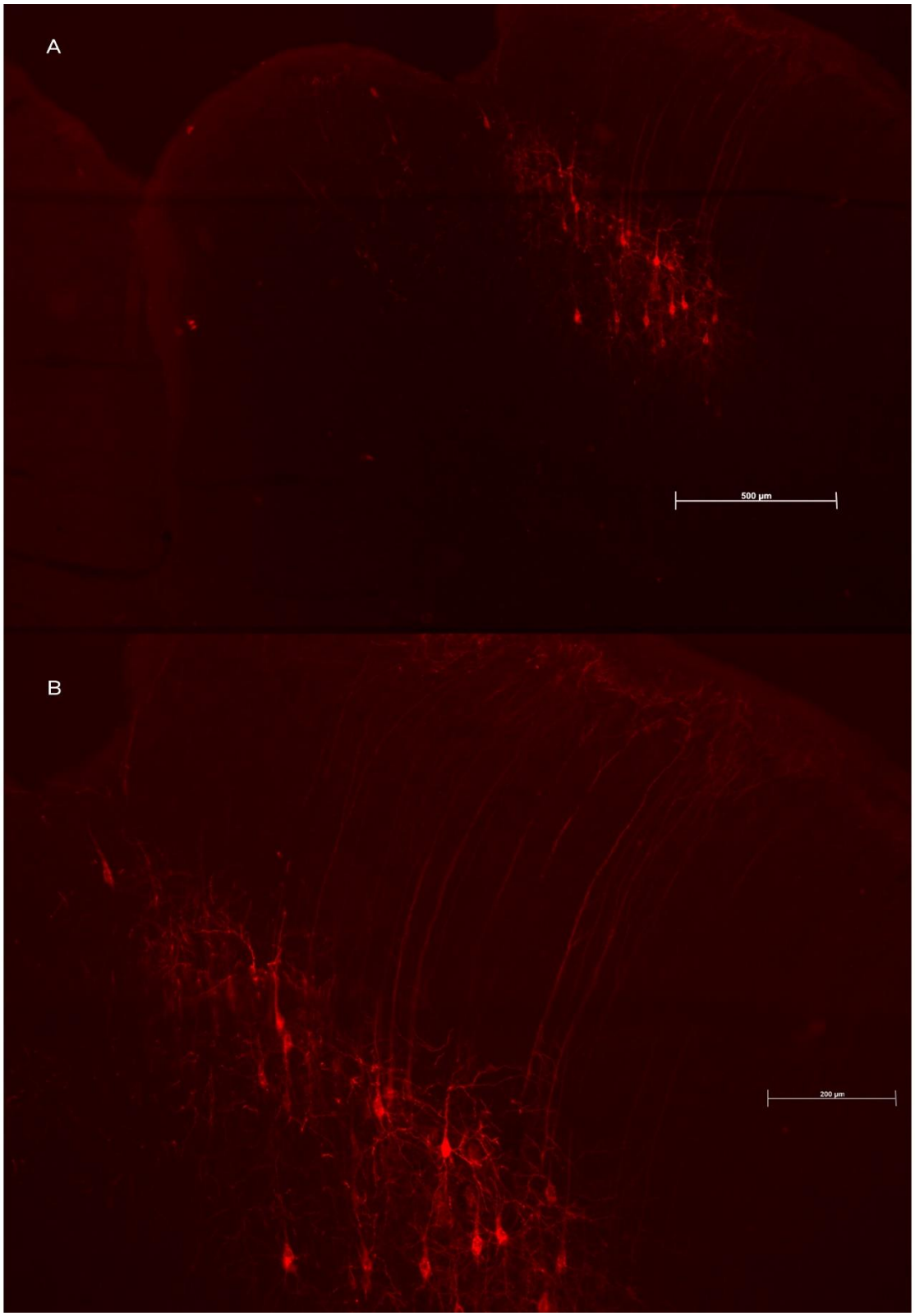
Support: This work was funded by a fellowship grant from Shriners Hospitals for Children SHC-84706.

Title: Enhancing forelimb recovery from cervical spinal cord injury in juvenile rats with the constitutive application of excitatory DREADDs

Authors: *R. D. SMIT, T. J. CAMPION, III, R. L. STINGEL, G. M. SMITH;
Shriners Hosp. Pediatric Res. Ctr., Lewis Katz Sch. of Med. at Temple Univ., Philadelphia, PA

Abstract: A hallmark found across mammalian species is that neonates recover more notably from spinal cord injury (SCI) compared to adults. Research in rodents indicates that the superior recovery, which ceases abruptly beyond postnatal day 10, is owed to the more robust regeneration of corticospinal neurons (CST) across the injury site whereas older animals are reliant on sprouting from uninjured subcortical populations. Few studies have simultaneously analyzed the discontinuation of axonal regrowth with aging and how each pathway's response translates to functional outcomes. Our aim is to define the specific regenerative and sprouting mechanisms of the CST and subcortical pathways after SCI in juvenile rats (14 days of age), and facilitate recovery through the constitutive activation of chemogenetic modifiers (DREADDs). This is the first study demonstrating successful co-infection of the CST in neonatal rats using a two-viral vector system where we simultaneously injected Cre-dependent-excitatory DREADD into the motor cortex and retroCre recombinase into CST axon terminals of the cervical spinal cord.

At postnatal day 5 rats received transcranial (motor cortex) injections of either non-Cre-dependent excitatory DREADD (hM3Dq), non-Cre-dependent inhibitory DREADD (hM4Di), control fluorescent (GFP), or the two-viral injection of Cre-dependent excitatory DREADD (AAV2-DIO-hM3Dq) + Cre (retroCre). At postnatal day 14, all animals received C5 hemisections. Subsequently, the constitutive activation of either excitatory or inhibitory DREADDs was achieved by treating diet gel with actuators (clozapine-N-oxide) for 30 days. Behavioral assays consist of isometric pull strength, IBB, single pellet reach, kinematics, and electrophysiological recordings. This study is exploring the age-dependent response to severe SCI and whether excitatory DREADDs could stimulate recovery during the infantile period. Thus far, the excitatory groups have demonstrated superior functional recovery of the forelimb compared to the control and inhibitory groups.



Co-infection of the motor cortex

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Poster

P365. Spinal Cord Injury: Therapeutic Strategies: In Vivo: Pharmacological

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Topic: C.11. Spinal Cord Injury and Plasticity

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Land Salzburg/IWB/EFRE 2014-2020 P1812596 "EV-TT"

Title: Enhancing functional recovery through intralesional application of extracellular vesicles in a rat model of traumatic spinal cord injury

Authors: P. ROMANELLI[1,2], L. BIELER[1,2], P. HEIMEL[3,4,5], S. SKOKIC[6], D. JAKUBECOVA[1,2], C. KREUTZER[1,2], P. ZAUNMAIR[1,2], T. SMOLCIC[6], B. BENEDETTI[1,2], E. ROHDE[7,8,9], M. GIMONA[9,10,8], D. HERCHER[3,5], M. DOBRIVOJEVIC RADMILOVIC[6,11], ***S. COUILLARD-DESPRES**[1,2,5];
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Abstract: Local inflammation plays a pivotal role in the process of secondary damage after spinal cord injury (SCI). We previously showed in a rat model that acute intravenous application

of extracellular vesicles (EVs) secreted by human umbilical cord mesenchymal stromal cells dampens the induction of inflammatory processes following spinal cord injury. However, systemic administration of EVs is associated with delayed delivery to the site of injury and the necessity for high doses to reach therapeutic levels. In this project, we compared the impact on the functional regeneration and structural integrity of EVs injected directly into the lesion site following spinal cord injury, with the former treatment in which EVs were injected intravenously.
Female Fisher 344 rats of 10 weeks of age received a 200 kdyn contusion on the spinal cord (Th8) which resulted in a moderate to severe incomplete spinal cord injury. The rats received acutely after lesion either an intra-parenchymal administration of vehicle or EVs, or an intravenous injection of EVs. Gene expression of pro and anti-inflammatory cytokines in the spinal cord were determined at 24 hours post-injury. Histological analyses were performed 2 and 8 weeks after contusion. Locomotor recovery was assessed until 8 weeks after contusion. MRI, diffusion tensor imaging (DTI) and microCT analysis were performed ex vivo 8 weeks after injury.
Intralesional EVs application was found to be the most efficient to improve locomotor function recovery during the first 8 weeks post-injury based on the BBB score and BBB sub-score. The acute intralesional application of EVs resulted in a marked reduction of pro-inflammatory cytokines expression measured at 24h post-tSCI. Even at 14 days post-injury, the accumulation of inflammatory cells around the lesion was significantly lower in rats treated with EVs. Furthermore, the deposition of collagen and CSPGs, as well as astrogliosis, were more attenuated by the intralesional application of EVs. Finally, 8 weeks post-injury, we measured with DTI that an acute intralesional EVs treatment could preserve more neuronal fibre bundles in the proximity of the lesion site, as compared to the vehicle administration
Our observation demonstrates that EVs are particularly potent to address the processes of inflammation and scarring when applied very early and close to the site of injury, and promote a better long-term motor function recovery. Moreover, an improvement of the lesion microenvironment and the additional structural sparing obtained with local EV treatment constitute valuable assets for follow-up interventions aiming for endogenous axonal regeneration or involving cell therapies

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Poster

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Program #/Poster #: P365.12

Topic: C.11. Spinal Cord Injury and Plasticity

Support: The Office of the Assistant Secretary of Defense for Health Affairs, through the Spinal Cord Injury Research Program under Award No. W81XWH-18-1-0807

Title: Isoflurane anesthesia, but not ketamine, blocks the induction of nociceptive induced hemorrhage after SCI in rats

Authors: *J. A. DAVIS, R. E. BAINE, M. M. TARBET, G. N. K. FAUSS, G. GIDDINGS, J. W. GRAU;
Texas A&M Univ., College Station, TX

Abstract: Spinal cord injuries are often accompanied by additional tissue damage (polytrauma), providing a source of pain (nociceptive) input. Research has shown that nociceptive stimulation soon after SCI impairs long-term recovery and increases tissue loss. This effect has been related to a pain-induced breakdown of the blood spinal cord barrier and hemorrhage at the site of injury. Using an animal model (rats), we have shown that inhibiting cellular activity at the site of injury (T10-11) with lidocaine blocks the adverse effect of noxious stimulation (Turtle et al., 2017, J Neurotrauma, 34, 1200). Other work suggests that this local effect depends, in part, on communication with rostral (brain) systems. Supporting this, nociceptive stimulation has no effect on tissue sparing in contused rats that received a rostral (T2) spinal transection (Reynolds et al., 2017, Frontiers Sys Neuro, 13.) or pharmacological transection with lidocaine at T2 (Davis et al, 2020, J Neurotrauma, 40, 9186). These observations led us to test whether treatment with a general anesthetic would have a protective effect. Pentobarbital delivered at a moderate to high (25-50 mg/kg, i.p.), but not low (12.5mg/kg), dosage blocked pain-induced hemorrhage in contused rats. Here we explore whether other anesthetics have a protective effect. Gas anesthesia was induced using isoflurane. A day after rats received a contusion injury at T10-11, they were placed in the induction chamber and delivered a 5% oxygen/isoflurane mixture or just oxygen. Anesthesia was reduced to 3% and half the animals in each condition received noxious electrical stimulation (shock) to the tail for 6 min. Three hours after shock, rats were sacrificed and one cm of tissue was collected surrounding the injury site. Pain input induced hemorrhage in non-anesthetized rats, but not in those treated with isoflurane. Ketamine was tested to examine the protective potential of dissociative, rather than anesthetic states. Interestingly, neither a high dose of i.p. ketamine (100mg/kg) alone, nor in combination with xylazine (12.5mg/kg), prevented pain-induced hemorrhage after SCI. Further work is being conducted to determine why these treatments yield divergent results.

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Poster

P365. Spinal Cord Injury: Therapeutic Strategies: In Vivo: Pharmacological

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Topic: C.11. Spinal Cord Injury and Plasticity

Support: National Institute of Neurological Disorders and Stroke (NS104422)

Title: Treatment with norepinephrine after spinal cord injury increases tissue loss and impairs functional recovery

Authors: *D. T. JOHNSTON, E. T. LOUT, R. E. BAINE, J. W. GRAU;
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Abstract: Prior work has shown that engaging pain fibers after spinal cord injury (SCI) by applying noxious electrical stimulation or the irritant capsaicin increases tissue loss and impairs long-term recovery (Grau et al., 2017, *J Neurotrauma*, 34, 1873). These adverse effects have been related to a breakdown in the blood-spinal cord barrier and the infiltration of blood (hemorrhage). Noxious electrical stimulation also induces an acute increase in blood pressure. The current studies examined the effects pharmacologically inducing hypertension with norepinephrine (NE) after SCI has on the induction of hemorrhage and recovery of locomotor function. In all experiments, male Sprague-Dawley rats received a contusion injury at the T10-11 vertebrae and had their baseline locomotor function (BBB) and blood pressure (BP) measurements assessed prior to receiving a subcutaneous (SQ) injection of either 0 or 0.5 mg/kg NE. In experiment 1, subjects received NE or vehicle and were allowed to recover for 21 days in which BP and BBB was assessed throughout and tissue was collected for histological analysis. Acute effects of pharmacologically induced hypertension were assessed in the following experiments. In experiment 2, subjects received NE or vehicle 1.5 or 6 hrs post-SCI to assess the window of time in which the spinal cord is most vulnerable to hypertension. In experiment 3, subjects received NE or vehicle at the beginning of treatment and then again 1.5 hr later to examine the effects of maintained hypertension. In experiment 4, subjects received either six-minutes of electrical stimulation to the tail or no shock followed by NE or vehicle to determine if a detrimental synergistic effect occurred when pain input was given in addition to pharmacologically inducing hypertension. In experiments 2-4, blood pressure measurements and BBB scores were assessed immediately, one, two, and three hours after treatment and then a 1-cm region of tissue encompassing the injury site was collected and assessed for hemorrhage and inflammatory markers. Treatment with norepinephrine undermined long-term recovery and increased tissue loss. Hypertension occurring 24 hours, but not 1.5 or 6 hr post-SCI was detrimental to locomotor recovery, but was not sufficient to induce hemorrhage. Hypertension and tail shock increased hemodynamic measures and undermine locomotor recovery in a synergistic fashion, however only shock treatment alone led to increased levels of hemorrhage. Additional studies are being conducted to assess the mechanism by which NE treatment is leading to a decrease in locomotor recovery.

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Poster

P365. Spinal Cord Injury: Therapeutic Strategies: In Vivo: Pharmacological

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P365.14

Topic: C.11. Spinal Cord Injury and Plasticity

Support: National Institute of Neurological Disorders and Stroke (NS104422)

Title: Brain-dependent processes fuel pain-induced breakdown of the blood-spinal cord barrier after spinal cord injury

Authors: *S. R. PARTIPILO, G. N. K. FAUSS, D. T. JOHNSTON, R. E. BAINE, J. A. DAVIS, J. W. GRAU;
Texas A&M Univ., College Station, TX

Abstract: Spinal cord injury (SCI) is often accompanied by other injuries which provide additional sources of pain input. Our laboratory has found that pain input soon after SCI, in the form of electrical stimulation or capsaicin injection, leads to an increase in inflammation and hemorrhage at the lesion site (Grau et al., 2017, J Neurotrauma, 34, 1873). The breakdown in the blood-spinal cord barrier (BSCB) likely proceeds this infiltration of blood and immune cells. Recently, we have found that a rostral spinal transection blocks this increase in hemorrhage seen after a spinal contusion injury, suggesting the involvement of brain systems in the exacerbation of secondary injury. Given this, the current study explores whether blood-spinal cord barrier (BCSB) permeability increases after pain input and if this is blocked by rostral transection. In experiment 1, adult male Sprague-Dawley rats were given a T12 spinal contusion injury. Twenty-four hours later, the rats received 6 minutes of intermittent electrical stimulation delivered to the tail or an equal period of restraint (no shock). Evan's Blue dye (2%) was administered (i.p.) immediately after treatment. Three hours later, the spinal cords were collected and processed for spectrophotometry. In experiment 2, rats received a T12 contusion and a subsequent T2 complete transection or sham surgery. Twenty-four hours later, rats in both groups either received 6 minutes of electrical stimulation or no shock. Evan's Blue dye was administered and spinal cords were collected and processed as described in experiment 1. It was found that shock after a spinal contusion increased Evans Blue dye extravasation in the spinal cord and that transection blocked this effect. These results suggest that pain input increases BSCB permeability after injury and that this is dependent on brain systems. Future studies will explore how pain input damages the BSCB and the brain systems/processes that drive this effect.

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Poster

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Program #/Poster #: P365.15

Topic: C.11. Spinal Cord Injury and Plasticity

Support: NIH Grant NS104422
DoD SCIRP Grant W81XWH-18-1-0807

Title: Estrous cycle modulates the harmful effects of pain after spinal cord injury (SCI)

Authors: R. E. BAINE[1], E. D. GILES[2], ***J. GRAU**[2];
[1]Texas A&M Univ., Bryan, TX; [2]Texas A&M Univ., College Station, TX

Abstract: In addition to deficits in sensory/motor function, spinal cord injuries (SCI) often induce chronic pain and other debilitating acute and chronic complications. The initial insult to the cord incites secondary injury processes by engaging maladaptive inflammation and fueling hemorrhage at the site of injury, expanding the lesion site over the following days to weeks. While sufficient alone to produce these effects, pain associated with accompanying injuries (polytrauma) exacerbate the damage inflicted by the original trauma and inhibit functional recovery. This painful (nociceptive) stimulation has been studied experimentally through the administration of intermittent electrical stimulation to the tail or intradermal capsaicin to the hindpaw. Given the greater incidence of SCI in males, our research has historically utilized male rats, while others have used female rats for their lower susceptibility to bladder infection. However, the rising incidence of SCI in females necessitates greater understanding of the role of sex in response to injury. In studies examining sex-dependent effects, multiple strategies are employed to incorporate sex as a biological variable and account for differences in gonadal hormones between males and females. These approaches are complicated by fluctuations in female sex hormones across the estrous cycle—a concern commonly mitigated by randomly assigning females to experimental treatments. Our results demonstrate how this approach may obscure the impact of changes in hormones across the estrous cycle on SCI outcomes. Prior to SCI, female rats were balanced according to their stage of estrous cycle at time of injury. Twenty-four hours after a moderate contusion injury at T10-11, female rats received nociceptive input (shock or capsaicin) or nothing. One set of animals (n=8) was sacrificed 3 hours following treatments, with locomotor function (BBB) and blood pressure assessed at each hour after stimulation. A one-cm section of tissue centered on the lesion site was collected and analyzed for hemoglobin content with immunoblotting. Another group of rats recovered over a period of 28-days and changes in locomotor function and estrous cycle were assessed. Tissue was collected and stained for lesion analysis. We found that pain, regardless of type, negatively impacted behavioral function and exacerbated hemorrhage in female rats. Importantly, the stage of estrous cycle at time of injury modulated both the response to injury and the adverse effects of

nociceptive input, suggesting that fluctuations in sex hormones play an important role in SCI outcomes.

Disclosures: R.E. Baine: None. **E.D. Giles:** None. **J. Grau:** None.

Poster

P416. Stimulus Features and Coding: Animal to Computer Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P416.01

Topic: D.04. Somatosensation – Touch

Support: NIH/NNDS R01 Grant NS107599
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Title: Global, low amplitude cortical state predicts response outcomes in a selective detection task in mice

Authors: *K. MARRERO[1], K. ARULJOTHI[2], B. ZAREIAN[2], C. GAO[3], Z. ZHANG[1], E. ZAGHA[2];
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Abstract: Spontaneous neuronal activity strongly impacts stimulus encoding and behavioral responses. We sought to determine the effects of neocortical prestimulus activity on stimulus detection. We trained mice in a selective whisker detection task, in which they learned to respond (lick) to target stimuli in one whisker field and ignore distractor stimuli in the contralateral whisker field. During expert task performance, we used widefield calcium imaging to assess prestimulus and post-stimulus neuronal activity broadly across frontal and parietal cortices. We found that lower prestimulus activity correlated with enhanced stimulus detection: lower prestimulus activity predicted response versus no response outcomes and faster reaction times. The activity predictive of trial outcome was distributed through dorsal neocortex, rather than being restricted to whisker or licking regions. Using principal component analysis, we demonstrate that response trials are associated with a distinct and less variable prestimulus neuronal subspace. For single units, prestimulus choice probability was weak yet distributed broadly, with lower than chance choice probability correlating with stronger sensory and motor encoding. These findings support a low amplitude, low variability, optimal prestimulus cortical state for stimulus detection that presents globally and predicts response outcomes for both target and distractor stimuli.

Disclosures: K. Marrero: None. **K. Aruljothi:** None. **B. Zareian:** None. **C. Gao:** None. **Z. Zhang:** None. **E. Zagha:** None.

Poster

P416. Stimulus Features and Coding: Animal to Computer Models

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EU FLAG-ERA JTC 2017 Project GRAFIN
TÜBİTAK 2211-A Scholarship
YÖK 100/2000 Scholarship

Title: Classification of epidural field potentials from the hindpaw representation in S1 cortex of awake behaving rats.

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Abstract: Cortical neuroprostheses are crucial to provide movement capability and somatosensory feedback for patients with severe disorders, such as spinal cord injury and amyotrophic lateral sclerosis. Mini- and micro-ECoG grids are promising for that purpose with a trade-off between less invasiveness and selectivity compared to penetrating microelectrodes. Here, we recorded epidural field potentials (EFPs) with a wide bandwidth (4-312.5 Hz) by using 16-channel Pt-Ir surface arrays (site diameter: 25 μm) implanted on the hindpaw representation in S1 cortex of two awake Wistar albino rats. The rats performed a very simple task by pressing a lever to start a trial, in which vibrotactile stimuli with randomly selected amplitudes (12-111 μm) and frequencies (40 or 80 Hz) were presented for 0.6 s on the glabrous skin of the hindpaws. In some trials, no stimulus was presented. Time-frequency analysis (Morlet wavelet transform) showed evoked activity mostly at the onset (0-0.2 s) of the stimulus duration, consistent with our previous acute experiments. Wavelet coefficients for gamma (25-70 Hz) and high gamma (70-125 Hz) bands were used in machine learning algorithms to classify frequency (3 classes) or amplitude (4 classes) of the stimulus offline. Average squared coefficients in two bands and from 10 working channels were input as 20 features into four different algorithms: multinomial logistic regression (MLR), k-nearest neighbor, linear discriminant analysis (LDA), and support vector machine. The data were used as a training and a test set with 8:2 ratio and with 5-fold cross-validation. Classification accuracies from all the test algorithms were ~0.4-0.5 for each rat, but MLR and LDA yielded slightly better results. Specifically, class-averaged precision, recall, and F1 scores were somewhat better for predicting amplitude (LDA: 0.53, 0.50, 0.50) versus frequency (LDA: 0.41, 0.43, 0.41) in Rat #3. However, the opposite was true in Rat #4 (amplitude LDA: 0.39, 0.40, 0.38; frequency LDA: 0.47, 0.46, 0.46). Studies are underway to include other frequency bands and refine feature selection in the machine learning algorithms.

Classifying stimulus information in cortical surface potentials can help to evaluate the effects of somatosensory feedback and improve the closed-loop performance of a neuroprosthesis.

Disclosures: D. Kilinç Bülbül: None. **B. Güçlü:** None.

Poster

P416. Stimulus Features and Coding: Animal to Computer Models

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Topic: D.04. Somatosensation – Touch

Support: NINDS Grant NS122333

Title: Computational stereognosis: how should tactile and proprioceptive signals be integrated to support object recognition?

Authors: *P. MATHIALAGAN[1], A. SOBINOV[1], N. S. CHEREPANOVA[1], E. OKOROKOVA[1], S. J. BENSMAIA[2];
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Abstract: When we grasp and manipulate an object, we vividly sense its three dimensional shape via sensory signals from the hand and forearm, an ability termed stereognosis. At each point of contact with the object, tactile signals convey information about its local contours, for example surface curvature or an edge. The global shape of the object is computed from proprioceptive signals about the conformation of the hand. These two streams of information - cutaneous and proprioceptive - are then integrated to give rise to stereognosis, a process about which virtually nothing is known. To shed light on the underlying computations, we simulated grasping of more than 50 everyday objects and geometric shapes in a physics engine and trained a simple neural network to recognize them based on the tactile and proprioceptive signals. In brief, objects were presented to the hand in varying positions and orientations and were grasped with all or a subset of digits. We then trained single-layer networks to recognize the grasped objects based on the hand joint angles (proprioception) and contact locations (tactile). First, we trained a fully connected model and found that this network could accurately recognize objects across differences in grasp shape. Then, we trained several networks with restrictions on the nature of their connectivity, while maintaining the same number of fitted parameters to allow for performance comparisons across models. The constraints were: (i) nodes only received either tactile or proprioceptive information, but never both (no cross-modality integration); (ii) nodes only received information from a single finger (no cross-digit integration); (iii) nodes only received information from neighboring fingers (limited cross-digit integration). We found that the fully connected model performed substantially better than models where modalities or digits were segregated, but on par with the model that integrated information only from the

neighbouring fingers. We conclude that stereognosis requires the integration of proprioceptive and cutaneous signals across digits and relate these findings to the known properties of neurons in higher order somatosensory cortices.

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Poster

P416. Stimulus Features and Coding: Animal to Computer Models

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Program #/Poster #: P416.04

Topic: D.04. Somatosensation – Touch

Support: NSERC

Title: A Bayesian observer model for tactile spatial perception

Authors: *S. DEHNADI[1], D. GOLDREICH[2];
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Abstract: The human brain is remarkably adept at producing an accurate perception of the world from noisy and ambiguous sensory information. The very success of perception suggests underlying neural mechanisms, not yet understood, that achieve accurate inferences from imprecise sensory inputs. Bayesian inference provides an optimal method for drawing perceptual conclusions from sensorineural activity, but this approach has scarcely been applied in studies of tactile perception. We explored whether a Bayesian observer with access to spike counts of simulated cortical neurons with Gaussian receptive fields (RFs) and Poisson firing rate variability could replicate fundamental aspects of human passive tactile spatial perception. We ran the Bayesian observer on a battery of tactile tasks that test spatial perception using punctate stimuli delivered to the skin, and we compared the Bayesian observer's performance to previously collected human participant data. The Bayesian observer broadly replicated human performance on three tasks that assess tactile spatial acuity: classic two-point discrimination (C2PD), sequential two-point discrimination (S2PD), and two-point orientation discrimination (2POD). The Bayesian observer's performance confirmed that C2PD is the least reliable method of assessing tactile acuity due to the presence of non-spatial cues and that S2PD provides the most sensitive measure of spatial acuity. The performance of the Bayesian observer decreased on these tasks when, unbeknownst to the decoder, jitter was applied to the position, width, and amplitude of the RFs. Still, the Bayesian observer was in many cases robust against jitter, suggesting that the brain may not need to keep track of the exact characteristics of its RFs in order to achieve accurate inferences. The Bayesian observer was additionally able to replicate two illusions of tactile spatial perception: an adaptation-induced repulsion illusion and an

anisotropy illusion in which horizontal distances are judged to be greater than equal vertical distances. The Bayesian observer replicated these illusions only when the decoder was unaware of the adaptation state of the neurons and of the smaller horizontal than vertical separation between RFs. Taken together, these results suggest that the brain may produce perceptual inferences using a decoder that is unaware of the precise characteristics of its inputs.

Disclosures: S. Dehnadi: None. **D. Goldreich:** None.

Poster

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Support: This work was supported by the EU Horizon 2020 research and innovation program under grant agreement 813713 (NeuTouch).

Title: Studying the contribution of tactile afferent classes to information encoding using large scale simulations

Authors: *M. A. CASAL[1,2], G. CORNIANI[3], S. PANZERI[4,1], H. P. SAAL[3];
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Abstract: The spiking activity of tactile afferents in the human skin has been widely investigated, from their responses to simple or complex tactile stimuli to the relationship between spiking activity, brain processing, and elicited touch sensations. The vast majority of the studies only explore the activity of individual or a handful of afferents due to technical limitations imposed by the commonly used acquisition tools. However, the glabrous skin of the human hand is innervated by approximately 17,000 afferents, and thus any natural mechanical interaction is expected to activate hundreds of receptors. The human glabrous skin is innervated by four classes of tactile afferents, which traditionally have been viewed as functionally segregated. This notion has lately been challenged with evidence that most stimuli excite all classes of afferents, and most tactile percepts are shaped by multiple submodalities. Still, our understanding of tactile information encoding on a population level remains primitive. Here, we exploit a published computational model, Touchsim [1], to simulate the activity of thousands of peripheral tactile afferents of three classes in response to precise and well-defined stimuli. To pinpoint the contribution of the entire neural population in the stimulus encoding, we first used Non-Negative Matrix Factorization (NMF) to linearly decompose single-trial population spike trains into non-

negative basis functions and coefficients. Then, we asked whether afferents of different classes and at different locations on the hand send redundant or complementary information to the brain. To this end, we fed a generalized linear model with the compact representation of the population activity found by NMF aiming at decoding stimulus features. We found that a significant portion of the stimulus information is redundant, i.e. it is shared among different afferent classes. Still, single classes provide unique information. Consequently, we observed that for every stimulus feature considered, the decoding performances improved when considering a combination of classes rather than single ones. Our results suggest that analyses based on single afferents might not unveil the complexity of the tactile coding strategies, which is driven by joined efforts of all the afferents classes even with relatively simple stimuli. These results will pave the ground for a deeper understanding of the convergence and integration of tactile submodalities in the brain. [1] H. P. Saal, B. P. Delhaye, B. C. Rayhaun, and S. J. Bensmaia, "Simulating Tactile Signals from the Whole Hand with Millisecond Precision," PNAS of the United States of America, vol. 114, 2017, pp. E5693-5702.

Disclosures: M.A. Casal: None. **G. Corniani:** None. **S. Panzeri:** None. **H.P. Saal:** None.

Poster

P416. Stimulus Features and Coding: Animal to Computer Models

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Program #/Poster #: P416.06

Topic: D.04. Somatosensation – Touch

Title: Establishing the ferret as a model for whisker-based somatosensory processing

Authors: *T. BURNETT, D. H. O'CONNOR, K. J. NIELSEN;
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Abstract: Rodent and primate models of somatosensation have enabled key insights into neural mechanisms of touch. However, translating findings across species poses major challenges due in part to differences in sensory organs and cortical organization. Rodent work has most commonly used the whisker system, allowing precise quantification of sensory input and access to well-defined circuits. Non-human primate models allow the direct study of hand-based sensing and have a more elaborated set of somatosensory cortical areas that more closely resembles that of the human. Putting findings from rodent and primate studies into a common framework would be aided by having an additional model system that relies on whiskers but has a cortical organization more similar to that of primates. Ferrets (*Mustela putorius furo*), an established animal model in vision and auditory research, could serve this role. Ferrets possess whiskers, similar to mice, and research suggests a cortical organization typical of carnivores, including multiple somatosensory areas beyond S1. While previous research has shown conservation of the leminscal touch pathway in ferrets, our current knowledge of ferret

somatosensory cortex (S1) is incomplete with respect to the somatotopy, response properties, and cortical borders. We address these knowledge gaps with the aim of establishing the ferret as a new model for investigating touch processing. In this project, we used tungsten electrodes and cytochrome oxidase staining to investigate the organization and borders of S1. Through cytochrome oxidase staining, we delineated the approximate borders of S1 and determined that ferrets lack a barrel cortex. The potential borders were then verified through electrophysiology. Receptive fields of individual neurons were recorded to determine somatotopy of S1. In our recordings, as expected we found receptive fields corresponding primarily to the contralateral body, but also observed bilateral receptive fields in the trunk representation. In agreement with previous studies, medial S1 possessed the representation of the body, and lateral S1 contained the representation of the face. Within S1, we found robust responses to whisker deflections and observed receptive fields that contained a clear principal whisker as well as multi-whisker surrounds. Together, our work serves as a foundation to establish the ferret as a new model of whisker somatosensation.

Disclosures: T. Burnett: None. **D.H. O'Connor:** None. **K.J. Nielsen:** None.

Poster

P416. Stimulus Features and Coding: Animal to Computer Models

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Topic: D.04. Somatosensation – Touch

Support: NIH T32 NS-115705-01-0623
Del Monte Institute for Neuroscience

Title: The Role of Parvalbumin-expressing Inhibitory Interneurons on Spike Synchrony Between Feature Encoding Neurons of the Somatosensory Cortex

Authors: *M. DUHAIN[1], K. WANG[1], M. GOMEZ-RAMIREZ[2];
[1]Neurosci., [2]Brain & Cognitive Sci., Univ. of Rochester, Rochester, NY

Abstract: Object sensing and manipulation (i.e., haptics) requires discriminating myriad of tactile features through the sense of touch. Key to this process are mechanisms of feature-based attention, which provide preferential processing of attended sensory features, while filtering out sensory information representing non-attended features. A previous study showed that selection of behaviorally-relevant features is represented in the synchronized spiking between neurons tuned for the attended feature. However, the underlying circuit mechanism generating this feature-specific spike synchrony effect is yet to be elucidated. We propose that this feature-specific spike synchrony effect is generated by a parvalbumin (PV) and pyramidal microcircuit, whose cells are tuned for the same tactile feature. We hypothesize that feature tuned PV

interneurons generate local inhibition in similarly-tuned excitatory cells, followed by a short window of high spiking probability that promotes co-activation of the feature tuned pyramidal ensemble. To test this circuit and its dynamics, we trained mice to perform a 2-alternative forced choice task requiring discrimination of vibration frequencies delivered to the forepaws. Animals were visually cued to discriminate stimuli in one paw, while ignoring all stimuli in the uncued location. We will make acute neural recordings in somatosensory cortex while animals perform this task. PV cells will be identified based on waveform classification analyses. We will determine whether correlated spiking between putative-PV and regular spiking cells is increased for pairs tuned for vibration features. This project aims to identify a novel functional circuit within somatosensory cortex that implements feature-based attention to enable preferential processing of attended features within local and cross-cortical areas.

Disclosures: M. Duhain: None. **K. Wang:** None. **M. Gomez-Ramirez:** None.

Poster

P452. Central Encoding of Self-Motion: Vestibular Signals

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Program #/Poster #: P452.01

Topic: D.06. Auditory & Vestibular Systems

Support: NSERC CGSM

Title: Neuronal sensitivity and variability mediate 3 parallel strategies for encoding natural translations in the primate vestibular system

Authors: *M. PROSTEBBY[1], J. CARRIOT[2], I. MACKROUS[3], M. J. CHACRON[3];
[1]McGill Univ. Integrated Program in Neurosci., Montreal, QC, Canada; [2]Physiol., [3]McGill Univ., Montreal, QC, Canada

Abstract: As we navigate and explore our environment, our motions can be described as a combination of linear translational motion and rotational (angular) motions. Everyday movements (e.g. walking, playing soccer) are composed of these two independent forms of motion which occur with characteristic statistics. Our previous investigations show that vestibular sensory neurons at the first stages of central processing are adapted to match their tuning curves to these characteristic statistics in the rotational component of natural motion. These neurons within the vestibular nucleus of the brainstem (VN) produce responses which are less redundant, and which are independent of frequency (i.e. temporally whitened). However, the responses of VN neurons which are sensitive to the translational component of natural motion (otolith VOs) have not been explored. Moreover, their increased heterogeneity in tuning curve profiles to artificial stimuli as well as differences in the sensitivity of their incoming afferents together comprise key differences which may change their coding strategy. Our study explores

whether, and if so, how the responses of otolith VOs in the VN of awake, behaving rhesus macaques are adapted to process naturalistic translations. Our results demonstrate that otolith VOs do not always employ efficient coding strategies matching natural statistics, instead responding as one of three classes ranging from strong low pass to optimally whitened. Thus, this study is the first to explore otolith VO responses and reveals 3 parallel encoding strategies used in the processing of translational motion which may relate to slow-varying head motions relative to gravity and faithful stimulus reconstruction needed for navigation and self-righting reflexes.

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Poster

P452. Central Encoding of Self-Motion: Vestibular Signals

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Topic: D.06. Auditory & Vestibular Systems

Support: NSF Grant # OIA-1920896
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Title: Natural statistics of head orientation relative to gravity in humans: implications for modeling spatial orientation

Authors: *C. SINNOTT[1], P. HAUSAMANN[2], P. R. MACNEILAGE[3];
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Abstract: The orientation of the head relative to gravity modulates both visual and vestibular sensory stimulation. In particular, the otoliths of the vestibular system are stimulated by the sum of linear acceleration driven by both self-motion and Earth's gravity. The nervous system must decompose this summed signal into gravitational and inertial components to generate independent estimates of orientation and linear self-motion. Various approaches have been proposed to model this process including frequency segregation, Bayesian estimation, and observer models. All of these approaches incorporate parameters representing prior knowledge of head orientation and motion statistics. While statistics of summed linear acceleration recorded by inertial measurement units (IMUs) have been reported previously, the separate statistics of gravitational and inertial components are generally not reported. In the current study we present measurements obtained using the Intel Realsense T265, a tracking camera that uses both visual and inertial data to generate robust estimates of both gravitational and inertial acceleration, i.e. head orientation and linear head motion. Ten clinically normal participants wore the T265 on

their heads during five hours of unstructured, everyday activity. The T265 was connected to a laptop worn in a backpack. Across all participants, distributions of head orientation were generally centered close to upright, as might be expected. Distributions were more variable in pitch than roll, and pitch distributions were skewed towards downward angles. Data were then separated into low- and high-velocity subsets to generate separate distributions for stationary and locomotor epochs. Distributions were generally more variable during locomotor than stationary epochs. These distributions were then incorporated as empirical priors in a Bayesian model that predicts biases in orientation perception that depend on shape and variability of the prior. The model using empirical priors generally predicted biases that were reduced relative to those observed in previous studies, but it did predict previously observed asymmetries in biases for pitch perception, consistent with pitch distribution skewness. In addition, power spectra of gravitational, inertial, and sum total linear acceleration revealed a crossing point just below 1Hz, with power of gravitational acceleration matching total acceleration power below this frequency, and power of inertial acceleration matching total acceleration above this frequency, providing an empricial basis for the cutoff frequency to be applied in frequency segregation models of vestibular processing.

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Poster

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Topic: D.06. Auditory & Vestibular Systems

Support: the National Natural Science Foundation of China Project
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the Shanghai Municipal Science and Technology Major Project

Title: Robust vestibular self-motion signals in macaque posterior cingulate region

Authors: *B. LIU[1,2], Q. TIAN[1,2], Y. GU[1,2];
[1]CAS Ctr. for Excellence in Brain Sci. and Intelligence Technology, Key Lab. of Primate Neurobiology, Inst. of Neuroscience, Chinese Acad. of Sci., Shanghai, China; [2]Univ. of Chinese Acad. of Sci., Beijing, China

Abstract: Vector-based navigation (also known as path integration) is essential for organisms to survive and explore the world, especially in the absence of effective external landmarks. Self-motion signals, which have been found to be ubiquitously distributed across parietal-temporal lobes, are thought to provide the most important idiothetic information for path integration. However, how they propagate to the hippocampal system for vector-based navigation is still up

in the air. We proposed that the information is transmitted via hubs including posterior cingulate cortex (PCC) and retrosplenial cortex (RSC) to the navigation circuit. Although numerous studies have indicated that posterior cingulate areas are involved in spatial tasks, it is unclear how their neurons represent self-motion signals. Providing translation and rotation stimuli to macaques on a 6-degree-of-freedom motion platform with a sophisticated virtual-reality system, we discovered robust vestibular responses in PCC with significant temporal and spatial tuning. The preference of horizontal plane with a tendency of leftward and rightward in translation indicating the potential heading discrimination function of PCC, which is further confirmed by PCA analysis. In rotation, the distributed preferred axis tends to be around pitch and roll, but not yaw, which indicating a potential role of azimuth tuning of head direction cells in the three-dimensional (3D) environment according to a previous work. Moreover, we decomposed temporal and spatial components of neural response by a combined 3D spatiotemporal model. The model captured data well and revealed multiple temporal components including velocity, acceleration, jerk, and position with spatial modulation. Compared to PCC, RSC contained moderate vestibular temporal modulations and lacked significant spatial tuning. Visual self-motion signals were much weaker in both regions compared to the vestibular ones. We conclude that macaque posterior cingulate region carries vestibular-dominant self-motion signals with plentiful temporal components that could be useful for path integration.

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Poster

P452. Central Encoding of Self-Motion: Vestibular Signals

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Program #/Poster #: P452.04

Topic: D.06. Auditory & Vestibular Systems

Support: CIHR Grant 162285
NIH Grant R01-DC002390
NIH Grant R01-DC018061

Title: Population coding of naturalistic self-motion in vestibular nucleus and ventral posterolateral thalamus

Authors: *M. MOHAMMADI[1], I. MACKROUS[1], G. MCALLISTER[1], J. CARRIOT[1], K. E. CULLEN[2], M. J. CHACRON[1];
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Abstract: The vestibular system detects head motion in space and provides information about self-motion perception as well as reflexes required for stabilization of posture and gaze. While

the physiology of the vestibular system has been well characterized via single-unit recordings, how the activity of neurons form perception and generate reflexes is not well understood as perception and behavior, in general, emerge from the collective activity of the population of neurons. Recent studies have investigated population coding in the vestibular pathway by recording simultaneously from multiple vestibular-only (VO) neurons in vestibular nuclei (VN). However, the stimuli used in these studies were either artificial or low in amplitude and thus strongly differed from natural self-motion stimuli that are characterized by higher amplitudes and a spectrum of frequencies. Accordingly, we investigated population coding in the vestibular system by recording simultaneously from multiple neurons in the vestibular pathway and characterizing their response during rest, artificial, and naturalistic head motion stimulation. In particular, we measured signal and noise correlations between VO neurons. VO neurons are thought to mediate vital reflexes and perception as they project to the spinal cord and thalamus, respectively. Furthermore, we will record from target neurons in the ventral posterolateral nucleus (VPL) during rest, artificial, and naturalistic head motion stimulation to determine how information is being decoded from neural populations in VN and whether/how it is influenced by correlation structure in VN. Our analysis so far shows that, under naturalistic stimulation, VO pairs displayed strong signal but negligible noise correlations thereby minimizing redundancy. These results are expected to provide critical insight for developing effective neuroprosthetics for patients with vestibular deficits.

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Poster

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Title: Coding strategies for representing natural self-motion through the vestibular pathway

Authors: J. CARRIOT[1], I. MACKROUS[3], K. E. CULLEN[4], M. J. CHACRON[2];
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Abstract: Understanding the neural mechanisms by which sensory input gives rise to behavior requires knowledge of how the sensory representations of this input evolve between successive

brain areas. Here we investigated how natural self-motion stimuli are represented in ascending vestibular pathways ranging from more peripheral to central cortical areas contributing to both vital reflexes as well as spatial perception. Recordings were made from neurons within the vestibular nuclei (VN), vestibular thalamic neurons within the ventral posterior lateral (VPL) Thalamus and neurons within the parieto-insular vestibular cortex (PIVC) during both artificial and naturalistic self-motion. Our results show that neurons within ascending vestibular pathways gradually become more optimized to natural stimuli. Further, our results demonstrate radical changes in neural coding strategy of self-motion across successive brain areas. Specifically, while VN neurons transmit information through a combination of changes in firing rate and precise spike timing, VPL and PIVC neurons instead represent self-motion exclusively through changes in firing rate. Finally, we found that information in this thalamocortical pathway, in contrast to postural reflex pathways, optimally represents the instantaneous head velocity. Taken together, our results suggest that dynamic information carried by precise spike timing is used to mediate vital reflexes in order to maintain posture, while information carried by firing rate is further optimized across brain areas in order to faithfully and optimally code a neural representation of instantaneous head velocity. Our results thus provide the neural basis for previous behavioral studies showing that self-motion perception is centered on head velocity rather than another sensory representation.

Disclosures: J. Carriot: None. **I. Mackrous:** None. **K.E. Cullen:** None. **M.J. Chacron:** None.

Poster

P452. Central Encoding of Self-Motion: Vestibular Signals

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P452.06

Topic: D.06. Auditory & Vestibular Systems

Title: Behaviourally dependent gating of sensory response for early vestibular processing in mouse

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Abstract: Vestibular afferents respond identically to externally-applied (vestibular exafference) and self-generated (vestibular reafference) movements. However, modulation at the next stage of vestibular processing occurring in the vestibular nuclei (VN) is markedly suppressed during self-generated movement via a cerebellar-based mechanism in both monkeys and mice (Roy & Cullen 2001; Medrea & Cullen, 2013; Brooks & Cullen, 2014). In particular, this suppression is specific to a class of neurons termed vestibular-only (VO) neurons that project to the spinal cord for mediating the vestibulo-spinal pathways and to the vestibular thalamus. Previous work done

in monkeys elegantly showed that when proprioceptive feedback resulting from self-generated motion is disrupted, VO neurons cannot distinguish between self-generated and externally-applied motions, thereby encoding both passive and active components of movement. To date, however, it remains unknown whether neurons in the VN of rodents show the behaviorally dependent gating of vestibular responses observed in a non-human primate model. We hypothesized that modulation in VO neurons in mouse, as in primate, will be suppressed when there is a match between the predicted and actual consequence of voluntary head-movement but not when there is a mismatch. Accordingly, we recorded the activity of VO neurons with the 960-channel high-density Neuropixel 1.0 probes when perturbations were applied to the mouse head while it made active head movements. Mice were trained to move their head to the left or to the right upon the presentation of an auditory cue. Head-restrained behavioral protocols (Beraneck & Cullen 2007; Medrea & Cullen, 2013) were used to characterize the passive vestibular sensitivity of VO neurons. Then to confirm neurons were insensitive to eye motion, we simultaneously recorded eye movement with an AMR magnetic field sensor. Finally, the responses of VO neurons were recorded during 3 conditions in which (1) the mouse was passively rotated, (2) the mouse actively rotated its head, and (3) the mouse head was subjected to an expected perturbation (adding load to the head) while making active head movements. Head velocity sensitivity of the VO neurons estimated from each condition was compared. Our preliminary results indicate for the first time that neurons in the VO neurons of mice robustly encode head velocity signal when a perturbation is introduced during head-free behaviour (a mismatch between expected and actual sensory feedback). Overall, this present study has specific implications for understanding how the brain ensures accurate posture, motor control, and spatial perception.

Disclosures: H. Chang: None. **A. Lawrence:** None. **K.E. Cullen:** None.

Poster

P452. Central Encoding of Self-Motion: Vestibular Signals

Location: Virtual Only

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Program #/Poster #: P452.07

Topic: D.08. Visual Sensory-motor Processing

Support: NIGMS of NIH Grant P20 GM103650
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Title: Visual-vestibular conflict detection with central versus peripheral visual stimulation

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Abstract: Stable perception depends on mechanisms which compare visual and vestibular cues to self motion and evaluate them for congruence. Conflict between signals can result in motion or simulator sickness. Here we investigate how characteristics of an observer's field of view (FOV) impact their ability to detect visual-vestibular conflict. Experiments were conducted using a head-mounted display (HTC Vive Pro Eye) which displayed a virtual optokinetic drum and tracked head and eye movements. In a 2x2 design, subjects performed one type of fixation behavior (head or scene-fixed) with one type of field of view restriction (peripheral or central) in each condition. Subjects fixated a target during the trial which remained either stationary relative to the environment (scene-fixed) or centered relative to their FOV (head-fixed). The FOV was restricted either by a virtual scotoma that encompassed the central ~40 deg of visual angle (peripheral stimulation) or reduced the FOV to ~40 deg (central stimulation) centered on the fixation point. During each trial, subjects made yaw head movements of between 15 and 50 deg over ~1.5 sec and the gain of the visual scene motion was manipulated to move slower or faster than the subject's head movement, and subjects were asked to report whether the environment appeared to be moving with (gain < 1) or against (gain > 1) their head movement. Fitting a psychometric function to the resulting data yields the gain perceived as stationary (PSE) and the range of gains that are compatible with perception of a stationary visual environment (JND). Our results show lower visual gain (low PSEs) in the head-fixed fixation conditions suggesting that smooth pursuit eye movements resulted in greater perceived speed of scene motion. Sensitivity to conflict was also increased (lower JND) in the scene-fixed and central stimulation conditions, suggesting naturalistic eye movements and the central visual field are important for detecting sensory conflict. Results from SSQ and discomfort score data show lower subjective levels of sickness in conditions where the periphery was obscured, consistent with the known effect of FOV restriction on simulator sickness.

Disclosures: S. Halow: None. **A. Hamilton:** None. **E. Folmer:** None. **P. MacNeilage:** None.

Poster

P452. Central Encoding of Self-Motion: Vestibular Signals

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Program #/Poster #: P452.08

Topic: D.06. Auditory & Vestibular Systems

Support: NIH Jackson lab MOUSE 90086979

Title: Neural variability, information encoding, and noise correlations in the mouse vestibular nuclei

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Abstract: In primates, regular semicircular canal afferents encode more average information about the stimuli across the behaviourally relevant frequencies ranging from 0 to 20 Hz compared to their irregular counterparts (Sadghei et al. 2007). At the next stage of vestibular stimuli processing, the vestibular-only (VO) neurons in the vestibular nuclei (VN), which contribute to the posture control and self-motion perception, show even lower information transmission capabilities. This occurs due to the higher variability in their responses when compared to even irregular afferent neurons (Massot et al., 2011). It has been suggested that the high neuronal variability of VO neurons is beneficial for the encoding of highly transient movement information. Indeed, recent studies have shown that these neurons use precise spike timing on the order of milliseconds to encode highly dynamic stimuli (Jamali et al. 2016). The mouse has become an increasingly popular system in neuroscience research. Single-unit recordings in the vestibular nuclei have shown that mice VO neurons show similar responses as the primates for low-frequency sinusoidal rotations but have lower sensitivities. To date, however, neither their information coding nor responses dynamics have been studied over the frequency range of natural head movements. We hypothesized that the high neuronal variability and lower sensitivities of mouse VO neurons would decrease their overall information capacity relative to monkey VO neurons. To test this proposal, we used the neuropixel probes to record the activity of VO neurons in the medial VN by subjecting the mice to a 1) broadband noise perturbation of 0-20 Hz frequency range 2) sinusoidal stimuli at 0.5 Hz in the yaw plane. As in monkeys, the mice VO neurons (n=3) in this study had lower coding fractions and higher gain and phase than did their afferent inputs. Our preliminary analysis further showed that the mouse VO neurons had similar mutual information density and coding fractions across the physiologically relevant frequency range of 0 to 20Hz. Finally, our analysis of population coding during sinusoidal stimulation using neuropixel probes revealed significant excess synchrony between the firing of mouse VO neurons (n=15 pairs; 0.0510±0.03), which was significantly greater than previously seen in the monkey using lower density probes. Taken together, our findings have important implications in understanding the neural coding strategies and information transmission in the neural circuitry of the mouse vestibular pathways.

Disclosures: A. Lawrence: None. **H.V. Chang:** None. **K.E. Cullen:** None.

Poster

P452. Central Encoding of Self-Motion: Vestibular Signals

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Program #/Poster #: P452.09

Topic: D.06. Auditory & Vestibular Systems

Support: NIH Grant R01-DC002390

Title: Effect of varying levels of sensory prediction error on the suppression of reafferent signals in the deep cerebellar nuclei

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Abstract: The ability to distinguish between self-generated (reafference) vs. externally-applied (exafference) sensory information is fundamental for ensuring accurate motor control as well as perceptual stability. This ability is particularly relevant in the context of the vestibulo-spinal reflexes, which are essential for providing postural responses to unexpected perturbations yet would impede voluntary movement. Accordingly, the brain suppresses vestibulo-spinal reflexes during active movements. To do this, it computes a prediction of the sensory consequences of the motor command, which is then compared with the actual sensory input. In the conditions where there is a match as would be the case during active movements, vestibulo-spinal reflexes pathways are suppressed (i.e. vestibular reafference is canceled). In contrast, in conditions when an unexpected head perturbation is experienced, there is a mismatch. As a result, the vestibular responses of neurons in the deep cerebellar nuclei and vestibular nuclei, which contribute to vestibulo-spinal reflexes, robustly encode head motion (i.e. vestibular exafference is not canceled; reviewed in Cullen and Zobeiri, 2021). To date, however, experiments have only considered conditions in which large perturbations were applied. This raises the question of whether systematically perturbing active head movements so they span movements both within and outside the range of natural variability will result in neurons encoding actively-generated vestibular inputs as reafference and exafference, respectively. To address this, we recorded neuronal responses of DCN neurons (rostral fastigial) during an experiment in which we randomly applied assistive or resistive perturbations to the head during a fraction of active head movement trials. The torques applied during these perturbation trials were calibrated, so the smallest perturbation changed head velocity within the band of natural variability, while the largest levels approximately doubled (assistive) or halved (resistive) head velocity. We found that on average DCN neurons showed reduced reafferent suppression in response to both the assistive and resistive perturbation. Notably, we observed increased sensitivity even for the small loads that changed head velocity within the range of natural variability. These findings suggest that even small externally-applied head movement perturbations result in a proportional shift in encoding from reafferent to exafferent. Taken together, these data reveal rules underlying cerebellar-based processing of sensory prediction error.

Disclosures: O. Zobeiri: None. **R.L. Mildren:** None. **K.E. Cullen:** None.

Poster

P452. Central Encoding of Self-Motion: Vestibular Signals

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Program #/Poster #: P452.10

Topic: D.06. Auditory & Vestibular Systems

Support: KAKEN grant 20H04286

Title: Tilt-translation discrimination in goldfish vestibulo-ocular reflex

Authors: *S. TADOKORO[1,2], Y. SHINJI[1], M. NAKANO[1], Y. HIRATA[1,3,4];
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Abstract: As stated by Einstein's equivalence principle, the otolith responds indistinguishably to an inertial motion (e.g., lateral translation) and a change in head orientation relative to gravity (e.g., roll tilt) (Fernandez et al., 1972). This is called tilt-translation ambiguity. Nonetheless, we can perceive these two self-motions distinguishably in the light as well as in the dark while producing compensatory vestibuloocular reflexes (VORs) differentially, i.e., horizontal (H) VOR for lateral-translation, and torsional (T) VOR for roll tilt (Koizuka et al., 1999). Therefore, the ambiguity must be resolved in the brain using extra-otolith signals (Merfeld et al, 1999; Angeraki et al., 1999). In parallel with the perception and VOR behavior, translation-selective cells have been found in the vestibular and cerebellar nuclei among other areas, while in the caudal vermis both translation-selective and tilt-selective Purkinje cells have been identified (Yakusheva et al., 2007; Laurens et al., 2013). However, it is still enigmatic how these brain areas solve the tilt-translation ambiguity problem. Here we investigated the VOR elicited by tilt-translation combined stimuli in goldfish to reveal the neural mechanism for solving the problem. Goldfish is one of the animal species most thoroughly understood for visual and vestibular behavior, and their underlying neuronal structure/function. Specifically, their angular VOR has been found to be induced robustly around all 3D axes. Presently we show that goldfish produce vertical (V) VOR in response to both roll tilt and lateral translation in the dark, appearing that their VOR does not distinguish the two motions. However, in lateral-eyed animals VVOR is appropriate for both motions if they are to stabilize the visual scene from the bottom. We hypothesized that they would stabilize the vision from the front or the side by producing H or TVOR, respectively if trained adequately by using visual-vestibular combined stimuli. Because optokinetic response (OKR) to visual stimulation in goldfish has been characterized only for horizontal rotation, we evaluated V and TOKR in preparation to test the hypothesis. We employed a custom-made virtual reality system that enables full-field optokinetic visual stimuli (OKS) around all 3D axes. We found that gains for V and TOKR in naïve animals are very small in contrast to robust HOKR. Nevertheless, their gains increased after prolonged exposure to OKS around each axis. Based on these results, we envision that goldfish VOR would distinguish tilt from translation after visual-vestibular combined training that updates internal models and Bayesian priors used to estimate self-motion.

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Poster

P452. Central Encoding of Self-Motion: Vestibular Signals

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Program #/Poster #: P452.11

Topic: D.06. Auditory & Vestibular Systems

Support: CIHR Grant 162285
NIH Grant R01-DC002390
NIH Grant R01-DC018061

Title: Second-order attribute of head motion is encoded at single-neuron level in the vestibular nuclei.

Authors: *I. MACKROUS[1], J. CARRIOT[1], K. E. CULLEN[2], M. J. CHACRON[1];
[1]McGill Univ., Montreal, QC, Canada; [2]Dept. of Biomed. Engin., The Johns Hopkins Univ., Baltimore, MD

Abstract: The vestibular system encodes head motion during our daily activities giving rise to vital autonomous reflexes as well as self-motion perception. Previous studies have shown that natural head motion stimuli are characterized by complex spatio-temporal attributes that differ from artificial stimuli (i.e., sinewaves). Specifically, natural self-motion stimuli consist of a fast time-varying waveform (i.e., first order) whose amplitude (i.e., second order or envelope) varies more slowly. Previous studies have shown that vestibular afferents can respond to the envelope only for higher amplitudes, yet behavioral studies have shown that human subjects can perceive changes even at low amplitudes. In order to investigate this apparent discrepancy, we recorded from downstream vestibular-only (VO) neurons within the vestibular nuclei in response to stimuli consisting of a noisy waveform whose envelope varied sinusoidally at lower frequency. Importantly, stimulus amplitude was kept low such as not to elicit static nonlinearities and thus no envelope response from afferents (e.g., rectification, saturation). Overall, we found that VO neurons robustly encoded the envelope with a gain that was independent of frequency. Interestingly, VO neural responses increasingly lagged the envelope stimulus as frequency increased. Computational modeling revealed that envelope responses in VO neurons were not due to static nonlinearities, instead it could be due to either a dynamic nonlinearity or are caused by external input. Further, we computed neural amplitude discrimination thresholds. Overall, our results show that the activities of~80 neurons are necessary to account for perception, which is much greater than what was found previously in the thalamus (Carriot et al., 2021). Overall, our results show that self-motion stimuli amplitude is encoded at single-neuron level in the vestibular nuclei and that amplitude discrimination begins at that level. This has critical implications for our understanding of how early processing of vestibular signal can encode second-order attributes of head motion that will further be used to ensure stable perception of the world and accurate motor control.

Disclosures: I. Mackrous: None. **J. Carriot:** None. **K.E. Cullen:** None. **M.J. Chacron:** None.

Poster

P452. Central Encoding of Self-Motion: Vestibular Signals

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Program #/Poster #: P452.12

Topic: D.06. Auditory & Vestibular Systems

Support: NSERC RGPIN-2020-05438

Title: Perceptual vestibular integration is influenced by the encoding vestibular end-organs

Authors: *A. CHEN, C. KUO, J.-S. BLOUIN;
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Abstract: The vestibular end-organs encode head linear and angular acceleration in space, relaying signals critical to our perception of self-motion. Our internal representation of self-rotation is thought to depend on semi-circular canal encoding and a central integration process (velocity storage). Velocity storage enables the prolongation of our internal representation of rotation after signals encoded by semi-circular canal afferents have adapted. Computational models explaining the velocity storage process all assume its operations are independent of the rotational axis. Physiological data, however, demonstrate that the axis of head rotation influences the perception of head rotation. The purpose of our study was to determine if the contribution of distinct vestibular end-organs influence the duration of perceived self-rotation to constant velocity stimuli. We recorded perceived rotation while varying the vestibular end-organs (semi-circular canals and otoliths) encoding head rotation by changing head orientation relative to an Earth-vertical rotational stimulus. First, we tested if the number or type of semicircular canal pair(s) activated were responsible for changes in velocity storage duration (n=16). Perceptual responses suggested that the number or type of semicircular canal pair(s) activated did not fully explain the results. We performed an additional experiment (n=9) to determine whether differential otolithic signals contribute to the perception of self-rotation by comparing three head orientations that altered otolith contributions to head rotation while maintaining similar semi-circular canal contributions. The perception of self-rotation was shorter when otolithic contributions were minimized. We propose a non-linear vestibular model discretizing the relative contribution of vestibular end-organs to explain our inference of self-rotation.

Disclosures: A. Chen: None. **C. Kuo:** None. **J. Blouin:** None.

Poster

P452. Central Encoding of Self-Motion: Vestibular Signals

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Support: NIH Grant DC015091
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American Otological Society

Title: Modulation of the vestibular nerve response by GABA-B activation

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Abstract: Vestibular end organs in the inner ear receive GABAergic inputs from efferent fibers and/or supporting cells. We have previously shown that activation of GABA-B receptors results in an excitatory effect on calyx nerve terminals. Here, we used *in vitro* patch clamp recordings from calyx afferent terminals to further explore the effect of baclofen, a GABA-B agonist, on the response properties of calyx terminals and nerve fibers. We injected a sweep of sines (0.01-100 Hz) or single sines (at different frequencies from 0.1 - 100 Hz) to the calyx before and after baclofen application. Following the activation of GABA-B receptors, there was an increase in the frequency range that calyces responded to (i.e., fired action potentials) and an overall increase in the gain of the response. The phase of this response decreased. Overall, the calyces responded faster and at higher rates, with stronger phase locking to the stimulus. Consistent with these findings, *in vivo* intracochlear injection of baclofen in anesthetized mice produced an increase in synchronized response of irregular afferents to a fast linear movement, as measured by vestibular sensory evoked potentials (VsEP). Intracochlear injection of a GABA-B antagonist (CGP 35348) had the opposite effect, suggesting the presence of spontaneous release of GABA in the vestibular periphery. Together, these results provide evidence for a modulatory role for GABAergic inputs in the vestibular periphery, which could modulate the phasic properties of irregular afferents.
These studies were funded by NIH DC015091 and DC019380 grants, and a research grant from the American Otological Society to SGS.

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Poster

P472. Functional Architecture of the Visual Cortex

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Program #/Poster #: P472.01

Topic: D.07. Vision

Support: MSCA-843379

Title: Spatial dimension of resonance phenomena in mouse primary visual cortex

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Abstract: Sensory cortices stimulated by rhythmic sounds, lights, or touch will respond in a rhythmic manner, with frequencies identical or harmonically related to the stimulus. Although such responses are preserved across species, reported temporal frequency tuning curves in human and mouse primary visual cortex (V1) do differ. In human EEG recordings (macroscale), V1 shows temporal frequency tuning with characteristic resonance peaks at around 10, 20, and 40 Hz, whereas only a single peak below 8 Hz is present in temporal tuning curves derived from multiunit and spiking activity (microscale) in mouse V1. By imaging transgenic animals expressing high temporal resolution fluorescent glutamate reporter iGluSnFR, we sought to characterize temporal frequency tuning in awake mouse V1 and its spatial distribution at the mesoscale. For this, we used a wide range of sine-wave modulated temporal frequencies (2-72 Hz). We found that mouse V1 responded to flickering light as high as 52 Hz - a limit dictated by the kinetics of iGluSnFR fluorescent reporter. Most importantly, temporal tuning curves showed resonance peaks at 8 Hz, 15 Hz, and 33 Hz, indicating that resonance phenomena in mouse V1 highly resemble that of humans. We also observed a variation in temporal frequency preference across V1: (1) lower flicker frequencies activated larger portions of V1 as compared to higher flicker frequencies; (2) all temporal frequencies elicited a retinotopic response in V1, with additional spatial peaks present at certain temporal frequencies. The latter finding suggests some spatial organization of temporal frequency responses in mouse V1. In conclusion, temporal frequency tuning with prominent resonance peaks in humans and in mice exhibit a high degree of similarity.

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Poster

P472. Functional Architecture of the Visual Cortex

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Program #/Poster #: P472.02

Topic: D.07. Vision

Support: NIMH R01MH116500

Title: Theta synchrony between mouse primary and lateromedial visual cortices following visual experience

Authors: *Y. TANG[1,2,3], A. A. CHUBYKIN[1,2,3];
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Abstract: Visual cortical circuits undergo dynamic functional and structural changes following visual experience. In the mouse primary visual cortex (V1), 4-8 Hz oscillations emerge following passive visual experience and persist beyond the visual stimulus time window. Such low-frequency activity has been associated with working memory. Its expression depends on the familiar spatial frequency content, suggesting that this resonant activity may involve functionally similar neuronal ensembles. Higher-order visual areas (HVA) in mice are also selective to distinct spatial and temporal features of the visual stimuli. We hypothesized that the persistent 4-8 Hz oscillations might be the underlying working memory process within V1 and the feature-specific HVAs. By recording in V1 and lateromedial area (LM) simultaneously using silicon probes in awake mice, we found stronger 4-8 Hz phase synchrony between these two areas following visual experience. This synchrony was higher when the visual stimulus spatial and temporal features were preferred by LM. V1 units that fired during the first two 4-8 Hz cycles were better phase-locked to the 4-8 Hz LM activity, while LM units that fired after but not during the first cycle were better phase-locked to the 4-8 Hz V1 activity. Furthermore, we found that V1 and LM units within the superficial layers showed higher correlated firing following the experience. These results demonstrated that visual experience induced persistent and synchronized 4-8Hz oscillations between V1 and LM. This finding suggests that the 4-8 Hz persistent oscillations emerging after experience may synchronize visual cortical areas based on their functional specificity to maintain information during working memory.

Disclosures: Y. Tang: None. **A.A. Chubykin:** None.

Poster

P472. Functional Architecture of the Visual Cortex

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Topic: D.07. Vision

Support: NIH Grant DP1-NS083063
HHMI

Title: A physiological and anatomical survey of the visual cortex in the northern treeshrew

Authors: *J. WEKSELBLATT[1], F. LANFRANCHI[3], M. SHAPIRO[2], D. Y. TSAO[1];
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Abstract: The macaque monkey has classically provided a key model for visual processing, due to its amenability to psychophysical tasks and well-delineated visual hierarchy. More recently, the rodent has emerged as a new model for visual processing, due to the availability of molecular and genetic tools. An ideal model system would satisfy both needs: amenability to complex psychophysical tasks and tractability for molecularly-based recording and perturbation techniques. Towards this goal, we have embarked on an effort to establish the northern treeshrew as a model organism for study of high-level object vision.A diurnal animal, the treeshrew has high visual acuity (>10x that of rodents), a cone-dominant retina, and a columnarly-organized visual cortex. Moreover, treeshrew cortex is lissencephalic, enabling increased optical access for imaging neural circuit dynamics compared to the macaque. Due to its small size (~150-200 grams) and relatively short reproductive and developmental cycles (6 weeks gestation, 4-6 months from birth to adulthood, 1-6 offspring per litter), the treeshrew offers experimental and genetic accessibility similar to rodents.We have begun high-throughput recordings using up to three simultaneously implanted neuropixel electrodes in different areas along the treeshrew visual hierarchy including primary visual cortex (V1), secondary visual cortex (V2), and inferotemporal cortex (IT). We report our initial results from over 1000 active recording sites per session in awake animals viewing various classical stimuli including simple oriented bars, Simonchelli textures, and a large number of objects. We find differential modulation across brain areas and stimulus sets, including replication of findings from primates suggesting a particular functional role for V2 in the representation of natural image structure. We also report our findings on connectivity patterns in visual cortex of treeshrew using a combination of retrograde tracing (cholera toxin subunit B), anterograde tracing (adeno-associated virus), and cytochrome oxidase staining. We have developed a cortical flatmount preparation which allows tangential slices through the entirety of each hemisphere, providing a full view of the projections to and from several cortical areas for which we have electrophysiological data.This work is supported by HHMI.

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Poster

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Topic: D.07. Vision

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R21 EY030297

Title: Intrinsic neural dynamics predict category selectivity in ventral visual cortex

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Abstract: The intrinsic dynamics of neural populations constrain the computational processes by which regions process information and the timescales over which they integrate information. Further, different domains of perceptual information require different information processing capacities. Therefore, we hypothesized that the intrinsic dynamics of ventral visual regions should predict the category of visual stimuli that those regions selectively process. We tested this hypothesis using 1,350 intracranial electroencephalography contacts implanted within ventral visual cortex of 41 patients with intractable epilepsy. Patients completed a 1-back task where they viewed pictures of bodies, words, faces, tools, houses, and phase-scrambled images. We then tried to predict which ventral visual contacts would demonstrate selective neural activity for each of the object categories using intrinsic dynamic features calculated from prestimulus activity. These dynamic features included measures of the distribution of prestimulus activity, its periodicity, autocorrelation, and power in canonical frequency bands. Across the patients, we could reliably predict whether contacts from a left-out patient would be selective for each of 6 object categories or not based on these intrinsic, prestimulus dynamics. Further, these intrinsic dynamics could differentiate between contacts selective for different object categories. This shows that, for example, word selective regions have different intrinsic dynamics compared to body selective regions in the ventral visual stream. Previous studies have demonstrated consistent differences in the dynamics of different brain regions across subjects. So, to ensure the dynamic features were not simply predicting the anatomical location of the contacts, which is also predictive of category selectivity, we compared category classifiers trained on the dynamic features to those trained on the anatomical location of the contacts. These analyses demonstrated that the predictive ability of the dynamic features was not purely driven by broad differences in dynamics across ventral visual regions. This suggests that subject-to-subject variability in the cortical localization of category selective regions may be related to variability in the dynamic properties of these regions across subjects. Several open questions remain, including the degree to which these intrinsic dynamics are conserved across spatially discrete regions selective for the same stimuli and how these intrinsic dynamics are influenced by connectivity with other brain circuits.

Disclosures: M.J. Boring: None. **R.M. Richardson:** None. **A.S. Ghuman:** None.

Poster

P472. Functional Architecture of the Visual Cortex

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P472.05

Topic: D.07. Vision

Support: NSFC 31971031
NSFC 31930053

Title: Laminar and columnar response profiles of color, disparity, and naturalistic texture in human early visual cortex

Authors: *H. AI[1], W. LIN[3], N. CHEN[1,2], P. ZHANG[3];
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Abstract: The early visual cortex can process a variety of basic visual features, such as color, disparity, and naturalistic statistical information. However, how these features are processed hierarchically in the visual system remains unclear. Using 7T BOLD fMRI at 1mm isotropic resolution, we investigated laminar and columnar response profiles of these basic features in human V1 and V2 by presenting three independent stimulus contrasts: (1) color versus luminance, (2) binocular disparity (3D) versus 2D control, (3) naturalistic texture versus spectrally matched noise. Different laminar profiles were observed. Compared to spectrally matched noise, naturalistic texture evoked a larger response in the deep cortical depth than in the middle and superficial depth, both in V1 and V2. On the other hand, color- and disparity-selective responses were stronger in the superficial depth than in the middle and deep cortical depth of V1, and no difference across different depth of V2. These results suggest that in V1 and V2, cortico-cortical feedbacks play important roles in the processing of naturalistic texture, while color and disparity representations might initially arise from V1 superficial layers and are further processed in V2 and beyond through the feedforward pathway. Color-selective thin and disparity-selective thick stripe columns can be robustly detected in V2. The contrast between naturalistic texture and noise showed no obvious columnar organizations. We further defined pale stripe ROIs in-between the thin and thick stripes. No response preference for naturalistic texture was found across the three types of stripes in V2. Altogether, these mesoscale response profiles reveal stronger feedback activity in V1 and V2 for naturalistic statistical visual processing compared to other basic features, without preference in V2 stripes.

Disclosures: H. Ai: None. **W. Lin:** None. **N. Chen:** None. **P. Zhang:** None.

Poster

P472. Functional Architecture of the Visual Cortex

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Topic: D.07. Vision

Support: Wu Tsai Neurosciences Institute Big Ideas Project
NIH 1 R21 EY030588-01

Title: Infants' sensory-motor cortices undergo microstructural growth coupled with myelination

Authors: *V. NATU[1], M. ROSENKE[1], H. WU[2], F. QUERDASI[1], H. KULAR[1], N. LOPEZ-ALVAREZ[1], M. GROTHEER[3], S. BERMAN[4], A. MEZER[5], K. GRILL-SPECTOR[1]; [1]Psychology, Stanford Univ., Stanford, CA; [2]Ctr. for Cognitive and Neurobiological Imaging, Stanford Universtiy, Stanford, CA; [3]Dept. of Psychology, Univ. of Marburg, Marburg, Germany; [4]Edmond and Lily Safra Ctr. For Brain Sci., Jerusalem, Israel; [5]Edmond and Lily Safra Ctr. for Brain Sci. (ELSC), The Hebrew Univ. of Jerusalem, Jerusalem, Israel

Abstract: The establishment of neural circuitry during early infancy is critical for developing visual, auditory, and motor functions. However, how cortical tissue develops postnatally is largely unknown. By combining T_1 relaxation time (T_1=1/R_1 relaxation rate) from quantitative MRI and mean diffusivity (MD) from diffusion MRI in 13 infants, we tracked cortical tissue development in infants across three timepoints (newborn, 3 months, and 6 months, *N*=10 at each timepoint). Lower T_1 and MD indicate higher microstructural tissue density and more developed cortex. Our data reveal three main findings: First, primary sensory/motor areas have lower T_1 and MD at birth than higher-level cortical areas. However, all primary areas show significant reductions in T_1 and MD from newborns to 6-months-olds, illustrating profound tissue growth after birth. Second, significant reductions in T_1 and MD from newborns to 6-month-olds also occur in all visual areas of the ventral and dorsal visual streams. Strikingly, this development was hierarchical across the visual streams: Earlier areas are more developed with progressively denser tissue at birth than higher-order areas, but higher-order areas had progressively faster rates of development. Finally, transcriptomic gene analyses comparing gene expression in postnatal (early infancy) vs. prenatal brain tissue samples showed significantly stronger postnatal vs. prenatal expression of genes associated with myelination, synaptic signaling, and dendritic processes. Our results indicate that these cellular processes may contribute to profound cortical tissue growth during early infancy observed with *in-vivo* MRI. These findings suggest a novel principle of postnatal maturation of sensory systems: development of cortical tissue proceeds in a hierarchical manner, enabling the lower-level areas to develop first to provide scaffolding for higher-order areas, which begin to develop more rapidly following birth to perform complex computations for vision and audition.

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Poster

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CRCNS US-German Research Proposal: Origins of distributed modular activity in the neocortex

Title: Uniform cortical stimulation in the early visual cortex evokes modular patterned activity through intracortical network interactions

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Abstract: Modular (columnar) activity is a fundamental mode of neural activity in the cortex of primates and carnivores. Work in ferret visual cortex has shown that already prior to visual experience, early spontaneous activity in ferret visual cortex is modular, revealing large-scale correlated networks that are predictive of future functional networks and exist in the absence of long-range horizontal projections and patterned feedforward activity. Computational models predict a striking ability of developing cortical circuits to self-organize into modular coordinated patterns of activity, but this prediction has not yet been directly tested during early development in vivo. Here, we combine widefield epifluorescence calcium imaging with excitatory optogenetics to simultaneously image and stimulate pyramidal neurons in layer 2/3 of developing ferret visual cortex (postnatal day 24-29). We found that optogenetic stimulation of the cortex with a large (~3 mm) spatially uniform stimulus lead to the rapid emergence of highly non-uniform, modular neural activity. The probability of evoking a modular pattern increased with optogenetic light intensity, suggesting a network activity threshold to generate structured activity from this spatially uniform input. Moreover, these modular opto-evoked events resembled spontaneous activity in their spatial structure and scale and revealed highly similar large-scale correlated networks as spontaneous activity. Notably, opto-evoked activity resided in a moderately low-dimensional subspace that is indistinguishable from that of ongoing spontaneous activity. In contrast, activity patterns elicited with visual stimulation are highly stereotyped and strongly low dimensional. Importantly, we found that modular opto-evoked events required activity propagating through local intracortical synapses for their emergence, and that modularity persisted even in the absence of feedforward input from the LGN. Together, this provides strong evidence that modular patterned activity is an emergent property from intracortical interactions through a self-organizing network. Furthermore, our approach provides a path towards studying the input-output transform of early cortical networks at the scale of several millimeters.

Disclosures: H.N. Mulholland: None. **M. Kaschube:** None. **G.B. Smith:** None.

Poster

P534. Cortical Planning and Execution: Behavior

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Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P534.01

Topic: E.04. Voluntary Movements

Title: Behavioral measurements of motor readiness in mice.

Authors: *E. MANGIN[1], J. CHEN[2], J. LIN[2], Y. HAO[3], N. LI[2];
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Abstract: Many movements occur too quickly for online correction; the brain plans goal-directed movements before they are executed. Classic experiments suggest the existence of a "state of readiness" which, once entered, permits rapid and precise execution of the prepared movements. In humans and nonhuman primate studies using delayed response tasks, reaction time (RT) decreases if subjects are given time to prepare for a movement before a Go cue. However, behavioral measurements of motor preparation are currently lacking in mice, an increasingly popular model system ideal for circuit dissections of motor preparation.
Here we examine non-body-restrained mice in an autonomous behavior system in which animals voluntarily engaged in two different delayed response tasks: a licking task involving the tongue and a reaching task involving the forepaw. In these tasks, animals planned their upcoming movement during a delay epoch (1.3s) and had to correctly lick or reach an instructed target after an auditory Go cue to receive a water reward. These behaviors were captured with high-speed videography and quantified using markerless tracking. We replicated classic experimental findings in mice, which demonstrated a RT savings with more preparation. On occasional probe trials, on which the Go cue arrived before the end of the delay epoch, mice were unprepared and thus slower to initiate the movement. RT decreased with increased delay duration before reaching a plateau. The shape of the RT curve thus reflected the degree of readiness. To date, we analyzed high-speed videos of the tongue, nose, and jaw in the licking task. Tongue movement was more precise with increased degree of readiness. Moreover, we found that the degree of readiness was reflected in the position of the jaw and nose before movement onset. Mice positioned their jaws and noses toward the instructed target and the direction of the upcoming lick movement could be read out from the jaw and nose position. The jaw and nose position predicted the RT in both control and probe trials and this relationship was common across mice, though idiosyncratic differences among animals were also found. These results provide behavioral signatures of motor preparation in mice and suggest body features as a viable way to measure readiness online. The measured readiness suggests a learned timing in which mice

anticipated the timing of the Go cue. We are currently examining these behavioral features in a dynamic preparation task in which mice rapidly select between and initiate licking or reaching movements.

Disclosures: E. Mangin: None. **J. Chen:** None. **J. Lin:** None. **Y. Hao:** None. **N. Li:** None.

Poster

P534. Cortical Planning and Execution: Behavior

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Program #/Poster #: P534.02

Topic: E.04. Voluntary Movements

Support: Israel Science Foundation
Science and Technology Ministry- Israel

Title: Disparity between default plan and default movement representation in the motor cortex

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[1]Biomed. Engin., Technion Israel Inst. of Technol., Haifa, Israel; [2]Physical Med. and Rehabil., Technion - Israel Inst. of Technol., Haifa, Israel

Abstract: Goal-directed behavior involves planning and execution of actions that incorporate information about the target, our bodies and the environment. Within this motor repertoire, we often perform some actions more than other, resulting in probability distributions over possible movements. Distribution of recent executed action affects movement representation in the primary motor cortex (M1) as revealed by biases in movements evoked by Transcranial Magnetic Stimulation (TMS). However, little is known whether M1 represents also the plans of these movements. In three behavioral-TMS experiments, we manipulated the distribution of executed movements and the distribution of upcoming target location and compared the default movement (DM)- the TMS-evoked thumb movement, and default plan (DP)- the distribution of movement begins prepared by default, in accordance with expectations about which movement would most likely be required. In Exp. 1 (n=20), we compare the baseline distribution of DM with DP using a time-response task in which participants heard a sequence of 4-tones (500ms apart) and were trained to initiate their thumb movement with the fourth tone. In a subset of trials, no target ever appeared but participants were still required to move, enabled us to assess participants' DP. We found two clusters of participants: one showed a large overlap between DP and DM whereas a second cluster showed remarkable disparity between the distributions. Next, we manipulated the distribution of recent movement by asking participants to intensively repeat a movement toward a specific direction (Exp. 2, n=15). We found that DMs were remarkably sensitive to prior distribution of movement whereas distribution of DP did not change. Lastly, we tested whether DP could be affected by distribution of the most expected movement (Exp. 3,

n=15). Here, we biased the distribution of upcoming targets by increasing the probability of target appearance in a specific direction, and found that distribution of DP, but not DM, was attributable to anticipation of upcoming movements. Overall, we demonstrated that DM in M1 reflects probability distributions of prior actions whereas DP reflects individual expectation for upcoming reaches.

Disclosures: A. Suleiman: None. **F. Mawase:** None.

Poster

P534. Cortical Planning and Execution: Behavior

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Program #/Poster #: P534.03

Topic: E.04. Voluntary Movements

Support: PAPIIT-DGAPA IN201518
CONACyT Fronteras de la Ciencia No. 846

Title: Hierarquical organization of corticomesencephalic projections

Authors: *V. LOPEZ-VIRGEN, M. MACIAS-RAMIREZ, P. RODRIGUEZ-MORENO, R. OLIVARES-MORENO, V. DE LAFUENTE, G. ROJAS-PILONI;
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Abstract: One major goal in neuroscience is to understand how different aspects of movements are regulated in parallel by the cerebral cortex. Output commands of cortical sensorimotor areas are driven by pyramidal tract neurons (PTNs) which projects to several subcortical structures. An open question is if PTNs are hierarchically organized generating different and specific output commands for sensorimotor integration. Here, using single cell 3D reconstructions in *In vivo* identified motor cortex PTNs projecting to mesencephalic red nucleus, or pons, we found morphological and electrophysiological characteristics for corticopontine and corticorubral neurons. Moreover, using photometry in both class of PTNs during a motor execution task, revealed a different relationship of this neurons with movement. The results indicates that PTNs are organized into different subsystems controlling in a coordinated manner distinct subcortical circuits related with different aspects of sensorimotor integration and thus, proper movement execution.

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Poster

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Program #/Poster #: P534.04

Topic: E.04. Voluntary Movements

Support: NIH R15NS116742
NSF 1912352

Title: Collapse of complexity of brain and body activity due to excessive inhibition and MeCP2 disruption

Authors: *J. LI, P. A. KELLS, A. C. OSGOOD, S. H. GAUTAM, W. L. SHEW;
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Abstract: Complex body movements require complex dynamics and coordination among neurons in motor cortex. Conversely, a long-standing theoretical notion supposes that if many neurons in motor cortex become excessively synchronized, they may lack the necessary complexity for healthy motor coding. However, direct experimental support for this idea is rare and underlying mechanisms are unclear. We address this problem in motor cortex of awake, freely behaving rats, comparing normal rats and a transgenic rat model of Rett syndrome (RTT), as well as pharmacologically inducing enhanced synaptic inhibition. We record single-unit neuronal activity with multi-electrode arrays in the deep layer motor cortex (M1). Simultaneously, we record the three-dimensional body movement of the rats by capturing representative beads on the rats with an infra-red camera system. First, we examined cortex-body interactions using spike triggered average body speed functions and CCA analysis. We found that both RTT and enhanced inhibition cause stereotyped, low dimensional cortex-body interactions. Next, we looked at synchrony of the neural activity, along with PCA-based analysis and spike triggered average population activity functions. Similarly, we found that synchrony is the lowest for normal rats. Both RTT and enhanced inhibition result in a significant higher synchrony, lower dimensional and stereotyped intracortical interactions. Last, we applied PCA-based analysis and B-SOiD analysis on the body movement data, and found that body activity is also less complex for RTT and enhanced inhibition. In summary, we found a collapse of complexity in the motor system for both RTT and enhanced inhibition. Reduced complexity was apparent in lower dimensional, stereotyped brain-body interactions, neural synchrony, and simpler behavior. Our results indicate that excessive inhibition leads to stereotyped, low-complexity relationships among neurons in motor cortex and body movements, and this imbalanced inhibition may contribute to motor dysfunction in Rett syndrome.

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Poster

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Topic: E.04. Voluntary Movements

Support: NSF grant 1558151

Title: Reaching errors resulting from the misestimation of arm position

Authors: C. BUNEO, *P. PHATARAPHRUK, P. VANGILDER;
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Abstract: The planning of visually-guided reaching movements requires the integration of visual and somatosensory inputs to estimate the arm's position. In some contexts (e.g., in dimly lit environments, during experimental manipulation of sensory feedback, or following neurological damage) the integration of visual and somatosensory inputs can lead to an arm position estimate that is biased and imprecise, leading to errors in movement planning and execution. To understand the neural correlates of this process we have developed a virtual reality based experimental paradigm for studying visual, proprioceptive, and tactile integration for arm position estimation in non-human primates. Animals reach from a defined start position to four targets in a semi-immersive VR environment that incorporates a fully-rendered monkey avatar arm. The avatar arm is viewed either in a position consistent with a real starting position ('veridical') or in a visually shifted position ('perturbed'). Veridical and perturbed arm positions are varied in separate blocks of trials. Behavioral analyses focus on changes in initial movement directions that are induced by perturbing the arm position. Here we report the results of computer simulations used to predict the behavioral consequences of perturbations in different directions and amplitudes. Two joint (shoulder and elbow) horizontal plane arm movements were simulated using an inverse model that first determined the joint torques corresponding to nominal endpoint trajectories. A forward model was then used to simulate movements in an open-loop fashion, i.e., feedback control was not included. Perturbations were simulated by introducing uncompensated bias and variance into the estimate of initial hand position. Simulations produced movement errors that were dependent upon movement direction, starting position, and perturbation direction and amplitude. For the primary start position used in our experiments, simulation results were consistent in several ways with behavioral data obtained from one animal performing reaching movements under visually perturbed conditions. These results provide important insights into the movement errors that result from misestimating arm position, which could prove useful for understanding corresponding errors that arise during investigations of the neural correlates of multisensory integration, as well as those arising from neurological damage.

Disclosures: C. Buneo: None. **P. Phataraphruk:** None. **P. Vangilder:** None.

Poster

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Program #/Poster #: P534.06

Topic: E.04. Voluntary Movements

Support: NSF Grant 1656882

Title: Posterior parietal area 5b neurons with somatosensory receptive fields on different segments of the forelimb act differently during locomotion

Authors: *W. U. NILAWEERA[1], I. N. BELOOZEROVA[2];
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Abstract: Posterior parietal cortical area 5b is a part of the cortical pathway that processes visual information while conveying it from visual to motor cortex (M1), equipping M1 with information for guiding limbs. During locomotion, neurons in M1 with somatosensory receptive fields (RF) on different forelimb segments behave differently (Stout & Beloozerova 2012). The source of this difference is unclear. Here we assessed the activity of the main cortical provider of visual information to M1, area 5b, to examine whether it differentially processes information regarding different segments of the forelimb and how this can influence locomotion-related activity of M1.Experiments were conducted on two cats walking on a flat surface and horizontally placed ladder. Accurate stepping on the ladder requires vision, whereas walking on the flat surface does not. The activity of 38 neurons with RF on the shoulder (SH), 29 with RF involving the elbow (EL), and 42 with RF on wrist/paw (WP) was analyzed, including 20 neurons that responded antidromically to electrical stimulation of M1, thus confirming projecting to M1, cortico-cortical neurons (CCs): 7 SH, 5 EL, 8 WP cells. We found that on the flat surface the group activity of SH cells, both CCs and neurons whose axon projection was unknown (noIDs), peaked during stance-swing transition, similar to that of M1 SH group (Stout & Beloozerova 2012). The activity of EL noIDs was steady over the step cycle, while that of EL CCs peaked at the swing, both different from the activity of M1 EL group. The discharge rate of WP noIDs was maximum during stance-swing transition, similar to that of M1 WP group, whereas, the activity of WP CCs peaked during swing-stance transition, an opposite phase. Thus, during vision-independent locomotion, area 5b may contribute to the stride-related activity modulation of M1 SH and possibly WP groups, but not the EL group. During ladder locomotion the group activity of SH noIDs was steady, while that of SH CCs had two maxima: during stance-swing and swing-stance transitions, both profiles different from that of M1 SH group. The activity of EL noIDs peaked during stance, later in the cycle than M1 EL group, which peaked during swing-stance transition. The activity of EL CC group was minimal during this latter phase. The activity of WP noIDs peaked during stance-swing transition, earlier than M1 WP group, which peaked during swing. The activity of WP CCs, however, was maximal later, during early stance when M1 WP group activity was depressed. Thus, it is unlikely that area 5b SH

group determines the activity of M1 SH cells during vision-guided locomotion, while EL and WP CCs may influence respective M1 groups via M1 inhibitory interneurons.

Disclosures: W.U. Nilaweera: None. **I.N. Beloozerova:** None.

Poster

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Topic: E.04. Voluntary Movements

Support: NSF grant 1558151

Title: Trimodal integration of visual, tactile, and proprioceptive cues influences reach planning in a virtual environment

Authors: P. PHATARAPHRUK, P. VANGILDER, *C. BUNEO;
Arizona State Univ., Tempe, AZ

Abstract: The planning of reaching movements requires integration of visual and proprioceptive cues to estimate the position of the arm before and during movement. Although bimodal (visuo-proprioceptive) integration has been well-characterized in human subjects, less is known about trimodal integration processes, e.g., for reaching movements performed when tactile information is initially present. Moreover, few studies have examined multisensory integration in non-human primates (NHP), which are more amenable to examination of the neural correlates of multisensory integration. Here, we used a semi-immersive virtual reality (VR) environment to study how visual, proprioceptive, and tactile cues about arm position are integrated prior to movement execution. A NHP was trained to reach from a single start position to four targets in VR while viewing a fully-rendered monkey avatar arm that was animated via a reflective-marker based motion capture system. Trials began with no visual feedback of the arm but once the starting position was cued and acquired, the avatar arm was displayed for 1-1.5 seconds prior to movement. The virtual arm appeared either in a position consistent with the physical location of the starting position (veridical vision (VV)), or shifted 6 cm toward the animal's body (perturbed vision (PV)). VV and PV conditions were varied on a trial-by-trial basis. In addition, tactile feedback was varied in separate blocks of trials by adding or removing a circular plastic button from the starting position. Evidence for multisensory integration was assessed by comparing mean reach directions at peak tangential acceleration across visual and tactile conditions. We observed target-dependent differences in initial reach directions between PV and VV conditions, regardless of tactile conditions. For targets with clear differences, initial reach directions tended to be more accurate (i.e. less deviated from cued target directions) on VV trials than PV trials. Notably, both the target-dependence and the direction of deviation on PV trials were largely

consistent with forward dynamics simulations of visually perturbed reaches. Initial reach directions also tended to be more accurate for reaches made with tactile feedback than without, with larger effects being observed for targets influenced by arm visual feedback. The results indicate that visual and tactile cues influence the planning of reaches performed in a virtual environment. The consistent nature of the observed effects suggest visual and tactile cues are integrated along with proprioception to form a more accurate estimate of initial arm position used in movement planning.

Disclosures: P. Phataraphruk: None. **P. Vangilder:** None. **C. Buneo:** None.

Poster

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Topic: E.04. Voluntary Movements

Support: NSF Grant 1558151

Title: Effects of Tactile Cues on Arm Movement Planning and Execution in a Virtual Environment

Authors: *P. VANGILDER, P. PHATARAPHRUK, C. BUNEO;
Arizona State Univ., Tempe, AZ

Abstract: During the planning of arm movements, visual and somatosensory inputs are combined to compute an estimate of the current arm position through a process known as multisensory integration. This estimate is used to determine the desired movement vectors as well as joint-based motor commands for action. Although multisensory integration for reaching has been extensively studied in humans, investigations involving non-human primates (NHP), which serve as an important model for understanding the underlying neural mechanisms, are relatively sparse. Here, we used a semi-immersive virtual reality (VR) environment paired with a motion capture system to study the integration of visual, proprioceptive, and tactile cues during reaching movements. Two NHPs were trained to wear a motion capture sleeve on their right arm, which served to animate a virtual replica of their own limb in real-time within the VR environment. The VR arm and environment were projected from a 3D monitor onto a mirror blocking vision of the animals' actual arm. After acquiring a central starting position, the animals made reaches to one of four targets in the horizontal plane during which visual feedback of the virtual arm was provided or removed on a trial-by trial basis. Physical properties of the virtual start and target positions were rendered to resemble those of real tactile cues, which were circular plastic buttons that were added or removed from the workspace in separate blocks of trials. Analysis focused on quantifying the variability in hand movement direction and distance

(relative to the starting position) as a function of movement extent (i.e., from 10-100% of movement on a given trial, in increments of 10%). We found that for all task conditions, directional variability decreased with movement extent, while conversely there was an increase in variability in distance traveled. Variability in direction and distance was reduced in conditions where tactile cues were provided, though this effect was more apparent during the later stages of the reaching movements. Reduced variability in direction and distance was also observed in conditions employing visual feedback but only when tactile cues were absent. These results show that the presence of tactile cues reduces the variability of reaching movements performed in a virtual environment. Moreover, the results suggest that tactile cues interact in a complex manner with ongoing visual and proprioceptive feedback to determine motor output.

Disclosures: P. Vangilder: None. **P. Phataraphruk:** None. **C. Buneo:** None.

Poster

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Topic: E.04. Voluntary Movements

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Title: Heritable differences in the expression level of the zinc transporter zip11 drive individual-to-individual variation in learned birdsong tempo

Authors: *D. METS[1], W. H. MEHAFFEY[1], B. COLQUITT[1], R. J. VELINE[1], M. S. BRAINARD[2];
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Abstract: Complex learned behaviors exhibit striking variation within populations, yet how heritable factors contribute to such inter-individual differences remains largely unknown. Here, we used genetic linkage analysis within a Bengalese finch population (*Lonchura striata domestica*) to investigate molecular and circuit mechanisms underlying heritable differences in the tempo of learned birdsong-a fundamental aspect of song performance suggested to be a target of sexual selection and known to be influence by experience, genetics, and their interaction. We identified a genomic locus encoding the zinc transporter ZIP11 and found that *zip11*(*SLC39A11*) transcript was expressed at higher levels in song control circuitry of faster singing birds. Reducing soluble zinc increased synaptic currents in motor circuitry and accelerated song, whereas reducing ZIP11 slowed song. Our results reveal a novel zinc-dependent mechanism that

modulates neural activity to drive differences in behavior and suggest that natural variation in learning may preferentially target modulatory processes rather than core neural machinery. Further, this work establishes genetic linkage and molecular manipulations in the songbird as a powerful system for studying the molecular and circuit level mechanisms that structure learned behavior and provides a framework for similar future studies. In particular, other aspects of song (syllable sequencing, amplitude, spectral flatness etc...) display heritability in this population of Bengalese finches and similar analyses of these phenotypes will provide insights into differing mechanisms influencing complex learned behavior.

Disclosures: D. Mets: None. **W.H. Mehaffey:** None. **B. Colquitt:** None. **R.J. Veline:** None. **M.S. Brainard:** None.

Poster

P534. Cortical Planning and Execution: Behavior

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P534.10

Topic: E.04. Voluntary Movements

Support: NSF Grant #1748523
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Title: Assessing how the introduction of daf-7 (e1372) in unc-33 affects rescue of dauer formation and locomotion in previously uncoordinated C.elegans.

Authors: *A. CRISTANCHO, R. VALTIERRA, M. C. MIRANDA, S. BECK, J. CANTU, A. VACIO, A. HOLGADO;
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Abstract: Recent studies show that the sixth-leading cause of death in the United States is Alzheimer’s disease with the number of deaths doubling between 2000 and 2018. Collapsin response mediator protein-2 (CRMP2) in humans, (UNC-33 in *C. elegans*) is a molecule that mediates axonal stability and degeneration. It is a potential drug target that could be used to treat Alzheimer’s and other neurodegenerative diseases, which can often be attributed in part to aging. In the aging process, CRMP2 becomes hyperphosphorylated, decreasing its functionality and leading to aging in neurons. *C. elegans* aging can be slowed by dauer formation, a developmental stage identified as a non aging stage. DAF-7 is the *C. elegans* homolog of transforming growth factor beta (TGFβ), which enables development in both humans and *C. elegans.* The mutation of *daf-7* leads to constitutive dauer formation, while the mutation of unc-33 leads to the inability to form dauers in response to environmental stress. Herein, we are introducing the *daf-7* mutation to an *unc-33* mutant to test for rescue of dauer formation.To this end, we created *unc-33; daf-7*

double mutants and assessed dauer formation. Double mutants were treated with 1% SDS to test for proper dauer formation after exposure to unfavorable conditions. Analysis of experimental data showed that *unc-33 (mn407); daf-7 (e1372)* double mutant had 94% survival to SDS treatment, compared to 0% in the *unc-33 (mn407)* mutant. The *unc-33 (e204); daf-7 (e1372)* double mutant had 89% survival, compared to 0% in the *unc-33 (e204)* mutant. In addition to dauer formation, we tested the rescue of locomotion and found that the distance traveled on plates was f 54.8 ± 5.3 nm in the Wild Type compared to 45.4 ± 8.6 nm in the *unc-33(e204); daf-7(e1372)* double mutant. When tested in liquid there were 189.3 ± 5.9 body bends/min in the Wild Type compared to 70.0 ± 3.1 body bends/min in the *unc-33(e204); daf-7(e1372)*double mutant. Based on these results, we conclude that the problem behind dauer formation in *unc-33* mutants lies within the DAF-7 pathway. Specifically, *unc-33* mutants have a problem suppressing DAF-7 signaling, leading to the continuation of reproductive programs even under unfavorable conditions. Results from the plate locomotion verify rescue in the *unc-33(e204); daf-7(e1372)* double mutant. In aging, where CRMP2/UNC-33 is hyperphosphorylated, the DAF-7 levels can be manipulated to increase lifespan. Animals that have an *unc-33* mutation concurrent with a *daf-7* mutation can live longer and potentially have slower neurodegeneration.

Disclosures: A. Cristancho: None. **R. Valtierra:** None. **M.C. Miranda:** None. **S. Beck:** None. **J. Cantu:** None. **A. Vacio:** None. **A. Holgado:** None.

Poster

P540. Cortical Planning and Execution: Neuroimaging

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Program #/Poster #: P540.01

Topic: E.04. Voluntary Movements

Support: NIH Grant P30 DA048742
MINNESOTA SPINAL CORD INJURY AND TRAUMATIC BRAIN INJURY RESEARCH GRANT

Title: Mesoscopic wide-field calcium imaging during a reaching behavioral task in mice.

Authors: *R. E. CARTER, E. FLAHERTY, C. HUFFMAN, A. SHEKHAR, T. J. EBNER; Neurosci., Univ. of Minnesota, Minneapolis, MN

Abstract: Reaching is a fundamental behavior exhibited by a number of organisms and involves complex limb movements coordinated and executed by many distinct brain areas. Wide-field, chronic calcium (Ca^{2+}) imaging through transparent polymer skulls is a relatively new technique that involves surgically replacing opaque cranial structures with transparent polymer windows. These windows allow for real-time visual imaging of brain activity across wide regions of the cortex in mice that are awake and behaving, allowing for innovative comparisons between brain

activity and behavior to be made. Thus, wide-field imaging of reach behaviors should be able to provide new insights into the brain regions involved with reach behaviors, as well as provide insights on how these brain areas work together in reach behaviors. We implanted our transparent polymer windows in Thy1-GCaMP6f mice that express GCaMP6f in pyramidal cells in layers II/III of the cerebral cortex. These mice were trained in a head-fixed, cued, reaching behavioral task. The task consists of a stimulus tone signifying the delayed presentation of a water droplet at the end of a spout in front of the mouse, requiring the mouse to reach out to obtain the water droplet. Mice were water-restricted to incentivize the reaching behavior. Following proficient reaching to the spout, simultaneous dual-wavelength wide-field Ca^{2+} imaging (20 Hz) and high-speed behavioral imaging (100 Hz) was performed during the behavioral task. Water rewards were delivered in either two paradigms over a 5-minute imaging session: 1) once every 30 seconds resulting in 8 deliveries per imaging session; or 2) in 1 of the 8 random deliveries, no water was delivered following the tone. Limb tracking during the reaching behavior was tracked using DeepLabCut, and the hemodynamic corrected Ca^{2+} signal was analyzed using Localized semi-Nonnegative Matrix Factorization (LocaNMF). In relation to the task, there were widespread changes in Ca^{2+} activity across much of the dorsal cerebral cortex that were coupled to both the tone and reach. Preliminary analysis shows that mice express a consistent functional connectivity map during the reaching task, between imaging sessions within the same day to across multiple days and mice. The functional connectivity evolves during the reach, with higher functional connectivity between the primary motor cortex, hind limb/forelimb region of the somatosensory cortex, and the retrosplenial cortex during the task compared to baseline behavior. These results demonstrate that the act of reaching widely engages the cerebral cortex, using a consistent, well-defined cortical network.

Disclosures: R.E. Carter: None. **E. Flaherty:** None. **C. Huffman:** None. **A. Shekhar:** None. **T.J. Ebner:** None.

Poster

P540. Cortical Planning and Execution: Neuroimaging

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Program #/Poster #: P540.02

Topic: E.04. Voluntary Movements

Support: NS111028
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T32-MH115688
University of Minnesota's MnDRIVE Initiative

Title: Wide-field calcium imaging of dynamic cortical networks during locomotion

Authors: *S. WEST[1], J. ARONSON[1], L. POPA[1], R. CARTER[1], K. FELLER[2], S. KODANDARAMAIAH[1], T. EBNER[1];
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Abstract: The cerebral cortex is widely activated during movement, including during locomotion. Evidence suggests that the cortex enters a new functional state compared to rest that enables the motor and sensory computations needed to accurately navigate the environment. However, the cortex-wide activity patterns underling this locomotion functional state have not been fully characterized. Here, we used widefield calcium imaging to evaluate the activity and functional relationships among cortical regions during locomotion. Clear polymer windows were implanted on 8 individual Thy1-GCaMP6f mice expressing the Ca^{2+} indicator GCaMP6f in layers II/III and V pyramidal neurons, allowing for wide field-of-view imaging of most of the dorsal cerebral cortex. Mice were head fixed on a freely moving disk treadmill that allowed free limb and body movements, including spontaneous walking. For each mouse, calcium imaging was recorded for 6-8 sessions, for a total of over 60 hours of mesoscopic data. Treadmill position was tracked with a rotary encoder, while behavior was tracked with infrared cameras. Paw movement was extracted using DeepLabCut. Mice spontaneously locomoted for 21.3 ± 10.4% of the time on the treadmill. Spatial Independent Component Analysis was used to identify independent functional regions. The mean fluorescence time series was extracted from each region, and this activity was regressed to behavior parameters as well as correlated to other cortical regions to compute the functional connectivity. Regressions show that treadmill and paw velocity explain a significant faction of the fluorescence variability across the cortex during locomotion, with the strongest effects at the onset and offset. Prior to the onset of locomotion, there is a large increase in correlation between most cortical regions. However, once a continued, steady state of locomotion is reached, the correlations between primary motor, somatosensory, and parietal regions decrease to levels lower than at rest. In contrast, the correlations from secondary motor (M2) and retrosplenial regions with most other regions increase compared to rest. At the locomotion offset, correlations increase between most regions before the network returns to the rest state. The results suggest the cerebral cortex passes through a transition state at locomotion onset, enters a new state during continued locomotion, then again passes through a transition state before returning to rest. Granger causality analysis shows that the most anterior regions of M2 Granger-cause activity in many other regions during locomotion, indicating anterior M2 may play a role in controlling the locomotion cortical state.

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Poster

P540. Cortical Planning and Execution: Neuroimaging

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Program #/Poster #: P540.03

Topic: E.04. Voluntary Movements

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Title: In vivo characterization of cortical noradrenergic activity during motor learning using an optical noradrenaline sensor in a mouse model of autism

Authors: *N. JONES[1], X. YIN[1], S. CHEN[2,1];
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Abstract: Clinical studies have reported that children with autism spectrum disorder (ASD) often exhibit difficulties with the acquisition of new motor movements; these motor symptoms can present earlier than social or cognitive indicators. However, the underlying mechanisms responsible for these motor learning disruptions remain unclear. Noradrenaline (NA) is garnering attention for its putative underlying role in ASD, in part, due to its powerful control over neuronal activity. Furthermore, recent perspectives highlight the abundance of ASD symptoms that can be viewed through the lens of inefficient neuromodulation by the locus coeruleus (LC) NA system. LC projects ubiquitously to cortex, and NA exerts neuromodulatory control on cortical activity. It has been well established that NA regulates motor learning and proper motor execution in the primary motor cortex (M1). Recent work in our lab revealed dysregulation in LC NA function as a culprit of motor learning delays, manifested in a behavioral phenotype by the 16p11.2 deletion mouse model (+/-) of ASD (Yin et al, 2021). Using adult male mice due to the predominance of ASD in males, we found that 16p11.2$^{+/-}$ mice exhibit delayed motor learning caused by a reduction of NA levels in M1, accompanied by elevated pyramidal neuron (PN) activity. Chemogenetic activation of LC rescued elevated M1 PN activity along with the delayed motor learning, underscoring the importance of M1 NA dynamics. Thus, to fully characterize changes in NA levels during motor learning in awake and behaving mice, I employed a newly developed optical NA sensor, combined with *in vivo* two photon imaging, to visualize spatiotemporal activation patterns of NA in M1. This allows us to characterize NA levels in the same M1 region throughout the entirety of the motor learning process. I found that M1 NA levels increase significantly during the initial learning phase in wild type (WT, n=7) and return to baseline levels as mice develop more consistent motor movements. This trajectory alludes to critical network modulation of M1 PNs by NA during the early phase of the motor learning process. In contrast, 16p11.2$^{+/-}$ mice (n=5) display an increase of NA levels in M1 in the later stages of learning, which corresponds with their delayed motor learning phenotype. These results align with our previous work to confirm a crucial role for LC NA in early motor learning, which, if disrupted, produces learning delays. This study provides a novel glimpse into the LC NA system in ASD, which can provide valuable insight for clinicians.

Disclosures: N. Jones: None. **X. Yin:** None. **S. Chen:** None.

Poster

P540. Cortical Planning and Execution: Neuroimaging

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Program #/Poster #: P540.04

Topic: E.04. Voluntary Movements

Support: National Science Foundation RII Track-2 FEC 1539068
Oklahoma Science and Technology Research and Development HR16-057
Institute for Biomedical Engineering, Science and Technology at the University of Oklahoma

Title: Correcting physiological noise in whole-head functional near-Infrared spectroscopy

Authors: *F. ZHANG, D. CHEONG, A. F. KHAN, Y. CHEN, L. DING, H. YUAN;
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Abstract: Functional near-infrared spectroscopy (fNIRS) has been increasingly employed to measure cerebral hemodynamics under normal and diseased conditions. However, fNIRS suffers from its susceptibility to superficial activity and systemic physiological noise. We have developed a new method, namely PCA-GLM (Zhang et al., 2021, doi: 10.1016/j.jneumeth.2021.109262), for denoising fNIRS recordings in a whole-head montage. To demonstrate the denoising performance, a high-density montage consisting of 109 long-separation channels and 8 short-separation channels was used for recording. In addition, auxiliary sensors were used to measure pulsation, respiration and head movements simultaneously. Our PCA-GLM method incorporates principal component analysis and general linear model to identify and remove a globally uniform superficial component, which is implemented in an automatic pipeline. The method was evaluated in experimental data acquired from a group of healthy human subjects during a visually cued motor task and further compared with four commonly used preprocessing methods in the literature, including a minimal preprocessing method and three other denoising methods. The results showed that after applying the proposed denoising method, the whole-head topography of fNIRS revealed focal activations in both the primary motor and visual areas. Further, quantitative evaluation of contrast-to-noise ratio, within-subject standard deviation and adjusted coefficient of determination showed that our PCA-GLM method yielded superior and robust denoising. In this study, an automatic method was established for removing physiological noise in whole-head fNIRS recordings. Our method can facilitate the clinical application of fNIRS in monitoring large-scale, network-level brain activities (Chen et al., 2020, doi: 10.3389/fnins.2020.560878; Khan et al., 2021, doi: 10.1088/1741-2552/abfdf9).

Disclosures: F. Zhang: None. **D. Cheong:** None. **A.F. Khan:** None. **Y. Chen:** None. **L. Ding:** None. **H. Yuan:** None.

Poster

P540. Cortical Planning and Execution: Neuroimaging

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P540.05

Topic: E.04. Voluntary Movements

Title: Neural correlation of simulated walking versus upper limb and lower limb movement: A fMRI study

Authors: *A. ALIVAR[1,2], S. SALEH[1,2], C. H. JIANG[3], G. YUE[1,2];
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Abstract: The neural mechanism of movement differs for upper and lower limbs. Such isolated movements have simpler neural properties than compound motor tasks requiring multi-limb coordination. We expect that isolated movement have different neural correlates than complex tasks. Previous functional imaging studies have compared upper and lower limb movements in various brain regions and none of them have reported differences in activation maps across motor areas compared to the intra-limb movement task like simulated walking. It is important to compare isolated and coordinated multi-limb movements to understand control mechanisms for guiding development of targeted rehabilitation strategies. Thus, this study aimed at evaluating and comparing activation maps during three motor tasks involving arm and leg movements performed in equal frequency and amplitude while functional magnetic resonance imaging (fMRI) data were obtained from 17 young and healthy subjects. The three motor tasks were performed as follows: alternating left and right arm movements, alternating left and right leg movements, and alternating left arm-right leg and right arm-left leg movements (simulated walking). General linear models (GLM) were used to identify the activation maps in each condition and compare the extent of activation within the motor control network to isolate upper extremity sub-region and lower extremity sub-region within the left and right motor cortices. In Simulated Walking compared to ARM, the activation map was highlighted in medial part of motor cortex (M1) ([x=-0.09,y=-30, z=57), p=6e-05, Z=6.48, k=1190) and compared to LEG, in lateral part f M1 ([x=28,y=-25, z=54), p=0.005, Z=5.32, k=556). Task specific functionally activated regions within M1 were used as seed region of interest (ROI) to perform Psychophysiological interactions (PPI) analysis and study the connectivity maps of M1 in each condition. Overall results show task specific activation map where simulated walking task recruits large activation within the motor cortex than arm and leg movement tasks. The significance of this study is to identify potential differences in the neural mechanism of each isolated and non-isolated limb movements and guide precise clinical treatments and rehabilitations.

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Poster

P540. Cortical Planning and Execution: Neuroimaging

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Program #/Poster #: P540.06

Topic: E.04. Voluntary Movements

Title: Assessing electrocortical changes in relative alpha and theta power over the frontal cortex during resting state in infants learning to reach

Authors: *A. JAYASEKARA[1], R. NISHIYORI[2], E.-R. SYMEONIDOU[2], R. XIAO[3], B. A. SMITH[2,4,5];
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Abstract: Electroencephalographic (EEG) theta activity in the frontal cortex has been associated with infant attentional control and information-encoding. Peaks in theta power have been observed over frontal electrode sites in infants with typical development (TD) during active toy-exploration and handling of objects. Increased frontal alpha power has been observed during periods of infant sustained attention towards dynamic visual stimuli. Emergence of frontal alpha and theta power both have been linked to infant modulation of attention and may contribute to development of goal-directed motor behavior. As infants learn to reach, we anticipate functional and structural cortical changes related to attention and motor learning. We expect these changes to differ between TD infants and infants at-risk (AR) for developmental disabilities. Here, we assessed longitudinal changes of relative theta (3-6 Hz) and alpha (6-9 Hz) power over 7 frontal electrode sites in 25 TD and 11 AR infants. We collected and analyzed EEG data during resting state, when infants looked at a glowing toy out of arms distance before and after a reaching assessment. Each infant was assessed once per month across 3-5 months as they learned to reach. Data collections took place between 38 and 230 days of gestation-adjusted age. We expected a positive correlation between age and frontal alpha/theta power in TD infants, yet not AR infants. We also calculated variance of gradients to assess within-group heterogeneity. In contrast to our hypothesis, our TD calculations showed no statistically significant correlation between age and relative theta ($r= -0.15$; $p= 0.11$) as well as alpha ($r= -0.16$; $p= 0.11$) power. AR also showed no significant correlations (theta: $r= -0.10$; $p= 0.26$; alpha: $r= -0.05$; $p= 0.38$). Variance in gradients for theta were 2.26E-7 for AR and 2.30E-7 for TD. For alpha, they were 7.62E-8 for AR and 9.13E-8 for TD. Although attention has been linked to infant motor skill acquisition, developmental trajectories are likely variable across infants. Additionally, attentional processes

can be modulated during the motor task, but this may not be as prevalent during resting state. These results are a first step in assessing potentially-distinct EEG profiles in TD/AR infants related to motor skill acquisition and emergence of attentional control in infant motor development. Further group analysis can potentially aid in advancing methods of early intervention for infants at-risk for atypical development. Moving forward we will refine our investigation by statistically testing for group differences.

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Poster

P540. Cortical Planning and Execution: Neuroimaging

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Program #/Poster #: P540.07

Topic: E.04. Voluntary Movements

Support: NRF-2020R1A2C3010304
HI17C1501

Title: Effects of High-Definition Transcranial Direct Current Stimulation on Upper Extremity Function and Hemodynamic Changes in Stroke Patients

Authors: *H. KIM[1,2], J. KIM[1,2], J. LEE[1], G. LEE[1], S. SHIN[1], W. CHANG[1], Y.-H. KIM[1,2,3]; [1]Dept. of Physical and Rehabil. Medicine, Ctr. for Prevention and Rehabil., Samsung Med. Ctr., Seoul, Korea, Republic of; [2]Dept. of Hlth. Sci. and Technology, SAIHST, [3]Dept. of Med. Device Mgmt. & Research, Dept. of Digital Health, SAIHST, Sungkyunkwan Univ., Seoul, Korea, Republic of

Abstract: This study aimed to investigate the effects of high-definition transcranial direct current stimulation (HD-tDCS) over the ipsilesional primary motor cortex (M1) on upper extremity function and cortical activation during the finger tapping task (FTT) in chronic stroke patients. Twelve-six chronic stroke patients (20 males; mean age: 59.4 ± 12.8 years) were enrolled. The inclusion criteria were (1) unihemiparetic stroke patients between the ages of 19 and 80 years, (2) patients with subcortical lesion stroke, and (3) patients capable of individual finger movements. Patients with contraindications to tDCS application were excluded. HD-tDCS (1 mA for 20 minutes, 41) was administered to the ipsilesional M1 using the Starstim 8 (Neuroelectrics®, Spain) for four days. Upper extremity function was assessed by the FTT, the Box and Block test, and the Fugl-Meyer assessment (FMA) before and after the interventions. The cortical activity was measured during the FTT with the affected hand using the NIRScout® functional near-infrared spectroscopy (fNIRS) system (NIRx Medical Technologies, Germany). fNIRS topomap using 67 channels covered partial areas of frontal, parietal, temporal, and

occipital cortices. Regions of interest (ROI) were determined as supplementary motor area (SMA), primary motor cortex (M1), and premotor cortex (PM) to investigate changes in cortical activation in the motor areas during the FTT. fNIRS data were analyzed using NIRS-statistical parametric mapping software package (NIRS-SPM) (KAIST, Korea). There were significant improvements in Box and Block test scores, the accuracy of the FTT, and the FMA scores after HD-tDCS intervention. The cortical activation was showed in M1 and SMA of bilateral hemispheres and PM in the affected hemisphere during the FTT with the affected hand before the interventions. After the interventions, only M1 and PM in the affected hemisphere were activated. The cortical activation during the FTT with the affected hand, which was observed in bilateral hemispheres before the interventions, was shifted to the affected hemisphere after the interventions. These results of the present study implies that HD-tDCS over the ipsilesional M1 in chronic stroke patients enhanced upper extremity function and effectively modulated cortical activation patterns. To confirm the better effect of HD-tDCS on rehabilitation, a comparative study using conventional tDCS is further needed.

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Poster

P540. Cortical Planning and Execution: Neuroimaging

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Program #/Poster #: P540.08

Topic: E.04. Voluntary Movements

Support: NIH R21 NS114816

Title: Brain changes associated with force control deficits during ankle dorsiflexion in individuals with Parkinson's disease

Authors: *J. CHUNG[1], A. BOWER[1], I. MALIK[2], J. P. MARTELLO[3], C. A. KNIGHT[1], J. J. JEKA[1], R. G. BURCIU[1];
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Abstract: A consistent observation from numerous functional magnetic resonance imaging (fMRI) studies is that during voluntary movements, the functional activity of the basal ganglia nuclei and the primary motor cortex (M1) is reduced in individuals with Parkinson's disease (PD) relative to healthy controls, and in several instances the reduced activity of the basal ganglia relates to the severity of motor symptoms. Although PD progresses to affect lower extremities, most fMRI studies focused primarily on identifying the neural correlates of upper extremity dysfunction. Thus, there is still a limited understanding of the brain circuitry

underlying lower limb symptoms in PD in part due to excessive head motion during leg movements in a supine position affecting the fMRI signal. Here we present preliminary results from a study that employs a force control paradigm to study brain changes during ankle dorsiflexion in individuals with and without PD. Seven PD (mean age: 65.8 ± 10.0; 4 males and 3 females) and fourteen healthy older adults (mean age: 62.3 ± 10.1; 6 males and 8 females) performed a visually cued isometric ankle dorsiflexion force production task at 15% of the maximum voluntary contraction (MVC). PD patients was tested following a 12-h withdrawal from antiparkinsonian medication and on the more affected side. Motor symptoms were assessed with the motor section of the MDS-sponsored revision of the Unified Parkinson's Disease Rating Scale (MDS-UPDRS-III). The tested side in controls was randomized. While both groups were able to produce 15% of MVC during the task (p=0.222), PD had a slower rate of force increase (p=0.044) and force decrease (p=0.022) than controls. The fMRI analysis revealed that compared to controls, PD had reduced brain activity during ankle dorsiflexion in the striatum, bilaterally, left globus pallidus, left M1 and left supplementary motor area. PD also had decreased force-related signal in the cerebellum, in vermis I-IV and the ipsilateral dorsal dentate nucleus. Our preliminary findings suggest that force control deficits during ankle dorsiflexion are associated in PD with extensive functional abnormalities that span both the basal ganglia- and cerebellar-cortical loops.

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Poster

P558. Neuroprosthetics: Other Motor Sensory Interfaces: E.G. Artificial Vision II

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Title: Modeling proprioceptive feedback evoked by intracortical microstimulation using topographical variational autoencoders

Authors: *J. SOMBECK[1], K. P. BLUM[2,1], N. D. SCHIMPF[3], A. A. FAISAL[5,6,7,8], L. E. MILLER[2,1,4,9];
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United Kingdom; [8]Data Sci. Inst., London, United Kingdom; [9]Shirley Ryan AbilityLab, Chicago, IL

Abstract: Tetraplegic patients can make visually guided reaches using efferent brain-machine interfaces (BMIs). However, the reaches are slow and imprecise, a bit like the deficits of people who can generate movement but have lost somatosensation. Restoring somatosensation will likely improve the use of BMIs. While intracortical microstimulation (ICMS) can provide touch feedback, using it for proprioceptive feedback has proven more difficult. In a single monkey in our lab, ICMS in area 2 of S1 biased the perception of the direction of a coincident bump in a direction predicated by the preferred direction (PD) of neurons recorded on the stimulated electrodes. In three other monkeys, the bias direction could not be predicted. The difference between touch and proprioception could be due to the topographical organization of the two areas. While touch has a distinct somatotopy, proprioceptive cortical maps do not, as receptors from many muscles need to be mapped onto the two-dimensional surface. Without a well-defined somatotopy, ICMS may activate neurons with very different encoding properties, resulting in perceived hand movements in unpredictable directions or even discordant effects at the hand and elbow. To explore the effect of topography, we used a topographical variational autoencoder trained to regenerate arm joint velocities via an 80x80 cortical map that included lateral inhibition, as we have done previously. This resulted in clusters of neurons with similar PDs, something like S1. We probabilistically activated neurons as a function of distance and hijacked the firing rates of activated neurons. We decoded joint velocities from cortical activity and used forward kinematics to compute hand cartesian velocities. We read out the effect of ICMS as the difference between hand velocity with and without ICMS. If the direction of this difference-vector was within 45° of the electrode's PD, we considered the effect to be predictable. We stimulated 300 randomly chosen electrodes with amplitudes from 15 μA to 40 μA. The percentage of electrodes with a predictable effect decreased from ~60% to ~40% with increasing magnitude, although the magnitude of the effect was quite small at small currents. In an effort to increase the magnitude of the effect while retaining its predictability, we stimulated sets of four electrodes with similar PDs simultaneously, each with 15 μA. Across 300 groups, the percentage with predictable effects was 70%, and the magnitude of the effect was nearly twice that of single-electrode stimulation at the same total current. With this model, we will explore the efficacy of various stimulation strategies for providing robust proprioceptive feedback.

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Poster

P558. Neuroprosthetics: Other Motor Sensory Interfaces: E.G. Artificial Vision II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P558.02

Topic: E.05. Brain-Machine Interface

Title: Myoelectric Closed-Loop Autoregressive Models for Predicting Ankle Kinetics and Kinematics Across Mobility Tasks

Authors: *J. J. EGANHOUSE[1], S. A. BEARDSLEY[2];
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Abstract: Numerous techniques have been proposed for controlling active lower limb prostheses during walking, however, predictive control systems that function across mobility tasks remain underdeveloped. The purpose of this study is to develop a closed loop predictive model that uses current muscle activity to predict future ankle angle and moment over time and across mobility tasks for use in an active transtibial prostheses. Kinematic, kinetic, and electromyography (EMG) data of lower limb movement obtained from six healthy ambulators was used to train a closed-loop nonlinear autoregressive (NARX) model to predict ankle angle and moment in response to changes in EMG during level (over ground) walking, stair ascent and stair descent (LW, SA, SD). Training data was assembled into 10 sets of LW, SA, and SD trials and separate NARX models were trained for each subject using a 10-fold cross-validation to characterize generalized performance across mobility tasks. All NARX models were configured with a 58.3 ms prediction interval, 120 ms sampling window, and 16 hidden units based on prior exploratory testing of the network structure. Batch training was used to ensure that models were trained on entire trials rather than randomly selected time points across trials. NARX model performance was characterized based on the type of feedforward input provided to the network which included: 3D shank velocity, and EMG signals obtained from the tibialis anterior, medial, and lateral gastrocnemius muscles (TA, MG, and LG respectively), and their combinations. Following training, the average root-mean-square errors (RMSE) of the model prediction across tasks varied with the type of input. RMSE for EMG only was 4.92° ± 1.394° and 0.154 ± 0.044 Nm/Kg, for shank velocity only was 2.93° ± 0.69° and 0.121 ± 0.0301 Nm/Kg, and for EMG plus shank velocity was 2.37° ± 0.58° and 0.084 ± 0.026 Nm/Kg . While network prediction of ankle angle and moment based on 3D shank velocity was generally better than EMG-only inputs, networks that combined 3D shank velocity and EMG inputs produced the best overall performance. The closed-loop model performance indicates that prior *estimates* of joint kinematics and kinetics can reliably predict gait when combined with external input(s) that convey user intent. Shank velocity input stabilized recurrent estimates of joint dynamics during cyclic movement, whereas EMG inputs improved prediction for transitions between walking conditions, where feedback of noncyclic user intent was most relevant. The results suggest that a NARX model combining shank velocity and EMG input could provide robust predictive control for use in active transtibial prostheses.

Disclosures: J.J. Eganhouse: None. **S.A. Beardsley:** None.

Poster

P558. Neuroprosthetics: Other Motor Sensory Interfaces: E.G. Artificial Vision II

Location: Virtual Only

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Program #/Poster #: P558.03

Topic: E.05. Brain-Machine Interface

Support: the National Key Research and Development Program 2017YFE0195500
NSFC 31371001

Title: Volitional Modulation of Temporally Precise and Structured Spiking Pattern in Motor Cortex

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Abstract: One of the most concerned and debated issues in neuroscience is how neurons communicate and convey information through spikes - firing rates or precise timing. There is accumulating evidence in favor of temporal coding. However, these pieces of evidence come dominantly from the sensory system. Little evidence is found in the motor cortex. Abundant studies have revealed the role of the motor cortex in driving muscles, which are often framed as a low-pass filter. Besides, the noisy nature of spiking time is inferred from the huge success of decoding kinematics by applying the Poisson rate code. These prevalent views underscore the validity and feasibility of coding with precise timing between neurons in the motor cortex. In this study, we designed a novel Brain-machine interface(BMI) paradigm, under which precisely timed spiking activity in the primary motor cortex (M1) was volitionally and robustly generated. We highlighted the feasible neural implementation of precisely timed spiking in the motor cortex that is hard to find and analyze under natural movements. Besides, we found the temporal offsets between the selected neurons were not uniformly distributed in the window of [0 15]ms defined by the BMI decoder. On contrary, neurons fire in smaller temporal offsets with higher probability, which further validates the temporal precision carried out in M1. To further understand how neurons in M1 can exhibit this temporally precise and structured pattern, we assigned pairs of neurons with various distances in between to be the “direct neurons” and then characterized the dynamics of “indirect neurons”. We found that traveling waves propagate through an area larger than the minimal area covering the “direct neurons”. Besides, the component of the propagation vector aligning to the “trigger-target” vector is larger than the component being perpendicular to that. Therefore, the traveling waves may serve to synchronize the neural activity of the recording sites. It was further supported by the finding that rats with longer distances between direct neurons performed worse than those with shorter distances. In addition, pronounced high gamma oscillation(60~90Hz) phase-locking to the discharging of direct neurons was found. High gamma oscillation phase locking has long been considered to arise from local E-I balance. This E-I balance may help to fine-tune the temporal structure by creating the “opportunity window”. In conclusion, our results suggest, in the primary motor cortex, temporal fidelity can be accomplished through the interplay between the global flow of excitation and local E-I balance.

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Poster

P558. Neuroprosthetics: Other Motor Sensory Interfaces: E.G. Artificial Vision II

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Program #/Poster #: P558.04

Topic: E.05. Brain-Machine Interface

Support: NIH Grant R01EY030569

Title: Minimally-invasive restoration of vision using photothermal stimulation of retinal neurons

Authors: *J. NIE[1], K. EOM[2], H. ALGHOSAIN[1], G.-M. GERBAKA[1], C. LANGEVIN[1], A. NEIFERT[1], X. GUO[1], V. COLVIN[1], J. LEE[1];
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Abstract: Age-related macular degeneration is an incurable eye disease leading to blindness due to photoreceptor degeneration. The current clinical retinal prosthetics to restore vision in blindness have too low resolution (worse than 20/200, legal blindness) to improve patients' daily-life functionality, and require complicated and often risky implantation surgery. Here, our study aims to prove a concept in blind model mice to restore higher resolution vision with a minimally-invasive approach with photothermal stimulation of retinal neurons. This novel approach has a potential of remotely activating retinal neurons with single-cell resolution, without either genetic manipulation or stem cell treatment. We intravitreally delivered Thy-1 antibody-coated gold nanorods (Thy-1-AuNRs) specifically targeting retinal ganglion cells (RGCs) and then stimulated RGCs with near-infrared (NIR) light, based on the well-studied phenomena of surface plasmon resonance and its photothermal effect. To validate the efficacy of this approach in blind Rho^{P23H} mice, we used single-cell fluorescent Ca^{2+} imaging *ex vivo* and electrophysiology *in vivo*. When we illuminated an NIR laser beam (780-nm wavelength, 15-μm beam diameter, 10-ms pulse, 33 Hz, 0.9-s duration) on individual cells of Thy-1-AuNR-delivered retinal explants, RGCs exhibited significant Ca^{2+} responses (0.16±0.04 % dF/F, n=163) while sham-delivered mice did not. In Thy-1-AuNRs-injected mice, visual cortical neurons were activated by NIR single-spot laser of retinal neurons (780-nm, single 100-ms pulse, 50 trials averaged), producing evoked potentials of 44.4±8.8 μV (n=9), but sham-delivered mice did not respond to NIR light. The amplitude of the NIR-evoked potentials was similar to visually evoked potentials in wild-type mice (49.4±14.2 μV, n=9; p=0.12). We proved the concept of activating retinal neurons with the minimally-invasive photothermal approach both ex vivo and in vivo. The result opens a possibility of restoring vision with an ideal pixel pitch (30 μm) while covering the macular visual field (10°) at an effective refresh rate (10 frames per second).

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Poster

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Title: Speech neuroprosthetic technology in a person with severe paralysis and anarthria

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Abstract: Speech is the most effortless, personal, and efficient mode of communication for almost every person. For individuals with severe paralysis who have lost the ability to speak, brain-computer interface (BCI) technology designed to restore communication has the potential to drastically improve their autonomy and quality of life. Remarkable progress has been made with existing communication BCI approaches, enabling users to type out intended messages letter-by-letter. A complimentary approach to restore communication to paralyzed individuals may be to decode speech directly from their brain activity as they attempt to speak. As part of a clinical trial, we implanted a custom 128-channel, high-density electrocorticography (ECoG) array over the surface of the speech motor cortex of a person with anarthria (loss of the ability to articulate speech) and spastic quadriparesis caused by brainstem stroke. We then collected brain activity as the participant attempted to say or silently mime a variety of utterances. Using deep artificial neural networks and natural-language models, we show that the participant's attempts to speak can be detected from brain activity and decoded into text in real time. We also characterize spatial, temporal, and spectral features of the cortical activity during the participant's attempted speech. Using brain activity collected over the course of more than 80 weeks, we show that chronic ECoG activity and discriminative spatial maps were relatively

stable throughout the study, facilitating consistent decoder performance without daily or weekly model recalibration. Our findings demonstrate that functional cortical representations of speech were present in a person after over 15 years of anarthria, enabling real-time text decoding directly from neural activity as the person attempted to speak using a clinically viable long-term neural interface.

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Poster

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Program #/Poster #: P558.06

Topic: E.05. Brain-Machine Interface

Support: William M. Wood Foundation, Bank of America Trustee

Title: Simulating the effect of asynchronous phosphene stimulation on artificial vision

Authors: N. MEITAL-KFIR, *J. S. PEZARIS;
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Abstract: Electrical micro-stimulation techniques used in visual prostheses are designed to restore visual functions following acquired damage or disease of the eye. Induced focal percepts, known as phosphenes, are achieved by applying localized electrical pulses to the visual pathway in a location that bypasses the impaired site. A given electrode elicits a single phosphene with a fixed receptive field. A complex artificial percept is achieved by coordinated electrical stimulation of multiple electrodes which translates the visual scene into an artificial percept made of a collection of individual phosphenes.Stimulation applied across a set of electrodes can take place simultaneously or may involve some form of temporal spread, resulting in asynchronous electrical pulses. The spatiotemporal pattern of activation across electrodes will determine the quality of the artificial percepts. Here, we describe an experiment to explore the temporal relationships between individual phosphenes and the impact of the electrical pattern on object binding and perception. We hypothesize that synchronous electrical stimulation will facilitate object perception as compared to asynchronous. A model system with sighted subjects, using virtual reality technologies, will be used to simulate how sight would appear to blind individuals with a visual prosthesis. We will assess the effect of temporal desynchronization of phosphene presentation on visual acuity in a simple sentence reading task based on the MNREAD test. Reading speed and accuracy will be tested at varying font sizes and under varying levels of phosphene temporal noise. The results of this study will be fundamental for

developing an efficient temporal pattern of stimulation and for the creation of functional prosthetic vision.

Disclosures: N. Meital-Kfir: None. **J.S. Pezaris:** None.

Poster

P558. Neuroprosthetics: Other Motor Sensory Interfaces: E.G. Artificial Vision II

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Program #/Poster #: P558.07

Topic: E.05. Brain-Machine Interface

Support: NIH Grant EY023336
NIH Grant NS103442

Title: Multi-electrode stimulation evokes consistent spatial pattern of phosphenes and improves phosphene mapping in blind subjects

Authors: *D. OSWALT[1], W. H. BOSKING[1], P. SUN[2], S. NIKETEGHAD[3], M. ARMENTA SALAS[4], U. K. PATEL[5], J. D. DORN[6], N. POURATIAN[7], M. BEAUCHAMP[1], D. YOSHOR[1]; [1]Neurosurg., Univ. of Pennsylvania, Philadelphia, PA; [2]Baylor Col. of Med., Houston, TX; [3]Bioengineering, Univ. of California Los Angeles, Panorama City, CA; [4]Neurosurg., UCLA, Los Angeles, CA; [5]Second Sight Med. Products, Inc., Sylmar, CA; [6]Second Sight Med. Prod, Sylmar, CA; [7]Neurosurg., Univ. of Texas, Southwestern, Dallas, TX

Abstract: Visual prostheses are envisioned to operate by combining phosphenes produced by stimulation of individual electrodes to produce a percept of an image. This strategy requires efficient and reliable methods for mapping individual phosphenes. We examined different mapping techniques in two blind subjects (BS1 and BS2) who are participants in the clinical trial for the Orion visual cortical prosthesis. Two fixation strategies were tested. For unimanual fixation, subjects placed their left index finger on a tactile fixation point. For bimanual fixation, subjects overlaid their right index finger atop their left index finger on the tactile fixation point. Two mapping strategies were tested. For absolute mapping, a single electrode was stimulated on each trial while for relative mapping, dynamic sequences of stimulation were delivered to three to five electrodes on each trial. Precision was quantified with 2DRMS. Bimanual fixation resulted in improved precision (lower 2DRMS) in phosphene localization compared to unimanual fixation (BS1: 4.0±2.6^o vs. 19±4.7^o, $p < 0.001$; BS2 4.1±2.0^o vs. 12±2.7^o, $p < 0.001$). Multi-point relative mapping had similar baseline precision as absolute mapping (BS1: 4.7±2.6^o vs. 3.9±2.0^o; BS2: 4.1±2.0^o vs. 3.2±1.1^o) but improved significantly when trial-to-trial variations were removed, revealing a consistently perceived and reported visual pattern across trials and sessions. The types of trial-to-trial variation observed were translational, rotational, and scaling variations, with translational errors accounting for a majority of the incongruity. Multi-point

sequences can capture basic retinotopic organization of visual cortex but tend to regularize distance between perceived phosphenes. Fitting a logarithmic map to these data can address this issue and generate a final map estimate. Relative mapping methods, combined with bimanual fixation resulted in the most precise and consistent estimates of phosphene location.

Disclosures: D. Oswalt: None. **W.H. Bosking:** None. **P. Sun:** None. **S. Niketeghad:** None. **M. Armenta Salas:** None. **U.K. Patel:** A. Employment/Salary (full or part-time):; Second Sight Medical Products. **J.D. Dorn:** A. Employment/Salary (full or part-time):; Second Sight Medical Products. **N. Pouratian:** None. **M. Beauchamp:** None. **D. Yoshor:** None.

Poster

P558. Neuroprosthetics: Other Motor Sensory Interfaces: E.G. Artificial Vision II

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Topic: E.05. Brain-Machine Interface

Support: NIH Grant EY023336
NIH Grant NS103442

Title: Modeling activation of early visual cortex and visual percepts produced by multi-electrode stimulation

Authors: *W. BOSKING, D. OSWALT, D. YOSHOR;
Neurosurg., Univ. of Pennsylvania, Philadelphia, PA

Abstract: Electrical stimulation of single electrodes implanted in visual cortex produces perception of a small flash of light known as a phosphene. Visual cortical prostheses (VCPs) are envisioned to operate by stimulating many electrodes at once to evoke perception of an image. However, in practice, results of multi-electrode stimulation have been difficult to predict. We developed a pipeline to predict the cortical activation and phosphenes that result from simultaneous multi-electrode stimulation. Our simulation framework builds on our previous model for predicting phosphene size based on current amplitude and the location of the electrode in the map of visual space (Bosking et. al., 2017). We expand on that model by simulating the resulting cortical activation produced by each electrode within a flat-map of the V1-V3 complex (Schira et. al., 2010). For each electrode, we assume a 2D-Gaussian profile of cortical activation. The center of each activation profile in visual space is set by the receptive field location for each electrode. The overall cortical activation for simultaneous stimulation of a group of electrodes is then calculated by linear addition of the activation profiles resulting from each electrode.This framework can be used to explore and predict the visual percepts evoked by VCPs with different implementations. For example, an array with a specific number of electrodes, and inter-electrode spacing, can be placed at a desired location over a portion of V1-V3. An input image can then be

fed to the model, simulating input from a camera to a VCP based on an array of this geometry. The stimulation current used for each electrode is set based on the gray scale value at each point in the image. Output from the model then provides a visualization of how the activation of each electrode ultimately leads to a full pattern of activity distributed over early visual areas, and to the visual percept predicted in response to the input image. Our model can be used to demonstrate the importance of balancing electrode spacing and stimulation currents for optimum performance of future VCPs. The equations used to predict cortical activation for each electrode, or the rules governing summation across electrodes, are modifiable to allow for flexibility in predicting visual percepts.

Disclosures: W. Bosking: None. **D. Oswalt:** None. **D. Yoshor:** None.

Poster

P558. Neuroprosthetics: Other Motor Sensory Interfaces: E.G. Artificial Vision II

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Topic: E.05. Brain-Machine Interface

Support: NIH Grant UH3NS103442

Title: Stability of phosphene thresholds and locations for a long-term cortical visual prosthesis - 3-year measurements with the Orion system

Authors: M. ARMENTA SALAS[1], U. K. PATEL[2], S. NIKETEGHAD[1], J. S. BELL[1], A. A. BARI[1], R. J. GREENBERG[3], D. YOSHOR[4], J. D. DORN[2], N. POURATIAN[5];
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Abstract: For the development of long-term visual prosthesis, it will be important to determine the safety and functionality of the system. After the pioneering work of Dobelle et al (2000) and Schmidt et al (1996), recent human clinical trials in and outside the U.S have started re-addressing these key questions. However, it will also be crucial to determine whether the systems can reliably generate visual percepts (i.e. phosphenes) after years of implantation. The Orion Early Feasibility Study sponsored by Second Sight (NCT03344848) has participants implanted for over three years, which allows us to start addressing some of these long-term stability questions. To test the system's ability to induce phosphenes, we measured thresholds at defined clinical time points (1, 6, 12 and 24/36 months) in the enabled electrodes, and semi-periodically for a subset of electrodes in four of the six subjects. These four subjects were recruited, implanted, and tested at the UCLA site. We used a modified binary-search algorithm to estimate thresholds, with the same stimulation parameters across subjects. We also captured the

phosphene locations at different time points, to evaluate whether the spatial map or the size of the phosphenes shifted over time. We were able to determine thresholds, at either 20 or 120 Hz, in most of the electrodes across all subjects (99.8%). There was a significant increase in thresholds from the first measurements to the last ones ($p < 0.01$), but phosphenes were still elicited within the system's stimulation ranges. The phosphene locations were mostly stable, although there was inter-session variability ($\mu_{shift} = 8°$, C.I. 0.6°). However, the gross locations within the upper and lower visual field were stable across most subjects. Three subjects had a significant increase in phosphene size from the first mapping measurements to the last one ($p < 0.001$). This increase correlated with the increase in thresholds ($\rho = 0.64$, $p < 0.001$), like that reported in sighted subjects (Bosking et al, 2017). These results are encouraging, since they highlight the ability to elicit phosphenes up to 3 years after device implantation, and well within safe stimulation parameters and the system's stimulation capabilities. Furthermore, the phosphenes are most likely perceived in the same location across the visual field, which means that once a map is determined from an initial fitting it could be sufficient for long-term use. Future experiments should address how video-mode functionality varies over time, and which behavioral or attentional aspects might affect this performance.

Disclosures: M. Armenta Salas: None. **U.K. Patel:** A. Employment/Salary (full or part-time):; Second Sight Medical Products. **S. Niketeghad:** None. **J.S. Bell:** None. **A.A. Bari:** None. **R.J. Greenberg:** F. Consulting Fees (e.g., advisory boards); Second Sight Medical Products. **D. Yoshor:** None. **J.D. Dorn:** A. Employment/Salary (full or part-time):; Second Sight Medical Products. **N. Pouratian:** None.

Poster

P558. Neuroprosthetics: Other Motor Sensory Interfaces: E.G. Artificial Vision II

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Topic: E.05. Brain-Machine Interface

Support: EU ERC FeelAgain Grant 759998
SNSF BRIDGE MYLEG Grant 193724

Title: Neuro-integration and bio-stability of polyimide-based electrodes implanted in human peripheral nerves

Authors: *G. VALLE[1], G. AIELLO[1], F. CIOTTI[1], P. CVANCARA[2], M. BUMBASIREVIC[3], T. STIEGLITZ[2], S. RASPOPOVIC[1];
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Abstract: In the last decades, the fascinating opportunity to communicate artificially with the brain using peripheral nerve stimulation (PNS) have been extensively explored in people with sensory-motor deficits. The use of innovative neural interfaces, enabling the interaction of robotics devices (e.g., limb prostheses) with the human somatosensory system, allows to restore the complex sense of touch and proprioception in people suffering from limb loss. These artificial sensory feedback was then successfully implemented in bionic prostheses and exploited by the users to accomplish different sensorimotor tasks with higher performance compared to conventional no-sensorized prostheses. In particular, multiple TIMEs (Transversal Intrafascicular Multichannel Electrode) have been implanted in the sciatic nerve of transfemoral amputees and utilized to restore sensory feedback in neuro-robotic leg prostheses. Although the provided benefits could have a tremendous potential impact on the patients' quality of life, they are strongly dependent by the electrodes and sensations stability mainly connected to the bio-integration of this neurotechnology over time. To this aim, here we unveil the characteristics of the artificial sensations restored using intraneural stimulation and we report the mechanisms of how they evolve during 3-month implants in human peripheral nerves. To do so, we assessed the electrically-evoked sensations of 3 transfemoral amputees implanted with multiple TIME in their tibial nerve. Firstly, we characterized in detail the perceptual thresholds, the sensation locations and qualities evoked by the neural stimulation at the brain level. Then, we evaluated the bio-integration of the device considering the time evolution of the evoked-percepts and their complex relationships, together with the tissue reactions and the electrode failures. The results of the neural stimulation collected during the months of implants together with the human nerve histologies and the electrode post-explant analyses allowed us to model the phenomenon using advanced computer simulations (Hybrid modelling). We considered both morphological and biophysical aspects of an human-machine connection exploiting neural interfaces and neuromodulation. Our results provide a comprehensive assessment of polyimide-based intra-neural electrode functionality, stability, and bio-integration and open a realistic pathway to refine and disseminate such implants in long-term clinical applications in humans. These neuroscientific evidences allow to optimize the neuroprosthetic device for the next applications.

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Poster

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Title: Model-based biomimetic encoding of proprioception for lower limb neuroprostheses

Authors: *F. CIOTTI[1], A. CIMOLATO[2], J. KLJAJIC[3], G. VALLE[1], S. RASPOPOVIC[1];
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Abstract: Restitution of the sense of proprioception to amputees is recognized as one of major challenges in the field of neuroprosthetics. Proprioception is essential in maintaining balance and performing motor tasks without visual aid: its absence limits the performance of the prosthetic users, their confidence, and ultimately increases the cognitive load required for the movement. Previous studies were successful in restoring tactile sensations through electrical nerve stimulation, but unfortunately reliable restoration of proprioception has never been achieved. We hereby propose a framework for the electrical stimulation of the peripheral nervous system leveraging experimentally validated models to elicit a physiological-like activity of muscle spindle afferents for different muscles simultaneously. In particular, a musculoskeletal model coupled with a Ia Prochazka muscle spindle transducer model modified to fit experimental human data is used to estimate the expected natural activity of muscle spindle afferents. The neurostimulation policy to replicate it is optimized by a computational model which estimates the effect of electrical nerve stimulation on each axon of the target nerve. A degree of complexity of anatomical reconstruction is higher of previously published models, including a complete 3D model of the nerves of interest of subjects with transfemoral amputation. The proposed stimulation system is evaluated against traditional methods for proprioception restoration such as linear charge modulation for knee angle encoding. The linear charge stimulation induces a response of muscle spindle afferents which is considerably different from the natural activity. The proposed solution is able to replicate the recruitment rate and frequency of the estimated physiological activity in terms of population average, with the limitation of synchronous activation as opposed to natural asynchronous point processes. We ultimately propose how we envision the system being implemented in the next generation of lower-limb neuroprostheses and show how it can be applied with minimal effort in other scenarios as for upper-limb prostheses.

Disclosures: F. Ciotti: None. **A. Cimolato:** None. **J. Kljajic:** None. **G. Valle:** None. **S. Raspopovic:** None.

Poster

P656. Threat, Fear, and Anxiety: Neural Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P656.01

Topic: G.06. Anxiety Disorders

Title: The effects of chronic cannabidiol (CBD) use in adolescent female mice on the prevalence of anxiety symptoms

Authors: *C. RECHSTEINER, E. MARTIN, T. LEAK, A. LACROSSE;
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Abstract: Cannabidiol (CBD) is a non-psychoactive compound derived from the cannabis plant, *Cannabis sativa.* Previous research has shown CBD's wide ranging therapeutic effects from antidepressant, antipsychotic, and anti-anxiety studies to preventing social-recognition deficit in an Alzheimer's disease model. However, there remains limited literature examining the effects of CBD treatment on anxiety levels in adolescent females. This is an important oversight as females have higher rates of diagnosed anxiety disorders compared to males. Furthermore, the effects from discontinuation of chronic CBD use has yet to be studied. Therefore, this *in vivo* study examines the effects of chronic oral consumption of CBD oil on anxiety symptoms with an adolescent, female mouse model. The current study also measures the effects of cessation from chronic CBD treatment on anxiety levels. We hypothesized that CBD use will decrease anxiety in adolescent users, and that the cessation of CBD use will potentiate levels of anxiety. In order to test our hypothesis, mice were divided into two groups, the CBD treatment group (n = 7) and a control group (n = 8). Based on the treatment group, mice received cheese containing 5 mg/kg CBD dissolved in olive oil or cheese containing olive oil only for 14 days to represent chronic use. Following treatment, all mice received cheese containing only olive oil for 10 days to represent discontinuation from treatment. The behavioral assays used in this study are the open field test and the elevated plus maze in order to measure anxiety symptoms in the mice at baseline (prior to beginning any treatment), after 14 days of chronic treatment, and again after CBD treatment was discontinued. All behavior data was digitally recorded and analyzed using an interrater reliability score of .80 or higher. Results indicate a greater decrease in anxiety symptoms in CBD treated mice compared to the control group after 14 days of treatment and after discontinuation. This suggests that CBD may have a lasting anxiolytic effect on adolescent, female mice. Future studies will investigate the significance of underlying neuronal changes due to CBD treatment in key brain regions relating to anxiety and cognitive ability in adolescent male and female mice.

Disclosures: C. Rechsteiner: None. **E. Martin:** None. **T. Leak:** None. **A. LaCrosse:** None.

Poster

P656. Threat, Fear, and Anxiety: Neural Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P656.02

Topic: G.06. Anxiety Disorders

Support: R21NS119991

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Brain & Behavior Research Foundation (NARSAD)
8G12MD007600
UPR MSC Chancellor's Office and Medicine Deanship
NSF PRCEN undergraduate and graduate Fellowships
NIH BP-ENDURE NEUROD-ID fellowship

Title: Evaluating the effects of glyphosate consumption on grooming behavior in rats

Authors: M. GONZÁLEZ-PEDRAZA, A. FIGUEROA-PÉREZ, M. CÁCERES-CHACÓN, H. HADDOCK-MARTÍNEZ, O. MARTÍNEZ-GUZMÁN, M. RIVERA-LÓPEZ, ***D. SIERRA-MERCADO**;
Anat. & Neurobio., Univ. of Puerto Rico Sch. of Med., San Juan, PR

Abstract: Glyphosate is the active ingredient in several commercial herbicides. Though initially considered safe for mammals, recent reports suggest that glyphosate administration at unusually high doses increases anxiety-like behaviors. However, few studies have evaluated the neurobehavioral effect of glyphosate exposure at the low concentration levels that the Environmental Protection Agency considers safe for human consumption. Grooming behavior, one of the most frequently performed behaviors in rodents, has been proposed as a measurement for states of generalized anxiety. Thus, we hypothesized that prolonged exposure to levels of glyphosate thought to be safe for humans will increase general anxiety-like behavior observed as abnormal grooming behavior in rats. To test this hypothesis, male Sprague Dawley Rats (n=13) had access ad libitum to drinking water containing glyphosate at a dose of glyphosate considered safe (2.0mg/kg) for 16 weeks, whereas control rats received filtered drinking water. After glyphosate exposure, rats were recorded in their home cage for 5 minutes. Videos were scored for total time spent grooming, as well as distance traveled and time where the rat was immobile. Preliminary data shows that glyphosate exposure does not affect the total number of grooming bouts (glyphosate: 0.67, control: 3.50; p=0.1268). Interestingly, however, glyphosate exposure decreased total distance traveled (glyphosate: 9.73, control: 15.79; p=0.0318) and increased the total time immobile (glyphosate: 429.30, control: 329.10; p=0.0364. Future directions include assessing the brain tissue for changes in biomarkers of neuronal activity in the bed nucleus of the stria terminalis, a brain region implicated in anxiety-like behaviors. The results of this study will allow us to elucidate the potential neurotoxic effects that glyphosate consumption may have on the brain, and how these effects could manifest as behaviors in rats.

Disclosures: M. González-Pedraza: None. **A. Figueroa-Pérez:** None. **M. Cáceres-Chacón:** None. **H. Haddock-Martínez:** None. **O. Martínez-Guzmán:** None. **M. Rivera-López:** None. **D. Sierra-Mercado:** None.

Poster

P656. Threat, Fear, and Anxiety: Neural Mechanisms

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Program #/Poster #: P656.03

Topic: G.06. Anxiety Disorders

Support: R01MH122712
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Brain and Behavior Research Foundation Young Investigator Award
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Title: Role of kappa opioid system in stress-induced anxiety-like behaviors in females

Authors: *A. KHURANA, C. BOUARAB, A. POLTER;
The George Washington Univ., Washington, DC

Abstract: Women are twice as likely as men to be diagnosed with a mood or anxiety disorder. Given that stress plays a significant role in the development of these disorders, sex differences in the response to stress are likely to be a critical factor in the enhanced vulnerability of females to these pathologies. Across both human populations and animal models, males and females exhibit divergent responses to stress at all levels, from molecular signatures to behavioral adaptations. Sub-chronic variable stress (SCVS) is a model of depression and anxiety in which female mice develop anhedonia and anxiety-like behaviors, but males do not.
Dysregulation of the mesolimbic reward circuitry is implicated in the pathophysiology of stress-related illnesses such as depression and anxiety. Dopaminergic (DA) neurons in the ventral tegmental area (VTA) are a major target of stress. Previous work has shown that acute stress leads to a days-long activation of kappa opioid receptors (KORs) in the VTA. We hypothesize that similar changes may underlie the female-specific behavioral deficits induced by SCVS. If this hypothesis is correct, pre-stress treatment with NorBNI (10mg/kg, ip) would prevent the stress-induced behavioral deficits observed in females. To test this, we used both male and female C57Bl/6J mice at 8-weeks old in a SCVS protocol (n=9 per group). Using dark-light box to assess anxiety-like behavior, we first confirmed that SCVS causes an increased latency to enter the light compartment in female mice only. Interestingly, this deficit is prevented by a pre-treatment with NorBNI. Next, we will inject the NorBNI post-stress to see if it can reverse this anxiety-like behavior in females. Together, this data indicates the important role of the kappa opioid receptor in the development of maladaptive behavioral responses in female mice.

Disclosures: A. Khurana: None. **C. Bouarab:** None. **A. Polter:** None.

Poster

P656. Threat, Fear, and Anxiety: Neural Mechanisms

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Program #/Poster #: P656.04

Topic: G.06. Anxiety Disorders

Support: NIH Grant R01MH123523
NARSAD Young Investigator Grant #22811

Title: Sex-specific effects of ventral hippocampal Egr1 expression on neuronal transcriptional programs and behavior

Authors: *D. ROCKS[1], I. JARIC[1], E. PURISIC[1], E. GALLO[1], J. M. GREALLY[2], M. SUZUKI[2], M. KUNDAKOVIC[1];
[1]Fordham Univ., Bronx, NY; [2]Ctr. for Epigenomic, Dept. of Genet., Albert Einstein Col. of Med., Bronx, NY

Abstract: Women have a two-fold increased risk for developing anxiety and depression disorders compared to men. Due to exclusion of female subjects from animal studies of the brain and behavior, the biological basis for this phenomenon is unknown. Clinical and epidemiological evidence indicate fluctuating ovarian hormones contribute significantly to this female-biased risk. Previously, we demonstrated that anxiety-like behavior changes in female mice over their estrous cycles. In our examination of the molecular basis for this behavioral phenotype, we demonstrated, for the first time, estrous-cycle dependent regulation of neuronal chromatin accessibility and gene expression in the ventral hippocampus, a brain region critical to emotion regulation and anxiety-like behavior in rodents. Analyses of these data indicated that Egr1, an estrogen-responsive immediate early gene product, is a putative regulator of these hormone-dependent changes in neuronal chromatin accessibility and gene expression within females. Here we aim to determine whether ventral hippocampal Egr1 expression mediates estrous-cycle dependent changes in chromatin accessibility, gene expression, and behavior. By measuring the behavior of female mice across the four phases of the estrous cycle, we found that females in the high-estrogenic (proestrus) phase have lower anxiety indices compared to females in the other, low-estrogenic phases. Serum estradiol levels positively correlate with a low-anxiety behavioral measure, further supporting an anxiety-reducing effect of estrogen. We also observed cycling expression of Egr1 and its putative target gene, Darpp-32, in the ventral hippocampus of females over the estrous cycle. In order to mechanistically link Egr1 expression to behavior, we delivered an adeno-associated virus carrying Egr1 into the ventral hippocampus of ovariectomized (OVX)-female and intact-male mice and performed anxiety- and depression-related behavioral tests. Without viral manipulations, we found that OVX females behave similarly to low-estrogenic (diestrus) females and exhibit higher anxiety indices compared to high-estrogenic (proestrus) females. Remarkably, overexpression of Egr1 in the ventral hippocampus of these OVX females reduces anxiety- and depression-related behavior and reproduces a proestrus-like behavioral phenotype. Our findings were sex-specific as we observed no behavioral changes in male animals. These results link Egr1 expression to estrous cycle-dependent gene regulation and behavioral plasticity and establish a foundation for the development of sex-specific treatments for anxiety and depression.

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Poster

P656. Threat, Fear, and Anxiety: Neural Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P656.05

Topic: G.06. Anxiety Disorders

Title: Viral infection of hypothalamic circuits: Implications for post-viral syndromes

Authors: *C. NIEMEYER[1], J. HASSELL, Jr[1], L. MERLE[1], B. BAXTER-BOLT[1], A. BUBAK[1], W. COLLOTY[1], D. RESTREPO[2], M. NAGEL[1];
[1]Univ. of Colorado Denver Sch. of Med., Aurora, CO; [2]Cell & Dev. Biology, Neurosci. Program, Univ. of Colorado Anschutz Med. Campus, Aurora, CO

Abstract: Renewed interest in the biological basis of post-viral syndromes (PVSs) has emerged during the COVID-19 pandemic because many individuals complain of persistent symptoms, including fatigue, anxiety, depression, and sleep disorders, which significantly affect quality of life. PVSs can also occur with other viruses, and interestingly, many of these symptoms can arise from hypothalamic dysfunction, leading to neuroendocrine, autonomic, and neurobehavioral changes. Understanding how viruses disrupt hypothalamic circuits to produce persistent clinical symptoms can potentially guide new therapies to prevent and treat specific features of PVSs. HSV-1 encephalitis (HSE), the most common cause of viral encephalitis, has been associated with PVSs, and can affect the hypothalamus. In humans with HSE, hypothalamic inflammation may be present; in HSV-1-infected rodents, viral antigen in the hypothalamus is seen and disruption of the hypothalamic-pituitary-adrenal axis is observed. However, detailed studies on how HSV-1 affects the hypothalamus, which can serve as a basis for how other viruses affect this structure and associated functions, are lacking. Here we investigate the pattern of viral antigen and the pattern of immune response in within the rostral caudal extent of the brain following intranasal inoculation of HSV-1 (10^5-10^6, McKrae strain) in C57bl/6 mice. We found the presence of HSV-1 DNA in the olfactory epithelium, olfactory bulb, and trigeminal ganglia, identifying possible routes of entry to the brain. We found striking specificity for subnuclei in the hypothalamus (bilateral dorsomedial, lateral hypothalamus, and paraventricular nuclei). Furthermore, we find extensive HSV-1 antigen in brainstem regions that project to hypothalamic subnuclei, including nucleus of the solitary track, and area postrema, suggestive of possible routes of infection to hypothalamic subregions. We also found extensive activation of iba-1 positive microglia in associated HSV-1 infected regions. Our anatomical results raise questions of whether functions associated with these infected hypothalamic regions are disrupted (including mood, appetite, sleep, and thermoregulation) and whether direct virus infection and/or

neuroinflammation are major contributors to pathology. Understanding mechanisms by which viruses affect hypothalamic circuits that may contribute to specific features of PVSs will provide the rationale for development of prognostic, preventive and treatment strategies.

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Poster

P656. Threat, Fear, and Anxiety: Neural Mechanisms

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Title: Glyphosate increases anxiety-like but not fear-like behaviors in rats

Authors: *M. CACERES-CHACON, A. FIGUEROA-PEREZ, H. HADDOCK-MARTINEZ, S. RODRIGUEZ-ROSADO, G. HERNANDEZ-BUSOT, M. RIVERA-LÓPEZ, O. MARTÍNEZ-GUZMAN, D. SIERRA-MERCADO;
Anat. and Neurobio., Univ. of Puerto Rico Sch. of Med., San Juan, PR

Abstract: Glyphosate-based herbicides (GBH) are the most commonly used herbicides worldwide. Glyphosate was initially considered safe for mammals because it acts by inhibiting a metabolic route not present in mammals. However, recent studies in rodents have shown that exposure to GBH at relatively high doses causes increased anxiety-like behavior. The Environmental Protection Agency has established a chronic reference dose for glyphosate of 2.0mg/kg daily, based mainly on toxicological studies that did not include behavioral assessments. Therefore, we aimed to evaluate the effect of prolonged oral consumption of GBH-contaminated drinking water on anxiety-like behavior. Furthermore, given the similarity in the neural circuit involved in anxiety-like behaviors and fear, we aimed to evaluate the effect of GBH on fear-like behaviors. To achieve this, rats were given a target dose of 2.0mg/kg daily of GBH in their drinking water. Control rats were treated in the same manner but received filtered drinking water. Of note, there were no differences in neither the amounts of water consumption, nor body weight between the groups ($p>0.05$). We assessed for anxiety-like behavior after 4

weeks of exposure using the open field test. No difference was observed in the amount of time spent in the center (control: 49.01, GBH: 47.68; p = 0.9051), or the number of entries into the center (GBH: 17.62, control: 17.33; p=0.9463). After 10 week of exposure, anxiety-like behavior was reassessed in the elevated plus maze where GBH caused a decrease in the amount of time spent in the open arms (GBH: 45.36, control: 103.7; p=0.0086), as well as a decrease in the number of entries into the open arms (GBH: 4.39, control: 9.50; GBH: 4.39; p=0.0214). Furthermore, we reassessed for anxiety-like behaviors after 14 weeks using the light/dark box. Interestingly, GBH reduced the percent of time spent in the light (control: 13.03, GBH: 3.32; 0.0173), suggesting that glyphosate may have a time dependent effect on anxiety-like behavior at low doses. Lastly, after 16 weeks of exposure, animals underwent delayed auditory fear conditioning and extinction. Contrary to our hypothesis, no differences in fear expression were observed during conditioning or extinction. Future directions include assessing other emotional memories and performing immunohistochemistry on brain regions involved in anxiety-like behaviors.

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Poster

P656. Threat, Fear, and Anxiety: Neural Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P656.07

Topic: G.06. Anxiety Disorders

Support: Alberta Innovates

Title: Mice with chronic, but not acute gut inflammation display generalized fear upon resolution of disease.

Authors: *C. E. MATISZ, M. PATEL, N. HONG, R. J. MCDONALD, A. J. GRUBER; Dept. of Neurosci., Univ. of Lethbridge, Lethbridge, AB, Canada

Abstract: Chronic inflammation in the body is a risk factor for mood disorders. This is particularly evident in inflammatory bowel diseases (IBD), which are highly comorbid with anxiety and other disorders, even during periods of IBD remission. A frequent feature of anxiety disorder is heightened threat assessment in benign contexts. Here, we test how the duration of gut inflammation evoked by dextran sodium sulfate (DSS) in mice affects such generalization using a fear to context paradigm. We found that neither an acute (5 days) or chronic (3x5 days) course of DSS-induced IBD affected learning or discrimination of safe/threating contexts at peak of disease. After remission of IBD symptoms one week later, however, the chronically-treated

animals exhibited equal amounts of freezing in both contexts. Their freezing behaviour was equivalent to other groups in the shock-associated context, but was greater in the benign context than other groups. Interestingly, all groups showed a preference for the benign context over the shock context. These data suggest that the repeated DSS treatment induced a generalization of fear to the benign context, even though learning & preference were intact.

Disclosures: C.E. Matisz: None. **M. Patel:** None. **N. Hong:** None. **R.J. McDonald:** None. **A.J. Gruber:** None.

Poster

P656. Threat, Fear, and Anxiety: Neural Mechanisms

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Program #/Poster #: P656.08

Topic: G.01. Fear and Aversive Learning and Memory

Support: NSF 2014862

Title: A streamlined approach to routine staging of the estrous cycle in behavioral studies

Authors: *G. M. RAIMONDI, A. K. ENG, M. A. BRITTING, M. P. KENNY, L. E. OSTROFF;
Univ. of Connecticut, Storrs, CT

Abstract: The amygdala is a central mediator of emotional responses, and the lateral amygdala (LA) circuit that underlies aversive auditory Pavlovian conditioning is among the most well-studied brain systems. However, decades of literature on its structure and function have focused largely on male subjects. Studies of sex differences have produced inconsistent results, especially when the female reproductive (estrous) cycle is taken into account. Estrous effects on fear- and anxiety-like behaviors have been reported, which makes the cycle a potential source of variability, but because tracking methods are subjective and not standardized, conflicting results are common between studies. We systematically compared several methods for monitoring the estrous cycle in rats, with the goal of finding a simple, reliable means of accounting for it in behavioral experiments such as Pavlovian conditioning. The 4-day cycle is traditionally divided into four stages of varying length that are defined by the appearance of vaginal epithelial cells. This method proved capricious; variability was high and some animals did not have regular cycles. Alternative methods such as vaginal resistance were unreliable. Rats are nocturnal, so the most variable phases of the cycle, which occur during the day, may not be relevant to behavior. Comparison across the light cycle revealed that samples taken in the dark cycle were extremely reliable, with nearly all rats having consistent 4-day cycles. This greatly simplified the tracking procedure, as only the most obvious day, the day of ovulation, needs to be detected. In addition, the common practice of excluding animals with irregular cycles was unnecessary. Using this

tracking method, we compared Pavlovian conditioning in males (n=24) and females (n=60) across the cycle, with all experiments performed in the dark. Relative to males and females on other days, females on the third day of the cycle showed less freezing behavior during training and testing. Quantification of c-Fos labeling after testing revealed that in males and females with equal freezing, females had more c-Fos cells in the intercalated cells of the amygdala but not in the LA. Although tracking the estrous cycle may not be necessary, it allows the cycle to be eliminated as a confound when sex differences are found, as in our c-Fos results. Overall, our approach is easier and less subjective than older methods and may allow improved reproducibility without undue effort in behavioral studies where the estrous cycle is of concern.

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Poster

P656. Threat, Fear, and Anxiety: Neural Mechanisms

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Topic: G.01. Fear and Aversive Learning and Memory

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Israel Science Foundation (ISF) Grant 1517/16

Title: Neural representations for distinct stress coping strategies in the rat

Authors: *G. RICHTER-LEVIN[1], C. MA[2], L. JING[3], L. XU[2];
[1]Univ. Haifa, Haifa, Israel; [2]Kunming Inst. of Zoology, Kunming, China; [3]Univ. of Haifa, Haifa, Israel

Abstract: Coping with stress is characterized by high individual variability, which is often attributed to either genetic background differences, sustained environmental conditions, or a combination of both. However, the neural mechanisms underlying coping style variability are yet poorly understood. Here we examined the impact of a single extended emotional challenge on coping style variability, and the associated involvement of the hippocampus, medial prefrontal cortex (mPFC) and periaqueductal gray (PAG). Sprague-Dawley rats (n = 170) were trained in an extended two-way shuttle avoidance (eTWSA) task for 7 days, and daily avoidance rates were measured. Forced swim test, elevated plus maze or Morris water maze were tested before or after eTWSA exposure. Excitotoxic lesion of the hippocampal dentate gyrus (DG) was performed by Ibotenic infusion. Transient pharmacological blocking of DG, mPFC or PAG was performed by

muscimol or CNQX+TTX infusion. Exposing rats to eTWSA was found to lead to naturally-developing dichotomous, not continuous, coping styles, we termed ‘Active’ or ‘Passive’. Prior emotional responses did not predict the developing coping style. Active coping was associated with beneficial outcomes including antidepressant effects and improved spatial learning. Passive coping led to impaired spatial retrieval. Active coping was abolished by lesioning or pharmacological blocking of the DG. Passive coping was prevented by blocking mPFC or PAG. A single exposure to a significant emotional challenge can lead, in otherwise healthy individuals, to dichotomous development of an active or passive coping style, with distinctive neural correlates, relating to emotional vulnerability or resilience.

Disclosures: G. Richter-Levin: None. **C. Ma:** None. **L. Jing:** None. **L. Xu:** None.

Poster

P656. Threat, Fear, and Anxiety: Neural Mechanisms

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Program #/Poster #: P656.10

Topic: G.01. Fear and Aversive Learning and Memory

Support: NINDS

Title: A signaled locomotor avoidance action is fully represented in the neural activity of the midbrain tegmentum

Authors: S. HORMIGO, B. SHANMUGASUNDARAM, J. ZHOU, ***M. CASTRO-ALAMANCOS**;
UConn Hlth. Sch. of Med., Farmington, CT

Abstract: Animals, including humans, readily learn to avoid harmful and threatening situations by moving in response to cues that predict the threat (e.g., fire alarm, traffic light). During a negatively reinforced sensory-guided locomotor action, known as signaled active avoidance, animals learn to avoid a harmful unconditioned stimulus (US) by moving away when signaled by a harmless conditioned stimulus (CS) that predicts the threat. CaMKII-expressing neurons in the pedunculopontine tegmentum area (PPT) of the midbrain locomotor region have been shown to play a critical role in the expression of this learned behavior, but the activity of these neurons during learned behavior is unknown. Using calcium imaging fiber photometry in freely behaving mice, we show that PPT neurons sharply activate during presentation of the auditory CS that predicts the threat - before onset of avoidance movement. PPT neurons activate further during the succeeding CS-driven avoidance movement, or during the faster US-driven escape movement. PPT neuron activation was weak during slow spontaneous movements but correlated sharply with movement speed and, therefore, with the urgency of the behavior. Moreover, using optogenetics, we found that these neurons must discharge during the signaled avoidance interval

for naive mice to effectively learn the active avoidance behavior. As an essential hub for signaled active avoidance, neurons in the midbrain tegmentum process the conditioned cue that predicts the threat and discharge sharply relative to the speed or apparent urgency of the avoidance (learned) and escape (innate) responses.

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Poster

P750. Cocaine: Cognitive and Behavioral Effects

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Program #/Poster #: P750.01

Topic: G.09. Drugs of Abuse and Addiction

Support: T32-AI-049815
P30 AI078498
P50 HD103536

Title: Assessing Combinatorial Effects of HIV Infection and Former Cocaine Dependence on Cognitive Control Processes: A Functional Neuroimaging Study of Response Inhibition

Authors: *K.-M. WAKIM-TAKAKI[1], E. G. FREEDMAN[1], M. TIVARUS[2], A. HEINECKE[3], J. J. FOXE[1];
[1]Neurosci., [2]Imaging Sci., Univ. of Rochester, Rochester, NY; [3]Neurosci., Maastricht, Maastricht, Netherlands

Abstract: Individuals with a diagnosis of co-morbid HIV infection and cocaine use disorder are at a higher risk of poor health outcomes. Active cocaine users, both with and without HIV infection, show clear deficits in response inhibition and other measures of executive function instrumental in maintaining drug abstinence --a factor which may complicate treatment. Neuroimaging and behavioral evidence indicate normalization of executive control processes in former cocaine users as a function of the duration of drug abstinence, but it is unknown to what extent a co-morbid diagnosis of HIV affects this process. To this end, we investigate the combinatorial effects of HIV and cocaine dependence on the neural substrates of cognitive control in cocaine-abstinent individuals with a history of cocaine dependence. Blood-oxygen level dependent signal changes were measured as 86 participants performed a Go/NoGo response inhibition task while undergoing functional magnetic resonance imaging (fMRI). Four groups of participants were selected based on HIV and cocaine-dependence status. Participants affected by both conditions demonstrated the lowest response accuracy of all participant groups. In a region of interest analysis, this reduced task performance was coupled with left putamen and midline-cingulate hyperactivation during response inhibition relative to participants with only one

condition. Results of a whole-brain analysis indicate response inhibition-related hyperactivation in the bilateral supplementary motor area, right hippocampus, bilateral superior temporal gyrus, right sensory association area, and right visual-motor area. These results indicate complex and interactive alterations in neural activation during response inhibition and highlight the importance of examining the neurocognitive effects of co-morbid conditions.

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Poster

P750. Cocaine: Cognitive and Behavioral Effects

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Program #/Poster #: P750.02

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH grant EB022015-03
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Title: Simultaneous, real-time, pharmacokinetic and pharmacodynamic (behavioral) measurements of psychoactive drugs in the brains of awake, ambulatory rats

Authors: *J. GERSON[1], K. HONEYWELL[2], N. EMMONS[3], J. GIBSON[3], K. PLAXCO[3], T. E. KIPPIN[3];
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Abstract: Psychoactive drugs exert their behavioral effects via concentration-dependent actions upon neural circuits. However, to date, the temporal resolution with which we can measure these compounds is *orders of magnitude* too slow to capture the concentrations associated with altered physiological processes. Thus motivated, our group has recently developed a new technology, electrochemical aptamer-based sensors (E-AB sensors) that can detect physiologically relevant concentrations of specific drugs *in-situ,* in awake, freely behaving animals. Procaine and cocaine are two psychoactive compounds that, although similar in their chemical structure, elicit opposing behavioral effects when administered. By adapting a previously presented aptamer we have improved the sensitivity of our E-AB sensor to both drugs by an order of magnitude, as well as enhanced drift characteristics observed in vivo. Here, we tested this new technology on rats that have been administered procaine intravenously and measured the in-brain concentrations of this drug while performing concurrent locomotion and physiological monitoring. We successfully measured procaine concentrations every 10 s in real-time and generated subject-specific full pharmacokinetic profiles which are compared to the pharmacodynamic (behavioral) profile. Increases in procaine concentration coincide with

decreased locomotion. We also show in vitro data suggesting a route forward for the detection of cocaine at physiologically relevant concentrations in rats. In conclusion, we have developed a novel, potentially revolutionary, technology that can measure the pharmacokinetics of psychoactive drugs within the brains of awake, freely behaving animals.

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Poster

P750. Cocaine: Cognitive and Behavioral Effects

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Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P750.03

Topic: G.09. Drugs of Abuse and Addiction

Support: R21 DA045335

Title: Local field potential oscillatory dynamics in prelimbic cortex to nucleus accumbens core circuitry after extended access cocaine self-administration and prolonged abstinence

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Abstract: Individuals with a substance use disorder often show increased rates of drug craving and seeking following abstinence from drug use. Further, neuroadaptations occur including changes in local field potential (LFP) activity that result from repeated drug use and abstinence. Work from our lab has shown that abstinence from cocaine self-administration results in a dampening of activity in several reward-related areas including the prelimbic cortex (PrL) and nucleus accumbens core (NAc core). Further, our recent work has also shown that transcranial alternating current stimulation can reverse cocaine-induced neural reductions in PrL-NAc core coherence and restore cocaine related deficits in behavioral flexibility. Here, we investigated the effects of extended access cocaine self-administration (6 hr/d for 2 weeks) and prolonged (1 month) abstinence on resting state (spontaneous) LFP activity using electrophysiological methods. Adult, male Sprague Dawley rats were trained to self-administer cocaine (n=10, 0.33 mg/inf, 6 h per session) or water (n=3) during 14 daily sessions, followed by 30 days of experimenter-imposed abstinence. LFP activity in the PrL and NAc core was recorded for 15 minutes at 3 different timepoints: before self-administration training began, after self-administration training was complete but before abstinence, and immediately following prolonged abstinence. Behavior was recorded during self-administration and in two extinction

sessions, one that occurred immediately after self-administration training, and the second after 1 month cocaine abstinence. Preliminary behavioral data indicates that cocaine rats exhibited a significant escalation in drug taking when comparing days 3 and day 14 (t_9= 2.902, p= 0.0175), and showed increases in extinction presses after prolonged abstinence. Preliminary LFP data suggests that rats with a history of cocaine exhibited an overall reduction in resting state PrL to NAc core coherence immediately following self-administration compared to controls. Ongoing studies are increasing the number of control (water) rats and examining relationships between LFP signaling and aspects of behavior such as escalation and incubation of drug seeking. Collectively, these data will provide insight into how a history of extended access cocaine self-administration and prolonged abstinence may alter LFP oscillatory firing dynamics within the PrL to NAc core circuit.
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Disclosures: M.L. Ngbokoli: None. **L.P. Edwards:** None. **R.M. Carelli:** None.

Poster

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Title: Cerebellar correlates of the transition from goal-directed behaviors to habits

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Abstract: Previous studies have demonstrated the ability of addictive drugs to create an imbalance between flexibility and repetition of behavioral patterns that can be biased towards incentive habits, becoming insensitive to devaluation. Our group found that the establishment of cocaine-induced incentive memory increases neural activity in the cerebellar cortex and cerebellar deep nuclei as well as the expression of perineuronal nets (PNNs) which are specialized extracellular matrix structures that stabilize synapsis. In the present research, we wonder whether a treatment with cocaine or alcohol in different phases of the learning process will increase insensitivity to reward devaluation and thus to facilitate the transition from goal-directed behavior to habit in natural rewarded behaviors. We also aimed to explore the cerebellar

correlates of this transition. Cocaine was administered every two days in ascending doses (2, 4 & 10 mg/kg; intraperitoneal injection (IP)) for 7 weeks. Different groups of adult male rats were exposed to intermittent voluntary alcohol consumption in ascending concentrations too (2,5, 5 & 10%) for 12 weeks. Animals were exposed to drug treatments before or after instrumental learning. Control groups were treated with saline or consumed water, respectively. Rats were trained in operant boxes to consume two kind of palatable pellets (banana and chocolate) in a random ratio schedule (RR10), then rewards were devalued by 1h of prefeeding and sensitivity to reward devaluation was assessed under extinction in two tests of 5 min. At the end of the last extinction test, rats were perfused and cerebella were processed for cFos and WFA immunofluorescence. Cocaine-treated animals maintained goal-directed patterns in all the conditions despite saline-treated rats showed insensitivity to reward devaluation when instrumental learning occurred before IP administration. However, in the alcohol groups (treatment before and after learning) a tendency towards insensitivity was observed. Related to cerebellar correlates, we found a reduction of the number of PNNs in lobule VIII of the cerebellum in alcohol pretreated rats. In conclusion, both alcohol groups showed a tendency to insensitivity to devaluation, but only saline treated rats exhibited this tendency when the treatment is administered after the learning process. On the other hand, cerebellar cFos and PNN expression depend on the drug and the cerebellar region evaluated.

Disclosures: P. Ibáñez-Marín: None. **R. Redón-Clofent:** None. **H. González-García:** None. **A. Sanchez-Hernandez:** None. **J. Guarque-Chabrera:** None. **M. Miquel:** None.

Poster

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Support: Whitehall Foundation

Title: Cocaine experience alters discrete aspects of risky decision-making within the Balloon Analogue Risk Task

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Abstract: The decision to endure risk in order to obtain reward progresses through a series of discrete cognitive processes: reward valuation, risk assessment, and feedback integration to update future expectations. While optimal decision making requires dynamic updating of anticipated costs and rewards, each facet of the decision is vulnerable to corruption, such as

impulsivity driven by prior long-term experience with drugs of abuse. Here, rodents complete the Balloon Analogue Risk Task (BART) to evaluate these discrete elements of risk-taking behavior and their vulnerability to prior life experiences, namely drugs and stress. In the BART task, when animals engage with a lever of learned reward probability, they can press that lever multiple times to cache sucrose pellets, with the knowledge that over-exploitation of the lever on any given trial can lead to a "bust", wherein all rewards are lost. Moreover, animals can choose between two levers of different reward and risk contingencies (i.e. "risky choice") and then press that chosen lever to their desired amount (i.e. "risky action").
While the neural circuitry underlying risky decision-making is known to include prefrontal cortical projections to the dorsomedial and ventral striatum (i.e. nucleus accumbens), differentiation in activity across the different aspects of the decision (e.g. risky choice versus risky action) remains poorly understood. Moreover, cortico-striatal activity can undergo dramatic remodeling within abstinence from drugs of abuse, such as cocaine, creating vulnerability to poor impulse control and risky behaviors. Here, animals self-administer cocaine (0.5 mg/kg/infusion) or saline over 14 short-access (2 hour) sessions and are then forced into home-cage abstinence. Animals are then trained to press one of two levers to obtain 40 mg sucrose pellets before receiving implants of electrophysiological arrays within prefrontal cortex and striatal areas. In male (but not female) rats performing our BART task, cocaine experience enhanced risky decision-making and correlates with differences in neural encoding within cortico-striatal circuitry.

Disclosures: M.C. Hillman: None. **E.C. Warnock:** None. **K.A. Siletti:** None. **M. Navarro:** None. **K.T. Brown:** None. **M.P. Saddoris:** None.

Poster

P750. Cocaine: Cognitive and Behavioral Effects

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Topic: G.09. Drugs of Abuse and Addiction

Support: NIDA R01-DA034696
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University of Minnesota Doctoral Dissertation Fellowship

Title: Reduced GIRK channel activity in prelimbic pyramidal neurons impairs extinction of cocaine-induced conditioned place preference in male, but not female, mice.

Authors: *T. ROSE, K. D. WICKMAN, E. MARRON;
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Abstract: Drug-induced neuroadaptations in the mPFC have been implicated in addictive behaviors. Repeated cocaine exposure has been shown to increase pyramidal neuron excitability in the prelimbic (PL) region of the mouse mPFC, an adaptation attributable to a suppression of $GABA_B$ receptor-mediated signaling through G protein-gated inwardly rectifying potassium/K^+ (GIRK) channels. Prior work in our laboratory found that selective suppression of GIRK channel activity in PL pyramidal neurons in drug-naïve mice enhanced the motor-stimulatory effect of cocaine, without affecting basal activity. The goal of this exploratory study was to examine the impact of GIRK channel plasticity in PL pyramidal neurons on affective and reward-related behaviors. Specifically, we used a viral GIRK channel ablation approach involving drug-naïve adult (7-8 wk) $Girk1^{fl/fl}$ mice and intra-PL delivery of viral Cre or control vectors to selectively reduce GIRK channel activity in PL pyramidal neurons. The behavioral impact of this manipulation was assessed in the elevated plus maze (EPM), forced swim test (FST), and cocaine-induced conditioned place preference (CPP) paradigm. GIRK channel ablation did not impact innate avoidance behavior in the EPM (male: p=0.177, n=16; female: p=0.963, n=9-14) or behavioral despair in the FST (male: p=0.533, n=16; female: p=0.881, n=9-14). While this manipulation did not alter the acquisition of cocaine-induced CPP (male: p=0.402, n=11-12; female: p=0.435, n=8-9), it impaired the extinction of cocaine-induced CPP in male (p=0.0098, n=11-12), but not female (p=0.166, n=8-9), mice. This finding suggests that cocaine-evoked GIRK channel plasticity in PL pyramidal neurons may facilitate prolonged drug-seeking behavior in male mice.

Disclosures: T. Rose: None. **K.D. Wickman:** None. **E. Marron:** None.

Poster

P750. Cocaine: Cognitive and Behavioral Effects

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Topic: G.09. Drugs of Abuse and Addiction

Support: T32AA007471

Title: The mPOA modulates sex differences in neural and behavioral responses to cocaine

Authors: *J. MARTZ, A. VASQUEZ, J. M. DOMINGUEZ;
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Abstract: Males and females respond differently to drugs of abuse; this difference is in part due to the influence of gonadal hormones such as estradiol. The medial preoptic area (mPOA) in the hypothalamus is an important integrator of neuroendocrine signaling and a key regulator of both natural and drug-related reward. While the mPOA is a sexually dimorphic structure and implicated in several behavioral sex differences, whether it is involved in the regulation of sex

differences in cocaine response remains unclear. To investigate this, we examined sex differences in cocaine-induced locomotion and neural activation in the mPOA of male and naturally-cycling female rats. We found a correlation between cocaine-induced locomotion and neural activation in the mPOA of both males and females. However, this relationship was only present in females in high estradiol phases of their estrous cycle, but not in females in low estradiol phases of their cycle. Using iontophoretic injections of the retrograde tract tracer Fluoro-gold into the ventral tegmental area (VTA) of male and female rats, we also explored possible sex differences in neural projections from the mPOA to the mesolimbic reward system. In particular, we analyzed sex differences in estrogen-receptor containing efferents. Results indicate that while projections from the mPOA to the VTA were comparable between males and females, projections that contained estrogen receptors were significantly greater in females than in males. Taken together, these findings further point to the mPOA as an important regulator of endocrine-mediated sex differences in cocaine response.

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Poster

P750. Cocaine: Cognitive and Behavioral Effects

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Program #/Poster #: P750.08

Topic: G.09. Drugs of Abuse and Addiction

Title: Housing environment affects anxiety-like behaviors but not cocaine-induced sensitization in female and male mice

Authors: A. SHAPIRO, V. KIMBLE, I. KEREN, S. NOBOA SALOBO, R. DIVEKAR, M. GIRONTA, M. J. KIM, J. WILHELM, *C. R. MCKITTRICK;
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Abstract: Environmental conditions have been shown to have a significant impact on behavior and mental health. In humans, social isolation and chronic psychosocial stress are risk factors for substance use disorders and for affective disorders such as anxiety and depression whereas environments providing social support and cognitive enrichment have been shown to ameliorate some of these adverse effects. Similar outcomes have been observed in animal models; however, the majority of these studies have been conducted only in male subjects. We investigated the effect of housing conditions on behavioral responsiveness to cocaine and on anxiety-like behavior in both female and male mice. Adult C57BL/6J mice were housed in environments that varied in social structure and cognitive enrichment: Impoverished (IE: individually housed with a single compressed cotton bedding sheet), Standard (SE: same-sex groups of 4 with aspen bedding and modest enrichment), or Enriched (EE: same-sex groups of 4 with aspen bedding, with multiple enrichments changed twice weekly). Animals were introduced to their respective

housing conditions 4 weeks prior to behavioral testing. For the sensitization study, locomotor activity was monitored in an 0.5 m^2 chamber for 10 minutes before and 50 minutes after i.p. administration of either saline or 20 mg/kg cocaine. On d0 of the sensitization protocol, half of the animals received saline and half received cocaine; all animals received cocaine on d7, d14 and d28 following the initial session. Locomotor sensitization was observed in all groups, with distance traveled increasing progressively with repeated intermittent administration of cocaine; however, there was no effect of environment or sex. Anxiety-like behaviors were assessed in the elevated plus maze (EPM) after animals had been drug-free for over one week. Behavior in the EPM was significantly affected by environment but not by prior cocaine exposure. Total distance traveled and distance traveled in the closed arms was significantly greater in the IE group compared to the EE group. The effect was greater in males than females, although there was more variability among the males, likely due to the fact that several male SE and EE groups had to be separated due to fighting. The observation that housing environment affected anxiety-related behaviors but not the sensitization response suggests that these behavioral processes involve distinct neural pathways. In addition, the robust behavioral sensitization to cocaine exhibited by both female and male mice supports the routine inclusion of females in studies of substance-use disorders.

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Poster

P750. Cocaine: Cognitive and Behavioral Effects

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Program #/Poster #: P750.09

Topic: G.09. Drugs of Abuse and Addiction

Title: Behavioral implications of cell-specific knockdown of D2 receptors on the cocaine response and the vulnerability for developing addictive-like behaviors

Authors: E. M. MURRAY, R. BOCK, A. MASKERI, I. BLEIMEISTER, M. BOCARSLY, V. A. ALVAREZ;
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Abstract: Cocaine is a monoamine transport blocker, which has shown to increase dopamine within the striatum. Dopamine in the striatum is important for motivation, motor function, and reinforcement. The actions of dopamine in the striatum are mainly driven by two types of receptors: D1 and D2. Clinical imaging studies showed that chronic cocaine abusers have lower levels of D2 receptor (D2R) availability within the striatum. A few studies showed that stimulant

abuse also correlates with low levels of D2R availability in the midbrain region. It is not yet clear how the D2R expressed in striatum or the midbrain contribute to cocaine taking behavior and the vulnerability to develop compulsive-like behaviors. Previous work using transgenic mouse models have shown that deletion of D2R within the striatum produces different behavioral outcomes, sometime even opposite effects, compared to D2R deletion in the midbrain. However, patients suffering from substance use disorders display lower levels rather than complete loss of D2R function, so knockdown of D2R is a more realistic manipulation which better mirrors the observations in the clinical condition. We hypothesize that lower levels of D2R in the striatum and the midbrain drive different alterations to the synaptic connectivity throughout the basal ganglia, which contribute to selected changes in cocaine driven behaviors. We also speculate that low levels of D2R in both striatum and midbrain might synergize to produce a vulnerable phenotype to cocaine abuse. The goal of this project is to test these hypotheses and dissect out the contribution of the D2R in the striatum and the midbrain to the behavioral response to cocaine. We generated transgenic mice with low levels of D2R in either the striatum (striatum-D2HET), the midbrain (auto-D2HET), or both (double-D2HET). We tested a battery of striatal-controlled behaviors and the response to cocaine. We also conducted intravenous cocaine self-administration (IV-SA) and compared the behavior among these three genotypes and littermate controls. Our data so far shows that mice with auto-D2HET and striatum- D2HET often display opposite changes in dopamine-related and cocaine-related behaviors. We also found that double-D2HET often compensate better than the region specific HETs and they show no evident alteration. Double-D2HET resemble auto-D2HET alterations on some behaviors while they mimic phenotype of striatum-D2HET on other behaviors. Completion of this project will give us a better understanding of the unique contributions of striatal and midbrain D2R to dopamine-dependent behaviors and the response to cocaine to better inform new treatment for substance use disorders.

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Poster

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Topic: G.09. Drugs of Abuse and Addiction

Title: Wild type and trpc4 knockout rats show no effect of cocaine on performance of reversal learning, while amphetamine disrupts reversal performance of both groups

Authors: *W. D. KLIPEC, B. POHL, M. VORHIES, L. GUCK, S. SEETHAMRAJU, M. JOHNSON, M. GEREAU, D. SHIVAPUJA, C. VARGAS, S. ABDULLAHI, J. MARTIN; Psychology & Neurosci., Drake Univ., Des Moines, IA

Abstract: Previous research in our lab showed that the deletion of the *trpc4* gene produced a brain-wide elimination of TRPC4 channels, including those within a subpopulation of TRPC4-bearing dopamine neurons in the ventral tegmental area (VTA). That research also showed that the *trpc4* knockout rat (KO), compared to normal (WT) rats exhibit reduced cocaine self-administration and reduced cell firing rates in VTA dopamine neurons, with no differences in simple or complex reversal learning using food or water rewards. More recently, we found that acute cocaine administration produces a significantly greater dose-dependent increase in early responding (impulsivity) in WT compared to KO rats maintained on a DRL reinforcement schedule. Here, we examined the effects of acute cocaine and amphetamine administration on the performance of a Y-maze reversal task, by switching between running toward the lighted alley and reversing to running to the unlighted alley. In Experiment 1, the effects of cocaine (0, 5, 10, and 15mg/kg, i.p.) on KO and WT rats were tested after a series of four reversals. We found no difference between KO and WT rats in the acquisition of the reversal task, or disruption in performance at any dose. To determine if the lack of a disruption in performance was due to overlearning the reversal, we conducted Experiment 2, using different KO and WT rats, administering both acute cocaine (0, 5, 10, and 15mg/kg, i.p.) and d-amphetamine (0, 1, 2, 3 and 4mg/kg, i.p.), tested after one reversal. Once again, we found no differences between KO and WT rats in the acquisition of the reversal task, or disruption in performance at any dose of either drug; however, amphetamine produced a significant increase in errors for both KO and WT rats. These results replicate the finding that KO and WT rats do not differ in learning a reversal task. Importantly, these results also show that the reduction in cocaine self-administration and increased impulsivity on the DRL task in KO compared to WT rats are specific to those tasks and do not represent a more general disruption of learning or performance. The finding that amphetamine but not cocaine disrupts performance in both KO and WT rats is interesting and warrants further research. In conjunction with our previous research, these findings suggest a potentially important role for the TRPC4 channels in cocaine addiction and dopamine disorders.

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Poster

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Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant DA044297
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Title: Isoform-selective PI3-kinase inhibition confers partial resilience to cocaine cessation-induced anxiety-like behavior

Authors: *B. R. BARBEE[1,2,3,4], S. L. GOURLEY[1,2,3,4];
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Abstract: Phosphoinositide 3-kinase (PI3K) is a multi-subunit signaling complex that phosphorylates phosphoinositides, membrane-embedded second messengers that are critical for synaptic and structural plasticity of neurons. Cocaine potentiates PI3K-Akt-mTOR cascade activity, and this activation persists beyond the period of drug exposure. The PI3K p110β isoform is neuronally enriched and able to control PI3K signal propagation, allowing for manipulation of PI3K activity in a more targeted manner than broad-spectrum PI3K inhibition. Cessation of cocaine use triggers anxiety-like behavior in humans and rodent models, and anxiety can be a causal factor in relapse. Here, we used viral-mediated gene silencing to reduce expression of p110β in the dorsomedial prefrontal cortex (dmPFC). Isoform-selective PI3K inhibition mitigated anxiety-like behavior triggered by acute cocaine. Interestingly, however, a history of repeated cocaine exposure occluded this resilience, presenting an opportunity to compare immediate-early gene expression between cocaine-vulnerable and cocaine-resilient mice. We examined 22 brain regions and found that resilient mice - those displaying less anxiety-like behavior - displayed lower immediate-early gene expression in the claustrum and lateral hypothalamus. We next found that chemogenetic stimulation of the claustrum induced anxiety-like behavior. Future studies will determine whether suppressing p110β in the dmPFC combats anxiety-like behavior via connections with the claustrum. Our findings suggest that isoform-selective PI3K inhibition mitigates cocaine cessation-elicited anxiety-like behavior, likely via coordinated brain regions and circuits.

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Poster

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Topic: G.09. Drugs of Abuse and Addiction

Support: P50-DA006634

Title: Modulation of cocaine self-administration behaviors with nicotine co-administration in rats

Authors: *M. H. DAWES, P. M. ESTAVE, S. ALBERTSON, S. R. JONES;
Wake Forest Univ. Sch. Med., Winston Salem, NC

Abstract: Prevailing preclinical models of cocaine use have not resulted in an FDA-approved treatment for cocaine use disorder, potentially due to a focus on cocaine use in isolation. It is known that clinical populations with cocaine use disorder are predominately polysubstance users, with high co-use of cocaine and nicotine. Nicotine has been shown to increase cocaine potency and enhance the reinforcing effects of cocaine, suggesting that examination of the combined use of cocaine and nicotine may provide a more translational model for cocaine use disorder. Prior studies suggest that non-contingent nicotine exposure is not sufficient to alter cocaine self-administration in rats; therefore, this experiment looked to examine if concurrent, co-administered cocaine and nicotine would increase responding for long access cocaine self-administration. Male Sprague Dawley rats were trained to self-administer either a concurrent combination of cocaine and nicotine (0.375 mg/kg/infusion cocaine + 0.03 mg/kg/infusion nicotine) or cocaine alone (0.375 mg/kg/infusion). After completing acquisition (defined as 2 consecutive days of 20 infusions), rats were tested on a long access, fixed ratio 1 schedule of reinforcement (6 hour sessions, unlimited infusions). Unexpectedly, rats self-administering combined cocaine and nicotine responded significantly less than rats self-administering cocaine alone during both acquisition and long access schedules. Further, rats with a history of concurrent cocaine and nicotine did not self-administer nicotine alone under the same long access self-administration conditions. Together, these data highlight the importance of nuance in nicotine self-administration paradigms, particularly context and environmental cues, and suggest that current widely-used preclinical models of cocaine administration may not fully recapitulate cocaine use in clinical populations.

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Title: N-ethoxycarbonyl-2-ethoxy-1,2-dihydroquinoline potentiates cocaine-induced locomotor activity in preweanling rats: role of 5-$HT_{1A/1B}$ receptors

Authors: A. TERAN, J. A. M. ROBINSON, G. I. RAMIREZ, C. G. KATZ, ***S. A. MCDOUGALL**;
Dept. of Psychology, California State Univ., San Bernardino, CA

Abstract: In adult rats and mice, dopamine (DA) receptor inactivation causes predictable behavioral effects. For example, the locomotor-inducing actions of D2 receptor agonists are almost completely eliminated if adult rats are pretreated with the peptide-coupling agent *N*-ethoxycarbonyl-2-ethoxy-1,2-dihydroquinoline (EEDQ). In preweanling rats, on the other hand, EEDQ does not attenuate the locomotor activating effects of D2 receptor agonists like *R*-propylnorapomorphine (NPA). More surprisingly, systemically administered EEDQ actually potentiates the cocaine-induced locomotor activity of male preweanling rats. Specifically, cocaine produces significantly more locomotor activity in EEDQ-treated male preweanling rats than vehicle-treated rats. It is uncertain why EEDQ leaves the NPA-induced locomotor activity of preweanling rats unaffected, while potentiating cocaine-induced locomotor activity, but the answer may involve serotonin (5-HT) system functioning. Specifically, NPA is a selective agonist at D2 receptors, whereas cocaine increases synaptic levels of both DA and 5-HT by blocking monoamine transporters. Two experiments were conducted to assess 5-HT involvement in this psychopharmacological effect. First, we extended previous findings by determining whether EEDQ potentiates the cocaine-induced locomotor activity of female preweanling rats. To this end, preweanling, adolescent, and adult female rats were pretreated with vehicle or EEDQ (7.5 mg) 24 hr prior to a test day injection of saline or cocaine (15 mg/kg). Second, to assess the importance of serotonergic involvement we examined whether EEDQ potentiated the locomotor activity produced by RU 24969 (a 5-$HT_{1A/1B}$ receptor agonist) and/or quinpirole (a D2/D3 receptor agonist). Results showed that EEDQ attenuated the cocaine-induced locomotor activity of adolescent and adult female rats, while potentiating the locomotor activity of female preweanling rats. EEDQ also potentiated the locomotor activity of preweanling rats treated with RU 24969+quinpirole, but not RU 24969 or quinpirole alone. These data indicate that EEDQ's ability to potentiate the cocaine-induced locomotor activity of preweanling rats is only manifested after co-activation of D2 and 5-HT_{1A}/$_{1B}$ receptors. If only DA receptors are stimulated (i.e., using NPA or quinpirole), a potentiated locomotor response is not evident. When considered together, these results show that preweanling and adult rats respond in a qualitatively different manner to EEDQ, and these age-dependent differences involve both DA and 5-HT system functioning.

Disclosures: A. Teran: None. **J.A.M. Robinson:** None. **G.I. Ramirez:** None. **C.G. Katz:** None. **S.A. McDougall:** None.

Poster

P771. Mechanisms of Attention: Animal Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P771.01

Topic: H.01. Attention

Support: F32NS112764
R01 NS034774-22A1
SFARI 633450

Title: Investigating the role of medial prefrontal cortex inhibitory neurons in attentional engagement

Authors: *B. FERGUSON[1], R. KADURU[2], J. R. HUGUENARD[3];
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Abstract: Disrupted attention presents as a comorbidity across several disease states, including but not limited to schizophrenia, autism, depression, bipolar disorder, Alzheimer's disease, and epilepsy. These impairments often persist even when hallmark symptoms respond to treatments. Thus, identifying neuronal circuits linked to successful and dysfunctional attention represents a significant public health concern. Attention is comprised of certain component processes, including sustained attention, selective attention, and attentional flexibility. The initial focusing of attention, precludes engagement of other components making it an important building block of many of our more complex behaviors. To explore this, we developed a protocol to capture this phenomenon in mice, referred to here as attentional engagement. Food-restricted mice were trained to initiate trials by nose poking in a center port and learned that responding correctly to a visual cue presented in one of two locations would result in a food reward. Using pseudorandom variation of cue lengths, we obtained a psychometric curve where intermediate cue lengths had variable detection rates that revealed relevant circuit components. With a combination of fiber photometry, optogenetics, and electrocorticography, we observed that the medial prefrontal cortex (mPFC) subserves attentional engagement with different neuronal subtypes playing distinct roles in specific task epochs. Dampening mPFC activity via chR2-mediated activation of VGAT-expressing neurons impairs performance during cue presentation, but not during the response period. Additionally, we determined that accuracy was linked to increases in cue-evoked parvalbumin-expressing interneuron activity, while somatostatin and vasoactive intestinal peptide-expressing neurons provide important error signals. Finally, electrocorticography of local field potentials indicated that increases in gamma oscillations help facilitate making the correct choice. Future directions involve investigating the contribution of long-range inputs to the mPFC, and their role in coordinating activity in distinct mPFC neuronal subtypes during attentional engagement.

Disclosures: B. Ferguson: None. **R. Kaduru:** None. **J.R. Huguenard:** None.

Poster

P771. Mechanisms of Attention: Animal Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P771.02

Topic: H.01. Attention

Title: Distinct coordinated activity patterns in the mPFC for exploratory versus defensive behaviors

Authors: *G. SHAH[1], J. POPOVITZ[3], S. P. MYSORE[2], H. M. ADWANIKAR[2]; [2]Psychological and Brain Sci., [1]Johns Hopkins Univ., Baltimore, MD; [3]GenEdit, San Francisco, CA

Abstract: Exploratory and defensive behaviors form a fundamental component of the behavioral repertoire of animals. Yet, how they are differentially signaled in neural representations remains largely unknown. To date, the dominant finding is the presence of individual neurons (or groups) that are selective for exploratory states versus for defensive states in the medial prefrontal cortex, (as well as the amygdala and the ventral hippocampus). The complexity of such affective states, multimodal characteristics of mPFC neurons and variability in single neuron selectivity suggests additional insights may be gained from investigating the coding by coordinated neural activity patterns - an approach that has proved effective in studies of other encoding problems in neuroscience. To test this, here, we examined the coordinated activity vectors of simultaneously imaged neural ensembles in mPFC of mice as they engaged in exploratory versus defensive behaviors in a variety of standard assays – the zero maze, plus maze and open field test. We measured neural activity in 100s of mPFC neurons using a miniature fluorescence microscope, and longitudinally tracked their Ca^{2+} dynamics as they traversed the exploratory or defensive zones of these paradigms. Our data reproduce the classic result that subsets of individual neurons are selective for these zones. Notably, however, they show that coordinated activity vectors of simultaneously imaged neurons (in n-d space) are distinct between the exploratory versus defensive behavioral states, with the cloud corresponding to exploratory zones being denser (more stereotypical patterns). These results are not accounted for purely by selectivity of individual neurons. Moreover, this representation in joint neural activity is maintained stably when animals are re-exposed to the assays (across weeks; unlike selectivity of individual neurons), and is consistent across assays (unlike selectivity of individual neurons). Surprisingly, sub-ensembles of neurons that are not individually selective also show these robust distinctions for the two behavioral zones in their coordinated activity. Results reveal a neural code in the mPFC that reliably signals the exploratory versus defensive behavioral states differentially, is stable across days, and is generalizable across behavioral paradigms.

Disclosures: G. Shah: None. **J. Popovitz:** None. **S.P. Mysore:** None. **H.M. Adwanikar:** None.

Poster

P771. Mechanisms of Attention: Animal Models I

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Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P771.03

Topic: H.01. Attention

Support: MH102211

Title: Corticotropin-releasing factor (CRF) acts within the mediodorsal thalamus to impair sustained attention.

Authors: *S. K. COOKE[1], S. E. NICOL[1], R. C. SPENCER[2], C. W. BERRIDGE[1];
[1]Univ. of Wisconsin Madison, Madison, WI; [2]Psychology, Univ. of Wisconsin, Madison, WI

Abstract: The prefrontal cortex (PFC) supports a variety of cognitive processes that guide goal-directed behavior. While PFC-dependent cognitive dysfunction is associated with multiple behavioral disorders, the treatment of this dysfunction is impeded by a limited understanding of the neural mechanisms underlying PFC-dependent cognition. Corticotropin-releasing factor (CRF) neurons and receptors are found in the PFC. However, their role in cognition has only recently begun to be examined. In prior studies we demonstrated that chemogenetic activation and inhibition of CRF neurons in the caudal dorsomedial PFC (dmPFC) of rats impairs or improves, respectively, working memory. These actions were dependent on local PFC CRF receptors, consistent with working memory impairing and improving actions of caudal dmPFC CRF receptor activation or inhibition, respectively. Additional studies demonstrated that activation/inhibition of caudal dmPFC CRF neurons also impairs/improves sustained attention. However, in contrast to working memory, this action was not dependent on PFC CRF receptors. Collectively, these observations indicate that PFC CRF neurons modulate distinct cognitive processes via divergent projection pathways. To date, the terminal fields involved in the sustained attention-modulating action of PFC CRF neurons are unknown. To initially approach this issue, we used an AAV5 viral vector system to express eGFP selectively in caudal dmPFC CRF neurons and examined the distribution of labeled axons outside the PFC. We observed particularly dense collections of varicosity-containing fibers in the medial and lateral aspects of the mediodorsal thalamus (MD) as well as the medial posterior parietal cortex, regions previously implicated in the regulation of attention. As a first step in determining whether either of these regions might contribute to the sustained attention actions of caudal dmPFC CRF neurons, we examined the effects of CRF infusion into these regions in animals engaged in an operant-based signal detection task of sustained attention. 500 nl infusions of CRF (50-250 ng) into the lateral MD elicited a dose-dependent impairment in sustained attention, as did inactivation of this region with the GABA-A agonist, muscimol (75 ng). Ongoing studies are examining the sustained attention actions of CRF infusions into the medial MD. In contrast to that seen in the MD, muscimol or CRF infusions into the parietal cortex had no significant effects on sustained attention. These results indicate the MD may be a site of action in the sustained attention impairing effects of caudal dmPFC CRF neurons. Future experiments will test this hypothesis.

Disclosures: S.K. Cooke: None. **S.E. Nicol:** None. **R.C. Spencer:** None. **C.W. Berridge:** None.

Poster

P771. Mechanisms of Attention: Animal Models I

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Program #/Poster #: P771.04

Topic: H.01. Attention

Support: NIH R34NS111653
Startup funds from the Johns Hopkins University

Title: Imaging sensory responses in the intermediate and deep layers of the superior colliculus

Authors: *A. BANERJEE[1], S. P. MYSORE[1,2];
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Abstract: The superior colliculus (SC) is a major sensorimotor hub in the vertebrate midbrain. Particularly, the intermediate and deep layers (SCid), which encode topographic maps of multisensory and motor space, are critical brain circuits involved in spatial target selection. However, an understanding of cell type-specific neural representations underlying SCid function is lacking because work to date has primarily employed electrophysiological techniques to measure SCid neural responses. Here, we present cellular-resolution calcium imaging of sensory responses in the mouse SCid using the *nVoke* microendocsope (Inscopix, Inc.). We first characterize visual response properties of CaMKII-positive neurons in SCid, and then investigate population-level signatures of stimulus competition by imaging SCid responses to distant competing stimuli. These results not only demonstrate the feasibility of imaging large-scale neural responses in SCid in a cell-type specific manner, but also shed light on the nature of competitive interactions underlying spatial stimulus selection in mammalian SCid.

Disclosures: A. Banerjee: None. **S.P. Mysore:** None.

Poster

P771. Mechanisms of Attention: Animal Models I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P771.05

Topic: F.02. Behavioral Neuroendocrinology

Support: NIH R01NS082179

Title: Neuroestrogen regulation of inhibitory synaptic transmission in genetically-identified auditory neurons

Authors: *H. KANG[1], L. REMAGE-HEALEY[2];
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Abstract: A prominent neuromodulator system found in forebrain auditory circuits of vocal learners such as humans and songbirds is neuronal aromatase, which provides locally-synthesized estrogens ('neuroestrogens'). In songbirds, local neuroestrogens fluctuate in the forebrain in response to song, and modulate neuronal firing states to shape song-evoked responses and auditory learning. However, the synaptic mechanisms and network motif that underlies these rapid actions are still unclear. To address this gap, we used a combination of cell-type specific viral vectors and whole-cell voltage clamp electrophysiology to investigate the synaptic mechanisms that account for acute neuroestrogen actions in songbird auditory forebrain. Specifically, we identified inhibitory synaptic transmission onto principal excitatory neurons as a primary target of neuroestrogen modulation. These findings and the results of ongoing experiments are thus far consistent with the hypothesis that neuroestrogens acutely modulate inhibitory synaptic transmission by regulating GABAergic input onto both excitatory and inhibitory cell types. Definitive evidence for how neuroestrogens specifically modulate genetically-identified targets of inhibition will clarify the mechanisms underlying neuroestrogen regulation of sensory processing in a vocal learning species. Furthermore, the principles extracted from this circuit modulation mechanism could then be applied to understand the neuromodulation of cortical sensory processing in other species and brain regions. Support from NIH R01NS082179.

Disclosures: H. Kang: None. **L. Remage-Healey:** None.

Poster

P771. Mechanisms of Attention: Animal Models I

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Program #/Poster #: P771.06

Topic: H.01. Attention

Support: NIH R34NS111653
NIH F32EY032776

Title: A midbrain inhibitory nucleus controls distracter suppression for spatial attention

Authors: *N. B. KOTHARI[1], W.-K. YOU[1], Q. ZHANG[1], S. LOGGIA[2], S. P. MYSORE[1]; [1]Johns Hopkins Univ., Baltimore, MD; [2]NEI, Bethesda, MD

Abstract: Selective spatial attention is the ability to preferentially process the highest priority (most important) spatial location, while simultaneously ignoring competing distracters. The neural circuit mechanisms underlying distracter suppression and target selection for spatial attention, however, are unknown. Here, in freely behaving mice engaged in a touchscreen-based primate-like task of visuospatial attention (Flanker task), we demonstrate that a midbrain inhibitory nucleus, called the pLTN (periparabigeminal lateral tegmental nucleus) controls distracter suppression. The pLTN, which consists of parvalbumin expressing (PV+) neurons, receives topographically organized input from the midbrain sensorimotor hub, the superior colliculus. In PV-cre mice trained on the flanker task, we expressed the cre-dependent DREADD, h4MD(Gi), bilaterally in pLTN. We found that inactivation of pLTN through CNO administration severely disrupted attentional target selection in the flanker task and produced hyper-distractibility. In addition, it also caused loss of reliability in selecting the attentional target by attenuating the categorical nature of psychophysical response curves measured as a function of relative priority between the target and flanker. Because our task design dissociates the locus of attention from the locus of behavioral report, we were able to rule out deficits of motor selection as an explanatory factor. Moreover, administration of CNO during target-alone trials did not alter performance, ruling out purely sensory deficits as an explanatory factor. Administration of saline instead of CNO to these animals does not alter target selection. The observed deficits in distracter suppression for target selection in the absence of purely perceptual deficits establish the midbrain pLTN as a critical hub for the control of distracter suppression for selective spatial attention.

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Poster

P771. Mechanisms of Attention: Animal Models I

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Program #/Poster #: P771.07

Topic: D.06. Auditory & Vestibular Systems

Support: R01DC018650
R00DC015014

Title: Identifyingnatural transitions from goal-directed to habit-like performance during sensorimotor learning in mice

Authors: *S. MOORE[1], Z. WANG[1,2], K. KUCHIBHOTLA[1,3];
[1]Psychological and Brain Sci. Dept., [2]Neurosci. Res. Undergraduate Program, [3]The Solomon H. Snyder Dept. of Neuroscience, Sch. of Med., Johns Hopkins Univ., Baltimore, MD

Abstract: Animals use different decision processes to efficiently adapt to complex environments. When operating in a goal-directed mode, animals deliberate amongst alternatives in a slow and cognitively demanding process. As the statistics of the environment become predictable, animals can shift to a faster and more automatic habit mode. The nature and timescale of the transition from goal-directed to habit-like performance during learning, however, remains poorly understood. Studying this transition is crucial for understanding adaptive behaviors and it may add to our knowledge of disorders like addiction and OCD. During learning, a challenge to identify the decision process (goal-directed vs habitual), is that the decision itself is a hidden variable that is not behaviorally observable. For one, performance outcomes (action rate and accuracy) are often similar when using either decision process. Secondly, in commonly used water-restriction paradigms, an animals' drive is based on a biological *need* linked to survival (hydration), so animals exhibit constantly high action rates (due to elevated motivation) and gradually improve accuracy (due to learning). We hypothesized that by shifting an animals' motivation from a *need* (for survival) to a *preference* (for palatability), we could use action rate variability as a behavioral indicator of goal-directed performance (highly variable) versus habit-like performance (stable) without impacting accuracy. Here we leveraged a recent protocol in which mice get *ad libitum* access to water with citric acid (CA), a non-palatable substance that fulfills the need for hydration. We compared CA mice with mice under two different water restriction protocols. All groups were trained in an auditory go/no-go task in which mice learned to lick in response to a tone to obtain a water reward and withhold licking in response to another tone to avoid a timeout. Our data shows that mice acquired task contingencies at similar rates in all three groups. Interestingly, throughout training, most CA mice initially showed high action rate variability, regularly shifting from epochs of high engagement (high action rate) to low engagement (low action rate). With time, CA mice exhibited an abrupt reduction in action rate variability, suggesting a shift to habit-like performance. This shift was not evident in water restricted mice. We are currently using pupillometry to link changes in action rate during the goal-directed phase with a standard biomarker of arousal. These data suggest that the transition from goal-directed to habit-like performance during learning is abrupt and may result from a winner-take-all decision process.

Disclosures: S. Moore: None. **Z. Wang:** None. **K. Kuchibhotla:** None.

Poster

P771. Mechanisms of Attention: Animal Models I

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Program #/Poster #: P771.08

Topic: H.01. Attention

Support: BBSRC (UK) [grant number BB/M010996/1]
Boehringer Ingelheim Pharma GmbH & Co. KG

Title: Using overtraining to abbreviate tests of attentional set-shifting

Authors: T. S. KNOTT[1], E. M. BOWMAN[1], V. J. BROWN[1], S. DEIANA[2], M. VON HEIMENDAHL[2], *D. S. TAIT[1];
[1]Sch. of Psychology and Neurosci., Univ. of St Andrews, St Andrews, United Kingdom; [2]CNS Dis. Res., Boehringer Ingelheim Pharma GmbH & Co.KG, Biberach, Germany

Abstract: The rodent intradimensional (ID)/extradimensional (ED) attentional set-shifting task has been used for over two decades to assess behavioral flexibility. However, a drawback of the classic ID/ED task is that there are seven learning stages, requiring three pairs of unique exemplars. It has previously been observed that overtraining improves reversal learning in multidimensional stimulus discriminations, with evidence suggesting this is due to increased attentional allocation to the predictive (relevant) stimulus dimension (Dhawan *et al*, 2019, *Behav. Brain Res*., 363**:** 45-52). If so, then it should be possible to assess attentional set-shifting using overtraining on a single stage to facilitate set-formation, substituting four of the usual seven stages. To investigate this, we inhibited medial prefrontal cortex (mPFC) function—a brain region implicated in mediating behavioral flexibility (Birrell & Brown, 2000, *JNsci*, 20:4320-4324). Twelve adult male Lister hooded rats received inhibitory designer receptors exclusively activated by designer drugs (DREADDs; AAV5-hSyn-hM4D(Gi)-mCherry—Bryan Roth, Addgene) bilaterally into mPFC. mPFC inhibition via the expressed receptors was induced by i.p. administration of Compound 21 (C21; 3mg/kg). Rats completed the standard 7-stage ID/ED task twice, following vehicle or C21 administration; and a 4-stage task, consisting of a compound discrimination, a reversal, a novel compound discrimination where the rewarded dimension was switched to the previously irrelevant, then a final reversal, four times: twice with thirty additional trials after reaching criterion (6 consecutive correct trials) in the initial compound discrimination, and twice without the additional trials, with vehicle or C21, in a pseudorandom order. Consistent with previous findings, mPFC inhibition impaired ED learning in the 7-stage task. In the 4-stage task, overtraining improved reversal learning performance and slowed learning when the rewarded dimension switched. Learning at that stage was slowed further by mPFC inhibition, but only when the rats were overtrained. Overall, our results indicate that overtraining to a compound stimulus drives focus to the relevant dimension, and that a subsequent stage where a shift is required is functionally equivalent to the ED stage of the classic 7-stage ID/ED test. In both the classic ID/ED task, and the abbreviated task, shifting performance was impaired by mPFC dysfunction. It is thus possible to reduce the number of task stages required to measure set-shifting.

Disclosures: T.S. Knott: None. **E.M. Bowman:** A. Employment/Salary (full or part-time):; University of St Andrews. **V.J. Brown:** A. Employment/Salary (full or part-time):; University of East London. **S. Deiana:** A. Employment/Salary (full or part-time):; Boehringer Ingelheim Pharma GmbH & Co.KG. **M. Von Heimendahl:** A. Employment/Salary (full or part-time):;

Boehringer Ingelheim Pharma GmbH & Co.KG. **D.S. Tait:** A. Employment/Salary (full or part-time):; University of St Andrews.

Poster

P779. Modeling and Behavior IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P779.01

Topic: H.03. Decision Making

Title: Rats adapt decision thresholds based on reward manipulation in an evidence accumulation task

Authors: *G. A. KANE[1], B. B. SCOTT[2];
[2]Psychological & Brain Sci., [1]Boston Univ., Boston, MA

Abstract: Animals, including humans, exhibit speed-accuracy tradeoffs in decision-making: the more time they spend deliberating or accumulating evidence, the more accurate their decisions, but at the cost of time and future opportunities. Finding the appropriate balance between speed and accuracy is critical for maximizing reward rate, but the neural mechanisms of choosing when to decide are not well understood. To determine whether rats could serve as a model to study the neural mechanisms of speed-accuracy tradeoffs, we investigated how rats choose to trade speed for accuracy in response to different task demands in a pulse-based evidence accumulation task. Rats self-initiated trials by nose poking in a center port and were then shown a series of light flashes in ports on their left and right according to a Bernoulli process (a weighted coin flip). Light flashes continued until rats executed a decision by nose poking in the left or right port. Rats were rewarded for choosing the side with the greater flash probability. Rats (n=9) were tested in their home cage with access to the task 24 hr/day; we collected an average of 1638 (st. dev. = 500) trials per rat per day using mild food restriction and 10% sucrose as reward. The most effective manipulation to encourage slower responses was to alter the reward landscape: in some conditions, rats were offered larger rewards for waiting longer to respond. In these conditions, rats not only exhibited slower responses, but also greater accuracy, suggesting that they accumulated more evidence while waiting. Accumulation to bound models fit to individual rats' behavioral choices and response times revealed that, in these conditions, rats exhibited a greater decision threshold (they required more evidence to commit to a decision), with no change in the rate at which rats accumulated evidence or in their non-decision time (time not related to evidence accumulation, such as the time to prepare and execute motor responses). Overall, these findings indicate that rats adapt their speed-accuracy tradeoff to task demands, providing a high-throughput model to study the neural mechanisms of adapting decision boundaries.

Disclosures: G.A. Kane: None. **B.B. Scott:** None.

Poster

P779. Modeling and Behavior IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P779.02

Topic: H.03. Decision Making

Support: European Research Council (ERC) under the European Union's Horizon 2020 research and innovation programme (grant agreement No.\ 758604)
ETH Zurich

Title: Rational inattention in mice

Authors: *N. GRUJIC, J. BRUS, D. BURDAKOV, R. POLANIA;
Swiss Federal Inst. of Technol. Zurich, Zurich, Switzerland

Abstract: Behavior exhibited by humans and other organisms is generally inconsistent and biased, and thus is often labelled irrational. However, the origins of this seemingly suboptimal behavior remain elusive. We developed a behavioral task and normative framework to reveal how organisms should allocate their limited processing resources such that there is an advantage to being imprecise and biased for a given metabolic investment that guarantees maximal utility. We trained mice to perform a novel choice task requiring them to make decisions based on ordinal comparisons of orientation stimuli spanning the whole sensory space. Additionally, we introduced trial-to-trial variability in the reliability of sensory stimuli and controlled the prior distribution of the sensory inputs in terms of presented orientations. During the experimental timeline, mice went through different novel stimulus-reward association environments. This allowed us to model the allocation of sensory resources across the orientation stimulus space in each stimulus-reward environment, and check whether it complies with the predictions of rational inattention theory.
We found that mice act as rational-inattentive agents by adaptively allocating their sensory resources in a way that maximizes reward consumption in each stimulus-reward association environment. Surprisingly, perception to commonly occurring stimuli was relatively imprecise. However, this apparent statistical fallacy implies "awareness" and efficient adaptation to their neurocognitive limitations. Interestingly, distributional reinforcement learning mechanisms efficiently regulate sensory precision via top-down normalization. Furthermore, we found that arousal systems, as tracked by pupil size, contain information about reward expectancy of orientation stimuli in a given sensory-reward association environment.
These findings establish a neurobehavioral foundation for how organisms efficiently perceive and adapt to environmental states of the world within the constraints imposed by neurobiology.

Disclosures: N. Grujic: None. **J. Brus:** None. **D. Burdakov:** None. **R. Polania:** None.

Poster

P779. Modeling and Behavior IV

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Program #/Poster #: P779.03

Topic: H.03. Decision Making

Support: NIDA DA041480
NIDA DA051598
State of Connecticut

Title: Model-free and model-based components of reinforcement-learning differentially mediate reversal learning in the rat

Authors: *S. GROMAN[1,2], S. L. THOMPSON[2], D. LEE[3,4], J. TAYLOR[5];
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Abstract: Reversal-learning paradigms have been extensively used to investigate flexible decision making in normal and abnormal states. However, our understanding of the mechanisms of reversal-learning remains limited. Differences in reversal learning performance have been attributed to impairments in value updating, state learning, and belief updating, but how these reinforcement-learning mechanisms relate to one another is not known. To elucidate those relationships, we assessed decision-making processes in both a three-choice, probabilistic reversal learning (PRL) task and in a multi-stage decision-making (MSDM) task in the same adult, male rats. Computational approaches were used to quantify value updating and state transitions in individual rats so that parameter values could be compared between the two decision-making paradigms. We find that reversal-learning performance is correlated with the prospective evaluation of reinforced actions (e.g., model-based learning) and, specifically, with the detection of the state transition between the acquisition and reversal phase. Retrospective evaluation (e.g., model-free learning), however, was related to differences in the degree to which action values were updated following rewarded and unrewarded outcomes. These data provide the direct evidence that model-free and model-based learning influence distinct aspects of adaptive choice behavior in a reversal-learning paradigm and showcase the independent of value updating from state detection. We then examined how these model-based and model-free computations could explain the reversal-learning impairments we have observed in rats following drug self-administration. We found that cocaine-induced reversal-learning deficits were due to selective impairments in the ability of animals to detect a state transition, whereas value updating following the state transition was not affected by cocaine self-administration.

Together, these data provide new insights into the reinforcement-learning computations that mediate flexible decision making in normal and pathological states.

Disclosures: S. Groman: None. **S.L. Thompson:** None. **D. Lee:** None. **J. Taylor:** None.

Poster

P779. Modeling and Behavior IV

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Program #/Poster #: P779.04

Topic: H.03. Decision Making

Support: NIMH R01 MH123661
NIMH P50 MH119569

Title: Sex differences in learning from exploration

Authors: E. KNEP[1], C. CHEN[1], B. A. EBITZ[2], *N. GRISSOM[1];
[2]Neurosci., [1]Univ. of Minnesota, Minneapolis, MN

Abstract: Sex-based modulation of cognitive processes could set the stage for individual differences in vulnerability to neuropsychiatric disorders. While value-based decision making processes in particular have been proposed to be influenced by sex differences, the overall correct performance in decision making tasks often show variable or minimal differences across sexes. Computational tools allow us to uncover latent variables that define different decision making approaches, even in animals with similar correct performance. Here, we quantify sex differences in mice in the latent variables underlying behavior in a classic value-based decision making task: a restless 2-armed bandit. In this task, animals chose from two options, each of which was associated with certain reward probabilities that changed randomly and independently over time. To maximize rewards, animals had to balance between exploiting a known good option with exploring better alternatives. While male and female mice had similar performance on reward acquisition, we found that they achieved this performance via different patterns of exploration by fitting a Hidden Markov model and reinforcement learning models. Male mice tended to make more exploratory choices overall, largely because they appeared to get "stuck" in exploration once they had started. Female mice tended to explore less, but they learned more quickly during exploration, indicated by elevated win-stay and lose-shift behaviors during exploratory trials. Together, these results suggest that sex exerts stronger influences on decision making during periods of learning and exploration than during stable choices. The neural mechanisms of exploration are thus poised to confer sex-modulated variability in cognition and potentially sex-modulated vulnerability to neuropsychiatric diagnoses.

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Poster

P779. Modeling and Behavior IV

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Topic: H.03. Decision Making

Support: DFG grant OT562/1-1

Title: Near-optimal time investments under uncertainty in humans, rats, and mice

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Abstract: Our scarcest resource is time. Humans and other animals must allocate their time on a vast number of different activities - from reading a paper, pursuing a degree, or foraging for food. Optimal, utility-maximizing, behavioral strategies must balance expected future gains with irrecoverable time investment. However, humans and other animals often succumb to a variety of cognitive biases that lead to suboptimal returns such as considering sunk costs when investing time to obtain future rewards. Comparative studies across species are important to understanding the evolutionary origins of irrational behavior. Here we re-examine the question whether humans, rats, and mice are able to invest their time optimally. First, by developing computational task models, we re-interpret previous reports that humans, rats, and mice succumb to the sunk cost fallacy as optimal behavior when the precise value of choices is uncertain (Sweis et al., 2018, Science). Our results demonstrate the importance of experimental control over the decision maker's reward uncertainty. Second, we behaviorally tested how optimal humans, rats, and mice invest time by controlling their uncertainty in obtaining future rewards. This allowed us to evaluate whether subjects scaled investment decisions in proportion to their confidence in obtaining a reward. Human or animal subjects committed to one of two choice alternatives with ambiguous sensory evidence, which enabled us to infer the subjects' confidence in making a correct choice. After making a choice, subjects invested time in their decision by waiting for randomly delayed reward for correct choices. We predicted optimal time investment for each trial using a statistical model of confidence and a non-parametric estimate of the subjects' mapping function between confidence and time investment. Crucially, this approach allowed us to isolate the contribution of confidence to investment behavior. Humans, rats, and mice invested time close to optimal model predictions, i.e., with low confidence noise, demonstrating that all species appropriately use sensory evidence to adjust investment behavior. In humans, explicit confidence reports predicted time investment, suggesting shared mechanisms for verbal and non-verbal confidence-guided behaviors. Our results demonstrate that humans, rats, and mice are

efficient economic agents, who optimize investment decisions by leveraging their sense of confidence in uncertain environments.

Disclosures: T. Ott: None. **P. Masset:** None. **M. Bosc:** None. **J. Sanders:** None. **A. Kepecs:** None.

Poster

P779. Modeling and Behavior IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P779.06

Topic: H.03. Decision Making

Support: Boston University Undergraduate Research Opportunities Program (UROP)

Title: Production of central tendency effects in interval timing tasks via reinforcement learning

Authors: *N. CHIULLI[1], G. KANE[2], B. B. SCOTT[3];
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Abstract: In interval time estimation and reproduction tasks, during which participants are asked to reproduce a range of time intervals over the course of many trials, participants exhibit a central tendency effect: their responses are biased toward the mean of previous sample times. Current literature suggests this behavior is an adaptive basis due to an awareness of and compensation for endogenous timing uncertainty (noise in the estimation of elapsed time); in other words, because the estimation of elapsed time is imprecise, estimates are combined with the prior expectation built from experience over multiple past trials to reduce uncertainty. While this hypothesis is widely accepted, other possible contributing factors have not been sufficiently ruled out. As the central tendency bias improves reward rate, we explored whether this effect could also be achieved by agents that seek to maximize reward using model-free reinforcement-learning (i.e. without knowledge of their endogenous timing uncertainty). We first used a discrete time Q-Learning algorithm in which, given a state (noisy sample interval), the model chose from a set of possible reproduction times and received reward based on how similar the chosen reproduction time (plus noise in the reproduction of the time interval) was to the sample interval. Similar to models that explicitly account for endogenous timing uncertainty, this Q-learning model with no knowledge of timing uncertainty exhibited a central tendency effect, and the central tendency bias grew with greater timing uncertainty. Next, we replicated these effects using an Actor-Critic model in which sample and reproduced times were mapped onto a continuous function rather than chosen from predetermined bins. The achievement of this behavior via model-free reinforcement learning algorithms indicates that it is possible to learn the actions (i.e. reproduction times) that maximize reward without explicit knowledge of

endogenous timing uncertainty. Future studies in humans and animals will be required to determine the extent to which these effects are due to awareness of and compensation for endogenous timing uncertainty or model-free reward maximization.

Disclosures: N. Chiulli: None. **G. Kane:** None. **B.B. Scott:** None.

Poster

P779. Modeling and Behavior IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P779.07

Topic: H.03. Decision Making

Support: NIH Grant NS118463

Title: A Deep Neural Network Model Adapts Flexibly to Different Opponent Strategies in a Competitive Game

Authors: *M. OSTROW[1], R. YANG[2], H. SEO[3];
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Abstract: In social interactions, theory of mind (ToM) is thought to be an inference in which an agent learns a model of other agents' intentions, goals, and reward function in order to predict future actions. ToM also allows for agents to generalize to novel social interactions. ToM has been modeled at the algorithmic level, with explicit modules that represent the postulated cognitive components of ToM. However, these models offer only limited insights into the potential neural mechanisms underlying ToM, largely due to the lack of linking hypotheses to the activity of complex neural networks. Recent applications of artificial neural networks (ANNs) in neuroscience research have shown that ANNs learning complex cognitive tasks often exhibit the spatiotemporal features similar to the neural activity observed in the brain, suggesting that ANNs might be a useful tool to understand how ToM might emerge in the distributed activity of biological neural networks. Here, we explore the potential of artificial neural networks to provide novel insights into the neural mechanisms underlying ToM.
Using a deep reinforcement learning actor-critic algorithm, we trained a recurrent neural network (RNN) to play a two-player competitive game against multiple opponents who generate their actions according to a set of higher-order strategies. We used a meta-learning approach inspired by past models of the prefrontal cortex as performing meta-reinforcement learning. We focused on two questions. *First*, can the network learn to play adaptively against a wide variety of opponents in its training set, and can it generalize to novel strategies of similar subtypes? *Second*, does adding an auxiliary task to model the opponents' action enhance the network's ability to adapt and generalize?

After training over one hundred different RNNs for between 300,000 and 3 million iterations to ensure significance of our results across random seeds and hyperparameters, we found that despite a lack of explicit ToM, the network can generalize well to new opponents. We next provided the models the ability to develop a rudimentary form of ToM by requiring it to make a prediction of the opponent's choice on each iteration of the game. Adding the explicit task did not significantly improve the network's ability to generalize. Our results demonstrate that RNNs can learn the strategies of the opponents in order to adaptively play a competitive game. They also suggest that explicit modules of ToM may not further enhance the network's ability to meta-learn and generalize in our present task. In future studies, we hope to further understand the potential neural mechanisms underlying ToM computations by analyzing network dynamics.

Disclosures: M. Ostrow: None. **R. Yang:** None. **H. Seo:** None.

Poster

P779. Modeling and Behavior IV

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Program #/Poster #: P779.08

Topic: H.03. Decision Making

Support: NSF grants #1658303
NSF grants 1850849

Title: Does pre-stimulus EEG power redict cognitive processes in perceptual decision making

Authors: *Q. SUN[1], M. D. NUNEZ[2], J. VANDEKERCKHOVE[1], R. SRINIVASAN[1]; [1]Dept. of Cognitive Sci., Univ. of California, Irvine, Irvine, CA; [2]Dept. of Psychological Methods, Univ. of Amsterdam, Amsterdam, Netherlands

Abstract: EEG activity prior to the onset of a stimulus may reflect different cognitive components of preparatory brain activity relevant to subsequent perceptual processes. 48 subjects performed a two-alternative forced-choice task by discriminating whether a Gabor patch presented with noise is high or low spatial frequency. Task difficulty was manipulated by the difference in spatial frequency (perceptual evidence) between the two choices. Participants performed the task in blocks of trials at 3 levels of difficulty (spatial frequency difference). A cue interval containing visual changing noise starts from 0.5 to 1.0 second before a stimulus onset. Response times increased as task difficulty increased. A standard Hierarchical Drift Diffusion Models (HDDM) were fitted using choice accuracy and reaction time to obtain estimates of the rate of evidence accumulation (drift rate) and the boundary separation between two choices. Hierarchical model parameters were estimated for each participant per condition, and showed that drift decreased as the task difficulty increased. Desynchronization in EEG oscillations was investigated using 5 frequency bands: theta (3-5Hz), low alpha (7-9Hz), high

alpha (11-13Hz), low beta (15-21Hz), and high beta (23-29Hz). Relative power of each frequency band was calculated by the ratio of the power of each electrode per trial to its corresponding baseline. Correlation matrices (channel x time) for each frequency band were generated by calculating the Pearson correlation coefficient between model parameters and relative power of every subject per condition, at each channel and each time point. Pre-stimulus interval is defined as 300ms prior to stimulus onset. Our results suggest that pre-stimulus alpha power reflects condition effects of drift rate, such that alpha power increases with task dmatlab.matifficulty as drift rate decreases. Correlations of boundary and EEG power in theta, low alpha, and high beta bands during pre-stimulus intervals primarily reflect individual differences, as these correlations show consistent patterns across conditions. These results provide a new perspective to use neural correlates to interpret diffusion model parameters. Preparatory activity in the prestimulus determines the rate of evidence accumulation and decision caution in perceptual decision making tasks.

Disclosures: Q. Sun: None. **M.D. Nunez:** None. **J. Vandekerckhove:** None. **R. Srinivasan:** None.

Poster

P985. New Approaches in Optogenetics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P985.01

Topic: I.08. Methods to Modulate Neural Activity

Support: DARPA N6600117C4012
NIH Grant U01NS090596

Title: High-density OLED-on-CMOS devices for implantable hyperlocal optogenetic stimulation in-vivo

Authors: *A. TAAL[1], I. UGUZ[1], S. HILLEBRANDT[2], C. MOON[2], C. KEUM[2], J. CHOI[1], E. POLLMANN[1], M. GATHER[2], K. SHEPARD[1];
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Abstract: The ability to precisely map neural circuits and to selectively manipulate region of interest is critical to understanding brain function. To that end, optogenetics has emerged as a great candidate due to its excellent spatiotemporal resolution and cell-specific targeting. Combined with simultaneous recordings of responsive neuronal micro circuitries, optogenetics manifests itself as a powerful tool for feedback regulated manipulation of particular brain regions and neural pathways. While these feedback loops are well-understood at the brain surface, the complexity of integration has obstructed such experiments in the deep brain. Accessing the deep

brain in a highly localized fashion for simultaneous stimulation and recording has thus remained elusive. We have been exploring fully implantable optoelectronic devices exploiting advances in complementary metal-oxide-semiconductor (CMOS) microelectronics and advanced heterogeneous integration as a means of accessing deep brain structures, bringing advantages over techniques using optical fibers or GRIN lenses. Here, we present an implantable CMOS shank probe containing 1024 organic light emitting diodes (OLEDs) on a 25-µm pitch. The OLEDs are fabricated directly on top of the CMOS integrated circuit, matching approaches used in advanced display technologies. Each of four 6-mm-long shanks carry 256 OLEDs, emitting 25nW per OLED from an active area of 400µm^2. The CMOS pixels support repetition rates beyond 50kHz and allow stimulation of the surrounding neural tissue with arbitrary patterns of individually addressable OLEDs over an area of 3200x800µm. Neurons as far as 50µm away can be reached, bringing the total addressable volume up to 0.1mm^2. To record the corresponding neural response, we combine the CMOS shank with a transparent electrode array (32 electrodes spaced at a 50-µm pitch) fabricated with 100-nm-thick PSS:PEDOT electrodes and indium-tin-oxide interconnect on a 4-µm-thick parylene substrate. The 20-by-20-µm electrodes have an impedance of 100KOhm at 1kHz with a baseline noise of 6.5µV rms over the 10Hz-to-3kHz band. During optogenetic stimulation, we observe artifact-free recording, which is not typical of electrical approaches In our experiments, we demonstrated optogenetic stimulation from the OLEDs by implanting them in the CA1 of Channelrhodopsin transgenic mice. We successfully manipulated neural populations down to single neuronal level and observed the effects of stimulation in real time. By adjusting the power of OLEDs, we seek to precisely control the field of stimulation and induce hyperexcitability in local populations.

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Poster

P985. New Approaches in Optogenetics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P985.02

Topic: I.08. Methods to Modulate Neural Activity

Title: Acousto-optics based simultaneous 3D imaging and photostimulation with temporal laser intensity modulation for precise temporal control of activity patterns at the level of individual neurons

Authors: *K. ÓCSAI[1,2], D. NAGY[3], G. SZALAY[3], C. CSUPERNYÁK[3], A. PLAUSKA[3], Á. SZEPESI[3], G. KATONA[1], B. RÓZSA[1,3];
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Abstract: Investigating the network dynamics of neuronal populations in the cortical areas involved in vision with photostimulation is one of the hot topics of the past few years. However, measurement protocols, visual paradigms, and experimental scenarios have all been restricted by the technological constraints of conventional microscope systems. Here we present a novel, 3D acousto-optical scanning-based photostimulation equipment which enables ms resolution precise temporal control of activity patterns at the level of individual neurons. To demonstrate the efficacy of the system we used it to replay activity on a set of neurons found to be responsible for coding certain visual stimulation. For performing this first we used fast multilayer acousto-optical frame scanning, which enables recording the activity of arbitrary cortical layers sequentially with high speed (40Hz with 512x512 pixel resolution, in a 500x500 µm areas). By this, we can follow the activity of hundreds of cells in multiple (3-11) areas of the cortex. Next, we developed novel analytic software modules to automatically detect responding neurons and to define among the acquired cell-level activity patterns those that are involved in the perception of specific visual stimuli (i.e., representing a latent task variable of a task). Optical stimulation is presented with a custom virtual reality system that provides binocular depth-perception and a fully immersive experience for small head-fixed. Finally, we photostimulate hundreds of somata simultaneously with 3D imaging. For precise temporal control of the activity on the cells selected to be stimulated we developed novel driver electronics of the AO system allowing us to arbitrarily modulate the stimulating laser intensity with a granularity of 30 µs. The achievable temporal resolution of the activity playback was found to be 100 ms, however, note that this is not limited by the stimulation method itself, but by the precision of the activity detection which was measured with GCaMP6f. Using this system, we showed that we can effectively stimulate and measure up to 250 cells simultaneously or can replay precise temporal dynamics on up to 20 cells with 100 ms temporal resolution.

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Poster

P985. New Approaches in Optogenetics

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Program #/Poster #: P985.03

Topic: I.08. Methods to Modulate Neural Activity

Support: NSF 2024270

Title: A flexible origami opto-electro array for in vivo optogenetic stimulation and electrophysiology recordings from dorsal root ganglion

Authors: *Y. GONG, W. LI;
Michigan State Univ., East Lansing, MI

Abstract: Developing a tool to better understand the relationship between spindle length-sensitive afferents from hip muscles and both interlimb coordination and gait selection is an active area of biomedical microelectrochemical systems research. Future tools must be flexible and configurable, given the evolving understanding of both neuroscience mechanisms and clinical outcomes. We describe a thin-film, three-dimensional (3D) opto-electro array with 4 addressable microscale light-emitting diodes for surface illumination of dorsal root ganglions (DRGs) and 9 penetrating electrodes for simultaneous recordings of light-evoked DRG activities. Emerging optogenetic stimulation provides distinct advantages over electrical stimulation, such as cell-type specificity, sub-millisecond temporal precision, rapid reversibility, and suppressed electrical artifacts. Importantly, using the origami design concept, the array can be fabricated directly on a silicon wafer in a two-dimensional (2D) configuration, which can transform into a 3D structure by folding. The device fabrication involves multiple steps of polymerization, photolithography, thermal evaporation of metal, wet etching of metal, and plasm etching of polymer. Polyimide is chosen as the substrate and packaging material, and a Ti/Au stack is used to form conductive patterns, e.g., electrodes, traces, and contact pads. During device release, the penetrating microelectrode probes were gravitational and buoyant lifted into a vertical position, and maintained vertically relying on both the plastic deformation of their base hinge regions and base package material. Currently, we have completed device fabrication, and will assemble the device with an external electronic headstage and test the integrated system in cat models to validate its capability for optogenetic stimulation and electrophysiology recordings from DRGs. This work will pave the way for neuroscience research of reversible modulation of proprioceptive afferent activities. The tool could also be used for delivering proprioceptive feedback to limb prosthetic users and/or patients with sensory neuropathies in the future.

Disclosures: Y. Gong: None. **W. Li:** None.

Poster

P985. New Approaches in Optogenetics

Location: Virtual Only

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Program #/Poster #: P985.04

Topic: I.08. Methods to Modulate Neural Activity

Support: NSF Grant 1512794

Title: Flexible Parylene photonic optoelectrodes for high-resolution neural recording and stimulation

Authors: *J. W. REDDY, M. YUAN, V. JAIN, Z. AHMED, M. CHAMANZAR;
Carnegie Mellon Univ., Pittsburgh, PA

Abstract: High resolution light delivery deep into the brain tissue with high spatial resolution, as well as collocated electrophysiology recording for local neural circuit interrogation are essential tools for experimental neuroscience. Optical techniques such as optogenetics or structural and functional imaging enable cell-type specific neural circuit analysis. Implantable photonic waveguide arrays can passively confine and guide light to target regions in the brain. Modern multimodal, microfabricated neural probes integrate light delivery and electrical recording functionality in a small footprint to minimize tissue damage. Flexible, polymer material platforms are preferred for chronic experiments to minimize damage to tissue due to tethering forces and micromotions. Here, we demonstrate a novel multimodal neural probe architecture consisting of flexible, biocompatible Parylene photonic waveguides for optogenetic stimulation and collocated recording electrodes for simultaneous electrophysiology readout. The Parylene photonic waveguides are broadband and operate over a wide range of wavelengths (λ; = 450, 532, 633, 680 nm). The input/output coupling via integrated micromirrors enables illumination volumes normal to the probe surface, collocated with the recording volume. We demonstrate 1.3 cm neural probes with 1-6 individually-addressable optical waveguide and 1-6 recording electrode channels in a linear array with 200 micrometer pitch along the probe shank. The 9 micrometer diameter electrodes show an impedance of 4.5 MOhm at 1 kHz, which is reduced to 106 KOhm at 1 kHz via PEDOT:PSS electroplating. We demonstrate compact optical packaging with optical fiber arrays and bare chip vertical cavity surface emitting laser (VCSEL) diodes, and electrical packaging with commercial neural amplifier headstages (Intan RHD2132). In vivo implantation of the flexible probes is demonstrated via attachment to a tungsten microwire shuttle using the polyethylene glycol (PEG) bioresorbable coating. The probe functionality is validated in vivo in mice.

Disclosures: J.W. Reddy: None. **M. Yuan:** None. **V. Jain:** None. **Z. Ahmed:** None. **M. Chamanzar:** None.

Poster

P985. New Approaches in Optogenetics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P985.05

Topic: I.08. Methods to Modulate Neural Activity

Support: Morton Cure Paralysis Fund Grant

Title: Response characteristics of hybrid optogenetic and electrical stimulation in rat peripheral motor nerves

Authors: E. M. MORAVEC[1,2], A. S. PAVLENKO[1], *J. WILLIAMS[1,3,2];
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Abstract: Optogenetic stimulation of peripheral motor nerves is a promising alternative to electrical stimulation for reanimating paralyzed muscles. This technique allows motor nerves to be stimulated with light after using a viral vector to induce expression of light sensitive ion channels, or opsins, along the nerve. Optical stimulation offers advantages over functional electrical stimulation including natural muscle recruitment order and stimulation target specificity. However, it is not as well characterized as electrical stimulation and suffers from challenges that may impact its clinical viability. The level of opsin expression and inherent opsin kinetics limit the strength of muscle responses to optical stimulation as well as the frequency at which these responses can be maintained. Improvement in both areas is essential for producing functional movements with optical stimulation. To address these issues, this study aimed to characterize EMG responses to optical stimulation, optimize optical stimulation parameters, and enhance optical EMG responses with subthreshold electrical stimulation. We injected the tibialis anterior muscle of rats with an adeno-associated virus (AAV) carrying the gene for channel rhodopsin (ChR2), and used stimulation of the corresponding peroneal nerve as the basis for the experiments. First, we found that EMG response latency after optical stimulation was a function of the intensity of optical stimulation but was not dependent on optical pulse width. Second, we found that optical stimulation intensity and pulse width also impacted EMG coupling to optical pulses at higher pulse train frequencies. Finally, subthreshold electrical stimulation paired with optical stimulation was able to modify EMG responses to both a single optical pulse and high frequency pulse trains, though the effect of combining these two modes of stimulation was highly dependent on their relative timing. The results of this study highlight unique challenges of optical stimulation while also providing pathways to address these issues so that optical stimulation may become a clinically viable option for muscle stimulation.

Disclosures: E.M. Moravec: None. **A.S. Pavlenko:** None. **J. Williams:** None.

Poster

P985. New Approaches in Optogenetics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P985.06

Topic: I.08. Methods to Modulate Neural Activity

Title: Coupling High-density SiNAPS CMOS-based Neural Recording Probes with Optogenetic Light Stimulation

Authors: *F. BOI[1], A. LOCARNO[2], J. F. RIBEIRO[1], G. ORBAN[1], R. TONINI[4], G. N. ANGOTZI[1], L. BERDONDINI[3];

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Abstract: The ability to selectively perturb and record hundreds to thousands of neurons across large brain circuits can provide the technological ground for innovative experimental tools to study brain circuits in health and disease. Although optogenetics is nowadays a widely used technique to study brain circuits by selectively controlling the activity of neurons through the expression expression of light-modulated opsins, electrode technologies for sub-millisecond recordings of light-evoked activities are typically limited to a few tens of neurons. With respect to conventional Si-probes realized only by MEMs processes, the recent introduction of implantable active neural probes based on microstructured monolithic complementary metal-oxide semiconductor (CMOS) devices (i.e. NeuroPixels, NeuroSeeker and SiNAPS) is opening new perspectives by enabling to scale up by several orders of magnitude the number of recorded neurons within and across multiple brain circuits. To this aim, previously reported studies have demonstrated the possibility of combining Neuropixels recordings with optogenetic stimulation of both excitatory and inhibitory neural populations in vivo. However, SiNAPS probes are built on a radically different circuit architecture that has not been shown yet in combination with optogenetic light stimulation. SiNAPS neural probes are based on a modular on-chip circuitry architecture and on DC-coupled front-ends. Rather than integrating front-end amplifiers at the base of the probe,as in NeuroPixels and NeuroSeeker, our SiNAPS probes integrate front-ends underneath each single electrode. This one-to-one correspondence between electrodes and amplifiers allows continuously recording at sub-millisecond resolution from all the integrated electrodes while defining an architecture that simplifies the realization of probes with different layouts and number of recording sites. Initially demonstrated with a 512 electrode single-shank probe layout, we recently presented a 4-shank version with 1024 electrodes (electrode pitch 28 µm) that was realized in standard 180 nm CMOS technology by “copy and paste” the basic circuit modules. Here, we will present and discuss first results obtained by combining optogenetics with electrical recordings using a 4-shank SiNAPS probe with 1024 simultaneously recording electrodes. Specifically, our preliminary recordings report light-evoked neural activity and confirm the possibility of exploiting SiNAPS probes for investigating optogenetic response dynamics with a large spatial coverage, both at the single-neuron and large-scale network level with an unprecedented spatiotemporal resolution.

Disclosures: F. Boi: None. **A. Locarno:** None. **J.F. Ribeiro:** None. **G. Orban:** None. **R. Tonini:** None. **G.N. Angotzi:** None. **L. Berdondini:** None.

Poster

P985. New Approaches in Optogenetics

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Program #/Poster #: P985.07

Topic: I.08. Methods to Modulate Neural Activity

Support: CURE Taking Flight Award
NIH F32NS116416

Title: Closed-loop optogenetic control of single unit phase locking to network oscillations in behaving mice

Authors: *Z. CHRISTENSON WICK, P. A. PHILIPSBERG, S. I. LAMSIFER, T. SHUMAN; Neurosci., Icahn Sch. of Med. at Mount Sinai, New York, NY

Abstract: The precise timing of neuronal spiking relative to the brain's endogenous oscillations (i.e., phase locking) has long been hypothesized to coordinate cognitive processes and maintain excitatory-inhibitory homeostasis. Though its causal role has never been tested, disruptions in phase locking have been described in several neurological diseases with associated cognitive deficits, including epilepsy. We predict that this loss of phase locking may prevent maintenance of excitatory-inhibitory homeostasis and exacerbate pathological neuronal activity such as seizures. Further, we predict that disruptions in phase locking may prevent effective information transfer between regions. We developed a closed-loop optogenetic system to bidirectionally control the phase locking of hippocampal neurons to ongoing endogenous theta oscillations. We performed in vivo silicon probe recordings in male and female head-fixed control and epileptic mice navigating a virtual track, identified the baseline phase preference of parvalbumin- or somatostatin-expressing interneurons within the hippocampus, and applied our closed-loop system to phase lock their spiking to the trough or peak of hippocampal theta. This closed-loop system uses real-time signal processing to detect the target phase and deliver bidirectional photo-manipulation within milliseconds; it is capable of targeting any phase of theta with high precision. Using this system in awake, behaving mice, we have altered the phase locked firing of hippocampal interneurons and have preliminary data suggesting that altering their phase locking may impact epileptiform activity. This closed-loop system outperforms existing tools and will enable us to continue investigating the causal role of precise single-unit phase locking to network-wide oscillations with many versatile applications.

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Poster

P985. New Approaches in Optogenetics

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Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P985.08

Topic: I.08. Methods to Modulate Neural Activity

Title: Hippocampal Power Spectrum Responses to Irregular Optogenetic Stimulation Parameter Spaces

Authors: *E. R. COLE[1], T. E. EGGERS[2], N. G. LAXPATI[2], M. J. CONNOLLY[3], M. C. GOMBOLAY[4], R. E. GROSS[2];
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Abstract: Introduction: Brain stimulation has become a fundamental tool for treating and understanding the mechanisms of neurological diseases, such as Parkinson's disease, epilepsy, and more. One key challenge in delivering effective brain stimulation is choosing the stimulation parameters (such as the amplitude, frequency, contact configuration, and pulse width) that induce a desired change in symptoms, behavior, or neural activity. Most clinical and translational studies use constant-frequency pulses of stimulation, but recent evidence suggests that irregular temporal stimulation patterns can produce unique changes in neural activity and also lead to better therapy in various clinical applications. Here, we comprehensively evaluate several optogenetic stimulation parameter spaces, report their differing effects on hippocampal LFP spectral activity, and present a machine learning strategy using Bayesian optimization to select parameters that induce a desired multi-band change in the power spectrum.
Methods: Sprague-Dawley rats (N=4) were prepared for pan-neuronal excitatory optogenetic stimulation via adeno-associated virus in the medial septum (hSyn-ChR2) and 16-channel LFP recording in the hippocampus, and we swept the following parameter spaces with grid and random search (>1000 trials per subject): Standard pulse train: Constant-frequency pulsatile stimulation.Nested pulse train: Pulsatile stimulation modulated by a slow on/off period.Sinusoid: Sinusoidal time-series of light power.Summed sinusoid: Sum of two different-frequency sinusoids.Poisson: Pulsatile pattern with an inter-pulse interval drawn from a Poisson distribution.
Results: Via dimensionality reduction analysis, we show that these parameter spaces produce similar effects on low gamma (32-50 Hz) activity, with key differences in driving theta (4-10 Hz) activity and modulating the combination of low and high-frequency activity throughout the power spectrum. We compare these induced spectral patterns to LFP activity recorded during two natural conditions (exploratory and quiescent behavior), and describe two strategies to represent the complete power spectrum as a single objective function for shaping multispectral neural responses: by computing hand-crafted features or learning features through dimensionality reduction on the power spectrum, then quantifying a distance metric between induced and desired features.
Conclusion: The results of this comprehensive evaluation of various parameter spaces can preliminarily guide the selection of stimulation parameters based on spectral biomarkers in translational and clinical applications.

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Poster

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Support: NIH R01 AA027214
NIH T32 AA07462

Title: The role of the anterior insular cortex to dorsolateral striatum in modulating binge alcohol consumption

Authors: *D. HAGGERTY, B. MUÑOZ, B. ATWOOD;
Indiana Univ. Sch. of Med., Indianapolis, IN

Abstract: Many drugs of abuse, including alcohol, affect neurotransmission in the dorsal striatum by altering glutamatergic signaling. For example, alcohol specifically ablates long-term synaptic depression (LTD) at glutamatergic synapses in the dorsolateral striatum (DLS). This specific type of LTD is mediated by μ-opioid receptors (mOP-LTD) exclusively on inputs from the anterior insular cortex (AIC). However, the behavioral effects of mOP-LTD and other types of drug-induced synaptic plasticity changes at AIC inputs to the DLS remain unknown. Here we show that photoexcitation of AIC to DLS projecting neurons alter consumption of binge-like levels of alcohol during Drinking in the Dark. Using wireless, in-vivo, closed-loop optogenetics to photoexcite AIC terminals in the DLS during alcohol consumption behaviors, we describe how modulating DLS inputs from the AIC can alter alcohol drinking behaviors, drinking microstructure, and decrease alcohol consumption levels across six weeks of Drinking in the Dark. Furthermore, photoexciting AIC inputs into the DLS only altered synaptic plasticity at AIC-DLS synapses that had been exposed to alcohol, not water during Drinking in the Dark sessions. These findings suggest that AIC to DLS projecting neurons may be uniquely sensitive to alcohol induced synaptic plasticity changes that correlate with alterations in alcohol consummatory behaviors. This work also indicates that insular cortex synapses in the DLS may be key drug targets for affecting the development of alcohol-induced neuroadaptations.

Disclosures: D. Haggerty: None. **B. Muñoz:** None. **B. Atwood:** None.

Poster

P985. New Approaches in Optogenetics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P985.10

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH grant UF1NS107574
NIH Grant F32-EY031977
NEI grant K99 EY029758-01
National Science Foundation Graduate Research Fellowship Grant No. DGE 1752814

Title: A transgenic mouse line for high-precision, all-optical manipulation and monitoring of neural activity

Authors: *H. A. BOUNDS[1], M. SADAHIRO[2], W. D. HENDRICKS[2], M. A. GAJOWA[2], I. A. OLDENBURG[2], K. GOPAKUMAR[2], D. QUINTANA[2], T. L. DAIGLE[3], H. ZENG[4], H. ADESNIK[2];
[1]Univ. Of California, Berkeley Neurosci. Grad. Program, Berkeley, CA; [2]Mol. and Cell. Biol., Univ. of California, Berkeley, Berkeley, CA; [3]Allen Inst. For Brain Sci., Seattle, WA; [4]Structured Sci., Allen Inst. for Brain Sci., Seattle, WA

Abstract: Establishing causal links between specific patterns of neural activity, sensory computation, and behavior requires the ability to simultaneously monitor and manipulate neural activity with cellular resolution. Systems that combine 2-photon calcium imaging and targeted photostimulation provide such an approach. These all-optical approaches require co-expression of a calcium indicator and a potent, soma-targeted excitatory opsin construct. This has so far been achieved in the mammalian brain through viral transduction. However, this invasive procedure results in focal, variable expression that often becomes toxic over time. To overcome these challenges, we developed and validated a Cre-reporter mouse ('Ai203') that conditionally expresses a soma-localized opsin, ChroME, fused directly to GCaMP7s. 1- or 2-photon illumination of expressing neurons *in vitro* and *in vivo* produces powerful activation comparable to viral expression. The novel soma-enriched, membrane-bound GCaMP7s is a potent calcium-sensing tool that provides sensitive measurements of neural activity. Mice expressing Ai203 in cortical excitatory neurons have normal visual responses in V1 neurons and normal performance in a cortex-dependent operant visual task, indicating that transgenic expression of the opsin and GCaMP7s does not compromise circuit function. Additionally, we demonstrate that the Ai203 line is useful for high-throughput, high-resolution 2-photon synaptic connectivity mapping. This new transgenic line will facilitate the study of neural circuits by providing a flexible, convenient, and stable expression mechanism for combined optogenetic manipulation and functional imaging of large neural populations.

Disclosures: H.A. Bounds: None. **M. Sadahiro:** None. **W.D. Hendricks:** None. **M.A. Gajowa:** None. **I.A. Oldenburg:** None. **K. Gopakumar:** None. **D. Quintana:** None. **T.L. Daigle:** None. **H. Zeng:** None. **H. Adesnik:** None.

Poster

P985. New Approaches in Optogenetics

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:00 AM - 11:00 AM

Program #/Poster #: P985.11

Topic: I.08. Methods to Modulate Neural Activity

Support: Swedish Research Council No 2016-06195
Lund University STYR grant 2015/1203
Lundbeck Foundation
Novo Nordisk Foundation

Title: Novel biocompatible probe design for optogenetics

Authors: *M. ANDERSEN[1,2], L. WALLMAN[2], L. PETTERSSON[2], O. KIEHN[1], J. SCHOUENBORG[2,3];
[1]Dept. of Neurosci., Univ. of Copenhagen, Copenhagen, Denmark; [2]Neuronano Res. Center, Dept. of Exptl. Medicine, Lund Univ., Lund, Sweden; [3]NanoLund, Lund Univ., Lund, Sweden

Abstract: Optogenetics has increased our knowledge of neural network function by its ability to selectively manipulate well-defined neuronal populations. However, reaching deep brain nuclei with light is difficult because of scattering and absorption of the emitted light, which makes implantation of optical fibers necessary. The implantation of stiff and rigid materials like glass and metal in the nervous system is known to result in a loss of nearby neurons and glial responses resembling infarct-like reactions, which likely strongly affects neuronal activity. It would therefore be valuable to develop highly flexible probes for optogenetics that can be used in combination with long-lasting electrophysiological recordings. The aim of the present work was thus to develop an implantable and biocompatible light guide in polydimethylsiloxane (PDMS) and to evaluate its light transmission properties and effects on the brain tissue in rats. We have designed and tested a mono-fiber pulled vertically from PDMS. The fiber was pulled from a drop of semi-cured Sylgard 184 (3:1 ratio) at room temperature. We focused on two diameters: 75 µm and 120 µm. Any desired diameter is, however, possible to construct using our method. The developed PDMS fibres were compared with optic glass fibers with respect to tissue reactions and neuronal responses to light in virally channelrhodopsin (ChR2) transfected rat hippocampus *in vivo*. Using immunohistochemistry of the implanted site we show an improved biocompability of PDMS over glass (core: pure silica (105µm), cladding: fluorine-doped silica (10µm)) of equal size fibers with respect to a lower ED1 staining close to the implant. Using extracellular acute recordings, we demonstrate that the developed single layer PDMS fibers (using the tissue as cladding) transmit enough light to activate ChR2 and generate action potentials in rat hippocampus *in vivo*. Our development and demonstration of *in vivo* functionality of highly biocompatible optical fibers provides the opportunity to deliver light with less damage to the area of interest. When combined with biocompatible recording electrodes and

transgenic technology these PDMS optical waveguides gives the ability to perform longitudinal single unit recordings from phototagged neurons in nearly intact areas.

Disclosures: M. Andersen: None. **L. Wallman:** None. **L. Pettersson:** None. **O. Kiehn:** None. **J. Schouenborg:** None.

Poster

P002. Neurogenesis and Gliogenesis: Proliferation I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P002.01

Topic: A.01. Neurogenesis and Gliogenesis

Support: Ministry of Science and Technology(110-2320-B-033 -001 -MY3)

Title: Maternal intake of Ganoderma tsugae benefits in brain development and cognitive function of the offspring in Sprague-Dawley rats

Authors: *C.-Y. TSENG;
Biomed. Engin., Chung Yuan Christian Univ., Chungli City, Taiwan

Abstract: Maternal malnutrition, exposure to environmental hormones, heavy metals, or environmental pollutants cause damage to fetal brain development. Antioxidants, anti-inflammatory agents, and nutraceuticals have been shown to protect or mitigate the adverse factors affecting maternal and child health. Our previous research showed that *Ganoderma microsporum* immunomodulatory proteins reduce the impacts of exposure to $PM_{2.5}$ during pregnancy in Sprague-Dawley (SD) rats on their offspring's brain development through immunomodulation. However, most of the literature focuses on how to reverse or prevent abnormal brain development, and few scholars explore whether these anti-inflammatory/antioxidants or nutraceuticals would benefit fetal brain development, promote neurogenesis, and improve nerve function? Humans may have retained attitudes about this research because of safety concerns during pregnancy, but literature search engines show few along with animal studies. Therefore, this study plan to take the lead in evaluating whether supplementing nutraceuticals during pregnancy can improve fetal brain development and nerve function. We adopted *Ganoderma tsugae* (GTDE) in this study, which has the best antioxidant function in Reishi, a typical and traditional Chinses medicine. We applied less than a thousand times the dose of no observed adverse effect levels of GTDE (200 μg/kg) during pregnancy. We found that maternal gavage of GTDE effectively enhances the rat fetal cortical neurogenesis, especially in the lateral ventricular/ventricular zone. The mRNA PCR microarray analysis of the fetal cortex shows nine genes that significantly increased due to maternal exposure to GTDE, including Bmp8a, Drd2, Egf, Gdnf, Hes1, Ndp, Pax2, Pax3, and Sox2. These genes play essential roles in neurogenesis. Moreover, the maternal application of GTDE promotes the

cognitive function of the offspring from working memory analysis. Here we present the first report showing that low doses of *Ganoderma tsugae* can effectively promote fetal brain development and offspring's cognitive function.

Disclosures: C. Tseng: None.

Poster

P002. Neurogenesis and Gliogenesis: Proliferation I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P002.02

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH Grant R15 CA235749

Title: Myomegalin regulates Hedgehog pathway by controlling PDE4D at the centrosome

Authors: *J. ZHANG[1], H. PENG[1], A. YA[1,3], W. MA[1], S. VILLA[1], S. SUKENIK[2], X. GE[1]; [1]Mol. and Cell Biol., [2]Chem. and Chem. Biol., Univ. of California, Merced, Merced, CA; [3]Mol. and Cell Biol., Dartmouth Col., Hanover, NH

Abstract: Mutations in the Hedgehog (Hh) signaling are implicated in birth defects and cancers, including medulloblastoma, one of the most malignant pediatric brain tumors. Current Hh inhibitors face the challenge of drug resistance and tumor relapse, urging new insights in the Hh pathway regulation. Our previous study revealed how PDE4D (Phosphodiesterase 4D) controls global levels of cAMP in the cytoplasm to positively regulate Hh signaling; in the present study we found that a specific isoform PDE4D3 is tethered to the centrosome by myomegalin, a centrosome/Golgi associated protein. CRISPR knockout of Myomegalin dislocates PDE4D3 from the centrosome, leading to local PKA over-activation and inhibition of the Hh signaling, leaving other PKA-related pathways unaffected. Targeting a FRET (fluorescence resonance energy transfer) based PKA sensor, AKAR4, to the centrosome, we detected a constitutively high local PKA activity, suppressing the Hh signaling activity. Myomegalin loss suppresses the proliferation of granule neuron precursors, and blocks the growth of medulloblastoma in mouse model. Our findings specify a new regulatory mechanism of the Hh pathway, and highlight an exciting therapeutic avenue for Hh-related cancers with reduced side effects.

Disclosures: J. Zhang: None. **H. Peng:** None. **A. Ya:** None. **W. Ma:** None. **S. Villa:** None. **S. Sukenik:** None. **X. Ge:** None.

Poster

P002. Neurogenesis and Gliogenesis: Proliferation I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P002.03

Topic: A.01. Neurogenesis and Gliogenesis

Support: Wellesley Summer Research Program

Title: Is the effect of serotonin on neurogenesis concentration dependent?

Authors: *A. AGYEMANG, B. S. BELTZ, P. CHAVES DA SILVA, A. EDWARDS; Neurosci. Program, Wellesley Col., Wellesley, MA

Abstract: Is the Effect of Serotonin on Neurogenesis Concentration Dependent?August Agyemang, Barbara Beltz and Paula Chaves da Silva*Wellesley College, 106 Central Street, Wellesley, Massachusetts, 02481vagyeman@wellesley.edu***Abstract**Adult neurogenesis, a process observed in both vertebrates and invertebrates, is the birth and integration of new neurons in the adult brain. In many crustaceans, including crayfish, neurogenesis occurs throughout life. Newborn neurons are produced by progenitor cells that congregate in a neurogenic niche. These cells proliferate, and descendants travel to proliferation zones in cell clusters 9 and 10 (CL9 and CL10) that innervate into the olfactory and accessory lobes. The niche progenitor cells originate from an extrinsic source. Studies indicate that the immune system supplies hemocytes that replenish these cells. Thus, the factors that affect the immune system, such as diet, exercise, and neurotransmitter activity, also affect neurogenic activity in the niche. One such neurotransmitter, the monoamine serotonin (5-HT), has been identified as a regulator of neurogenesis. 5-HT influences the attraction of hemocytes to the niche as well as the proliferation of the precursor cells in CL10. Serotonin (5×10^{-9}M)results in an increase in hemocyte counts in the blood and niche cell counts compared with control conditions. With knowledge of this interaction, this study investigates the effects of varying concentrations of serotonin on the early stages of neurogenesis *in vivo*; total hemocyte counts (THC), the total number of niche cells, and the number of proliferating cells in the niche will be assessed. *Procambarus acutus* crayfish were incubated in different concentrations of5-HT, [5×10^{-10}M, 5×10^{-8}M, 5×10^{-6}M], for 36 hours.Additionally, the animals were injected with bromodeoxyuridine (BrdU), a proliferation marker, 12 hours after incubation. We hypothesize that as 5HT concentration increases, so will neurogenic activity in the niche.These studies will test whether serotonin influences the crayfish niche in a concentration-dependent manner.

Disclosures: A. Agyemang: None. **B.S. Beltz:** None. **P. Chaves da Silva:** None. **A. Edwards:** None.

Poster

P002. Neurogenesis and Gliogenesis: Proliferation I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P002.04

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH grant EY07060
NIH grant EY07003

Title: Il10 and inflammatory resolution are critical element of photoreceptor regeneration in zebrafish

Authors: *M. NAGASHIMA, P. F. HITCHCOCK;
Univ. of Michigan Dept. of Ophthalmology and Visual Sci., Ann Arbor, MI

Abstract: In the retina of zebrafish, acute inflammation is required to initiate reprogramming and proliferation of the intrinsic stem cells, Müller glia. Reprogrammed Müller glia divide asymmetrically to produce retinal progenitors, which rapidly proliferate and differentiate to replace the ablated neurons. Chronic inflammation, however, is detrimental. Mechanisms governing the resolution of inflammation during the regeneration of photoreceptors remain to be elucidated. Il10 is a classical anti-inflammatory cytokine that plays a critical role in resolving inflammation and promoting wound healing. In this study, we examine the expression and function of il10 in the zebrafish retina following the death of photoreceptors. We also assess how dysregulation of inflammatory resolution affects multi-step process of retinal regeneration. Following photolytic lesion, *il10* is significantly upregulated by 2 days post lesion and returns to baseline by 5 days. In the absence of Il10, expression of the pro-inflammatory cytokines, tnf-a, tnf-b, and il1b, are significantly elevated. These data are consistent with that il10 is a potent anti-inflammatory cytokine and indicate that Il10 suppresses the production of pro-inflammatory cytokines following acute death of photoreceptors. At 5 to 7 days post lesion, when in wild-type animals photoreceptor progenitors exit the cell cycle, proliferation in the il10 mutants remains significantly elevated. These results suggest that il10-mediated inflammatory resolution determines the magnitude of proliferation among the Müller glia-derived progenitors following the acute death of photoreceptors.

Disclosures: M. Nagashima: None. **P.F. Hitchcock:** None.

Poster

P002. Neurogenesis and Gliogenesis: Proliferation I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P002.05

Topic: A.01. Neurogenesis and Gliogenesis

Support: Reed College start-up (to KLC) and student fellowships
Wellcome Trust (SW and HR)

Title: Visual input promotes proliferation and survival of stem and progenitor cells in the zebrafish optic tectum

Authors: O. L. HAGEN[1], Y. KIM[1], E. KUSHKOWSKI[3], C. G. ROBERTS[2], H. ROUSE[4], M. VARGA[5], S. W. WILSON[4], ***K. CERVENY**[1];
[1]Biol., Reed Col., Portland, OR; [2]Biol., Reed Col., PORTLAND, OR; [3]Univ. of Chicago, Chicago, IL; [4]Univ. Col. London, University College London, United Kingdom; [5]ELTE Eötvös Loránd Univ., Budapest, Hungary

Abstract: In zebrafish, visual system growth and development is a life-long process. Specialized stem cell niches established early in eye and brain development support life-long growth of each retina and corresponding optic tectal lobe. Coordinated growth of these tissues ensures that accurate retinotopic mapping is maintained as new neurons are added as the visual system grows. Studies of adult fishes have provided evidence that survival, proliferation, and differentiation of cells in the optic tectum is correlated with retinal innervation, raising the possibility that retinal axons provide crucial information to control stem and progenitor cell behaviors in the optic tectum. To explore how retinal innervation influences optic tectum growth in larval zebrafish, we examined brains lacking innervation from (i) both eyes as a result of the *lakritz* mutation and (ii) one eye as a result of surgical eye removal, and then compared them with wild-type larvae with two eyes. We observed elevated cell death in non-innervated and denervated optic tecta lobes only after 6 days post-fertilization (dpf). In addition, we uncovered that optic tecta lacking innervation contained fewer *sox2+* stem cells and a smaller proportion of proliferating progenitors. To test how much light-evoked retinal activity contributes to proliferation and survival in the optic tectum, we reared fish from 1-9 dpf in the dark and then asked how optic tectal cells in innervated and non-innervated tecta behave. We found that innervation was still able to promote survival and proliferation, albeit to a lesser extent, when fish were reared in the dark. Taken together our data led us to examine whether trophic factor(s) were being provided by the optic nerve, irrespective of light-evoked retinal activity. Combining bulk RNA-Sequencing and candidate gene approaches, we identified innervation-dependent Wnt/B-catenin pathway activity in larval zebrafish optic tecta, raising the possibility that a Wnt, may travel along the optic nerve and serve as a trophic cue for tectal stem and progenitor cells.

Disclosures: O.L. Hagen: None. **Y. Kim:** None. **E. Kushkowski:** None. **C.G. Roberts:** None. **H. Rouse:** None. **M. Varga:** None. **S.W. Wilson:** None. **K. Cerveny:** None.

Poster

P002. Neurogenesis and Gliogenesis: Proliferation I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P002.06

Topic: A.01. Neurogenesis and Gliogenesis

Support: TETFund Grant
FRC Grant
Supervisors incentive

Title: Age-related distribution of oligodendrocytes in the brain of the male japanese quail (curtonix japonica)

Authors: *A. M. SHEHU[1,2], P. MAZENGENYA[3,1], P. NKOMOZEPI[4], A. O. IHUNWO[1]; [1]Sch. of Anatom. Sci., Univ. of the Witwatersrand, Johannesburg, South Africa; [2]Human Anat., Federal Univ. Dutse, Jigawa, Nigeria; [3]Col. of Med., Ajman Univ., Ajman, United Arab Emirates; [4]Human Anat. and Physiol., Univ. of Johannesburg, Johannesburg, South Africa

Abstract: Gliogenesis involves the generation of new glial cells (astrocytes and oligodendrocytes) from neural stem cell or progenitor cells (NSPC) in both developing and adult individuals. Oligodendrocytes are cells accountable for myelin production and responsible for maintaining the electrical impulse through the myelin sheath in neuronal axons. However, there is a paucity of literature on the life history changes in the expression of oligodendrocyte transcription factor 2 (OLIG2) in the avian species. We use myelin silver stain and oligodendrocyte transcription factor 2 (OLIG2) to study the distribution of oligodendrocytes, and in the juvenile, sub-adult and adult brain. The results showed that the myelin stain exhibited an increase in the density of myelinated fibres as development progresses but showed a decline in the adult brain. There was a high density of positive Olig2 immunoreactivity (OLIG2-ir) in the white matter and fibre tracts (optic tectum, optic tract, anterior and posterior commissure and tectal ventricle) arranged in rows while those found in the grey matter are scattered (pallium, Subpallium and hippocampus). A few moderate OLIG2-ir were observed in the caudal area of the ventricular zone. The density level of the OLIG2-ir oligodendrocytes in the quail brain corresponds to the regions where the myelination level is on the high. The presence of OLIG2 in the Japanese quail brain indicates that the protein plays a critical role in the production, development and differentiation of oligodendrocytes. In conclusion, the three week juvenile and six week sub-adult stages presented the highest density and more regions for oligodendrocytes progenitor cells. The oligodendrocytes transcription factor 2 in the Japanese quail brain similarly decline with post-hatching age.

Disclosures: A.M. Shehu: None. **P. Mazengenya:** None. **P. Nkomozepi:** None. **A.O. Ihunwo:** None.

Poster

P002. Neurogenesis and Gliogenesis: Proliferation I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P002.07

Topic: A.01. Neurogenesis and Gliogenesis

Support: State of Pennsylvania CURE grant
Charles Henry Leach Fund

Title: Hippocampal neural stem cell proliferation and survival are increased in the adult brain during a viral infection

Authors: *M. N. CHANDWANI[1], Y. S. KAMTE[1], M. E. HEMERSON[2], L. A. O'DONNELL[1]; [1]Dept. of Pharmacol., Duquesne Univ. Grad. school of Pharmaceut. sciences, Pittsburgh, PA; [2]High Point Univ., High Point, NC

Abstract: Viral infections in the adult central nervous system (CNS) can cause physiological changes despite successful viral control. Neural stem cells (NSCs) may be altered during an infection either by direct infection and/or the anti-viral immune response, potentially contributing to changes in neurological function. To define the components of the anti-viral immunity that regulate NSCs, we utilized a mouse model of CNS measles virus (MV) infection. In this model, human CD46+ (a MV receptor) is expressed only on mature neurons, restricting MV to neurons and sparing the NSCs from infection. Using this model, we are assessing the effects of the anti-viral immune response on NSC activity in the adult brain in the context of successful control of the virus. Two-month-old mice were infected with MV, and flow cytometry was conducted at 7 and 60 days post infection (DPI) to quantify the NSC, neuronal, and glial populations. NSC numbers (Nestin+) increased at 7 and 60 dpi in the hippocampus. NSC proliferation (Nestin+BrdU+) trended towards an increase during and after infection, implying long-term changes in NSC activity. Mature neurons (βIII-tubulin+) were reduced at 7 dpi, possibly due to infection-mediated cell death, but then recovered to basal levels by 60 dpi. Neurogenesis (Doublecortin+BrdU+) was increased in the hippocampus at 7dpi, which may account for the recovery of mature neurons at 60 dpi. NSC proliferation and neurogenesis increased at 7 dpi in the SVZ, but both proliferation and neurogenesis were reduced at 60 dpi compared to uninfected controls. Thus, hippocampal NSC activity was increased at 7 and 60 dpi, whereas SVZ NSC activity was increased at 7 dpi but was reduced at 60 dpi. The SVZ has a higher viral load as compared to the hippocampus at 7 and 60 dpi, which could be driving these region-dependent differences in the outcome in NSCs. Immune cells such as microglia, T-cells, and B-cells were increased early in infection (7 dpi); however, only resident memory cytotoxic T-cells remain at 60 dpi after the infection is resolved. In the absence of the adaptive immunity (CD46+/Rag2-KO), NSCs increased during infection similarly to CD46+ mice. Microglia were upregulated in both CD46+ and CD46+/Rag2KO mice, and positively correlate with hippocampal NSCs and negatively correlate with the SVZ NSCs, suggesting that microglia may regulate NSC activity during infection. Current studies address the role of microglia in modulating NSC activity *in vivo* and *in vitro.* We are also assessing the long-term effects of the infection on learning and memory. These studies emphasize that NSCs experience enduring changes due to a successful anti-viral immune response in the adult brain.

Disclosures: M.N. Chandwani: None. **Y.S. Kamte:** None. **M.E. Hemerson:** None. **L.A. O'Donnell:** None.

Poster

P002. Neurogenesis and Gliogenesis: Proliferation I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P002.08

Topic: A.01. Neurogenesis and Gliogenesis

Support: Supported by the UTHSC Neuroscience Institute

Title: Preterm birth compromises hippocampal neurogenesis

Authors: *I. ISKUSNYKH[1], N. FATTAKHOV[1], A. ZAKHAROVA[1], L. MUKHAMETZYANOVA[1], R. BUDDINGTON[2], V. CHIZHIKOV[1];
[1]Univ. of Tennessee Hlth. Sci. Ctr., Memphis, TN; [2]Univ. of Memphis, Memphis, TN

Abstract: Preterm birth, especially prior to 34 weeks of gestation, is associated with disrupted development of the central nervous system and is a leading cause of childhood disability. The lower hippocampal volumes of children that were born preterm are of clinical interest because of the association with varying degree of memory impairment. We explored the molecular and cellular mechanisms of the lower hippocampal volumes using preterm pigs delivered via C-section at day 105 of gestation (91% of term and relevant to 32-week preterm infants) and reared in NICU settings for 10 days to term equivalent age. Newborn term pigs served as controls. Each experimental group included n=8 animals of both sexes. Ki67+ progenitors and Prox1+ neuronal populations were identified in the hippocampal dental gyrus by immunohistochemical staining and counted using unbiased stereological method. The numbers of Ki67+ and Prox1+cells in the dental gyrus of preterm pigs were decreased compared to control pigs. Proliferation of neural progenitors in the hippocampus of preterm pigs was reduced as well. The disturbances in dental gyrus neurogenesis may contribute to the long-term memory impairment. Importantly, the reduced neurogenesis in preterm pigs occurred without the presence of necrotizing enterocolitis or other diseases that frequently occur after preterm birth and are known to indirectly disrupt brain development. Our findings highlight the preterm pig as a translational model to study the consequences of premature birth on brain development, the underlying molecular mechanisms, and interventions aimed to mitigate the disrupted development and long-term disabilities. Supported by the UTHSC Neuroscience Institute.

Disclosures: I. Iskusnykh: None. **N. Fattakhov:** None. **A. Zakharova:** None. **L. Mukhametzyanova:** None. **R. Buddington:** None. **V. Chizhikov:** None.

Poster

P007. Neuronal Differentiation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

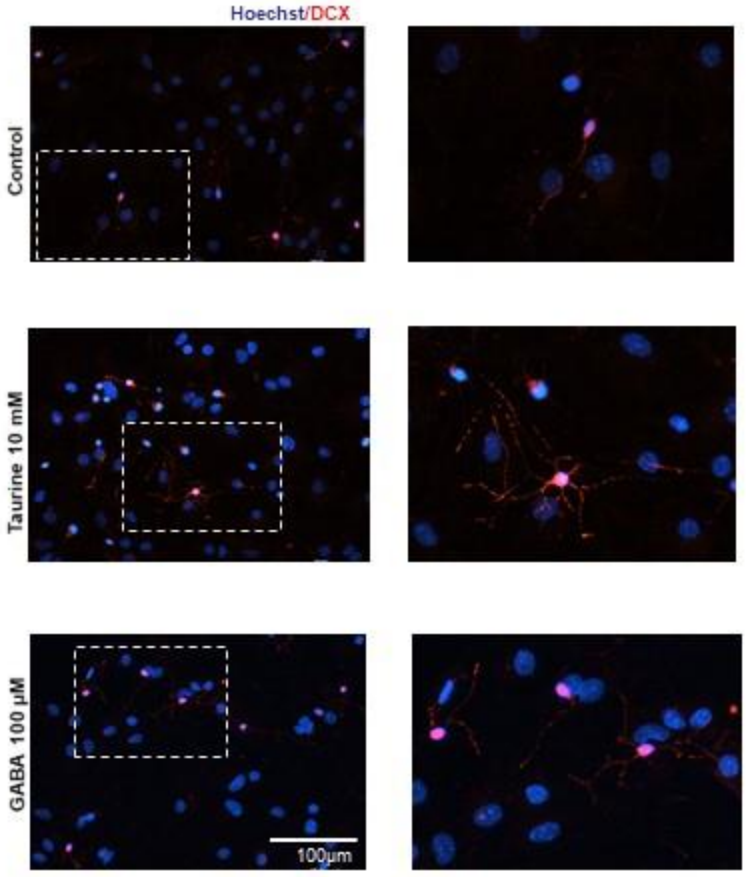
Program #/Poster #: P007.01

Topic: A.01. Neurogenesis and Gliogenesis

Title: Subventricular neural progenitor cells differentation is modulated by taurine via gaba receptors interaction

Authors: *N. GUTIÉRREZ[1], E. GRIEGO[1], E. J. GALVAN[1], L. OCHO-DE LA PAZ[2]; [1]CINVESTAV Sur, Mexico City, Mexico; [2]Facultad de Medicina, UNAM, Mexico City, Mexico

Abstract: Neurogenesis occurs throughout mammals' lives, in the regions composed of the hippocampal subgranular zone and the subventricular zone (SVZ) of lateral ventricles, and multiple extrinsic factors regulate this process. Taurine, a neuroactive factor, plays a central role in the proliferation, differentiation, and migration of neural progenitor cells. However, the mechanism of action of taurine is not well understood. Thus, we explored the effect of taurine on the differentiation process of SVZ neural progenitor cells through the interaction with GABA receptors. Immunofluorescence assays were performed on cells isolated from SVZ of CD1 mice (P8) in the absence or presence of taurine. Our results show that neural progenitor cells express Sox-2 and Nestin, both progenitor cell markers; however, no signal for DCX was observed, confirming that cells obtained from the SVZ are neural progenitors. In cultures exposed to differentiation conditions with taurine, the number of DCX+ cells was increased. Morphometric analysis revealed a significant difference in cell morphology. Compared to control and GABA-treated cells, taurine-treated cells exhibited a significant number of secondary and tertiary neurites, increased dendritic branching, and marked complexity in dendritic arborization. Taurine actions were sensitive to picrotoxin, indicating active participation of $GABA_A$ receptors. Also, we measured the electrophysiologic properties of the control, GABA-, and taurine-treated cells with patch-clamp whole cell recordings. Taurine-treated cells developed an electrophysiologic behavior (passive and active properties) akin to central neurons. Our results provide information regarding the role of taurine as a morphogen in the neurogenic processes throughout the interaction with GABA receptors and their role as a central player in the maturation processes of NPCs into functional neurons. This work represents an advance in the morphometric effect and suggests a functional effect of taurine in the neuronal differentiation process.



Disclosures: N. Gutiérrez: None. **E. Griego:** None. **E.J. Galvan:** None. **L. Ocho-de la Paz:** None.

Poster

P007. Neuronal Differentiation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P007.02

Topic: A.01. Neurogenesis and Gliogenesis

Support: Spectrum Health-Michigan State University Alliance Corporation
Autism Research Initiative
R00-NS096108
R00-NS096108-S1

Title: Impact of conditional Wac loss of function on GABAergic interneuron development

Authors: A. M. STAFFORD[1], H.-H. KIM[2], D. M. AUTIO[2], H. C. RUDOLPH[1], L. E. MARTINETTI[2], A. PAUL[3], S. R. CRANDALL[2], **D. VOGT**[1];
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Abstract: Multiple genes that incur an increased risk of an autism diagnosis have been identified, however, some are still without studied animal models to uncover their impact on brain function. As more genes that contribute to autism are studied, distinct cellular hubs have been identified, including synaptic dynamics, nuclear regulation and cellular signaling. The latter is of particular interest, as there are several monogenic syndromes caused by genes implicated in autism, with known cellular signaling pathways that could be manipulated in future therapeutic approaches. One such syndrome, DeSanto-Shinawi syndrome (DESSH), is caused by mutations in the WW domain-containing adapter with Coiled Coil (*Wac*). Through its protein domains, *Wac* has been shown to positively regulate MTOR activity, one of the main signaling hubs that harbors many other genes implicated in autism. Wac can also mediate events within the nucleus. However, the function of *Wac* in the brain has not been studied.
We generated a conditional deletion of *Wac* in brain inhibitory cortical interneurons (CINs), a critical cell type known to be impacted by MTOR activity and implicated in autism. Our data reveal a critical role for *Wac* in regulating the proper number of CINs that populate the developing and adult cortex, with distinct impacts on different CIN subgroups. In addition, we found evidence for changes in brain excitability and neuron morphology that could underlie some of the cognitive symptoms associated with DESSH. Overall, our results provide a new genetic mouse model to study potential inroads into DESSH and cellular hubs implicated in autism biology.

Disclosures: A.M. Stafford: None. **H. Kim:** None. **D.M. Autio:** None. **H.C. Rudolph:** None. **L.E. Martinetti:** None. **A. Paul:** None. **S.R. Crandall:** None. **D. Vogt:** None.

Poster

P007. Neuronal Differentiation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P007.03

Topic: A.01. Neurogenesis and Gliogenesis

Support: Brain and Behavior Research Foundation NARSAD Young Investigator Award
McNair Scholars Program
Murchison Summer Research Fellowship

Title: Differentiation of HT22 cells, an immortalized hippocampal-derived neuronal cell line, increases degrees of neuronal characteristics

Authors: E. FARRELL[1], J. CLARK[2], M. HORN[1], J. VALADEZ[1], C. WILLIAMS[2], *G. M. BEAUDOIN, III[3];
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Abstract: HT22 cells are an SV40 transformed, immortalized mouse hippocampal neuron cell line. They are commonly used to study glutamate induced toxicity. Because they are immortalized, HT22 cells act very differently from true hippocampal neurons. HT22 cells have been used in multiple studies under the assumption that they operate similarly to primary cultured neurons, but that assumption may be invalid. We are characterizing the neuronal qualities of HT22 cells that are grown with sera and after switching to a neuron supporting media with N2 or B27 supplements. Additionally, the effects of nerve growth factor (NGF) and retinoic acid (RA) to further induce and support neuronal characteristics is being tested. The media supplements N2 and B27 are consistently used in the culturing of primary neurons and the differentiation of immortalized neurons. NGF has been shown to induce differentiation and structural changes in HT22 cells into neuronal cells. RA has been shown to regulate the CaMKIIα promoter. Differentiation is characterized via a variety of metrics. Activity of a late stage neuronal differentiation promoter, CaMKIIα, is assessed via expression of green fluorescent protein (GFP) and measured via confocal imaging. Because CaMKIIα is a neuron-specific promoter, quantifying its expression across differentiation conditions provides insight into how neuronal the cells become. Complexity of cell morphology, through extension of neurites, is evaluated via neurite analysis methods, including sholl analysis and tip counting. Cell cycle progression and apoptosis/necrosis levels are quantified using propidium iodide and annexin V staining and analyzed via flow cytometry. These cell cycle analyses are performed because differentiated neurons exhibit higher levels of apoptosis and are mitotically arrested. Finally, the maturation of a stable hyperpolarized membrane potential and expression of voltage-gated ion channels is assessed through electrophysiological recordings. Overall, results indicate that HT22 cells differentiated with a combination of N2 supplement, RA, and NGF show the highest degrees of neuronal characteristic when compared to undifferentiated cells. Additionally higher rates of apoptosis have been observed in cells differentiated with B27 supplement alone, however, rates of apoptosis appear to decrease with the addition of NGF and RA.

Disclosures: E. Farrell: None. **J. Clark:** None. **M. Horn:** None. **J. Valadez:** None. **C. Williams:** None. **G.M. Beaudoin:** None.

Poster

P007. Neuronal Differentiation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P007.04

Topic: A.01. Neurogenesis and Gliogenesis

Support: JSPS Kakenhi 20KK0338

Title: Transcriptional and epigenetic regulation of neuronal differentiation and function reveals key epigenetic phenomena

Authors: *Y. ZHOU, S. RASHAD, K. NIIZUMA;
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Abstract: Neuronal differentiation is a complex process. Although transcriptomic changes may be one of the key factors contributing to this complexity, a comprehensive analysis of transcriptomics and post-transcriptomics, including RNA stability, alternative splicing, and RNA binding proteins (RBPs) during this process is rarely explored. In order to decipher the whole landscape of transcriptomics and post-transcriptomic regulation underlying neuronal differentiation *in vitro*, we performed RNA sequencing and bioinformatic analyses on neural-like cells from SH-SY5Y human neuroblastoma cells, induced by retinoic acid and BDNF. Well-differentiated SH-SY5Y cells exhibited extensive neurite outgrowth and connections, and express mature neuronal markers. 728 differentially expressed genes (DEGs) were identified in differentiated cells compared to undifferentiated cells and Gene Ontology (GO) analysis revealed most DEGs were enriched in synaptic signaling and cell morphogenesis pathways, which was consistent with morphological changes of differentiated cells. At the RNA stability level, GO analysis showed a number of pathways including electron transport chain and glutathione metabolic process were differentially regulated. Mature neurons are known to be highly sensitive to oxidative stress. Our results suggest that this heightened sensitivity is regulated at the RNA stability level, and in return mRNA modifications and translation, rather than the transcriptional level. Differential exon usage analysis revealed enrichment in cellular component morphogenesis and cell junction organization, which was in line with DEGs enriched pathways. However, further analysis of local splice variants (LSV) showed splice type specific gene enrichment patterns that may affect neuronal function via distinct LSV dependent mechanisms. Importantly, genes with retention intron, alternative 5 and 3 prime splicing site exclusively clustered in the translation and translation initiation pathway, implicating the potential effect of AS on translation following neuronal maturation. Splice motif analysis was able to reveal enriched motifs for RBPs that regulate various splice types and can be further correlated to distinct functions or expression of these RBPs in neuronal differentiation and maturation. This study systematically explores the transcriptomics, epigenetics, and potential post-transcriptomics during neuronal differentiation, indicating an important role of RNA stability and AS in regulating neuronal differentiation and neuronal function and behavior, which can be a new insight into the molecular mechanism of neuronal differentiation.

Disclosures: Y. Zhou: None. **S. Rashad:** None. **K. Niizuma:** None.

Poster

P007. Neuronal Differentiation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P007.05

Topic: A.01. Neurogenesis and Gliogenesis

Support: R01EY030458-02S1

Title: Dopaminergic interneuron diversity is restricted by the Kinase LKB1

Authors: *R. MACKIN, J. LIANG, C. BURGER, M. SAMUEL;
Baylor Col. of Med. Dept. of Neurosci., Houston, TX

Abstract: The mammalian central nervous system is comprised of thousands of distinct neuronal types, but even neurons within apparently uniform populations can display key functional differences. In particular, subtypes of dopamine neurons modulate circuits supporting motor function, cognition, and vision, and dysfunction or loss of these cells is associated with cognitive defects. Understanding what restricts the formation of closely related dopaminergic neurons may shed light on the unique susceptibility of subsets to disease. Using the murine retina, we show that the serine-threonine kinase LKB1 acts as a restrictive cue that limits dopaminergic neuron diversity. In wild type animals, dopaminergic amacrine interneurons are sparse, reside only in the inner nuclear layer, and are OFF laminating cells, responding to light decrements. Removing LKB1 from the retina markedly and specifically increases the numbers of dopaminergic amacrines (DACs) but not other amacrine cell types. Corresponding alterations in DAC cell body and synapse lamination accompany these changes. Subsets of newly generated DACs reside in the ganglion cell layer but retain amacrine cell markers and send neurites to the ON sublamina. DAC cellular and laminar alterations persist into adulthood. This process appears to be intrinsic to AC precursors, as removing LKB1 from AC fate specified cells but not from other neurons produces similar results. Together, these data suggest that a kinase cascade controlled by the serine-threonine LKB1 initiates neuron subtype specification and migration to restrict the development of diverse dopamine neuron subsets. Further, because newly generated ON DACs appear alike in many respects to their OFF laminating counterparts they may constitute a newly formed dopaminergic paramorphic pair.

Disclosures: R. Mackin: None. **J. Liang:** None. **C. Burger:** None. **M. Samuel:** None.

Poster

P007. Neuronal Differentiation

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Program #/Poster #: P007.06

Topic: A.01. Neurogenesis and Gliogenesis

Support: Fondecyt 1191152
NIH R01-HD097665
FRAXA Research Foundation
ANID 21161044

Title: Scanning the RCOR1 corepressor on active chromatin: Is it actually a silencer of neuronal genes?

Authors: C. A. RIVERA[1,2,3], H.-G. LEE[1,2], A. LAPPALA[1,2], V. NOCHES[3], M. OLIVARES-COSTA[3], M. SOJBERG[3], J. T. LEE[1,2], *M. E. ANDRES[3];
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Abstract: The transcriptional corepressor RCOR1 (CoREST) is recruited by the transcription factor REST to silence neuronal genes in non-neuronal tissues and neural stem cells. RCOR1 constitutes a chromatin-modifying complex as it allows the LSD1 and HDAC1/2 enzymes to erase transcriptionally-permissive histone modifications such as H3K4me2/1 and acetylation. We hypothesized that this complex must be actively acting in heterochromatin regions. Thus, we tested this hypothesis using bioinformatics, chromosome modeling, high-resolution microscopy, and biochemical studies in non-neuronal cells. Paradoxically, metagenomic analyses of RCOR1 chromatin occupancy showed exclusion of RCOR1 peaks from heterochromatin and enrichment at transcriptionally active chromatin of highly expressed genes. Furthermore, biochemical fractionations and high-resolution microscopy showed that RCOR1 was mainly enriched in euchromatin regions governed by histone acetylation. We also explored molecular mechanisms to characterize the role of RCOR1 in active regions by following its impact on transcriptional dynamics on short time scales, leading us to propose that RCOR1 has a novel and non-canonical role as a transcriptional dampener in euchromatin domains.

Disclosures: C.A. Rivera: None. **H. Lee:** None. **A. Lappala:** None. **V. Noches:** None. **M. Olivares-Costa:** None. **M. Sojberg:** None. **J.T. Lee:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); co-founder of Translate Bio and Fulcrum Therapeutics. F. Consulting Fees (e.g., advisory boards); Advisor to Skyhawk Therapeutics. **M.E. Andres:** None.

Poster

P007. Neuronal Differentiation

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Program #/Poster #: P007.07

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH Grant TL1TR001871
Schmidt Family Foundation
Chan Zuckerberg Biohub
UCSF Program for Breakthrough Biomedical Research

Title: Fate plasticity of interneuron specification

Authors: *M. A. MOSTAJO-RADJI[1], W. R. MANCIA LEON[2], A. BREEVOORT[2], L. ZHOU[2], M. T. SCHMITZ[2], Y. PEREZ[2], A. ELOR[1], S. TUMMALA[1], M. G. ANDREWS[2], D. TEJERA[2], M. TEODORESCU[1], S. KURNIAWAN[1], D. HAUSSLER[1], A. KRIEGSTEIN[2], A. ALVAREZ-BUYLLA[2], A. A. POLLEN[2];
[1]Univ. of California Santa Cruz, Santa Cruz, CA; [2]Univ. of California San Francisco, San Francisco, CA

Abstract: Competing genetic programs underlie the generation of neuronal subtypes of the mammalian central nervous system. To date, the extent to which extrinsic cues influence these programs remains unknown. Using medial ganglionic eminences (MGEs), which give rise to populations of interneurons (INs) of different developmental timelines, we modeled the contribution of the environment in shaping and maintaining neuronal identity. First, we transplanted mouse MGE progenitors into 2D and 3D mouse and human cortical, MGE and thalamic co-culture models, including dissociated cells, organoids, organotypic and conditioned cultures. We detected different proportions of somatostatin (SST) or parvalbumin (PV)-positive IN descendents across models. We discovered that transplantation into 3D models of human, but not mouse, corticogenesis leads to the accelerated and efficient differentiation of PV-positive INs in a nonautonomous manner. We observed additional aspects of PV-identity, including increased soma size, the upregulation of molecular markers of PV maturation, the repression of SST-specific markers and the establishment of perineuronal nets. Finally, transplantation of early postmitotic SST-positive interneurons into human cortical organoids reprograms the molecular signature of transplanted INs to PV. Altogether, our work supports a model of multipotent progenitors in the MGE, whose cell bias is defined and refined by the environment.

Disclosures: M.A. Mostajo-Radji: E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Provisional patent. **W.R. Mancia Leon:** None. **A. Breevoort:** None. **L. Zhou:** None. **M.T. Schmitz:** None. **Y. Perez:** None. **A. Elor:** None. **S. Tummala:** None. **M.G. Andrews:** None. **D. Tejera:** None. **M. Teodorescu:** None. **S. Kurniawan:** None. **D. Haussler:** None. **A. Kriegstein:** None. **A. Alvarez-Buylla:** None. **A.A. Pollen:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Provisional patent.

Poster

P007. Neuronal Differentiation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P007.08

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH R01MH124870
Ohio State University Fellowship

Title: The transcription factor Satb2 mediates differentiation and circuit integration of pyramidal neurons in CA1

Authors: *M. HANSON, P. NAGARAJAN, J. WESTER;
Neurosci., Ohio State Univ., Columbus, OH

Abstract: Hippocampal pyramidal cells (PCs) in CA1 are essential for learning and memory. These cells can be parsed into deep (near stratum oriens) and superficial (near stratum radiatum) sublayers, each with distinct morphologies and intrinsic membrane properties. Furthermore, PCs in CA1 form biased circuits with local inhibitory interneurons. In particular, deep cells receive stronger inhibitory postsynaptic currents (IPSCs) from parvalbumin-expressing (PV+) basket cells than superficial cells. The mechanisms that regulate the differentiation of PCs into deep or superficial subtypes are currently unknown. Here, we test the hypothesis that the transcription factor *Satb2* is critical for differentiation of PCs in CA1. *Satb2* is necessary for the specification of neocortical pyramidal cell types and is preferentially expressed in superficial PCs of the hippocampus. We used the Emx1-cre mouse line to conditionally knock out (cKO) *Satb2* from pyramidal cells during early development (E10). We then used whole-cell patch clamp recording in acute hippocampal slices of juvenile mice to investigate differences between deep and superficial PCs and how early loss of *Satb2* altered their developmental trajectories. Among superficial PCs, we found that conditional *Satb2* knockout resulted in an increase in voltage sag and concomitant depolarization of the resting membrane potential. This depolarization of superficial PC membrane potential significantly reduced the difference between deep and superficial cells observed in control mice. To test biases in PV+ basket cell input, we further crossed our mice to a PV-tdTomato reporter line and performed paired whole-cell recordings between identified deep and superficial pyramidal cells and PV+ basket cells. Strikingly, we found that loss of *Satb2* abolished the bias in PV+ basket cell input strength to deep versus superficial PCs, as the amplitude of IPSCs observed in superficial cells increased to match those recorded in deep cells. Our data show that *Satb2* is necessary for the differentiation of CA1 PCs and their integration into circuits with local inhibitory interneurons dependent on their subtype identity.

Disclosures: M. Hanson: None. **P. Nagarajan:** None. **J. Wester:** None.

Poster

P007. Neuronal Differentiation

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Program #/Poster #: P007.09

Topic: A.01. Neurogenesis and Gliogenesis

Support: The Pet Trust Fund
NSERC

Title: Activity-dependent neurogenesis and expression of microRNAs in human cerebral spheroids

Authors: *T. PARMENTIER[1], F. M. K. JAMES[2], E. HEWITSON[1], C. D. C. BAILEY[1], N. WERRY[1], S. D. SHERIDAN[4,5], R. H. PERLIS[4,5], T. G. KOCH[1], L. GAITERO[2], M. PERREAULT[1], J. LALONDE[3], J. LAMARRE[1];
[1]Biomed. Sci., [2]Clin. studies, [3]Mol. and Cell. Biol., Univ. of Guelph, Guelph, ON, Canada; [4]Ctr. for Quantitative Health, Ctr. for Genomic Med. and Dept. of Psychiatry, Massachusetts Gen. Hosp., Boston, MA; [5]Dept. of Psychiatry, Harvard Med. Sch., Boston, MA

Abstract: Neurogenesis is influenced by both internal and external cues during brain development and the addition of new neurons postnatally. These intrinsic and environmental factors shape neurogenesis partly through the modulation of local network activity, which in turn releases several neurotransmitters and growth factors. An important consequence of this activity-induced neurogenesis is illustrated in epilepsy where seizure-induced neurogenesis participates in the progression of epilepsy and the behavioural comorbidities. The lack of accessibility to the human brain at the cellular and molecular levels *ante mortem* impedes our understanding of the exact mechanisms of activity-induced neurogenesis, which could help identify mechanisms of epilepsy and potential targets for its treatment. Using cerebral spheroids derived from a human induced pluripotent stem cell line, we investigated the effect of increased neuronal activity on neurogenesis. We hypothesized that, when subjected to the pro-epileptic drug 4-aminopyridine (4AP), cerebral spheroids would demonstrate increased neurogenesis. Here, we show that 4AP increases neuronal activity but does not elicit seizure-like events in 3-month-old cerebral spheroids as measured with multi-electrode arrays, calcium imaging and expression of activity-dependent genes known to be dysregulated in human epileptic tissue. However, an increase in new neurons and a decrease in new glial cells formed after 4AP exposure is observed, suggesting that activity-induced neurogenesis can be recapitulated in human cerebral spheroids. We then explored a potential mechanism for this activity-induced neurogenesis by measuring the expression of several microRNAs that are involved in neurogenesis and dysregulated in epileptic tissue. We identified a decrease in the expression of miR-135a, a miRNA previously implicated in exercise-induced neurogenesis. MiR-135a is predicted to regulate the SMAD2/3 and BDNF pathways, and therefore represents a promising candidate for the regulation of activity-induced neurogenesis. Together, the results of this study suggest that cerebral spheroids are a suitable model to study some aspects of activity-induced neurogenesis, a phenomenon previously described in only the postnatal brain of humans and other animals.

Disclosures: T. Parmentier: None. **F.M.K. James:** None. **E. Hewitson:** None. **C.D.C. Bailey:** None. **N. Werry:** None. **S.D. Sheridan:** None. **R.H. Perlis:** None. **T.G. Koch:** None. **L. Gaitero:** None. **J. Lalonde:** None. **J. LaMarre:** None.

Poster

P007. Neuronal Differentiation

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Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P007.10

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH Grant 1R21MH125367

Title: Molecular and morphological heterogeneity of immature neurons within the human paralaminar amygdala

Authors: *C. E. PAGE[1], S. BIAGIOTTI[1], M. A. NASCIMENTO[2], S. F. SORRELLS[1]; [1]Dept. of Neurosci., Univ. of Pittsburgh, Pittsburgh, PA; [2]Neurolog. Surgery, Univ. of California San Francisco, San Francisco, CA

Abstract: The amygdala is central to processing emotional stimuli and volumetric changes in amygdala nuclei are hallmarks of both neurotypical development and neuropsychiatric disorders. In humans, the amygdala increases in total volume and neuron number during childhood. We recently characterized a novel form of neurodevelopment in the paralaminar nuclei (PL) of the amygdala where a population of immature neurons delay their maturation and may contribute to postnatal amygdala growth. These immature neurons are found at all ages (birth-77 years), though the majority develop into excitatory neurons during childhood and adolescence (birth-19 years). When immature, they are small, have compacted chromatin, and express the immature markers doublecortin (DCX) and polysialylated neural cell adhesion molecule (PSA-NCAM), which they stop expressing as they mature. During this investigation we also noticed diversity in morphology and marker expression among these cells. The majority of immature PL neurons are connected to large axonal projections which we mapped across the medial temporal lobe. In addition, a small subset are not connected to projections and instead have migratory morphology. As they mature, PL neurons express markers indicative of several different neuronal identities, suggesting additional heterogeneity in the types of neurons formed or possibly different stages of their maturation. We also re-analyzed single nuclei RNA-sequencing (snRNA-seq) data from amygdala samples at 4-15 years of age and find additional evidence of heterogeneity of the immature population. Within the cluster of immature neurons from these samples we further identify five distinct subpopulations. We conclude that, rather than being a homogenous population, some immature neurons in the human PL develop into projection neurons while others retain a migratory phenotype, and different subsets express distinct molecular markers.

Understanding the heterogeneous identities of these neurons is essential for gaining insight into their neurotypical function and potential alteration in neuropsychiatric disease.

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Poster

P007. Neuronal Differentiation

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Program #/Poster #: P007.11

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH-R21AG057974
NIH-R25NS080687
HRD-1906130

Title: Presence and possible function of miRNA-8 in the echinoderm radial nerve cord

Authors: *Y. MIRANDA-NEGRON[1], A. MORALES-RIVERA[2], E. QUESADA-DIAZ[2], R. GARCIA-ROSARIO[2], S. CRUZ-GONZALEZ[2], J. E. GARCIA-ARRARAS[3];
[1]Biol., Univ. de Puerto Rico, Rio Piedras, PR; [2]Univ. Of Puerto Rico, Rio Piedras, PR; [3]Dept Biol., Univ. Puerto Rico, Rio Piedras, PR

Abstract: For decades, scientist have employed extensive resources to elucidate important gene regulators involved in the process of nerve tissue regeneration. Regenerative competent organisms have been studied to learn how to overcome the regeneration limitations present in mammals. One of these organisms is our model system, the sea cucumber, *Holothuria glaberrima*. Here we explore the possible role of miRNA-8 as a molecular driver involved in the regeneration of the radial nerve cord (RNC) of this echinoderm. MicroRNA-8 (miRNA-8) has been shown to be responsible for cell proliferation, neuroblast transition and differentiation to neuron-like cells in the nervous system of various animal species. We now report the identification of an echinoderm miRNA-8 homolog stem-loop in transcriptomes of *H. glaberrima*. The sequence is highly similar to that of the sea urchin *S. purpuratus* showing 82% (61/74) identity. Moreover, a comparison of the holothurian miR-8 stem-loop with that of the human miR200a sequence showed 58% (54/93) identity and two possible mature miRNAs sequences with an identity of 77 % to miR-200a and 86% to miR200c of humans. Using RNA sequencing techniques and a novel *in vitro* protocol of RNC explants we show that miRNA-8 is differentially expressed in the holothurian RNC following injury. Changes in miRNA-8 take place during the period that corresponds to the dedifferentiation of radial glia-like cells, the initial process in the regeneration of the holothurian nervous system. Ongoing experiments are focused on the knockdown of miRNA-8 in the RNC explants as to determine its role during

regeneration. The effect of miRNA-8 depletion is being determined by cellular proliferation, dedifferentiation, and apoptosis analyses. Our results using this miRNA-RNAi technique will provide important technical and theoretical information on the role of miRNA-8 and toward explaining the amazing regenerative capacities of echinoderms.

Disclosures: Y. Miranda-Negron: None. **A. Morales-Rivera:** None. **E. Quesada-Diaz:** None. **R. Garcia-Rosario:** None. **S. Cruz-Gonzalez:** None. **J.E. Garcia-Arraras:** None.

Poster

P060. Animal Models of Developmental Disorders Other Than Autism II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P060.01

Topic: A.07. Developmental Disorders

Support: NIAAA Grant R21 AA026613-01
NIAAA Grant R01 AA027269-01

Title: Representation of mPFC axonal efferents in the thalamic nucleus reuniens in a rodent model of FASD

Authors: *I. SMITH, Z. GURSKY, A. KLINTSOVA;
Univ. of Delaware, Newark, DE

Abstract: Alcohol exposure (AE) during the prenatal period can result in fetal alcohol spectrum disorders (FASDs), a prevalent yet completely preventable developmental disorder. One of the prominent deficits of FASDs is impaired executive functioning (EF). Spatial working memory (SWM), an integral part of EF, relies on the coordination of activity between the medial prefrontal cortex (mPFC) and hippocampus (HPC) by the thalamic nucleus reuniens (Re), a structure that has been shown to be damaged following high-dose binge AE in a rodent model of third trimester alcohol exposure (Gursky, Savage, & Klintsova, 2019). Layer 5 mPFC neurons do not project directly to HPC, but rather communicate with the HPC via a disynaptic pathway where they first synapse with neurons in Re, and axons of those neurons go on to make contacts with CA1 neurons in HPC. This experiment investigated the effect of binge AE (5.25 g/kg/day, two doses 2 hours apart) during postnatal days 4-9 (PD4-9) on prefrontal axonal projections to Re in male and female Long Evans rat pups. Between PD 42-49, 400 nL of a fluorescent anterograde viral tracer (AAV5-CAG-tdT) was infused into the left mPFC of AE rats (n=10) and sham intubated (SI) rats (n=7). After three weeks survival on PD70, animals were perfused, and brains were extracted and postfixed followed by sectioning on cryostat in preparation for fluorescence microscopy. Hoechst33342 was applied to stain cell nuclei in order to locate areas of interest. Unbiased stereology was performed using the StereoInvestigator software (MBF Biosciences) to estimate injection volume, Re volume (Cavalieri probe), and cumulative length

of mPFC-originating axonal terminals within Re (Spaceballs probe). No significant main or interaction effects of sex were found, thus data were collapsed across sex for remaining analyses. Independent samples t-tests revealed significant main effects of postnatal treatment on Re volume [$t_{15} = 2.372$, $p = 0.0315$] and cumulative length of mPFC-originating axon terminals in Re [$t_{14} = 3.198$, $p = 0.0064$]. AE reduced the cumulative length of mPFC-originating axon terminals in Re, as well as overall Re volume. This ongoing experiment will help to better understand the structural basis for how AE effects mPFC-Re-HPC connectivity and subsequent EF deficits associated with FASDs. This study is supported by NIAAA1 R21 AA026613-01 and 1 R01 AA027269-01 to AYK.

Disclosures: I. Smith: None. **Z. Gursky:** None. **A. Klintsova:** None.

Poster

P060. Animal Models of Developmental Disorders Other Than Autism II

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Program #/Poster #: P060.02

Topic: A.07. Developmental Disorders

Support: COBRE: MRI Pilot Funding 2P20GM103653-06
NIH/NIAAA R01 AA027269

Title: A running start: histologically exploring the therapeutic potential of adolescent aerobic exercise to recover corpus callosum myelination after prenatal alcohol exposure

Authors: *E. K. BRENGEL, K. A. MILBOCKER, G. L. LEBLANC, A. Y. KLINTSOVA; Psychological & Brain Sci., Univ. of Delaware, Newark, DE

Abstract: Fetal Alcohol Spectrum Disorders (FASD), a group of related developmental disorders caused by prenatal alcohol exposure (AE), are reported in 1 of every 20 live births in the U.S. (May et al., 2014). Prenatal AE may result in a range of physical, cognitive, and behavioral symptoms including a deficit of executive function, for which the heavily myelinated interhemispheric fibers of the corpus callosum (CC) are critical. Myelination of the central nervous system occurs throughout life via the production and growth of oligodendrocytes. AE-induced disruption of CC myelination, which continues into adolescence, has been observed in clinical and preclinical studies (Newville et al., 2017; Gautam et al., 2014). Conversely, upregulation of CC myelination has been observed in response to aerobic exercise in adolescence (Tomlinson et al., 2018). This study utilizes immunohistochemistry (IHC) of brain tissue samples to investigate the effects of developmental AE and an adolescent wheel-running intervention (WR) on oligodendrocyte-lineage cell quantity in CC in a preclinical rodent model of FASD. Male and female Long-Evans rat pups received AE via intragastric intubation (5.25 g/kg/day) on postnatal days (PD) 4-9 during the brain growth spurt, a time of rapid

neurodevelopment that occurs during the third trimester of the human pregnancy. Sham-intubated controls underwent identical procedures without liquid administration. On PD 30-42, half of all rats were housed in cages with free access to WR; the other half remained socially housed in standard cages. Brains were perfused on PD42 with 4% paraformaldehyde and collected for IHC. Tissue was stained with CC1-Alexa Fluor594 antibodies (Millipore, OP-80; ThermoFisher, A21203) for quantification of mature oligodendrocytes (mOL) in 40 μm coronal sections using microscopic imaging and unbiased stereology (MBF SteroInvestigator). One-way ANOVA of preliminary data collapsed across treatment and sex groups demonstrates that there is no significant effect of WR intervention on mOL quantity in CC ($F_{1,16} = 0.045$, $p = 0.840$, n = 8-12). The sample will be expanded to eight groups ($n = 12$) to allow for analysis of WR, AE, and sex as independent variables. In addition, the CC oligodendrocyte precursor population, which is most immediately upregulated by aerobic exercise, is being quantified. Alongside IHC approaches, our lab obtained Diffusion Tensor Imaging scans before and after WR to assess changes to CC myelination longitudinally (submitted as a separate abstract). Taken together, these findings will help elucidate the mechanisms by which CC development is disrupted by AE and may be rescued by adolescent aerobic exercise.

Disclosures: E.K. Brengel: None. **K.A. Milbocker:** None. **G.L. LeBlanc:** None. **A.Y. Klintsova:** None.

Poster

P060. Animal Models of Developmental Disorders Other Than Autism II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P060.03

Topic: A.07. Developmental Disorders

Support: NIH/NIAAA R01AA027269-01

Title: Spatial working memory is impaired in adult male and female rats after binge alcohol exposure in a third - trimester model of FASD

Authors: *S. KIM, A. Y. KLINTSOVA, A. L. GRIFFIN;
Psychological and Brain Sci., Univ. of Delaware, Newark, DE

Abstract: Alcohol exposure (AE) during pregnancy is known to affect executive functioning later in life, including working memory (WM). In rodents, spatial WM is dependent upon the medial prefrontal cortex (mPFC) and hippocampus (HC). Previous work has demonstrated that these two structures are vulnerable to AE during the brain growth spurt, which occurs during 3rd trimester of human pregnancy and the first two postnatal weeks in rodents. Our lab has shown that thalamic nucleus reuniens (Re) is damaged in a rodent model of human 3rd trimester AE. We have also demonstrated that Re is critical for mPFC-HC oscillatory synchrony and spatial

working memory (SWM). The current study investigated the effects of early AE on delay dependent SWM ability in adulthood in male and female rats. We predicted that AE would lead to SWM impairment as WM demand increased.
Alcohol was administered on postnatal days 4-9 (PD4-9) as ethanol solution in milk formula in a dose 5.25 g/kg/day via intragastric intubations in a binge manner (AE group). Control animals were intubated without liquid (sham intubated (SI) group). Both male and female rats were included in the study. On PD90, rats were trained on a continuous alternation (CA) T-maze task. Upon reaching a choice accuracy criterion of 80% on CA for two consecutive days, we introduced increasing delays between trials (DA) in order to enhance SWM demand. The rats underwent 6 DA sessions, consisting of 12 each of 10-second (s), 30 s, and 60 s delay trials given in a pseudorandom sequence.
Our results show that male and female animals in both AE and SI groups learned the CA task at similar rates: no sex difference was observed. However, on the DA task, the SI group showed consistent improvement across sessions on the 30-s delay trials, whereas the AE group did not (postnatal treatment x day interaction: $F(5,195)=2.70$, $p = .02$). For the 10 s delay trials, both AE and SI groups showed improvement across days (main effect of day: $F(5,195)=7.93$, $p < .001$), whereas for the 60 s delay trials, both groups performed uniformly poorly across days. There was no main effect of sex ($F(1,37)=.029$, $p=.479$)
These findings support the hypothesis that binge AE during the brain growth spurt produces SWM deficits that depend on the level of SWM demand. This study will lead to a better understanding of the impact of HC-Re-mPFC circuitry damage due to developmental AE on cognitive function later in life.

Disclosures: S. Kim: None. **A.Y. Klintsova:** None. **A.L. Griffin:** None.

Poster

P060. Animal Models of Developmental Disorders Other Than Autism II

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Program #/Poster #: P060.04

Topic: A.07. Developmental Disorders

Support: 2P20GM103653-06
NIH/NIAAA R01 AA027269

Title: Running to Repair: Does aerobic exercise intervention in adolescence mitigate the effect of prenatal alcohol exposure on myelination?

Authors: *K. MILBOCKER[1], G. L. LEBLANC[1], E. K. BRENGEL[1], S. K. HEKMATYAR[2], C. F. FERRIS[3], A. Y. KLINTSOVA[1];
[1]Psychological and Brain Sci., [2]Ctr. for Biomed. and Brain Imaging, Univ. of Delaware, Newark,

DE; [3]Dept Psychology, Northeastern University, Ctr. for Translational NeuroImaging, Boston, MA

Abstract: 1 in 20 infants born annually in the U.S. has been exposed to alcohol prenatally which could produce cognitive deficits categorized as Fetal Alcohol Spectrum Disorders (FASD) (May et al., 2018). Cognitive processing impairments resulting from corpus callosum (CC) abnormalities including reduced axonal myelination are well-described in adolescents with FASD (i.e., Wozniak et al., 2019; Jacobson et al., 2017). Mammalian CC myelination, which begins in childhood and continues into adolescence, is imperative for optimizing axonal conduction and cognitive processing capacity. Diffusion tensor imaging scanning and immunocytochemical analysis were used to investigate the effects of: 1) prenatal AE and 2) exercise, a stimulator of myelinogenesis, in adolescence on CC myelination in a rodent model of FASD. Male and female Long-Evans pups were AE via intragastric intubation twice daily (5.25 g/kg/day) or served as sham-intubated controls during the rodent brain growth spurt (Dobbing & Sands, 1979). From PD 30-42, half of all rats had free access to running wheels for aerobic exercise while the second half remained in the homecage (n = 8-12, counterbalanced for sex/postnatal treatment group/intervention group). Scans were acquired twice longitudinally (PD 30 and 42) in all rats to assess alterations to CC myelination within-subject. Following scanning, tissue was collected for quantification of oligodendrocyte lineage cells and concentration of myelin basic protein in CC. Histological results will be presented at this meeting in the poster by Brengel et al. Longitudinal analysis of scan data shows that forebrain and CC volume increase between PD 30 and 42 ($F_{1, 75} = 29.0$, $p < .001$) and exercise stimulates forebrain growth. Cross-sectional analysis at each time point shows that AE significantly reduces CC volume compared to controls ($p < .01$). Since the effect of postnatal treatment on CC volume is mediated by forebrain volume, it is likely that AE and exercise intervention have opposing effects on CC growth across adolescence. Further, it was observed that radial diffusivity, interpreted as a noninvasive measure of myelination, decreases with age in CC ($p < .01$) as expected. However, radial diffusivity remains higher in AE rats compared to controls at both time points ($p < .01$), ultimately decreasing to pre-exercise (control) levels by PD 42. These findings confirm that the trajectory of CC myelination is delayed in cases of late term prenatal alcohol exposure. Adolescent exercise intervention supports whole and regional brain growth but has not been shown to mitigate alcohol-induced alterations to myelination using longitudinal neuroimaging.

Disclosures: K. Milbocker: None. **G.L. LeBlanc:** None. **E.K. Brengel:** None. **S.K. Hekmatyar:** None. **C.F. Ferris:** None. **A.Y. Klintsova:** None.

Poster

P060. Animal Models of Developmental Disorders Other Than Autism II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P060.05

Topic: A.07. Developmental Disorders

Support: 5R01MH119435-02
5R01MH122471-02

Title: The consequence of patient-derived anti-NMDA receptor autoantibodies in neural circuit development and subsequent behavior

Authors: *J. ZHOU[1], A. GREENFIELD[2], R. LOUDERMILK[2], C. BARTLEY[2], B. TRAN[2], H. WANG[2], M. WILSON[2], S. PLEASURE[2];
[1]Univ. of California San Francisco, San Francisco, CA, CA; [2]Univ. of California, San Francisco, San Francisco, CA, CA

Abstract: Anti-NMDA receptor autoantibodies cause severe behavioral deficits and prominent abnormal movements in pediatric patients, indicating disruption of sensory-motor circuits. A key structure for integrating sensory-motor information is the callosal connection between primary somatosensory cortices (S1). We have demonstrated that genetic disruption of NMDA receptor and antibody-mediated loss of function lead to defects in the somatosensory callosal projection in mice. We showed that disruption of axon guidance functions of the EPHRIN-B/EPHB pathway due to loss of NMDAR is the likely mechanism of this phenotype (Zhou et al., *eLife*, 2021). To further address whether exposure to patient-derived anti-NMDA receptor autoantibodies during callosal development disruptes callosal connectivity in S1, we generated anti-NMDA receptor monoclonal antibodies from the cerebrospinal fluid (CSF) of an anti-NMDA receptor encephalitis patient and identified two mAbs against two different subunits of NMDA receptor - mAb1 is against the NR2A subunit, mAb3 is against the NR1 subunit. Injections of either of these antibodies during callosal development in mice disrupts the ordered somatosensory callosal circuit. Injection of mAb1 caused small changes in the callosal projection but injection of mAb3 caused dramatically increased callosal innervation in the somatosensory cortex. Most interestingly, the disruption of a developmentally patterned circuit by the two patient-derived anti-NMDA receptor autoantibodies causes sensory-motor deficits in fine movement when these mice grew to adulthood. In particular, mAb3-treated mice show much worse performance in sensory-motor coordination tasks than mAb1-treated mice. This is the first demonstration that sensory-motor circuit deficits caused by patient-derived autoantibodies during development, cause subsequent sensory-motor behavior deficits in adulthood. This is relevant to the effects of this syndrome in pediatric and newborn patients and may also shed light on the protracted neurocognitive deficits experienced by anti-NMDA receptor encephalitis patients even after resolution of the acute phase of the illness.

Disclosures: J. Zhou: None. **A. Greenfield:** None. **R. Loudermilk:** None. **C. Bartley:** None. **B. Tran:** None. **H. Wang:** None. **M. Wilson:** None. **S. Pleasure:** None.

Poster

P060. Animal Models of Developmental Disorders Other Than Autism II

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Program #/Poster #: P060.06

Topic: A.07. Developmental Disorders

Support: Duke University SRP ES010356

Title: Neonicotinoid insecticides cause long-lasting neurobehavioral effects following embryonic exposure in zebrafish

Authors: *A. B. HAWKEY, Z. R. HOLLOWAY, E. D. LEVIN;
Duke Univ. Med. Ctr., Durham, NC

Abstract: Neonicotinoid compounds are commonly used insecticides which have become increasingly popular following growing concern for older generations of insecticides, such as organophosphates. As with other replacement compounds, these insecticides were introduced to provide an effective alternative with reduced health risks for non-target species. However, like nicotine and organophosphates, neonicotinoids exert their toxicity through well-conserved agonistic actions at nAChR. Given the established neurotoxicity of nicotine and other cholinergic toxicants, particularly during development, developmental neurotoxicity studies are needed to identify the nature and persistence of neonicotinoid toxicity in vertebrate species. The current study was performed to evaluate the neurobehavioral toxicity of two neonicotinoid insecticides using developing zebrafish as a model system. Fertilized zebrafish eggs were placed into glass petri dishes and exposed to vehicle (0.1% DMSO), clothianidin (CLO) (1-100uM) or dinotefuran (DIN) (1-10uM) from 5-120h post-fertilization. Neurobehavioral tests were conducted at larval (6 days), adolescent (10 weeks), and adult ages. Although these compounds produced no significant behavioral impairments in larval fish, phenotypic changes emerged over development. A low concentration of CLO (1uM) led to reduced anxiety-like diving in the novel tank test in adolescence, while the highest concentration (100uM) produced enhanced anxiety-like diving in adulthood. When tested in adulthood, developmental exposure to the highest concentration of CLO caused reduced locomotor activity in the novel tank overall, prior tap in the tap test, and in the predator avoidance assay, as well as increased avoidance of a fast predator cue. DIN exposure during early development produced no significant effects on behavior in adolescence. When the fish were tested in adulthood, the lower DIN concentration (1uM) caused enhanced diving responses in the novel tank, while both concentrations (1-10uM) caused reduced locomotor activity during the initial minutes of the predator avoidance test. These data suggest that neonicotinoid insecticides may carry some of the same risks for vertebrates posed by other classes of insecticides, and that these risks may include adverse neurobehavioral outcomes that emerge later in life. This work was supported by the Duke University Superfund Research Center (ES010356).

Disclosures: A.B. Hawkey: None. **Z.R. Holloway:** None. **E.D. Levin:** None.

Poster

P060. Animal Models of Developmental Disorders Other Than Autism II

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Program #/Poster #: P060.07

Topic: A.07. Developmental Disorders

Support: Translating Duke Health Neuroscience Initiative Grant
MH111684
DA047258
MH117429
NARSAD young investigator grant 25163
MH117851
GM007171

Title: Deciphering the neuronal role of the WASH complex and its influence on cognition and movement

Authors: *J. L. COURTLAND[1], T. W. A. BRADSHAW[2], G. WAITT[3], E. J. SODERBLOM[3], T. HO[3], A. RAJAB[6], R. VANCINI[4], I. KIM[7], S. H. SODERLING[5];
[1]Neurobiology, MSTP, [2]Neurobio., [3]Proteomics and Metabolomics Shared Resource, [4]Pathology, [5]Cell Biol., Duke Univ. Sch. of Med., Durham, NC; [6]Burjeel Hosp., VPS Healthcare, Muscat, Oman; [7]Anat. and Neurobio., Univ. of Tennessee Hlth. Sci. Ctr., Memphis, TN

Abstract: Multiple neurodevelopmental and neurodegenerative disorders are driven by disruptions in intracellular trafficking between organelles in neurons. One branch of this trafficking network, the endo-lysosomal pathway, is thought to tightly regulate many neuronal processes, including the dynamic recycling of synaptic receptors and the homeostatic regulation of membrane lipid composition. This idea is supported by numerous accounts of human neurological diseases that develop from the disruption of endo-lysosomal processes. In particular, mutations in subunits of the endosomal WASH complex, Strumpellin and SWIP, are implicated in motor and cognitive disorders, but the cellular etiologies of these associations are unknown.To explore the role of the WASH complex in neurons, we first sought to identify the molecular interactors of WASH in mouse brain. We utilized *in vivo* BioID (iBioID) to delineate the neuronal WASH complex proteome in wild-type mouse brain, revealing a rich network of endosomal proteins. Comparison of this network to previously reported WASH interactors in other cell types demonstrated the conservation of WASH function in endosomal trafficking within the mouse nervous system. Then, to uncover how dysfunction of endosomal SWIP leads to disease, we generated a novel mouse model of a human *WASHC4*$^{c.3056C>G}$ mutation. We performed quantitative spatial proteomics analysis of SWIP^{P1019R} mouse brain, which demonstrated that this mutation destabilizes the WASH complex and causes significant perturbations in both endosomal and lysosomal pathways. We then utilized cellular and histological analyses to confirm that SWIP^{P1019R} results in an endo-lysosomal disruption *in vitro*

and *in vivo*. We also found indications of neurodegeneration in SWIP^{P1019R} mice that appeared selective for the motor cortex. To determine if these structural changes produced functional effects in SWIP^{P1019R} brain, we conducted behavioral assays in wild-type and mutant SWIP^{P1019R} littermates at adolescence and adulthood. These studies revealed that SWIP^{P1019R} not only impacts cognition, but also causes significant progressive motor deficits in mice. By analyzing human SWIP^{P1019R} patient data, we found that this *WASHC4* mutation produces similar movement deficits in humans. Combined, our research provides the first molecular analysis of WASH complex function in the mammalian nervous system, and supports the model that WASH complex destabilization, resulting from SWIP^{P1019R}, drives cognitive and motor impairments via endo-lysosomal dysfunction in the brain.

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Poster

P060. Animal Models of Developmental Disorders Other Than Autism II

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Program #/Poster #: P060.08

Topic: A.07. Developmental Disorders

Support: Japan Society for the Promotion of Science
Hydrocephalus Association Innovator Award
the Center for Clinical and Translational Science and Training at the University of Cincinnati funded by National Institutes of Health (NIH) Clinical and Translational Service Award (CTSA) program under Award Number 2UL1TR001425-05A1
Rudi Schulte Research Institute

Title: The anti-inflammatory agent bindarit attenuates the impairment of neural development through suppression of microglial activation in a neonatal hydrocephalus mouse model

Authors: *E. IWASAWA[1], C. SHULA[1], F. N. BROWN[1], F. KAHN[1], S. LEE[3], T. BERTA[3], K. J. CAMPBELL[2], F. T. MANGANO[1], J. GOTO[1];
[1]Div. of Neurosurg., [2]Developmental Biol., Cincinnati Children's Hosp. Med. Ctr., Cincinnati, OH; [3]Anesthesiol., Univ. of Cincinnati, Cincinnati, OH

Abstract: Neonatal hydrocephalus presents with various degrees of neuroinflammation and long-term neurological deficits in surgically treated patients, provoking a need for additional medical treatment. We previously reported elevated neuroinflammation and severe

periventricular white matter damage in the *progressive hydrocephalus* (*prh*) mutant which is caused by a point mutation in the *Ccdc39* gene. These mutants exhibit ependymal cilia defects and altered cilia-mediated cerebrospinal fluid flow and develop into severe hydrocephalus within the first postnatal week. In this study, we demonstrate that suppression of microglial activity using the anti-inflammatory small molecule, bindarit, significantly inhibits white matter edematous injury and supports neural cell maturation of the early postnatal *prh* brain. Bindarit treatment inhibited nuclear localization of p65 in amoeboid-shaped *P2ry12+* microglia and upregulation of pro-inflammatory genes including *Ccl2, Ccl3, C3aR, Tyrobp, Trem2, Tlr4* and *Cd86*. While bindarit only mildly reduced the ventricular volume, it greatly improved microglial morphology, myelination, interneuron morphogenesis, and vascularization of the *prh* mutant. In addition, the treatment attenuated thinning in cortical layers II-IV and supported excitatory synapse formation in the *prh* mutants, while protecting the ramified-shaped homeostatic microglia from cell death in the developing cortex. These data implicate the active involvement of pro-inflammatory microglia in the developmental defects observed in neonatal hydrocephalic brains. Collectively, we revealed that suppressing microglial-induced inflammation is crucial for supporting white matter and neuronal maturation in neonatal hydrocephalus.

Disclosures: **E. Iwasawa:** A. Employment/Salary (full or part-time):; Japan Society for the Promotion of Science. **C. Shula:** None. **F.N. Brown:** None. **F. Kahn:** None. **S. Lee:** Other; The National Institutes of Health under the Award Number NS121946. **T. Berta:** Other; The National Institutes of Health under the Award Number NS121946. **K.J. Campbell:** None. **F.T. Mangano:** None. **J. Goto:** Other; Hydrocephalus Association, National Institutes of Health Clinical and Translational Service Award, Rudi Schulte Research Institute.

Poster

P060. Animal Models of Developmental Disorders Other Than Autism II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P060.09

Topic: B.04. Ion Channels

Support: T32 DA03980
R01 MH118934

Title: Fgf13 regulation of postnatal cortical development and function

Authors: *S. LIN[1], A. GADE[2], H. WANG[2], A. RAJADHYAKSHA[1], G. S. PITT[2];
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Abstract: Fibroblast growth factor homologous factor 13 (FGF13) is a non-canonical member of the fibroblast growth factor (FGF) superfamily that is not a secreted growth factor and functions intracellularly. Long-studied for its role in regulating voltage-gated sodium channels in brain and

heart, the full complement of FGF13 molecular functions is undefined. C**linical studies revealed that patients with disruptions or mutations in the *Fgf13* gene have early onset cognitive impairment and febrile seizures. FGF13 also has two major alternatively-spliced isoforms in the cortex, one which is predominantly expressed in excitatory and the other in inhibitory cells.** To study cell-type specific FGF13 expression in the cerebral cortex, I generated mice lacking FGF13 throughout CamkIIα-expressing glutamatergic or *Gad2*-expressing GABAergic interneurons. Mice heterozygous for *Fgf13* in interneurons suffer epileptic seizures, consistent with the human phenotype, and hemizygous knockout mice die perinatally. Mice with *Fgf13* deficiency in glutamatergic cells exhibit behavioral abnormalities but survive through adulthood. To further define the functional roles of FGF13, I conducted a battery of behavior assays to identify differences between knockout mice and wildtype littermates. My findings reveal distinct roles of *Fgf13* in the two cell types, likely mediated by distinct molecular mechanisms. **These data show that differential regulation of *Fgf13* via alternative splicing generates distinct proteins with different neuronal functions.**

Disclosures: S. Lin: None. **A. Gade:** None. **H. Wang:** None. **A. Rajadhyaksha:** None. **G.S. Pitt:** None.

Poster

P060. Animal Models of Developmental Disorders Other Than Autism II

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Program #/Poster #: P060.10

Topic: A.07. Developmental Disorders

Support: This study was approved by CONACYT (doctoral scholarship awarded to DACR 960415)

Title: Maternal restraint stress during pregnancy increases neonatal status epilepticus severity in rats

Authors: *D. A. CRUZ ROJAS[1], K. P. RAMOS RIERA[1], L. BELTRAN PARRAZAL[2], C. MORGADO VALLE[2], M. L. LOPEZ MERAZ[1];
[1]Inst. de Investigaciones Cerebrales, [2]Univ. Veracruzana, Xalapa, Mexico

Abstract: Evidence suggests that stress during pregnancy promotes acute and long-term consequences in offspring, which include molecular, cellular, and behavioral alterations. The goal of this study was to evaluate the effect of maternal restraint stress during pregnancy on neonatal *status epilepticus* (SE) in rats. Gestational stress was induced by immobilization of pregnant Wistar rats from gestation days 12 to 20 during two 2 h sessions, twice a day (stress rats, n = 4); control rats (n=4) were kept in standard housing conditions. The body weight and height of pups were evaluated from postnatal day (P) 3 to P7. In P7, SE was induced with the

lithium-pilocarpine model (LiCl 5mEq/kg, i.p.; pilocarpine hydrochloride 320mg/kg, s.c.). The behavioral evaluation was performed considering a modified scale to that described by Torolira et al. (2019): Stage 1 = motor hyperactivity, head, and forelimbs clonus; stage 2 = Clonic activity with loss of posture and rotational movements; stage 3 = sustained generalized seizures, running, and vocalization. The results showed that the neonates from stressed mothers (n = 31) showed a lower weight [males: 10.85 ± 0.80 g; females: 10.48 ± 0.82 g) and length [males: 6.55 ± 0.25 cm; females: 6.42 ± 0.30 cm] than those from control mothers (n = 20) [males: 13.94 ± 0.95 g and 7.22 ± 0.43 cm, respectively; females: 13.18 ± 1.05 g and 7.58 ± 0.54 cm, respectively]. Males born from stressed mothers (n=18) showed decreased latency to stage 1 (10.71 ± 1.63min), stage 2 (15.42 ± 1.90min) and stage 3 (15.77 ± 1.77min) seizures compared with those born from control mothers (n=14; 17.78 ± 1.80min, 35.58 ± 6.48min, 39.48 ± 10.22min, respectively). Males from stressed mothers also had a higher frequency of stage 1 (29.82 ± 5.05) and stage 3 (17.67 ± 3.72) seizures, and longer duration to stage 3 seizures (2.73 ± 044min) than control males (14.69 ± 4.23, 5.45 ± 1.77 and 1.32 ± 0.40min, respectively). Regarding female pups, those born from stressed mothers (n=13) showed decreased latency to stage 2 seizures (16.69 ± 2.94min) compared with control females (n=11; 30.81 ± 7.68min). In addition, female pups from stressed mothers showed increased number (33.54 ± 6.08) and duration of stage 2 seizures (0.40 ± 0.08min), as well as longer duration of stage 3 seizures (2.98 ± 0.76min) when compared with female control rats (12.45 ± 2.0, 0.15 ± 0.02min and 0.92 ± 0.35min, respectively). In conclusion, maternal restraint stress during pregnancy affects pup development and increases the severity of neonatal SE.

Disclosures: D.A. Cruz Rojas: None. **K.P. Ramos Riera:** None. **L. Beltran Parrazal:** None. **C. Morgado Valle:** None. **M.L. Lopez Meraz:** None.

Poster

P060. Animal Models of Developmental Disorders Other Than Autism II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P060.11

Topic: A.07. Developmental Disorders

Support: R01 NS113824 (Wang) NIH (NINDS) 9/2019~9/2024 Cortical development and pathogenesis in DEPDC5-related epilepsies
U54NS117170 (Project 3 lead) NIH (NINDS) 2020-2025 Epilepsy Multiplatform Variant Prediction (EpiMVP): Project 3: Cell Systems to Pre-Clinical Models

Title: Pv^+ interneurons are non-cell autonomously dysregulated in cerebral cortex with conditional knock-out of Depdc5 in pyramidal neurons.

Authors: *T. YANG, S. HU, M. KAO, W.-Z. CHANG, Y. WANG;
Univ. of Michigan, Ann Arbor, MI

Abstract: MTOR-related malformation of cortical development (MCD) is the most common cause of medically refractory epilepsy in children. It is still unclear which cell subtypes with MTOR hyperactivation contribute to the pathogenesis and epileptogenesis of MCD. Clinical and experimental data have suggested that MTOR hyperactivation in dorsal progenitors-derived cells, but not ventral progenitors derived interneurons, generates pathognomonic cellular changes, overall cortical overgrowth, and severe epilepsies. However, studies have also reported impaired inhibitory pre-synaptic transmission as well as the reduction of interneuron number in MTOR-associated MCD, raising a critical question how genetically intact interneurons are impaired. In this study, we used *Depdc5* conditional knockout mice lines to show that *Depdc5* deletion in cortical dorsal progenitor, but not in ventral progenitors, is necessary and sufficient to generates thickened cortex, cytomegalic neurons, and severe epileptic seizures. Astrocytes-specific deletion of *Depdc5* failed to produce these pathognomonic features of MTOR-related MCD. We further found that perineuronal nets (PNN) and parvalbumin (PV) + interneuron (IN) are impaired non-cell autonomously through microglial-mediated neuroinflammation in a response to the downregulated microglial inhibitory molecule, CD200, and the severely disrupted homeostasis in mutant cortical excitatory neurons. Therapeutically, treatment with Everolimus restored both cell-autonomous pyramidal neuron and non-cell autonomous PV+ IN impairments, and rescued severe seizures, suggesting critical roles of PV+ INs in the epileptogenesis of MTOR-related MCD.

Disclosures: T. Yang: None. **S. Hu:** None. **M. Kao:** None. **W. Chang:** None. **Y. Wang:** None.

Poster

P129. Inhibitory Neurons and Circuits

Location: Virtual Only

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Program #/Poster #: P129.01

Topic: B.08. Intrinsic Membrane Properties

Support: 1U19NS107616-02

Title: Brief synaptic inhibition persistently interrupts firing of fast-spiking interneurons

Authors: *S. CHAMBERLAND, M. HANANI, R. EGGER, E. R. NEBET, S. B. LARSEN, K. EYRING, R. W. TSIEN;
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Abstract: Input-output operations performed by neurons rely on their ability to integrate synaptic activity with their intrinsic electrical properties, a process thought to be constrained by the duration of synaptic events. At variance with this classical view, we find that the sustained firing of CA1 hippocampal fast-spiking (FS) parvalbumin-expressing interneurons (PV-INs) is persistently interrupted for several hundred milliseconds following brief GABAAR-mediated

inhibition. A single presynaptic neuron could interrupt PV-INs, occasionally with a single action potential (AP), and reliably with brief bursts of APs. Experiments and a computational model revealed that the persistent interruption of firing maintains neurons in a depolarized, quiescent state through a cell-autonomous mechanism dependent on an inactivating $K_v1.1$-mediated current and largely inactivated Na^+ channels. Strikingly, interrupted PV-INs are highly responsive to Schaffer collateral inputs, with subthreshold stimuli at rest becoming suprathreshold during the interruption. We show that the persistent interruption of firing represents a powerful disinhibitory mechanism that favors spike generation in CA1 pyramidal cells. Overall, our results demonstrate that neuronal silencing can far outlast brief synaptic inhibition owing to a well-tuned interplay between neurotransmitter release and postsynaptic membrane dynamics, thus impacting circuit function.

Disclosures: S. Chamberland: None. **M. Hanani:** None. **R. Egger:** None. **E.R. Nebet:** None. **S.B. Larsen:** None. **K. Eyring:** None. **R.W. Tsien:** None.

Poster

P129. Inhibitory Neurons and Circuits

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P129.02

Topic: B.08. Intrinsic Membrane Properties

Support: from the National Natural Science Foundation of China (81761128011), the Shanghai Municipal Science and Technology Major Project (No. 2018SHZDZX01) and ZJLab
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Title: Electrophysiological diversity of human pyramidal neurons and interneurons in L2/3 of the temporal lobe

Authors: *C. LUPASCU[1], M. HUANG[2], M. JIANG[3], Q. HE[4], Y. SHU[4], F. KAMP[1], Y. YU[2], M. MIGLIORE[1];
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Abstract: The electrophysiological properties of neurons strongly determine information processing in the networks they form. Usually these properties are extracted from in vitro recordings from animals, especially rodents. However, the ultimate goal is to understand how human neurons work. The problem is that, for obvious reasons, there are only rare recordings from human neurons. In this work, have studied the electrophysiological properties of pyramidal and fast spiking interneurons in Layers 2/3 of the human temporal lobe. Whole-cell patch clamp recordings were obtained from human cortical L2/3 neurons collected from 21 pyramidal cells from 7 males and 10 females and from 9 interneurons from 5 males and 4 females. A total of 491 traces were used for the analysis.
We first compared our data with the few electrophysiological features available from published datasets recorded from the same region (Deichter et al. 2017, Chameh et al. 2021, Berg et al. 2020 and Beaulieu-Laroche et al. 2018), and confirmed that they are consistent with those extracted from our traces. We then analysed several features, and we found statistically significant differences between males and female pyramidal neurons for the resting membrane potential (M: -68.530 ± 4.966 mV, F: -71.731 ± 3.017 mV, p = <0.001), the mean firing frequency (M: 28.743 ± 17.343 Hz, F: 39.246 ± 29.009 Hz, p=0.032), the instantaneous frequency of first ISI (M: 90.593 ± 48.082 Hz, F: 156.567 ± 108.828 Hz, p= <0.001) and the minimum voltage between spikes (M: -54.823 ± 4.036 mV, F: -51.637 ± 8.241 mV, p=0.004). For interneurons, features that significantly differ between males and females were the resting membrane potential (M: -65.806 ± 4.502 mV, F: -73.883 ± 4.562 mV, p = <0.001), the mean frequency (M: 87.386 ± 52.759 Hz, F: 58.523 ± 50.696 Hz, p=0.025) and the input resistance (M: 70.020 ± 55.934 MΩ, F: 185.999 ± 159.454 MΩ, p = <0.001). Females pyramidal neurons showed higher mean frequency and input resistance compared to males, whereas female interneurons showed lower mean frequency compared to males. Preliminary results also suggest that some of the features changes with age. These results are the first assessment of electrophysiological features of fast spiking interneurons of the human temporal cortex. They are being used to implement detailed biophysical models of L2/3 temporal cortex neurons and interneurons able to reproduce the intrinsic variability of excitability exhibited by these cells.

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Poster

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Program #/Poster #: P129.03

Topic: B.08. Intrinsic Membrane Properties

Support: R01MH106460
R01MH111601

Title: Diurnal variation in parvalbumin interneuron excitability in mouse prefrontal cortex

Authors: *J. BURNS[1], Y. H. HUANG[2], C. A. MCCLUNG[3];
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Abstract: Approximately twenty-four hour rhythms in behavior and physiology are widely conserved across the species and can be observed at the cellular and molecular level as changes in gene expression and cellular function. Previous studies in the prefrontal cortex (PFC) have shown that parvalbumin (PV) interneurons, which play a key role in regulating the excitation/inhibition balance and have been implicated in cognitive function, display diurnal changes in both PV expression and perineuronal nets (PNN). Given that studies have also found changes in PV cell excitability linked to PNN presence and overall diurnal changes in the excitation/inhibition balance have been observed in other cortical regions, we investigated diurnal changes in PV cell excitability, membrane properties, and the excitatory drive to these cells. Male and female adult G42 mice were sacrificed at ZT 5-6 or 17-18 (lights on ZT 0) and slices were prepared for whole cell patch electrophysiology. PV cells in the medial PFC were selected for recording. Excitatory drive was measured as spontaneous excitatory postsynaptic currents to PV cells in the presence of picrotoxin. To measure PV cell excitability and membrane properties, current pulses were injected via the recording electrode (500ms; -60 to 300pA); responses were averaged across 3 repeats. We find that PV cells show reduced excitability in the dark phase, with fewer action potentials per current injection as well as diurnal variation in membrane properties. These changes in PV cell excitability may have a profound impact on the overall excitation/inhibition balance of the PFC. Moreover, PV cells have been heavily implicated in psychiatric diseases and previous research in our lab has shown robust circadian reprogramming of the transcriptome in the PFC in schizophrenia. Therefore, through better understanding the normal diurnal changes in the physiology of these cells, we may gain insight into how these processes are disrupted in disease.

Disclosures: J. Burns: None. **Y.H. Huang:** None. **C.A. McClung:** None.

Poster

P129. Inhibitory Neurons and Circuits

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Program #/Poster #: P129.04

Topic: B.08. Intrinsic Membrane Properties

Support: NIH R56 AG072473
Emory ADRC
Autifony Therapuetics

Title: Biophysical alterations dampen excitability of cortical PV interneurons and contribute to network hyperexcitability in early Alzheimer's

Authors: *M. J. M. ROWAN[1], J. DIMIDSCHSTEIN[3], V. J. OLÁH[2], A. M. GOETTEMOELLER[2];
[1]Cell Biol., Emory Univ. Sch. of Med., Atlanta, GA; [2]Cell Biol., Emory Univ. Sch. of Med., ATLANTA, GA; [3]Broad Inst., Cambridge, MA

Abstract: AD is characterized by synaptic dysfunction and degradation, ultimately resulting in memory loss and dementia. The molecular and cellular mechanisms responsible for the initiation of this neurodegenerative progression remain poorly understood. A prevailing theory underlying the etiology of AD is that toxic, protein-rich accumulations result in neuronal death and eventual cognitive decline. However, increasing evidence from both human clinical trials and animal models suggests that these aggregates are not the causal determinates of disease initiation. Thus, uncovering alternative molecular and cellular mechanisms involved in the initiation of neurodegeneration is crucial. A guiding concept for this study is that circuit overexcitation may act as an initial determinant of future neurodegeneration in AD. Prior studies have uncovered that cortical overexcitation and intermittent epileptiform discharges emerge in the pre-plaque stages of AD. Multiple studies now suggest that this circuit overexcitation is due to changes in action potential (AP) firing of GABAergic inhibitory interneurons. 'Fast-spiking' parvalbumin (PV) interneurons appear to be particularly vulnerable to changes in AP firing in human APP (hAPP)-expressing mouse models. However, the molecular and biophysical mechanisms responsible for these changes remain largely unexplored. Here we used a novel, PV-specific AAV labeling technique, to perform targeted whole-cell patch clamp recordings in pre-plaque 5xFAD mice and controls. We found that the AP firing was severely dampened in cortical PV neurons from 5xFAD mice. PV-neuron specific qPCR found no changes in the mRNA levels of several Kv and Nav channels necessary for AP firing at this stage. However, outside-out patch clamp recordings uncovered significant alterations in the biophysical properties of Kv3 channels from 5xFAD mice. Using dynamic clamp and biophysical models, we found that the observed modifications in Kv3 channel activity could fully recapitulate changes in AP firing and waveform observed in 5xFAD mice. This study indicates that direct ion channel modulation precedes changes in gene expression, which is sufficient to modify AP firing in GABAergic interneurons in an early-stage hAPP model.

Disclosures: M.J.M. Rowan: None. **J. Dimidschstein:** None. **V.J. Oláh:** None. **A.M. Goettemoeller:** None.

Poster

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Program #/Poster #: P129.05

Topic: B.08. Intrinsic Membrane Properties

Support: NIH R56 AG072473
Emory ADRC

Title: Examining mechanisms of human APP expression on AP firing in cortical GABAergic interneurons

Authors: *A. M. GOETTEMOELLER, M. J. M. ROWAN;
Cell Biol., Emory Univ. Sch. of Med., Atlanta, GA

Abstract: Alzheimer's Disease (AD) causes a neurodegenerative cascade resulting in severe memory loss in elderly patients. To date, effective preventative interventions remain elusive. Current treatments often focus on amyloid plaques; however, plaque pathology occurs relatively late in the disease. New studies from AD patients and mouse models have identified cortical hyperexcitability prior to amyloid accumulation or plaque pathology. Hyperexcitability, which is associated with neuronal and synaptic pathology, is typically prevented by inhibition from local-circuit GABAergic interneurons. Interestingly, recent literature has shown that distinct interneuron subtypes are functionally impaired at this early phase in APP-expressing mouse models, including 'fast spiking' parvalbumin (PV) interneurons. Preliminary studies from our group also show altered PV interneuron firing in young 5xFAD mice before plaque formation. Together, this suggests that PV interneurons become dysfunctional independent of extracellular amyloid accumulation during AD. Here we utilized an AAV viral approach to express human APP (hAPP) and several of its downstream products in the cortex of adult wild-type or PV-Cre mice. Using ex-vivo patch-clamp electrophysiology, we observed altered firing phenotype in PV interneurons following short-term viral induction of WT hAPP, which mirrored findings from pre-plaque 5xFAD mice. These findings suggest that abnormal hAPP processing or expression, independent of extensive amyloid accumulation, represents an alternative cellular mechanism responsible for cortical circuit hyperexcitability observed in early AD.

Disclosures: A.M. Goettemoeller: None. **M.J.M. Rowan:** None.

Poster

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Program #/Poster #: P129.06

Topic: B.08. Intrinsic Membrane Properties

Support: RO1 MH100510
NSF GRFP

Title: Dysfunction of novel low-threshold spiking hippocampal interneuron in Fragile X syndrome

Authors: *L. T. HEWITT, G. J. ORDEMANN, D. H. BRAGER;
Inst. for Neurosci., The Univ. of Texas at Austin, Austin, TX

Abstract: Fragile X syndrome (FXS) is the most common inherited form of intellectual disability and the leading monogenetic cause of autism. FXS patients exhibit an increased incidence of sensory hypersensitivity, epilepsy, and anxiety in tandem with other cognitive impairments. Recent data suggests that dysfunctional inhibitory/excitatory balance in the brain may be a major contributor to the debilitating cognitive deficits in FXS. Previous studies, focused almost exclusively on excitatory pyramidal neurons, identified multiple impairments of voltage-gated ion channels in the hippocampus of the *fmr1* knockout mouse. Whether there are similar ion channel changes in hippocampal inhibitory interneurons remains largely unknown. Hippocampal interneurons can be classified based on firing pattern: fast spiking (FS) and low-threshold spiking. We recently showed that low-threshold spiking interneurons in the CA1 stratum oriens form two distinct groups based on interspike interval and subthreshold properties (Hewitt et al., 2021). Voltage clamp recordings revealed that the density of the hyperpolarization non-selective cation channel (I_h) was significantly different between the two populations. We refer to these two groups as oriens-lacunosum moleculare (OLM) and Low-Threshold High I_h (LTH) cells (Hewitt et al., 2021). We made whole-cell current clamp recordings from FS, OLM, and LTH neurons in hippocampal slices from adult wild type and *fmr1* knockout mice. We found no differences in subthreshold properties or action potential frequency for FS and OLM cells between wild type and *fmr1* KO mice. By contrast, resting membrane potential was depolarized and input resistance was lower in *fmr1* KO LTH cells. Additionally, action potential frequency was decreased in the *fmr1* KO compared to wild type LTH cells. Consistent with a potential difference in I_h, the effect of the h-channel blocker ZD7288 on input resistance was significantly larger in *fmr1* KO LTH cells compared to wild type. Given the diversity of interneuron function between subtypes, these results suggest aberrant properties of LTH cells may alter hippocampal function in FXS.

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Poster

P129. Inhibitory Neurons and Circuits

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Program #/Poster #: P129.07

Topic: B.08. Intrinsic Membrane Properties

Support: FRM DEQ20180839583

Title: Long lasting reduction of intrinsic excitability in O-LM interneurons is mediated by endocannabinoid dependent up-regulation of Kv7 channels

Authors: *S. INCONTRO, M. SAMMARI, *N. ANKRI, M. RUSSIER, J. FANTINI, D. DEBANNE;
Aix-Marseille Univ., Marseille, France

Abstract: KCNQ-Kv7 potassium channels are found at the axon initial segment of pyramidal neurons where they control cell firing and resting membrane potential. While Kv7 channel redistribution along the AIS has been demonstrated in homeostatic plasticity, their contribution to functional plasticity is still unclear. We show here for the first time that induction of long-term synaptic depression (LTD) in oriens lacunosum moleculare (O-LM) interneurons results in the synergistic expression of long-term depression of intrinsic excitability (LTD-IE) mediated by a gain of function of Kv7 channels. Both LTD and LTD-IE were found to involve endocannabinoid biosynthesis for their induction. Stimulation of CB1 receptor induced pre-synaptic LTD whereas direct interaction of endocannabinoids with post-synaptic Kv7.2/3 channels induced LTD-IE, thus showing the importance of lipids in the regulation of Kv7 channels. Using electrophysiology combined with pharmacological tools, cell biology, molecular modeling and microtensiometry, we demonstrate the direct activation of Kv7 channels by endocannabinoids. The long lasting reduction of the intrinsic excitability can have important consequences on the role of O-LM interneurons in the feedback inhibition and in the control of hippocampal network activity.

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Poster

P183. Molecular Mechanisms: Brain Aging, and Neurodegeneration II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P183.01

Topic: C.01. Brain Wellness and Aging

Support: NIH Grant R01AG066027

Title: Single-cell transcriptomics reveals region and cellularsubclass-specific changes with age in the mouse brain

Authors: *K. JIN, C. VAN VELTHOVEN, K. SMITH, J. GOLDY, N. DEE, B. TASIC, H. ZENG;
Allen Inst. For Brain Sci., Seattle, WA

Abstract: The mammalian brain is a highly heterogeneous tissue that is composed of a diverse collection of neuronal and non-neuronal cell types that vary in function and composition across different regions of the brain. Single-cell genomics methods are rapidly expanding our definition of cell types as well as our knowledge of their properties and functions. However, how these cell types change with age is not yet fully understood. Here we present a comparison between old and young adult mice via single cell transcriptomics. Our dataset contains 820,560 aged (18 month) and 364,644 young adult (2 month) cells analyzed by single-cell RNA sequencing on the 10x v3 Genomics platform. This dataset spans 16 different brain regions, including areas within the isocortex, hippocampal formation, cerebral nuclei, hypothalamus, midbrain, and hindbrain. Upon label transfer from previous RNA-seq datasets, we detect diverse cellular subclasses across all 16 regions including both neuronal and non-neuronal types. When equivalent subclasses are compared in young adult and aged animals for differentially expressed (DE) genes, we find more DE genes in non neuronal compared to neuronal cells. These non-neuronal subclasses include astrocytes, microglia, oligodendrocytes, vascular/leptomeningeal cells, endothelial cells, and smooth muscle cells. We find expected gene ontology (GO) terms enriched among DE genes in certain non-neuronal subclasses, including enrichment of adaptive immune response terms across microglial populations, and axon and myelin sheath related-terms across oligodendrocyte populations. We also find enrichment of a wide range of GO terms across different neuronal populations, including terms specific to synaptic structure, axonogenesis, mitochondrial function, respiration, and others. Overall, we observe that changes in the molecular landscape of the aged brain vary widely at the cellular subclass level, with the greatest number of genes and GO terms changing in non-neuronal types. As analysis of this tremendous dataset progresses, we will continue to uncover novel insights about specific cell types and regions of the mammalian brain.

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Poster

P183. Molecular Mechanisms: Brain Aging, and Neurodegeneration II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P183.02

Topic: C.01. Brain Wellness and Aging

Support: BT/PR17550/MED/30/1755/2016

Title: Association of Single nucleotide polymorphisms ARMS2-rs 10490924, RAD51B-rs8017304 and VEGF-rs 699947 and their protein levels in Age Related Macular Degeneration.

Authors: *P. BATTU, K. SHARMA, M. RAIN, R. SINGH, A. ANAND;
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Abstract: Association of Single nucleotide polymorphisms ARMS2-rs 10490924, RAD51B-rs8017304 and VEGF-rs 699947 and their protein levels in Age Related Macular Degeneration. Priya Battu[a], Kaushal Sharma[a], Manjari Rain[a], Ramandeep Singh[b], Akshay Anand[a#]a.Neuroscience Research Lab, Department of Neurology, Post Graduate Institute of Medical Education and Research, Chandigarhb.Advanced Eye Centre, Post Graduate Institute of Medical Education and Research, ChandigarhAge-related macular degeneration is a neurodegenerative retinal disease affecting the elderly worldwide. Single nucleotide polymorphisms have been implicated in this disease, but studies in the context of these SNPs in the Indian population are lacking. Also, how protein levels of the studied genes vary with the genotype remains elusive in AMD. In this study, we have investigated whether SNPs in ARMS2-rs 10490924, RAD51B-rs8017304 and VEGF-rs 699947 and their protein levels are associated with AMD in the Indian population. A case-control study consisting of 265 AMD patients and 78 unaffected was conducted at Advanced Eye centre PGIMER, Chandigarh. ELISA was used for the estimation of proteins in the serum of the participants. Taqman genotyping assay was used for SNP genotyping of the samples. We found that the TT variant of the ARMS2 genotype was significantly associated with AMD (p=.001). But for the other two genes, we did not find any significant differences. Also, the levels of ARMS2 and VEGF were increased in AMD compared to controls.Whereas, RAD51B levels were decreased in AMD as compared to controls. Hence, we conclude that TT is a risk variant for AMD in our studied population and varying levels of these proteins among the cases and controls show that these proteins might be implicated in the pathophysiology of AMD.
Conflict of interest: NoneEthical Statement: The study was conducted after taking approval from Institute ethical committee, PGIMER, Chandigarh, India.
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Disclosures: P. Battu: None. **K. Sharma:** None. **M. Rain:** None. **R. Singh:** None. **A. Anand:** None.

Poster

P183. Molecular Mechanisms: Brain Aging, and Neurodegeneration II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P183.03

Topic: C.01. Brain Wellness and Aging

Support: Institution startup

Title: Histone H2A ubiquitination resulting from Brap loss of function connects multiple aging hallmarks and accelerates neurodegeneration

Authors: *Y. FENG[1], C. LOWE[1], Y. GUO[2], A. A. CHOMIAK[2], Y. HONG[3], E. S. MONUKI[4]; [1]Uniform Services Univ. of the House Sci., Bethesda, MD; [2]Northwestern Univ., Chicago, IL; [3]Univ. of Turku, Turku, Finland; [4]UC Irvine Sch. of Med., Irvine, CA

Abstract: Aging is an intricate process that is characterized by multiple hallmarks including stem cell exhaustion, genome instability, epigenome alteration, impaired proteostasis, and cellular senescence. While each of these traits is detrimental at the cellular level, it remains unclear how they are interconnected to cause systemic organ deterioration. Here we show that abrogating Brap, a BRCA1 associated protein important for neurodevelopment, results in cellular senescence with persistent DNA double-strand breaks and elevation of histone H2A mono- and poly-ubiquitination (H2Aub). The high H2Aub initiates histone proteolysis, leading to both epigenetic alteration and proteasome overflow. When these changes occur in mice carrying Brap deletions in the brain, they promote neuroinflammation, impair proteostasis, accelerate neurodegeneration, and shorten lifespan. We further show H2Aub is also increased in human brain tissues of Alzheimer's disease. These data together suggest that chromatin aberrations mediated by H2Aub act as a nexus of multiple aging hallmarks.

Disclosures: Y. Feng: None. **C. Lowe:** None. **Y. Guo:** None. **A.A. Chomiak:** None. **Y. Hong:** None. **E.S. Monuki:** None.

Poster

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Topic: C.01. Brain Wellness and Aging

Support: NIA AG057558
NIA AG051807
NIA AG054349
NIH R25 GM 55246-24

Title: Age-related DNA methylation signatures in the dorsal hippocampus of male and female mice

Authors: *C. A. CHINN[1], H. REN[2], J. MORIVAL[2], Q. NIE[2], M. A. WOOD[3], T. DOWNING[2]; [1]Neurobio. and Behavior, [2]UC Irvine, Irvine, CA; [3]Neurobio. and Behavior, Univ. of California Irvine, Irvine, CA

Abstract: DNA methylation is a well-characterized epigenetic modification involved in numerous molecular and cellular functions. Methylation patterns have also been associated with aging mechanisms. However, how DNA methylation patterns change within key brain regions involved in memory formation in an age- and sex-specific manner remains unclear. Here, we performed reduced representation bisulfite sequencing (RRBS) from mouse dorsal hippocampus - which is necessary for the formation and consolidation of specific types of memories - in young and aging mice of both sexes. Overall, our findings demonstrate that methylation levels within the dorsal hippocampus are divergent between sexes during aging in genomic features correlating to mRNA functionality, (particularly in 3'UTRs) and in gene regulatory elements. In transcription factor binding site motifs, we found hypomethylated regions to have a neural activity related signature (AP-1, FOS/JUN), in contrast to hypermethylated regions, which contained transcription factors significant during developmental processes. These results define age-related changes in the methylome across genomic features and build a foundation for investigating potential target genes regulated by DNA methylation in an age- and sex-specific manner.

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Poster

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Topic: C.01. Brain Wellness and Aging

Support: T32 AG00096
T34GM136498
5R01MH101491-05

Title: Crest: a keystone regulator in nucleosome remodeling and histone acetylation in memory formation and aging

Authors: *F. GARCIA, R. G. SERRANO, E. KRAMER, M. WOOD;
Univ. of California, Irvine, Irvine, CA

Abstract: One hypothesis proposed to underlie age-dependent cognitive decline and memory formation impairment is that the epigenome becomes altered with age, leading to alterations in gene expression required for long-term memory processes. Recent studies have highlighted a transcription factor, CREST (Calcium RESponsive Transactivator), as a keystone coordinator of two different epigenetic mechanisms, histone acetylation and nucleosome remodeling. CREST has two distinct domains, one of which binds CBP (CREB-Binding Protein, a potent histone

acetyltransferase) and the other BRG1 (Brahma related gene 1, an ATPase and main enzymatic component of BAF nucleosome remodeling complex), leading to the coordination of histone acetylation and nucleosome remodeling. Surprisingly, there is almost nothing known about the role of CREST in synaptic plasticity and memory. Mutations in the human *Crest* gene give rise to Amyotrophic lateral sclerosis (ALS; aka Lou Gehrig's disease), which is a progressive nervous system disease. My research will test the hypothesis that CREST is essential for adult- and age-dependent memory formation, synaptic plasticity, and activity-dependent gene regulation. To test this hypothesis, we have generated both *Crest*-FLOX and *Crest* heterozygous knockout genetically modified mice, as well as adeno-associated viruses (AAVs) that express wildtype CREST, or mutants of CREST. We characterized hippocampal long-term potentiation (LTP, a form of synaptic plasticity), long-term memory, and gene expression during the consolidation phase of memory formation. In addition, using a computational screening of motifs in CREST peptide, we found a candidate phospho-Tyrosine conserved in mouse and human mapping to the proximity of residues necessary for CREST-CBP interaction, which may provide insight into how CREST is regulated. Lastly, using RNA seq data generated by the U-54 MODEL-AD initiative here at UCI, we found that *Crest* mRNA expression is progressively decreasing over the span of of the aging mouse hippocampus, suggesting a potential age-dependent mechanism by which reduced CREST expression leads to memory dysfunction. Together, completion of these studies will determine whether CREST regulates synaptic plasticity and memory formation and age-related cognitive decline along with a means of understanding how CREST may be responding to activity-dependent regulation via the coordination of histone acetylation and nucleosome remodeling.

Disclosures: F. Garcia: None. **R.G. Serrano:** None. **E. Kramer:** None. **M. Wood:** None.

Poster

P183. Molecular Mechanisms: Brain Aging, and Neurodegeneration II

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Program #/Poster #: P183.06

Topic: C.01. Brain Wellness and Aging

Support: NIH R01 AGO33605

Title: 17b-estradiol regulates mir-9-5p and mir-9-3p stability and function in the aged female rat brain.

Authors: *M. LINSCOTT[1], C. KIM[1], S. FLURY[1], M. ZHANG[1], T. R. PAK[2];
[1]Loyola Univ., Chicago, IL; [2]Dept Cell and Mol. Physiol., Loyola Univ. Chicago Stritch Sch. of Med., Maywood, IL

Abstract: The “timing hypothesis” speculates that the ovarian hormone 17β-estradiol (E_2) is neuroprotective within a narrow window of time following menopause, suggesting that there is a biological switch in E_2 action that is temporally dependent. Our previously published work suggests that microRNAs (miRNAs), a class of small non-coding RNAs, can post-transcriptionally regulate many mRNA targets and therefore could underlie this temporal switch in E_2 action in the aging female brain. Specifically, we found that the ability of E_2 to regulate miRNAs in aged rats was dependent on the length of time between ovarian hormone depletion (via ovariectomy) and the subsequent timing of replacement E_2 treatment. However, while E_2 treatment upregulated levels of mature miR-9-5p and miR-9-3p in the rat hypothalamus, the primary and precursor levels remained unchanged, suggesting that this upregulation was not mediated by an increase in transcription. Here, we focused on mir9-5p and mir-9-3p, miRNAs derived from the same primary transcripts which our previous work found to be differentially regulated by E_2 in the hypothalamus and hippocampus of aged female rats. Therefore, we hypothesized that E_2 maintains miR-9-5p and -3p transcript levels by altering either their stability or activity. To study the stability of miRNA in aged female brain, we developed biochemical assays in cell lines and in rat brain tissue lysate. Interestingly, we found that E_2 treatment stabilized miR-9-3p, but not miR-9-5p, in the rat hypothalamus and this stabilization was dependent on age of treatment. Through polysome fractionation experiments, we found that E_2 was able to shift polysome loading of miRNAs, which repressed translation of the predicted mRNA targets. Moreover, miR-9-5p and -3p transcripts appeared to occupy different fractions of the polysome profile, indicating differential subcellular localization. Together, these studies demonstrate the significance of a miRNA-mediated switch that coordinates the neuroprotective effects of E_2 in the aged female brain.

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Poster

P183. Molecular Mechanisms: Brain Aging, and Neurodegeneration II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P183.07

Topic: C.01. Brain Wellness and Aging

Support: NIH RO1 AG033605

Title: Absolute quantification and functional analysis of hippocampal phosphorylated ERβ amino acid residues in a menopause model.

Authors: *S. FLURY[1], M. ZHANG[1], C. KIM[1], W. C. CHUNG[2], J. KIRK[1], T. R. PAK[1]; [1]Loyola Univ. Chicago, Chicago, IL; [2]Biol. Sci., Kent State Univ., Kent, OH

Abstract: Menopause, characterized by a depletion of circulating 17β-estradiol (E2), can coincide with serious symptoms, particularly neurological. This can range from memory problems to mood alterations, such as an increase in anxiety and depression. Hormone therapy (HT) can be used to alleviate the symptoms of menopause. However, the time post menopause in which HT is implemented greatly alters efficacy of the treatment. This clinical observation raised the possibility that estrogen receptor (ER) signaling could be altered through changes in the receptor itself following long periods of estradiol deprivation and/or increasing age. ERβ is known to mediate E2 effects on anxiety, therefore we hypothesized that age and length of E2 deprivation differentially alter phosphorylated ERβ in the brain leading to changes in ERβ function. In this study we used absolute quantification mass spectrometry (AQUA-MS) to quantify the levels of phosphorylation at four ERβ residues; S105, S176, S200 and Y488. AQUA-MS quantification was performed using human hippocampal samples (21-83 years old) as well as hippocampal samples from our rat model of menopause. We observed that absolute phosphorylation levels were similar at all of the sites in human and rat models. Specifically, S105 was highly phosphorylated regardless of E2 treatment or age; S176 phosphorylation was decreased by age alone; S200 was lowly phosphorylated and further decreased by age in humans and E2 deprivation in rats, respectively, and Y488 levels were moderate and unaltered by age or E2. To address the functional consequences of ERβ phosphorylation, we used electrophoretic mobility shift assays (EMSA) and dual luciferase assays in combination with phospho-mimetic and phospho-nulls for each residue. The EMSA results showed that the phosphorylation status of ERβ S176 and S200 greatly inhibited receptor binding to DNA. Moreover, ERβ function at minimal promoters was not always consistent with what we predicted from the DNA binding, suggesting that the proteins recruited to ERβ upon DNA binding are altered based on phosphorylation status. To our knowledge, this is the first study to map absolute quantification of phosphorylated ERβ at specific amino acid residues in the brain. Further, we demonstrate changes across the lifespan and propose functional changes in ERβ signaling that could impact the success of HT in postmenopausal women.

Disclosures: S. Flury: None. **M. Zhang:** None. **C. Kim:** None. **W.C. Chung:** None. **J. Kirk:** None. **T.R. Pak:** None.

Poster

P183. Molecular Mechanisms: Brain Aging, and Neurodegeneration II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P183.08

Topic: C.01. Brain Wellness and Aging

Support: CNU Startup Funds
CNU Summer Scholars Student Grants
CNU Independent Research Student Grants

Title: Non-cell-autonomous intestinal occluding junction modulation in aging and disease

Authors: C. SESSIONS, K. PAULEY, ***A. SALAZAR**;
Christopher Newport Univ., Newport News, VA

Abstract: Aging is a process marked by a continuous decline in multiple physiological functions. Intestinal barrier function, for example, is tightly linked to longevity in *Drosophila melanogaster* and other organisms. We have previously shown that altered expression of occluding junctions in the guts of fruit flies can lead to various hallmarks of aging, including modulation of intestinal homeostasis, variations in microbial dynamics, changes in immune activity, and alterations in lifespan. Loss of a specific occluding junction, Snakeskin (Ssk), leads to rapid and reversible intestinal barrier dysfunction, altered gut morphology, dysbiosis, and a dramatically reduced lifespan. Remarkably, restoration of Ssk expression in flies showing intestinal barrier dysfunction rescues each of these phenotypes previously linked to aging. Intestinal up-regulation of Ssk protects against microbial translocation following oral infection with pathogenic bacteria. Furthermore, intestinal up-regulation of Ssk improves intestinal barrier function during aging, limits dysbiosis, and extends lifespan. Additionally, perturbing barrier function in the gut has non-cell-autonomous impacts, including alterations in the brain and muscle. Moreover, these analyses add more information about the impact of the gut on tissue outside the gut and begin to address communication between the gut and the brain and muscles in disease models. These findings indicate that intestinal occluding junctions may represent prolongevity targets in mammals, in addition to their possible roles in intestinal dysfunction, aging, and disease.

Disclosures: C. Sessions: None. **K. Pauley:** None. **A. Salazar:** None.

Poster

P183. Molecular Mechanisms: Brain Aging, and Neurodegeneration II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P183.09

Topic: C.01. Brain Wellness and Aging

Support: Wellcome Trust

Title: Maintenance of axonal morphology during aging is regulated by the microtubule cytoskeleton

Authors: *R. GOSLING, N. SANCHEZ-SORIANO;
Inst. of Systems, Mol. and Integrative Biol., Univ. of Liverpool, Liverpool, United Kingdom

Abstract: Axons are cable-like projections of neurons that electrically wire the nervous system. Although microscopic in diameter, these slender structures can extend up to a metre long and

often need to be maintained throughout the lifetime of an organism. This makes them particularly vulnerable to the pathological processes of ageing and neurodegenerative disease (ND), yet the mechanisms driving these are not fully understood. Axon morphology, structure and physiology are reliant on densely packed parallel bundles of microtubules (MT) within the axonal core. Not surprisingly, dysfunctional MT networks are strongly linked with neurodegeneration. In addition, deterioration of axonal MT bundles has been observed in the ageing primate brain. All these cumulative findings have prompted us to propose that aberrant MT regulation during ageing may drive age-related neuronal deterioration. We use *Drosophila melanogaster* to investigate MT regulation in mature axons, and to understand how this process changes during ageing. We have identified an array of MT regulators necessary for the maintenance of MT bundles in axons and synapses from mature neurons within the adult brain. Excitingly, their manipulation leads to structural axonal and synaptic defects which are characteristic of the ageing brain. Moreover, we find that the activity of several MT regulators decreases with age and that boosting some of these factors not only improves MT decay but also axonal and synaptic atrophy during ageing, thus suggesting that aberrant MT regulation is a key factor in age-dependent neuronal decay.

Disclosures: R. Gosling: None. **N. Sanchez-Soriano:** None.

Poster

P211. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P211.01

Topic:

Support: Edson Foundation Seed Grant
NIH R01 AG062500
NIH R01 AG059627

Title: Glyphosate crosses the blood-brain barrier; A driver of neuroinflammation and a potential risk factor for Alzheimer's Disease

Authors: *J. K. WINSTONE[1,2], K. V. PATHAK[3], R. SHARMA[3], M. DONNAY[1], J. WHITE[1], M. J. HUENTELMAN[4], P. PIRROTTE[3], R. VELAZQUEZ, JR[1,2];
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Abstract: The prevalence of Alzheimer’s Disease (AD) is rapidly increasing in the US with an expected 14.8 million cases by 2050. Evidence suggests that risk factors for AD include both heritable and non-heritable environmental components. Recently, pesticides have gained much

attention as environmental toxins due to the potential hazard they pose to human health. Glyphosate, the active ingredient in the broad-spectrum herbicide Roundup, is the most heavily applied pesticide worldwide. The recent rise in glyphosate application to corn and soy crops has a strong, positive correlation with increased death rates due to AD and other dementias. Notably, glyphosate has been shown to cross the blood-brain barrier (BBB) *in vitro* but has yet to be verified *in vivo.* Additionally, its potential detrimental effects on AD-related pathologies remain elusive. Here, we examined whether glyphosate crosses the BBB in both male and female 4-month-old C57/bl6 laboratory mice. Mice received either 125, 250, 500 mg/kg/day of glyphosate or a vehicle control via oral gavage for 14 days. Post-mortem brain tissue analysis revealed that glyphosate infiltrated brain tissue in a dose dependent manner. We then analyzed brain expression levels of tumor necrosis factor α (TNFα), a key inflammatory cytokine that is elevated in peripheral tissue following glyphosate exposure. Notably, TNFα has been shown to induce cell death and exacerbate amyloid-β plaque burden in the brains of mouse models and patients with AD. Our data shows a significant upregulation of TNFα in brain tissue post glyphosate exposure. Future work will focus on examining whether glyphosate exposure exacerbates cognitive deficits and associated AD neuropathology in mouse models of AD. Thus far, this data highlights the need for further exploration into the long-term consequences of glyphosate exposure in relation to AD and has broad implications regarding the health of the general population.

Disclosures: J.K. Winstone: None. **K.V. Pathak:** None. **R. Sharma:** None. **M. Donnay:** None. **J. White:** None. **M.J. Huentelman:** None. **P. Pirrotte:** None. **R. Velazquez:** None.

Poster

P211. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P211.02

Topic: C.05. Tauopathies, Tau-dementias, and Prion Diseases

Support: NIA Grant R01AG062500
NIA Grant R01AG059627
NINDS Funding U24NS072026
NIA Grant P30 AG19610
Arizona Department of Health Services (Contract 211002, Arizona Alzheimer's Research Center)
Arizona Biomedical Research Commission (Contracts 4001, 0011, 05-901 and 1001 to the Arizona Parkinson's Disease Consortium
Michael J. Fox Foundation for Parkinson's Research

Title: Identification of retinoblastoma binding protein 7 (Rbbp7) as a mediator against tau acetylation and subsequent neuronal loss in Alzheimer's disease and related tauopathies

Authors: *N. DAVE[1], A. S. VURAL[1], I. S. PIRAS[2], W. WINSLOW[1], L. SURENDRA[1], J. K. WINSTONE[1], T. G. BEACH[3], M. J. HUENTELMAN[2], R. VELAZQUEZ[1];
[1]Neurodegenerative Dis. Res. Ctr., Biodesign Inst. At Arizona State Univ., Tempe, AZ; [2]Neurogenomics, Translational Genomics Res. Inst., Phoenix, AZ; [3]Civin Lab. for Neuropathology, Banner Sun Hlth. Res. Inst., Sun City, AZ

Abstract: Tau hyperphosphorylation and neurofibrillary tangle formation contribute to the extensive neuronal death in Alzheimer's disease (AD) and related tauopathies. Recent work has identified that tau acetylation promotes tau phosphorylation. Tau acetylation at lysine 280 (K280) has been shown to be a prominent site resulting from increased expression of the lysine acetyltransferase p300. The exact upstream mechanisms mediating p300 expression remain elusive. Additional work suggests that dysregulation of epigenetic proteins may contribute to acetylation and hyperphosphorylation of tau. Here we identified Retinoblastoma-binding Protein 7 (Rbbp7), the histone-binding subunit of the Nucleosome Remodeling and Deacetylase (NuRD) complex, as an upstream mediator of tau acetylation. Rbbp7 chaperones chromatin remodeling proteins to their nuclear histone substrates, including histone acetylases and deacetylases. Rbbp7 binds to p300, suggesting a role in modulating tau acetylation. We interrogated Rbbp7 in post-mortem brain tissue, cell lines and mouse models of AD. We found reduced Rbbp7 mRNA expression in AD cases, a significant negative correlation with CERAD (neuritic plaque density) and Braak Staging (pathogenic tau inclusions) and a significant positive correlation with post-mortem brain weight. We also found a neuron-specific downregulation of Rbbp7 mRNA in AD patients. Rbbp7 protein levels were significantly decreased in 3xTg-AD (12 month) and PS19 (8.5 month) mice compared to respective NonTgs, but no decreases were found in APP/PS1 (12 month) mice lacking tau pathology. Interestingly, tau acetylated at lysine K280 was observed in 2-month-old PS19 mice, preceding tau phosphorylation; significant reduction of Rbbp7 in PS19 mice compared to NonTg also occurs at 2 months of age, corresponding with early changes in tau acetylation. Rbbp7 deficiency, tau acetylation at K280 and hyperphosphorylation of tau at Ser202/Thr205 (AT8) were evident in the older PS19 mice. This suggests that Rbbp7 downregulation and tau acetylation at lysine K280 may be early events in tau pathogenesis. In vitro, Rbbp7 overexpression rescued TauP301L-induced cytotoxicity in immortalized hippocampal cells and primary cortical neurons. In vivo, hippocampal Rbbp7 overexpression rescued neuronal death in the CA1 of PS19 mice. Mechanistically, we found that increased Rbbp7 reduced p300 abundance, tau acetylation at lysine 280 and tau phosphorylation at AT8 and AT100 sites. Collectively, these data identify a novel role of Rbbp7, protecting against early events in tau pathogenesis, and highlight its potential as a therapeutic target in AD and related tauopathies.

Disclosures: N. Dave: None. **A.S. Vural:** None. **I.S. Piras:** None. **W. Winslow:** None. **L. Surendra:** None. **J.K. Winstone:** None. **T.G. Beach:** None. **M.J. Huentelman:** None. **R. Velazquez:** None.

Poster

P211. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P211.03

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R01AG059627
NIH Grant R01AG062500

Title: Adulthood dietary choline deficiency results in organ dysregulation, metabolism dysfunction, and subsequent Alzheimer's disease.

Authors: *A. K. DECKER[1], W. WINSLOW[1], E. A. BLACKWOOD[3], A. BILAL[3], I. MCDONOUGH[1], J. K. WINSTONE[1,2], S. TALLINO[1,2], O. ESPINOSA[1], P. SARETTE[4,1], C. GLEMBOTSKI[3], R. VELAZQUEZ, JR[1,2];
[1]Neurodegenerative Dis. Res. Ctr. at the Biodesign Inst., [2]Sch. of Life Sci., Arizona State Univ., Tempe, AZ; [3]Translational Cardiovasc. Res. Ctr. and Dept. of Intrnl. Med., [4]Col. of Med. Phoenix, Univ. of Arizona Col. of Med., Phoenix, AZ

Abstract: Alzheimer’s Disease (AD) rates are rising as the aging population increases. Dietary nutrients play a key role in mediating mechanisms associated with body and brain function. Choline, a nutrient found in common foods is required to produce acetylcholine, a neurotransmitter responsible for memory and muscle control. Choline also builds cell membranes, donates methyl groups for epigenetic mechanisms that regulate gene expression, and reduces homocysteine (Hcy), a toxic amino acid. Elevated Hcy is linked to health issues such as obesity, impaired glucose metabolism, and cardiac pathology, which are all risk factors for AD. While humans and rodents can produce endogenous choline, these levels fail to meet bodily demands, requiring dietary consumption. In 1998, the U.S. established an adequate intake (AI) of daily dietary choline for adult women (425mg/day) and men (550mg/day). However, epidemiology studies show that ~90% of Americans fail to reach the daily AI. Thus, it is imperative to determine whether choline deficiency at adulthood increases body dysfunction and AD risk. Here, we placed the 3xTg-AD mouse model of AD and NonTg controls on a diet that was either choline normal (ChN) or deficient (Ch-) from early adulthood throughout life (3-12 months of age). Mice were assessed for motor function using the rotarod task at 10 months of age followed by a glucose tolerance test to assess glucose metabolism. Heart, liver and brain tissue were subsequently harvested to assess metabolic and neuropathological changes with choline deficiency. The Ch- diet induced motor deficits, increased weight significantly, and impaired glucose metabolism in both 3xTg-AD mice and NonTg, with 3xTg-AD Ch- mice showing greater deficits. Additionally, we found evidence of cardiac pathology and fatty liver disease in the Ch- diet groups. In 3xTg-AD mouse brains, a Ch- diet elevated the levels of toxic oligomeric and insoluble Amyloid-ß (Aß) levels in the hippocampus and throughout the cortex. Collectively, these results highlight that a choline deficient diet could be a risk factor for motor

deficits, metabolic dysfunction and subsequent AD and suggest that simply modifying one's diet to include adequate choline is critical to offset the risk of developing these pathologies.

Disclosures: A.K. Decker: None. **W. Winslow:** None. **E.A. Blackwood:** None. **A. Bilal:** None. **I. McDonough:** None. **J.K. Winstone:** None. **S. Tallino:** None. **O. Espinosa:** None. **P. Sarette:** None. **C. Glembotski:** None. **R. Velazquez:** None.

Poster

P211. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P211.04

Topic:

Support: Academy of Finland
Olav Thon Foundation, Norway

Title: Preclinical evaluation of drug treatment options for sleep-related epitleptic spiking in Alzheimer's disease

Authors: N. JIN, I. GUREVICIENE, ***H. TANILA**;
Univ. Eastern Finland, Kuopio, Finland

Abstract: *Aims.* Epileptic spiking without motor manifestations can be found in 30-50% patients with early AD. This number may still be an underestimate as this spiking occurs almost exclusively during sleep and may limit to the deep temporal lobe. Further, AD patients with subclinical epileptiform activity have about twice faster cognitive decline during a follow-up of 3-5 years (Vossel K et al. Ann. Neurol. 2016; Horvath A et al., EEG Clin Neurophys. 2021). Treating epileptic spiking during sleep may thus offer a way to slow down AD progression, but so far there is no published clinical study on the topic, calling for preclinical testing to guide the choice of optimal drug treatments.
Methods. Ten male APP/PS1 mice (3-5 months) were implanted bilaterally with cortical screw electrodes and hippocampal staggered triple wire electrodes to record local field potentials. Sleep was monitored by overhead video-recording and neck EMG wire electrodes. We tested the standard AD-drugs donepezil and memantine as well as anti-epileptic drugs levetiracetam and lamotrigine that have been recommended as drugs of choice in AD patients with epilepsy, In addition, we assessed amyloid-beta reduction by the gamma-secretase inhibitor semagacestat and anti-inflammatory approach with the tetracycline derivative minocycline at doses with no overt motor manifestations. The analysis focused on hippocampally generated giant spikes and was done within-subject in reference to a repeated vehicle injection.
Results. Preliminary data suggest that levetiracetam decreases while lamotrigine unexpectedly

increases sleep-related giant spikes in APP/PS1 mice. Other tested drugs seem to have no significant effect.

Disclosures: N. Jin: None. **I. Gureviciene:** None. **H. Tanila:** None.

Poster

P211. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P211.05

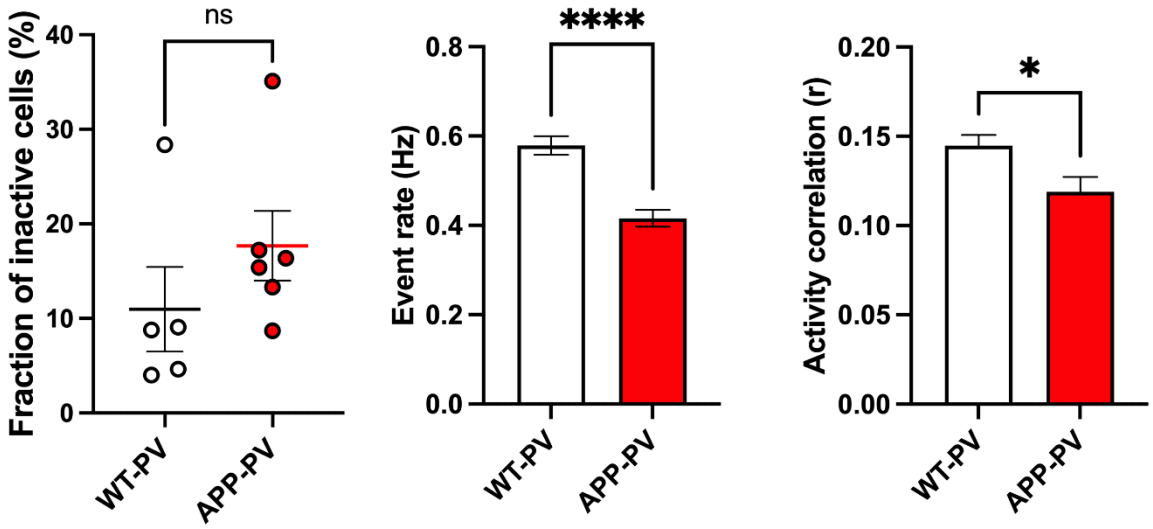
Topic:

Support: NIH Grant 1RF1AG061774-01

Title: Characterization of interneuronal activity in living APP/PS1 mice using genetically encoded calcium indicators

Authors: *M. ALGAMAL[1], A. N. RUSS[2], L. OBAID[3], B. J. BACSKAI[5], K. KASTANENKA[4]; [1]Neruorology, [2]Neuorology, [3]Neurol., [4]Massachusetts Gen. Hosp., Charlestown, MA; [5]Dept Neurol., Mass Gen. Hosp., Charlestown, MA

Abstract: Alzheimer's disease (AD) is characterized by neuronal network dysfunction. Alzheimer's patients exhibit disruptions in neuronal activity that have been linked to cognitive decline. Similarly, amyloidosis animal models, including APP/PS1 (APP) mice, show amyloid-beta-induced aberrant neuronal activity. However, the mechanism underlying these aberrations is not understood. Here, we explored the relationship between amyloid-beta and calcium transients of excitatory neurons as well as two major classes of inhibitory interneurons, parvalbumin and somatostatin interneurons. To this end, the genetically encoded calcium indicator GCaMP6s was targeted to excitatory cortical neurons through CAMKII promoter in 8-12 months-old APP mice and age-matched non-transgenic littermates (WT). Similarly, jGCaMP7s was targeted to parvalbumin or somatostatin interneurons in the somatosensory cortices using Cre-Lox technology with viral delivery. A month later, cortical neurons were imaged through cranial windows under anesthesia using high-resolution two-photon microscopy. The spontaneous calcium transients in distinct neuronal populations were monitored and compared between genotypes. Analysis of calcium events revealed that parvalbumin interneurons are hypoactive, while somatostatin interneurons were moderately hyperactive in APP relative to WT mice. In addition, all neurons, regardless of class, showed disrupted neuronal synchrony in APP relative to WT mice. Finally, proximity to amyloid-beta plaques correlated with neuronal activity in excitatory and somatostatin interneurons. Thus, amyloid beta differentially affects calcium transients of distinct cortical neuronal populations and ultimately leads to network failure in Alzheimer's disease.



PV interneurons are hypoactive in APP/PS1 mice

Disclosures: M. Algamal: None. **A.N. Russ:** None. **L. Obaid:** None. **B.J. Bacskai:** None. **K. Kastanenka:** None.

Poster

P211. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P211.06

Topic:

Support: NIA Grant R01AG062500
NIA Grant R01AG059627

Title: Unbiased proteomic analysis reveals dietary choline deficiency-induced changes to neurodegeneration-relevant pathways in 3xTg-AD mouse model of Alzheimer's disease

Authors: *S. TALLINO[1,3], A. DECKER[1], N. DAVE[1], J. SANDLER[2], T. KARR[2], R. VELAZQUEZ[1,3];
[1]ASU-Banner Neurodegenerative Dis. Res. Ctr., [2]Biosci. Mass Spectrometry Facility, Arizona State Univ. Biodesign Inst., Tempe, AZ; [3]Sch. of Life Sci., Arizona State Univ., Tempe, AZ

Abstract: Alzheimer’s disease (AD) is an increasingly prevalent and costly health burden, creating an urgent need for mechanistic insight into modifiable environmental risk factors, such as diet, to offset disease. The dietary nutrient choline is found in a variety of foods; while adequate intake values were established in 1998, recent reports suggest 90% of Americans do not

meet these requirements. Choline plays an integral role in diverse neurodegeneration-relevant pathways such as acetylcholine biosynthesis, myelination, synapse formation, neurogenesis, and epigenetic modification. Thus, inadequate dietary choline may be a potential risk factor for AD. Here, we investigated hippocampal proteomic alterations induced by choline deficiency in the 3xTg-AD mouse model of AD and nontransgenic (NonTg) controls to identify molecular pathways linking diet to AD pathology. 3xTg-AD and NonTg mice were fed either a choline-normal (ChN) or choline-deficient (Ch-) diet from 3 to 12 months of age (n=4 animals per genotype per diet). Hippocampal tissue was harvested and label-free quantitation (LFQ) was performed using high-resolution mass spectrometry. LFQ revealed significant changes in relative protein abundance in 3xTg-AD and NonTg mice as a result of Ch- diets (>4,500 proteins identified). In particular, feeding 3xTg-AD mice Ch- diets resulted in significant changes in relative abundances of 491 proteins compared to 3xTg-AD mice on ChN diets (2-fold change; FDR <0.05). Of proteins with decreased abundance, gene set enrichment analysis (GSEA; hypergeometric test) using STRING (v11.0) revealed significant enrichment of tau-family protein pathways (FDR <0.03) as well as in pathways related to mitochondrial RNA processing (FDR <0.04) and scaffold protein binding (FDR <0.004); of proteins with increased abundance, we observed significant enrichment of pathways related to cytoskeletal organization and binding (FDR <0.004) as well as myelination and calcium homeostasis (FDR < 0.05). Similarly, comparison between NonTg mice showed the Ch- diet resulted in significant protein abundance changes in 202 proteins (2-fold change; FDR <0.05). Of proteins with decreased relative abundance, GSEA revealed significant enrichment of neurogenesis, neurodevelopment and metabolic pathways (FDR <0.05); myelination and vesicular transport pathways were significantly enriched amongst the proteins with increased relative abundance (FDR <0.05). Collectively, these results highlight that inadequate dietary choline alters AD-relevant biochemical pathways in the hippocampus of NonTg mice as well as in 3xTg-AD mice and may be a modifiable risk factor in AD.

Disclosures: S. Tallino: None. **A. Decker:** None. **N. Dave:** None. **J. Sandler:** None. **T. Karr:** None. **R. Velazquez:** None.

Poster

P211. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P211.07

Topic:

Support: NIA Grant R01AG062500
NIA Grant R01AG059627
Arizona State University Edson Foundation

Title: Dysregulated expression of retinoblastoma binding protein homologs 4 and 7 in Alzheimer's Disease

Authors: *J. M. JUDD[1], I. S. PIRAS[3], N. DAVE[1], D. F. MASTROENI[2], M. J. HUENTELMAN[4], R. VELAZQUEZ, JR[5];
[1]Neurodegenerative Dis. Res. Ctr. at the Biodesign Inst., [2]Biodesign Neurodegenerative Res. Inst., Arizona State Univ., Tempe, AZ; [3]Translational Genomics Res. Inst., Phoenix, AZ; [4]Neurogenomics, The Translational Genomics Res. Inst., Phoenix, AZ; [5]Neurodegenerative Res. Ctr., Biodesign Inst. At Arizona State Univ., Tempe, AZ

Abstract: Epigenetic dysregulation has been identified as a mechanism contributing the pathogenesis seen in Alzheimer's Disease and a related class of disorders known as the tauopathies. Chromatin remodeling complexes, such as the Nucleosome Remodeling and Deacetylase Complex (NURD), contribute to epigenetic regulation. NURD and other complexes, contain retinoblastoma binding (Rbbps) proteins, which act as chaperones and make histones available for modification. Recently, we identified Rbbp7 as a mediator against tau pathology observed in Alzheimer's disease (AD) and related tauopathies. We found evidence that rescuing Rbbp7 in the PS19 mouse model of tauopathies reduces tau acetylation of lysine K280 and phosphorylation of Ser202/Thr205 (AT8). Notably, Rbbp7 and Rbbp4, share 92% homology, are expressed in many shared chromatin remodeling complexes implicated in diseased aging and neurodegeneration. Given the homology of Rbbp4 and Rbbp7, we examined their mRNA expression levels in AD post-mortem brain tissue and aged-matched controls. We used two cohorts of post-mortem brain tissue obtained from the Arizona Study of Aging and Neurodegenerative Disorders/Brain and Body Donation Program. Cohort one included a total of 56 brain samples of young (n = 21, M = 35.4yrs), aged (n = 18, M = 80.7yrs) and AD (n = 17, M = 86.7yrs) patients, and the area examined was the entorhinal cortex (ENT). Cohort two included a total of 187 brain samples (n = 89 AD, M = 84.7yrs; n = 98 age-matched controls, CTL, M = 85.0yrs), and the area examined was the middle temporal gyrus (MTG). The ENT and MTG were selected because these two brain regions are affected in AD; brain samples underwent microarray analysis to identify mRNA expression differences. Subject age, gender, and postmortem autopsy intervals were not significantly different per groups. Quantitative analysis revealed that Rbbp4 is significantly upregulated in the ENT of patients with AD relative to young and aged controls. In the MTG, we found increased Rbbp4 mRNA expression in AD patients compared to controls. Further analysis of MTG tissue showed a significant positive correlation between Rbbp4 mRNA expression with neuritic plaque density in AD brains, but a non-significant positive correlation between Rbbp4 and Braak staging. Interestingly, we found that Rbbp4 and Rbbp7 are negatively correlated in AD post-mortem brain tissue, illustrating that as Rbbp4 goes up, Rbbp7 goes down. Collectively, these results highlight a potential association of Rbbp4 in AD related pathologies.

Disclosures: J.M. Judd: None. **I.S. Piras:** None. **N. Dave:** None. **D.F. Mastroeni:** None. **M.J. Huentelman:** None. **R. Velazquez:** None.

Poster

P211. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P211.08

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH grant R21AG067024
NIH grant R01AG072893
NIH grant P30AG066508

Title: Reduced striatal gray matter volume and apathy in Alzheimer's disease: a meta-analysis and experimental confirmation

Authors: *S. CHAUDHARY, S. ZHORNITSKY, H. CHAO, C. VAN DYCK, C.-S. LI;
Yale Med. Sch., New Haven, CT

Abstract: Abstract: Affecting nearly 70% of patients with Alzheimer’s Disease (AD), apathy is associated with higher morbidity and reduced quality of life. Basal ganglia, anterior cingulate and orbitofrontal cortical atrophy is noted in association with the severity of apathy. However, the findings varied across studies and it remains unclear whether subdomains of apathy may involve distinct neuroanatomical correlates. We thus performed a literature search and meta-analysis of the extant findings. The results of a total of six eligible studies of 184 AD and 56 healthy controls (HC) showed convergence toward smaller basal ganglia volumes in AD. To replicate these findings, we recruited 19 patients with AD or mild cognitive impairment (MCI) and 25 age- and gender- matched healthy controls (HC) for T1 weighted anatomical magnetic resonance imaging. Apathy was assessed using apathy evaluation scale (AES) with the sub-domains - cognitive, behavioral and emotional apathy - separately scored. We performed voxel-based morphometry to examine the gray matter volumes (GMV) of the putamen, caudate and nucleus accumbens in region of interest analyses. The analysis revealed right putamen atrophy to be significantly ($p<0.05$ FWE-corrected, small volume correction) associated with higher apathy, especially the total, behavioral, and emotional AES scores. The association remains significant even after controlling depression as assessed by Beck Depression Inventory II (BDI). However, after controlling both BDI and Mini Mental State Examination (MMSE, cognition) scores, right putamen GMV was significantly associated with only the emotional AES subscore. The findings suggest potential influence of depression and cognition in apathy. Right putamen atrophy may be a neural marker of behavioral and emotional apathy. A larger sample size will be needed to further confirm these preliminary findings.

Disclosures: S. Chaudhary: None. **S. Zhornitsky:** None. **H. Chao:** None. **C. van Dyck:** None. **C. Li:** None.

Poster

P211. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P211.09

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: Stichting Alzheimer Onderzoek (SAO-FRA-20180003)
Scientific Research Flanders (FWO- G048917N)
Scientific Research Flanders (FWO-G045420N)

Title: Altered basal forebrain activity during recurrent patterns of brain activity in 4-month-old TgF344-AD rats

Authors: M. VAN DEN BERG, M. ADHIKARI, M. VERHOYE, *G. A. KELIRIS;
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Abstract: Alzheimer's Disease (AD) is a severe neurodegenerative disorder that leads to brain network dysfunction and cognitive decline. Changes in functional networks at symptomatic stages of AD can be captured using resting state fMRI (rsfMRI) in patients and animal models. In this study, we used a rat model manifesting the full-spectrum of human AD-pathology to identify if spatiotemporal network alterations are present at an early, pre-symptomatic stage of AD. RsfMRI data were collected from four-month-old TgF344-AD (TG) rats (N=15) and wildtype (WT) littermates (N=11) on a 9.4T preclinical MRI scanner. Preprocessing included normalization, filtering and global signal regression. Recurrent, quasi-periodic patterns (QPP) of whole brain activity during a time window (3.6 seconds/6 image volumes) were extracted from the group-level image series[1]. The resulting QPPs were hierarchically clustered based on spatiotemporal similarity to identify robust clusters and their highest occurring QPPs (rQPPs). Cluster-averaged occurrence rates and voxel-wise activations of matched rQPPs were compared between groups using a two-sample t-test. QPP analysis resulted in two clusters of QPPs per group involving two anti-correlated RS networks, the lateral cortical network and the default mode network. Cluster wise-occurrence rates were significantly lower for both clusters in TgF344-AD animals ($p= 0.0427$ & $p= 0.0449$). Spatially, the co-activation of the basal forebrain (BFB) and the cingulate cortex was significantly reduced in the TG rats. The BFB is an important orchestrator of whole-brain network activity [2], and is one of the first regions affected by AD in humans[3]. Alterations in BFB activity might contribute to differences in whole-brain spatiotemporal properties of QPPs as is observed in this study. Spatiotemporal changes in QPPs together with altered 'orchestrating' BFB activity could be a potential signature to identify early onset changes at the network level. Further histological analysis will be performed to gain more insight into the disease-mechanisms driving the changes in QPPs.
References: [1]. Belloy, M., Naeyaert, M., Abbas, A., Shah, D., Vanreusel, V., Auderkerke, J., Keilholz, S., Keliris, G., Van der Linden, A., Verhoye, M. (2018). NeuroImage 180, 463-484. [2]. Nair, J., Klaassen, A., Arato, J., Vyssotski, A., Harvey, M., Rainer, G. (2018). PNAS 115(6),

1352-1357. [3]. Hall, A., Moore, R., Lopez, O., Kuller, L., Becker, J. (2008). Alzheimer's & Dementia 4(4), 271-279

Disclosures: M. van den Berg: None. **M. Adhikari:** None. **M. Verhoye:** None. **G.A. Keliris:** None.

Poster

P211. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P211.10

Topic: C.02. Alzheimer's Disease and Other Dementias

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Title: Investigating whether fractional anisotropy is associated with reduced reaction time cost on an attentional control task

Authors: N. S. MCKAY[1], A. DINCER[1], V. MEHROTRA[1], ***D. A. HOBBS**[1], A. J. ASCHENBRENNER[1], D. BALOTA[1], R. C. HORNBECK[1], J. HASSENSTAB[1], J. C. MORRIS[1], T. L. S. BENZINGER[1], B. A. GORDON[2];
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Abstract: Background Previous research has shown that with age and disease, the integrity of white matter (WM) structures can decline. One such measure of WM integrity is fractional anisotropy (FA), where lower values are often associated with disrupted WM. Similarly, previous work has highlighted that attentional control also declines with age and disease. It was therefore of interest to investigate whether FA was associated with cognitive ability in an aging population. To assess this, we derived a measure of attentional control performance from the Consonant-Vowel, Odd-Even (CVOE) task.
Methods246 participants aged 42-87 years (*M* = 69, *SD* = 8.5), who were enrolled in studies at the Knight ADRC, were included in this analysis. Participants were classified as cognitively normal (CDR = 0, *n* = 185), or cognitively impaired (CDR > 0, *n* = 61). All participants completed an MRI and the CVOE task. Tract-Based Spatial Statistics (TBSS) were run on FA

maps calculated for the participants. We used reaction time (RT) cost, the burden caused by the most difficult aspects of this task, as our behavioral measure of interest. This measure was calculated as the difference between RTs within pure vs mixed trial blocks.
ResultsUsing TBSS, we found that the relationships between FA and RT-cost were significantly different for each group (Figure 1). More specifically, we found no relationship between RT-cost and FA in our cognitively normal individuals ($r = 0.14$, $p = 0.052$; Figure 2). In contrast, we do observe a significant negative relationship between RT-cost and FA in our cognitively impaired individuals ($r = -0.64$, $p < 0.01$; Figure 3).
ConclusionOverall, we found a strong relationship between WM integrity and cognitive performance, as measured by the CVOE task. That is, as integrity becomes more compromised, participants become slower at the task when in the mixed versus the pure blocks within the CVOE task. Importantly, this relationship is only observed for individuals that are cognitively impaired. Taken together, our results add to a body of literature that report a relationship between WM integrity and cognitive performance, but hint that further development of more sensitive cognitive tasks may be needed to fully understand this relationship.
Figure 1. TBSS relationship between fractional anisotropy and reaction time cost for our two groups (CDR = 0, CDR > 0).
Figure 2. Scatterplot depicting the relationship between fractional anisotropy and reaction time cost in individuals who are cognitively normal (CDR = 0).
Figure 3. Scatterplot depicting the relationship between fractional anisotropy and reaction time cost in individuals who are cognitively impaired (CDR > 0).

Disclosures: N.S. McKay: None. **A. Dincer:** None. **V. Mehrotra:** None. **D.A. Hobbs:** None. **A.J. Aschenbrenner:** None. **D. Balota:** None. **R.C. Hornbeck:** None. **J. Hassenstab:** None. **J.C. Morris:** None. **T.L.S. Benzinger:** None. **B.A. Gordon:** None.

Poster

P211. Mechanisms of Synaptic Dysfunction in Alzheimer's Disease: In Vivo Models II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P211.11

Topic: C.02. Alzheimer's Disease and Other Dementias

Support: NIH Grant R21AG065819

Title: Affective behavioral phenotyping and imaging functional network connectivity in Alzheimer’s disease mouse models

Authors: *M. GRUDNY, M. FEBO;
Psychiatry Dept., Univ. of Florida, Gainesville, FL

Abstract: Apathy presented as impaired motivation and reduced affective expression, is commonly diagnosed in patients with dementia, including Alzheimer's disease (AD). Evidence suggests that in the initial pre-cognitive stage of Alzheimer's disease the communication between brain areas involved in reward and memory processing becomes dysregulated. Extracellular plaque deposits of the β-amyloid peptide (Aβ) are a primary pathological hallmark used to diagnose and characterize AD, however the role of this pathology in the affective symptoms of AD is not fully understood. The present preliminary study investigated affective behavioral phenotypes exhibited by amyloidosis mouse models and expands on prior magnetic resonance imaging (MRI) studies examining functional and structural brain abnormalities in mouse models of AD. Specifically, the behavioral phenotypes were investigated with an open-field test, sociability and preference for social novelty test, and sucrose preference test. Young (5-7 m.o.) male and female 5XFAD, CRND8, and their sex- and age- matched wildtype counterpart mice (BxD, B6, B6D2F1/J, and B6C3F1/J) were tested. One-way ANOVA indicated a strain x mutation effect in total distance ($p < 0.05$) and margin distance ($p < 0.05$) during open field between B6C3F1/J and CRND8 mice. Additionally, One-way ANOVA indicated a strain x strain effect in total distance ($p < 0.05$) and margin distance ($p < 0.05$) between BxD 5XFAD and CRND8 mice. The unpaired t-test did not indicate any difference in social interaction and recognition between wildtype and 5XFAD mice on a BXD background. One-way ANOVA indicated a strain x strain effect in sucrose preference ($p < 0.001$) between B6 and BxD mice and a strain x strain effect in sucrose preference ($p < 0.05$) between B6 and 5XFAD mice. These results suggest that even at an early timepoint, prior to significant Aβ burden and cognitive impact, CRND8 mice have significantly increased locomotion, primarily in margin distance traveled. This effect is seen when comparing to B6C3F1/J and 5XFAD mice. Sucrose preference was significantly elevated in B6 mice when compared to both BxD and 5xFAD mice. There was no effect observed in the sociability or preference for social novelty in these mice. However, the lack of effect observed in this task could be caused by the early time point of this study. Overall, these behavioral results show that there is some differential impact of Aβ mutations and background genetics dependent on the phenotype observed. Additionally, these mice were scanned using functional MRI and will be longitudinally studied at an aged time point to discern the impact aging and Aβ pathological progression has on brain function.

Disclosures: M. Grudny: None. **M. Febo:** None.

Poster

P584. Vocal/Social Communication: Avian II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P584.01

Topic: F.01. Neuroethology

Title: The neural basis of spectral prosody in avian vocal duets

Authors: *A. LEMAZINA, L. TROST, M. GAHR, S. HOFFMANN;
Behavioural Neurobio., MPI for Ornithology, Starnberg, Germany

Abstract: Turn-taking requires participants to control and adjust their vocal production in regards to conversation cues received from a partner. It has been suggested that birds pay attention to the prosodic structure to coordinate their song. We hypothesize that prosodic cues, such as changes in the “intonation pattern” of song elements, facilitate duet coordination by predicting the boundaries of the partners‘ turns. In songbirds, vocal production is controlled by an interconnected network of brain nuclei, the so-called vocal control system. One of these nuclei, HVC, plays a crucial role in arranging individual vocal emissions into sequences. We hypothesize that if prosodic cues are used by songbirds in vocal turn-taking, the production of song elements that differ in prosodic pattern should be controlled differently by HVC. To tackle these questions, we used duet singing White-browed Sparrow Weavers (*Plocepasser mahali*) as animal model. Pairs of *P. mahali* sing antiphonal duets by precisely alternating their vocalizations. We recorded the individual vocal and HVC activity in both partners of duetting pairs, and found that *P. mahali* duet songs are indeed organized into spectral prosodic patterns. The maximum frequency contour of male birds’ song syllables could be classified either as rising (R) or as falling (F) pattern. The pattern of the maximum frequency contour of female birds' song syllables could also be divided into two groups: low (LM) - and high (HM) - frequency modulated syllables. Based on these prosodic patterns, we constructed a markov chain for each individual song to determine if there are underlying duetting rules to guide the turn-taking in the duet song. Female prosodic types LM and HM showed greater than expected transition probabilities to male prosodic types R and F, respectively. In addition, we found that in male *P. mahali*, HVC activity was different depending on the prosodic pattern of produced syllables. Median spike time relative to syllable onset was significantly higher in F-type syllables than in R-type syllables. This indicates, that the right HVC is active later during the production of F-type syllables than during the production of R-type syllables. While in R-type syllables, maximum frequency is rather low at the beginning of the syllable, in F-type syllables, maximum frequency is rather low at the end of the syllable. Given that low frequencies are produced by the right syrinx, lateralization of vocal motor control might explain our observations. Our study demonstrates the existence of spectral prosodic cues in birdsong and suggests song lateralization as the underlying neural mechanism for prosody generation.

Disclosures: A. Lemazina: None. **L. Trost:** None. **M. Gahr:** None. **S. Hoffmann:** None.

Poster

P584. Vocal/Social Communication: Avian II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P584.02

Topic: F.01. Neuroethology

Title: Context-dependent control of singing behavior in wild nightingales (Luscinia megarhynchos)

Authors: *G. COSTALUNGA, D. VALLENTIN;
Max Planck Inst. for Ornithology, Seewiesen, Germany

Abstract: Adapting vocal outputs depending on different contexts is an essential feature of learned interactive vocal exchanges, like complex human conversations. Like humans, songbirds are one of the few vocal learning animals. Among them, nightingales are an exceptional example performing sophisticated interactive singing with conspecifics. Every night during their breeding season, male nightingales sing hundreds of different songs engaging in vocal duels against conspecifics to defend territories and attract females. This vocal sparring consists of epochs of turn-taking and overlapped singing with a rival. Additionally, nightingales are able to perform song-matching, listening to and repeating the song sung by their opponents. Although the naturalistic vocal behavior of nightingales is well described, if and how these birds are able to adjust vocal strategies while singing against different opponents remains unknown. To tackle this question, we conducted field experiments exposing wild nightingales to playbacks of an unknown conspecific (CON playback) and of the bird's own songs (BOS playback). We investigated the temporal aspects of the bird's responses to the two playback contexts. The duration of the songs produced by the birds remained unaltered, however the birds increased gap durations and reduced song frequency when exposed to the BOS playbacks. Moreover, we found that nightingales changed the timing of their singing behavior by reducing the number of overlapping songs and the number of consecutive overlaps when exposed to the BOS playbacks compared to the CON playbacks. We then analyzed the song-matching proficiency in the two playback conditions. We found that the birds increased their song-matching behavior while singing against their own songs compared to the CON playbacks. Interestingly, we observed that nightingales were also able to anticipate the upcoming playbacks while counter-singing against the BOS playbacks. The rate of anticipations and matches were higher than the expected chance level based on their song repertoire size. In both conditions, the duration of matches and anticipations of the birds were correlated with the duration of the target songs of the playbacks. Taken together, these results show that nightingales are able to adjust their vocal outputs depending on the context. The birds adapt their temporal performance by changing the timing of their songs in respect to the vocal partner' singing behavior and adjust the content of their responses by singing context-specific songs imitating and anticipating the simulated opponent. Our findings reveal a novel form of context-dependent flexibility in birds' singing behavior.

Disclosures: G. Costalunga: None. **D. Vallentin:** None.

Poster

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Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P584.03

Topic: F.01. Neuroethology

Support: RF1 MH117809-01

Title: Chunks of birdsong: Zebra finch motor thalamus preferentially activates HVC neurons representing the onset of song elements

Authors: *F. W. MOLL[1], A. CORREDERA ASENSIO[1], D. KRANZ[2], M. A. LONG[1];
[1]Neurosci. Inst., NYU Sch. of Med., New York, NY; [2]Harvard Univ., Cambridge, MA

Abstract: Thalamic input to forebrain motor centers has been shown to be essential for the execution of simple movements, but its role in more complex behaviors is poorly understood. We explore this issue in the context of the zebra finch courtship song, a well-studied skilled motor behavior consisting of a learned, stereotyped sequence of typically 3-7 syllables approximately 200 ms in length that function as the minimal effective 'chunk' of singing behavior. The song is represented on a moment-to-moment basis by a sequence of sparse activity in the forebrain premotor circuit HVC which receives input from the motor thalamic nucleus Uvaeformis (Uva). Previous lesion studies have indicated that Uva is necessary for singing, but the precise role of Uva in song production remains undefined. Two main models have been proposed: (1) Uva is part of a distributed feedback loop in which rapid cycling of activity through several brain regions is necessary to advance the song on a moment-to-moment basis. (2) Song-relevant sequences are mediated by local interactions within HVC, initiated by inputs from thalamus.

To address these models, we perturbed either HVC or Uva activity in singing birds using bilateral electrical stimulation. In both cases, most stimulation events resulted in song truncations. In stark contrast to HVC stimulation, which is capable of stopping the song at arbitrary moments, Uva stimulation led to selective truncation of the song at syllable boundaries. This result suggests that thalamic activity may be critical for the initiation of syllables.

To test this idea, we asked whether Uva preferentially targets a functional subgroup of HVC neurons active at the beginning of syllables. Using in vivo calcium imaging, we identified a small set of HVC cells reliably activated by focal Uva stimulation and designated them as putative thalamorecipient neurons. We then measured the activity of these neurons during singing. We found that most of putative HVC thalamorecipient neurons became active at the beginning of song elements, suggesting a wiring scheme favorable of thalamic activation of forebrain circuity capable of driving behaviorally-relevant vocal chunks. Taken together, our results indicate that thalamic inputs can provide important temporal structure for skilled motor performance.

Disclosures: F.W. Moll: None. **A. Corredera Asensio:** None. **D. Kranz:** None. **M.A. Long:** None.

Poster

P584. Vocal/Social Communication: Avian II

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Program #/Poster #: P584.04

Topic: F.01. Neuroethology

Support: NIH F31 NS116933-01
Simons Collaboration on the Global Brain

Title: Understanding the song-production pathway using sleep replay

Authors: *M. ELMALEH[1], F. MOLL[2], D. KRANZ[4], A. CORREDERA ASENSIO[1], M. A. LONG[3];
[1]NYU Langone Med. Ctr., New York, NY; [2]Neurosci. Inst., NYU Sch. of Med., New York City, NY; [3]Neurosci. institute, NYU Sch. of Med., New York, NY; [4]Neurosci., Harvard Univ., Boston, MA

Abstract: Complex behaviors often require the coordination of multiple brain regions, complicating our ability to determine the contribution of local network mechanisms to their execution. During song production in the zebra finch, premotor sequences within the forebrain region HVC may result from some combination of local connectivity or long-range inputs originating from the motor thalamus (nucleus uvaeformis, or Uva). Because lesions to either structure abolish song vocalization, we examine network 'replay' activity during sleep using high-density recording methods and can reconstruct precise song related events. Sleep replay slows when HVC is cooled, demonstrating that it provides temporal structure for these events. Surprisingly, we find that reconstructed sleep sequences continue to span the duration of the song following Uva lesions, supporting a model in which local HVC connections are sufficient to propagate these dynamics. Additionally, we perform simultaneous bilateral recordings which reveal that, while the two hemispheres are precisely coordinated during song production, the cortical regions of this pathway can operate independently during sleep. Because Uva is the only site of bilateral feedback in the song-production pathway, the import of thalamic input during song may be to coordinate the local premotor sequence propagating networks. Our results highlight the power of examining offline activity for studying behaviorally relevant circuit organization.

Disclosures: M. Elmaleh: None. **F. Moll:** None. **D. Kranz:** None. **A. Corredera Asensio:** None. **M.A. Long:** None.

Poster

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Topic: F.01. Neuroethology

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DFG (German Research Foundation) - Project number 459524793

Title: Inhibitory regulation of vocal timing: Integrating auditory-evoked and vocal-premotor activity in the songbird forebrain.

Authors: *J. BENICHOV[1], P. NORTON[2,3], M. PEXIRRA[1], S. SCHREIBER[2,3], D. VALLENTIN[1];
[1]Max Planck Inst. for Ornithology, Seewiesen, Germany; [2]Inst. for Theoretical Biol., Humboldt Univ., Berlin, Germany; [3]Bernstein Ctr. for Computat. Neurosci., Berlin, Germany

Abstract: The ability to regulate vocal timing is a fundamental aspect of communicative interactions for many species, including conversational speech among humans. During these interactions, the production of vocalizations is often influenced by perceived acoustic signals from other individuals. Little is known, however, about the neural circuitry that enables incoming auditory information to affect if and when a vocal reply is produced. To approach this problem, we generated a spiking network model informed by behavioral and electrophysiological data from zebra finches engaged in call-response interactions. The model incorporates the activity of single neurons in the forebrain vocal premotor nucleus HVC prior to vocal production and in response to call playbacks. By simulating the interplay between premotor and auditory-evoked activity, the model accounts for experimentally-induced changes in call timing and predicts that two patterns of inhibition can regulate vocal-motor responses to a heard call. In one regulatory scenario, the strength of production-related premotor inhibition translates into plasticity in vocal response delays. In the other scenario, fast transient interneuron activity in response to auditory input results in the active suppression of call production while a call is heard. The model provides a parsimonious framework by which auditory-vocal transformations can give rise to social coordination in the form of vocal turn-taking. More broadly, it highlights multiple roles of local inhibition within a sensorimotor circuit for behavioral modulation at different time scales.

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Poster

P584. Vocal/Social Communication: Avian II

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Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P584.06

Topic: F.01. Neuroethology

Support: NSF Award EF-1822476

Title: Neural mechanisms for the robustness of the timing network in songbird HVC against cooling

Authors: *A. KHARE, D. M. SEDERMAN, D. Z. JIN;
Dept. of Physics, Pennsylvania State Univ., University Park, PA

Abstract: Understanding the robustness of neural computation against perturbations such as temperature fluctuations is important for systems neuroscience. Birdsong in the zebra finch has exquisite timing features, controlled by an ultra-sparse sequential bursting of projection neurons in the premotor nucleus HVC (proper name) [1]. Experimental and modeling works suggest that a feedforward network within HVC generates sequential bursts [2]. Focal cooling of HVC slows down the song [3]. The effect of temperature on any biological process is quantified using a Q10 temperature coefficient. Most cellular and synaptic processes have Q10 values close to 2.0, and cooling by 10 degrees Celsius typically leads to a 100 percent change in time constants. In contrast, cooling HVC by 10 degrees Celsius in the zebra finch slows down the song only by 30 percent (Q10 ~ 1.3). Hence the burst sequence in HVC is much more temperature resilient than the cellular processes. This difference in Q10's of the song tempo and the cellular and synaptic processes led to a dilemma: Can a network entirely localized within HVC generate timing signals that are more robust against cooling than the cellular and synaptic processes? [3] Here we resolve this problem by computational modeling of a feedforward network within HVC and testing this model for temperature changes. An important aspect of our work involves incorporating axonal delays [5]. Signal transmission along the axons takes up a significant part of the network activity. Unmyelinated axons in HVC, like many such axons across species [6], should have a Q10 of approximately 1.25. This contributes significantly to the temperature robustness of the network. Moreover, our analysis of a single neuron's dynamics shows that the spike responses of HVC projection neurons have a Q10 smaller than the cellular and synaptic Q10's. A combination of these two aspects leads to a Q10 close to 1.3-1.5 for burst propagation time in the network. We also propose how a significant drop in the firing rate of inhibitory neurons could further lower the network Q10. Our work shows that local neuronal networks can be robust to temperature changes, despite higher sensitivity of cellular and synaptic processes to temperature.
References:[1] Hahnloser et al (2002)[2] Long et al (2010)[3] Long et al (2008)[4] Hamaguchi et al (2016)[5] Egger, Tupikov, et al., (2020)[6] Swadlow et al (1981)

Disclosures: A. Khare: None. **D.M. Sederman:** None. **D.Z. Jin:** None.

Poster

P584. Vocal/Social Communication: Avian II

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Program #/Poster #: P584.07

Topic: F.01. Neuroethology

Support: UCSF Discovery Fellows Program

Title: Vocal motor commands engage respiratory brainstem dynamics for call production in songbirds

Authors: *E. A. KISH, K. YACKLE, M. S. BRAINARD;
UCSF, San Francisco, CA

Abstract: Some of the evolutionarily oldest behaviors are generated by rhythmic central-pattern generators (CPGs) in the brainstem and spinal cord. Complex behaviors driven by evolutionarily newer brain regions like the cortex and midbrain often build on these simpler rhythmic behaviors, turning simple behaviors like walking and breathing into complex ones like dancing and vocalizing. Vocalization is a complex behavior used by many species for communication. Vocal motor commands from cortex and midbrain generate sound through vital respiratory machinery, such as the diaphragm and larynx, that is otherwise under the control of CPGs in the respiratory brainstem. Studying how vocal motor commands are coordinated with the ongoing dynamics of the respiratory brainstem can give us a deeper understanding of how descending motor commands interact with CPGs to produce complex behaviors. Bengalese finch calls provide a unique opportunity to study how vocalizations are coordinated with respiratory dynamics since they are produced one at a time, allowing the close study of how individual vocal commands interact with ongoing respiratory dynamics. By pairing audio recordings of vocalizations with air sac pressure recordings of breathing, we found that calls can interrupt breathing at any phase of the respiratory cycle. This indicates that the call-motor command can overcome the ongoing dynamics of the respiratory brainstem. Moreover, calls reset the respiratory rhythm, which means that calls directly interact with the respiratory CPG. Using electrical stimulation of midbrain vocalization nucleus DM to evoke calls, we discovered that the generation of the call respiratory waveform is dependent on a dynamical system. Moreover, our preliminary results from electrical stimulation of the respiratory brainstem indicate that a dynamical system in the respiratory brainstem is sufficient to produce an expiratory vocalization following a large inspiration evoked by electrical stimulation of the respiratory brainstem. Finally, we found evidence that local respiratory dynamics are not just driven by call commands but can also exert influence on call timing. We found a bias of call timing with respect to the respiratory cycle, in that calls are more likely to occur during the inspiratory phase of breathing. Taken together, these results show that top-down motor commands generated by the midbrain can override pre-existing brainstem respiratory dynamics to generate calls, but these local brainstem dynamics can in turn influence the production and timing of calls. Other complex behaviors may similarly depend on a precise interplay between top-down motor commands and local CPG dynamics.

Disclosures: E.A. Kish: None. **K. Yackle:** None. **M.S. Brainard:** None.

Poster

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National Institutes of Health 1R01NS099375

Title: Extreme fine motor control of birdsong by recruiting the smallest, weakest and fastest motor units.

Authors: *I. ADAM[1], A. MAXWELL[1], H. ROESSLER[1], E. B. HANSEN[1], M. VELLEMA[2], J. BREWER[1], C. P. ELEMANS[1];
[1]Univ. of Southern Denmark, Odense M, Denmark; [2]Max Planck Inst. for Ornithology, Seewiesen, Germany

Abstract: Songbirds are an excellent model system to understand the acquisition and maintenance of auditory guided fine motor skill learning, however very little is known about the motor pool controlling the muscles of the vocal organ, the syrinx. The control resolution of all animal behavior is set by the smallest possible force step when recruiting the muscles involved in that behavior. This in turn depends on the number of motor units (MU), their size distribution as well as their muscle-specific force. To measure the MU size distribution, we developed and applied a new method using spatiotemporally resolved calcium imaging on cross-sections of intact and living syrinx muscles. Additionally, we determined how force drives fundamental frequency (f_o) using an in vitro syrinx preparation. Combining both data sets with measurements of muscle stress allowed us to determine the control resolution of f_o, an important vocal parameter, in zebra finches. We show that the syrinx motor pool is made up of extremely small MUs, 40-50% of MUs innervated 3 muscle fibers and 13-17% even consisted of only one single muscle fiber. In combination with the lowest specific stress known in skeletal vertebrate muscles (5 mN/mm^2), we found that the major muscle controlling f_o is equipped with small force steps of 50 mHz to 7.3Hz per MU. In conclusion, the song system possesses the highest possible control resolution in vertebrates. We suggest that this evolved in response to strong selection on finely graded vocal output. We furthermore propose that high-resolution control was one of the features contributing to the successful radiation of songbirds by expanding their vocal space allowing for diversification of song ultimately leading to speciation.

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Poster

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UBACYT Grant 20020170100220BA (Argentina)

Title: Dynamical system model for the neural activity of singing canaries (Serinus canaria)

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Abstract: Vocal production in songbirds is a key topic regarding the motor control of a complex, learned behavior. Birdsong is the result of the interaction between the activity of a neural network specifically dedicated to song production and learning (known as the “song system”), the respiratory system and the vocal organ. These systems interact and give rise to precise biomechanical motor gestures which result in song production. Telencephalic neural nuclei play a key role in the production of motor commands that drive the periphery, and while several attempts have been made to understand their coding strategy, difficulties arise when trying to understand neural activity in the frame of the song system as a whole. In this work, we report neural additive models embedded in an architecture compatible with the song system to provide a tool to reduce the dimensionality of the problem by considering the global activity of the units in each neural nucleus. This model is capable of generating outputs compatible with measurements of air sac pressure during song production in canaries (*Serinus canaria*). In this work, we show that the activity in a telencephalic nucleus required by the model to reproduce the observed respiratory gestures is compatible with electrophysiological recordings of single neurons in freely behaving animals.

Disclosures: A. Amador: None. **C.T. Herbert:** None. **S. Boari:** None. **G.B. Mindlin:** None.

Poster

P662. Human Fear and Aversive Learning and Memory

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P662.01

Topic: G.01. Fear and Aversive Learning and Memory

Support: NIH R00MH106719

Title: Neural reinstatement reveals divided organization of fear and extinction memories in the human brain

Authors: *A. C. HENNINGS[1], M. MCCLAY[2], M. R. DREW[3], J. A. LEWIS-PEACOCK[4], J. E. DUNSMOOR[5];
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Abstract: Neurobiological research in rodents has revealed that competing experiences of fear and extinction are stored as distinct memory traces in the brain. This divided organization is adaptive for mitigating overgeneralization of fear to related stimuli that are learned to be safe, while also maintaining threat associations for unsafe stimuli. Whether a similar division exists in the human brain remains unclear. Here, we used a hybrid form of Pavlovian conditioning with an episodic memory component to identify overlapping multivariate patterns of fMRI activity associated with the formation and retrieval of fear versus extinction. In healthy adults (N=24), distinct regions of the medial PFC and hippocampus showed selective neural coding for fear and extinction memories. Specifically, the vmPFC showed specificity for extinction memories, while the dACC displayed specificity for fear memories. This dissociation was absent in participants with PTSD symptoms (N=24). Like healthy adults, individuals with PTSD did display reinstatement of fear memories in the dACC. However, extinction memory reinstatement was also observed in the dACC, representing a misallocation of extinction memories. Similar to the double dissociation observed in the mPFC of healthy adults, we also observed a double dissociation in fear and extinction reinstatement along the long axis of the hippocampus. Specifically, the anterior hippocampus exhibited selective reinstatement of extinction memories while the posterior hippocampus exhibited selective fear memory reinstatement. Again, this pattern was dysregulated in individuals with PTSD, such that no extinction memory reinstatement was observed in the anterior hippocampus. The divided neural organization of fear and extinction may support flexible retrieval of context-appropriate emotional memories, while their disorganization may promote overgeneralization and increased fear relapse in affective disorders.

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Poster

P662. Human Fear and Aversive Learning and Memory

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Program #/Poster #: P662.02

Topic: G.01. Fear and Aversive Learning and Memory

Support: the University of Zürich's Clinical Research Priority Program for the CRPP "Synapse& Trauma"

Title: Can you remember the context? Quantification of human context fear conditioning

Authors: *Y. XIA[1,2], J. WEHRLI[1,2], D. R. BACH[1,3,4];
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Abstract: Contextual fear conditioning is a paradigm commonly used to study aversive learning and the formation of trauma memory in humans. We sought to optimise the quantification of contextual fear memory recall from psychophysiological measures. We first used an existing virtual reality paradigm (Kroes et al. 2017). Participants learned the context-shock association on their first visit; memory retention was then tested one week later under extinction. Measures of conditioning and retention were derived from startle eye-blink responses (SEBR) and autonomic responses, such as skin conductance responses (SCR) and pupil dilation. We first exploratively studied whether memory acquired from this paradigm can be retained for 7 days with N = 23 healthy participants. Although self-reports on valence, arousal and shock expectancy showed successful conditioning and memory retention, model-based analysis of psychophysiological responses did not reveal any differences between the contexts. These negative results were replicated in an independent confirmatory experiment with N = 28 healthy participants. We then adapted a configural fear conditioning paradigm (CFG) using five static room images as conditioned stimuli (CSs; Stout et al. 2019). One of the images (CS+) was reinforced by unconditioned stimulus (US) during memory acquisition with a reinforcement rate of 83%. In two independent experiments with N = 29 and N = 24 healthy human participants, aversive memory was acquired with stronger SCR and pupil dilation responses to CS+ than to CS-. One week later, memory was retained, reflected by stronger SEBR and pupil dilation responses to CS+. Overall, we established a contextual fear conditioning paradigm in healthy humans and verified memory retention over 7 days, based on psychophysiological responses.

Disclosures: Y. Xia: None. **J. Wehrli:** None. **D.R. Bach:** None.

Poster

P662. Human Fear and Aversive Learning and Memory

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Program #/Poster #: P662.03

Topic: G.01. Fear and Aversive Learning and Memory

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Brain & Behavior Research Foundation (NARSAD)
8G12MD007600
NSF PRCEN graduate fellowships
BP ENDURE NEURO-ID fellowships
PRCTRC Pilot Project Program

Title: Effects of Concussive-like Brain Injury on Fear Behaviors in Rats

Authors: *G. HERNÁNDEZ-BUSOT, M. RIVERA-LÓPEZ, O. MARTÍNEZ-GUZMÁN, M. CÁCERES-CHACÓN, D. SIERRA-MERCADO;
Anat. & Neurobio., Univ. of Puerto Rico Sch. of Med., San Juan, PR

Abstract: Each year 40 million people worldwide suffer from traumatic brain injury, mainly in the form of concussions. Human studies have linked concussions with the development of fear-related disorders such as post-traumatic stress disorder (PTSD). However, the relationship between concussions and fear behaviors remain unclear and animal studies show conflicting results. To evaluate the potential link between concussions and fear behaviors, a biological link must be examined using reliable injury models and behavioral tests. Given that failure to extinguish fear is a hallmark of PTSD, we hypothesized that a concussive brain injury will impair fear extinction using Pavlovian conditioning in rats. To address this gap, rats underwent fear conditioning where they learned that a tone predicts a shock. Freezing behavior to the conditioned tone, an index of fear characterized by lack of movement save those necessary for breathing, was quantified. Afterwards, anesthetized rats received either a concussive-like injury or sham injury. Following recovery from concussive-like injury, rats underwent extinction where they learn that the tone no longer predicts a shock. Results showed no significant difference ($p>0.05$) between concussive-like ($n=9$) and sham ($n=8$) groups, suggesting that concussive brain injury does not affect the ability to extinguish fear. Future studies will aim at examining changes in neuronal activity in brain regions relevant to fear behaviors such as the amygdala using cFos immunohistochemistry.

Disclosures: G. Hernández-Busot: None. **M. Rivera-López:** None. **O. Martínez-Guzmán:** None. **M. Cáceres-Chacón:** None. **D. Sierra-Mercado:** None.

Poster

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Program #/Poster #: P662.04

Topic: G.01. Fear and Aversive Learning and Memory

Support: Wellcome Trust Fellowship

Title: The role of hippocampal-prefrontal theta oscillations in human contextual fear conditioning

Authors: *I. MARTIN, D. BUSH, J. BISBY, A. CASTEGNARO, R. ADAMS, N. BURGESS; UCL, London, United Kingdom

Abstract: Rodent studies have implicated the hippocampal and medial prefrontal network (HPC-mPFC) in anxiety and spatial fear learning. Both HPC and mPFC theta power, along with HPC-mPFC theta coherence, are increased in anxiogenic environments. Interestingly, however, mPFC theta power is specifically increased in the safe areas of a dangerous environment (Adhikari et al., 2010). These findings may aid in the understanding of underlying mechanisms of anxiety and development of treatments. Thus, we sought to replicate and extend these key findings in humans. We used a magnetoencephalography (MEG) contextual fear conditioning paradigm in which 22 participants were asked to freely navigate to and 'pick' flowers in a circular virtual reality (VR) environment. Flowers in one half of the environment were associated with a 'bee sting' and 50% chance of electric shock . In the other half of the environment, flowers were never associated with a shock. When participants picked a flower, they were asked to rate their expectancy of shock on a scale of 1-10. Given prior findings, we focused our analyses on the longest continuous period of movement towards flowers, and on the theta band. Expectancy ratings were significantly higher in the dangerous half of the environment, showing that participants successfully learned the task contingencies. Based on the grand average power spectrum, and previous literature, we analysed low (2-5 Hz) and high (6-10 Hz) theta bands separately, comparing oscillatory activity between trials in the safe and dangerous halves of the environment. This revealed increased 6-10 Hz power over frontal sensors in the safe half, and increased 6-10 Hz power over middle-temporal sensors in the dangerous half of the environment. Increased theta power in the dangerous half was localized to right hippocampus (rHPC); while increased theta power in the safe half was localized to medial prefrontal cortex (mPFC). Using rHPC as a seed, we examined differences in 6-10 Hz phase coupling and found increased rHPC-dorsolateral PFC coupling in the dangerous half of the environment. Our findings offer important translational support for the roles of HPC and mPFC across rodents and humans in particular that HPC-PFC communication is important in processing danger. Our study is also the first to replicate mPFC theta power increases in the safe areas of dangerous environments in humans. The findings are consistent with recent work describing threat dependent modulation of hippocampal theta power (Khemka et al., 2017) and offer insight into the neural correlates of contextual fear in humans (Suarez-Jimenez et al., 2018).

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Poster

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Program #/Poster #: P662.05

Topic: G.01. Fear and Aversive Learning and Memory

Title: Anticipatory brain activity during Pavlovian fear conditioning is modulated by the controllability of threat

Authors: *L. R. NELSON[1], A. BOLARAM[1], J. B. PURCELL[1], H. E. DARK[1], D. K. GREY[1], A. J. KNIGHT[2], D. C. KNIGHT[1];
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Abstract: The ability to control the threats we encounter has important implications for resilience to stress. Chronic stress exposure can lead to emotional dysfunction and the development of anxiety disorders. Therefore, examining anticipatory neural responses to controllable versus uncontrollable threats during Pavlovian fear conditioning may provide new insight into the processes underlying emotional dysfunction. We hypothesized the ability to control threat would influence the anticipatory functional magnetic resonance imaging (fMRI) signal response within the amygdala and prefrontal cortex (PFC), which are brain regions that underlie fear conditioning. This study investigated anticipatory neural responses (i.e., conditioned responses; CR) to threats (i.e., unconditioned stimuli; UCS) that were either controllable or uncontrollable using a yoked Pavlovian fear conditioning procedure. Participants (n = 26 [21 females]; mean age = 23.62) were presented visual stimuli (green and yellow squares) as conditioned stimuli (CSs; 8 second duration). One CS (CS+; 20 trials) was always paired (100% pairing rate) with a loud white noise UCS (100dB; 0.5 to 6 second duration). The other CS (CS-; 20 trials) was presented without the UCS (0% pairing rate). The UCS was also presented alone on some trials (20 trials). Stimuli were presented during a Controllable Condition (CC) and an Uncontrollable Condition (UC). During the CC, participants could terminate the UCS by pressing a button, while during the UC, the duration of each UCS was yoked to the UCS durations chosen by a prior participant during their CC. Functional MRI data were collected using a Siemens 3T Prisma scanner and analyzed using Analysis of Functional NeuroImages (AFNI) software. ANOVA revealed that learning-related changes in the fMRI signal response to the CS+ and CS- varied with the controllability of the UCS. More specifically, a significant interaction was observed between CS type (i.e., CS+ versus CS-) and controllability (i.e., CC versus UC) within the amygdala, ventromedial PFC, dorsomedial PFC, ventrolateral PFC, and posterior cingulate. These results suggest that learning-related changes in anticipatory brain activity (i.e., the CR) are impacted by the ability to control impending threats (i.e., the UCS). These brain regions may play an important role during fear conditioning by maintaining

anticipatory representations of control over an impending threat. These findings suggest that threat controllability may impact the way we form associations between a CS and UCS, which could ultimately have implications for the development and subsequent treatment of stress and anxiety-related disorders.

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Poster

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Program #/Poster #: P662.06

Topic: G.01. Fear and Aversive Learning and Memory

Title: Probing the role of human primary sensory cortex in acquisition and consolidation of aversive memory with transcranial magnetic stimulation

Authors: K. E. OJALA[1], M. STAIB[1], S. GERSTER[1], C. C. RUFF[2], ***D. R. BACH**[3]; [1]Computat. Psychiatry Res., Psychiatric Univ. Hosp. Zurich, Zurich, Switzerland; [2]SNS-Lab, Univ. of Zurich, Zurich, Switzerland; [3]Univ. Col. London, London, United Kingdom

Abstract: Aversive conditioning is often thought to rely primarily on the amygdala with no essential contribution from cortical areas. More recently, however, it emerged that rodents require primary sensory cortex for learning to discriminate complex stimuli that predict threat from those that predict safety. It is not firmly known whether this is also the case for simple conditioned stimuli, and in humans. Here, we used continuous theta-burst transcranial magnetic stimulation (cTBS) to investigate the role of primary sensory cortex in discriminative threat conditioning in humans. Because of its clear lateralisation, we chose the somatosensory domain as a model system. N = 68 participants received cTBS immediately before conditioning in the CS-contralateral (experimental) or CS-ipsilateral (control) hemisphere. They were then trained with simple (location) and complex (location pattern) somatosensory conditioned stimuli (CS) and aversive electric stimulation as US. After overnight consolidation, we measured threat memory retention using fear-potentiated startle. We found that memory retention was attenuated in the experimental compared to the control group. There was no evidence for a difference in this effect between simple and complex stimuli. During initial acquisition, learning was quantified using pupil dilation and skin conductance responses. We found no evidence that cTBS had an immediate effect on initial learning in these measures, or on CS identification. This suggests a stronger effect of cTBS on consolidation than on initial stimulus processing or short-term learning. We propose that non-invasive stimulation of sensory cortex may provide a new avenue for interfering with threat memories in humans.

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Poster

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Program #/Poster #: P662.07

Topic: G.01. Fear and Aversive Learning and Memory

Support: MH107444
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MH121735

Title: Neuroticism/negative emotionality is associated with increased reactivity to uncertain threat in the bed nucleus of the stria terminalis

Authors: *S. E. GROGANS[1], J. HUR[3], M. G. BARSTEAD[4], A. S. ANDERSON[5], S. ISLAM[6], H. C. KIM[2], M. KUHN[7], R. M. TILLMAN[1], A. S. FOX[8], J. F. SMITH[1], K. A. DEYOUNG[1], A. J. SHACKMAN[9];
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Abstract: Neuroticism/negative emotionality (N/NE)—the tendency to experience greater negative affect—is a central dimension of temperament and personality. Elevated N/NE has profound consequences for public health and wealth, drawing the attention of researchers and clinicians. Yet the underlying neurobiology has remained enigmatic. Here, we used a suite of neuroimaging techniques to test the hypothesis that N/NE is associated with heightened reactivity to threat-of-shock in the extended amygdala (EA)—including the central nucleus of the amygdala (Ce) and bed nucleus of the stria terminalis (BST)—in 220 young adults selectively recruited from a pool of nearly 7,000 phenotyped individuals to capture a broad spectrum of N/NE. To clarify specificity, parallel analyses were performed using data from an emotional-faces paradigm. While ‘threatening’ faces are potent triggers of amygdala activity and widely used in on-going biobank projects, they do not elicit robust negative affect and do not represent a trait-relevant challenge. A multiband pulse sequence, best-practice data processing techniques, spatially unsmoothed data, anatomically defined regions-of-interest, and a cross-validated

robust-regression framework enabled unbiased hypothesis testing with unprecedented anatomical resolution. Results revealed that N/NE is associated with heightened BST activation during aversive anticipation (p=0.049), and this association is uniquely evident when the timing of aversive stimulation is uncertain (p=0.004). BST reactivity to uncertain threat remained predictive of N/NE in models excluding BST reactivity to certain threat (p=0.003) or including Ce reactivity to uncertain threat (p=0.002). Other findings suggest that relations between the N/NE phenotype and BST function are indirect and reflect increased threat-elicited arousal (p=0.011). In contrast, N/NE was unrelated to Ce reactivity to threat-of-shock and BST/Ce reactivity to emotional faces (ps>0.05). While it is tempting to treat different 'threat' paradigms as interchangeable probes of brain function, this has rarely been examined. Follow-up analyses revealed no evidence of between-task convergence in the Ce (p>0.05) and marginal evidence in the BST (p=0.059). A large, well-phenotyped sample and pre-registered, best-practices approach enhance confidence in the robustness and translational relevance of these results. Together, these observations have implications for theory and practice and lay the groundwork for the longitudinal and mechanistic studies that will be necessary to determine causation and, ultimately, to develop improved interventions for extreme N/NE.

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Poster

P662. Human Fear and Aversive Learning and Memory

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Program #/Poster #: P662.08

Topic: G.01. Fear and Aversive Learning and Memory

Support: UAB

Title: Predictability and controllability alter neural reactivity to threat

Authors: *A. BOLARAM, J. B. PURCELL, H. E. DARK, D. K. GREY, A. J. KNIGHT, D. C. KNIGHT;
Univ. of Alabama, Birmingham, Birmingham, AL

Abstract: Emotional resilience to stressful events depends, in part, on our ability to predict and control threats in the environment. Thus, determining the neural response to predictable and controllable threats, may provide new insights into brain processes that underlie resilience to emotional dysfunction. The present study assessed the functional magnetic resonance imaging (fMRI) signal of participants (N=26, [20 female], Mean Age = 23.62) who completed a differential fear conditioning procedure designed to investigate the neural response to predictable

and unpredictable threats under controllable and uncontrollable conditions. Two visual stimuli (green and yellow squares) served as conditioned stimuli (CSs; 8 second duration) and a loud, 100 dB white noise served as the unconditioned stimulus (UCS; 0.5-6.0 second duration). One CS (CS+) was always (100% pairing rate) paired with the UCS (Predictable threat; 20 trials). The other CS (CS-; 20 trials) was never paired with the UCS (0% pairing rate). In addition, the UCS was presented alone on some trials (Unpredictable threat; 20 trials). Stimuli were presented during both Controllable and Uncontrollable conditions. The color of a UCS expectancy rating bar (blue or red) indicated whether the UCS was controllable or uncontrollable. During Controllable trials participants could terminate the UCS (Controllable threat) by a button press. During Uncontrollable trials the button press did not terminate the UCS (Uncontrollable threat). To ensure UCS durations were equivalent across Controllable and Uncontrollable conditions, the duration of the Uncontrollable UCS was yoked with the duration of a prior participant's Controllable UCS. Skin conductance response and UCS expectancy served as behavioral indices of threat-related processing during the task. Functional MRI data were collected using a 3T Siemens Prisma scanner. Repeated measures ANOVA revealed a significant interaction between predictability and controllability in the threat-related fMRI signal response within the dorsomedial, dorsolateral, and ventrolateral prefrontal cortex, middle and posterior cingulate, and inferior parietal lobule. Follow up analysis revealed that across trial blocks, activity in these regions was greater for predictable threats that were uncontrollable than predictable threats that were controllable. The present findings demonstrate that threat predictability and controllability alters the neural response within brain regions that support emotion regulation processes, which may have important implications for susceptibility/resilience to internalizing disorders.

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Poster

P736. Alcohol Intake and Preference: Mechanisms I

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Program #/Poster #: P736.01

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH AA020919
NIH DA035958

Title: Cd5 knockout mice exhibit alterations in measures of ethanol sedation and consumption

Authors: *A. J. PAYNE[1,2], J. D. OBRAY[3], B. M. WILLIAMS[2], C. A. SMALL[2], S. M. BAUSERMAN[2], J. T. YORGASON[2], J. G. EDWARDS[2], K. S. WEBER[2], S. C. STEFFENSEN[2]; [1]Noorda Col. of Osteo. Med., Pleasant Grove, UT; [2]Brigham Young Univ., Provo, UT; [3]Med. Univ. of South Carolina, Charleston, SC

Abstract: Cluster of differentiation 5 (CD5) is expressed on the surface of T and a subset of B cells (B1 cells) and is a negative regulatory molecule that alters lymphocyte activation pathways. It has been found that CD5 expression is altered during chronic ethanol (EtOH) use. Specifically, the number of CD5+ B cells is reduced during active alcoholism and returns to normal after withdrawal. Given the apparent sensitivity of these cells to chronic EtOH consumption and the role of some lymphocytes in regulating the development of inhibitory GABA synapses, we assessed the impact of a CD5 genetic knockout (CD5KO) on measures related to EtOH reward and sedation. We found that CD5KO mice display decreased EtOH consumption compared with wild-type C57BL/6J controls in a 24-hour access two bottle choice drinking paradigm. Additionally, CD5KO mice displayed considerable resistance to the EtOH-mediated decrease in locomotor activity at low doses (0.5 - 2.0 g/kg) and EtOH-induced sedation at high doses (4.0 g/kg). We found that there was no difference in spontaneous dopamine (DA) release events between CD5KO mice and wild type controls, and evoked DA release was also measured. Additionally, we found decreased levels of DA transporter (DAT) RNA transcripts in the nucleus accumbens (NAc) in the CD5KO mice. There was no evidence of a CD5-dependent effect in the EtOH-induced inhibition of GABA neuron firing rate in the ventral tegmental area (VTA). These results suggest that CD5KO mice are less sensitive to some of the effects of EtOH and that peripheral neuroimmune mechanisms involving CD5 may be a factor in EtOH consumption and sedation.

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Poster

P736. Alcohol Intake and Preference: Mechanisms I

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Program #/Poster #: P736.02

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant P20GM113109

Title: The impact of early alcohol exposure and differential rearing on operant responding for alcohol and synaptic plasticity.

Authors: *T. MOSER[1], B. C. HOWATT[1], K. ESCHLIMAN[1], I. MALI[1], S. BOSSMANN[2], M. CAIN[3];
[1]Kansas State Univ., Manhattan, KS; [2]Univ. of Kansas, Kansas City, KS; [3]Psychological Sci., Kansas Sate Univ., Manhattan, KS

Abstract: The experiences in adolescence can drastically affect how the brain forms in early adulthood. An isolated rearing environment and early alcohol exposure both negatively alter brain development. With either of these experiences, a decrease in synaptic plasticity can occur caused by a concentration change in metabolites (i.e., homoscysteic acid) that cause more neuroinflammation resulting in a reduction of synaptic plasticity. Our current study examines how differential rearing and early alcohol exposure affect alcohol seeking and synaptic plasticity using nuclear magnetic resonance (NMR) to measure markers for synaptic plasticity in adulthood. We hypothesize that enriched rats that have intermittent adolescent alcohol exposure (IAEE) during adolescence will have lower responding for alcohol in an operant paradigm. We also hypothesize that enrichment will have a higher homocysteic acid concentration than standard condition (SC) or isolated condition (IC). In the current study, we raised male and female Long Evans rats in Enriched (EC), SC, and IC conditions during adolescence. During the rearing period, twelve injections of 20% alcohol (w/v; ip) or saline were administered every other day. Rats were then trained to lever press in an operant chamber. After training was completed, rats responded for 10% alcohol on two fixed-ratio (FR-1 and FR-5) schedules and a progressive ratio (PR) schedule. Following a two-week rest period, a challenge injection of alcohol (1.5 g/kg) or saline was administered and blood was collected for a blood alcohol concentration (BEC) quantification and brains were collected for NMR to measure homocysteic acid concentration. The results indicate that enrichment and sex influence responding for 10% alcohol in the operant paradigm; however, IAEE treatment has no effect. Enriched female rats responded the least for 10% alcohol across the different schedules tested with IC and SC female rats responding more. Enrichment decreased the BEC compared to SC and IC rats regardless of sex and prior alcohol exposure. Preliminary results of the NMR imaging suggest that male isolated rats that received an acute injection of alcohol or in the saline control group (alcohol naïve) have lower concentrations of homocysteic acid in the medial prefrontal cortex. In summary, our results indicate that enrichment in early adolescence decreases responding for alcohol on a variety of operant schedules, decreases BEC following IAEE, and helps maintain higher levels of homocysteic acid in the mPFC.

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Poster

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R01AA027653

Title: Disturbances in the anatomical location and phenotype of peptide neurons induced by low-dose embryonic ethanol exposure in zebrafish and rats

Authors: *A. D. COLLIER, G.-Q. CHANG, N. KHALIZOVA, N. YASMIN, S. CAMPBELL, M. FAM, S. F. LEIBOWITZ;
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Abstract: Prenatal alcohol exposure (PAE) is well known to disturb neuronal development and behavior. Previous work from this lab and others has shown in rats and zebrafish that low-moderate concentrations of ethanol exposure during gestation increases the density of neurons expressing the peptides, hypocretin/orexin (Hcrt) and melanin-concentrating hormone (MCH), which are most concentrated in specific areas of the hypothalamus and known to promote alcohol consumption. With studies in rats showing Hcrt neurons under normal conditions to be neurochemically heterogeneous and perhaps functionally distinct based on their anatomical location, we used time-lapse live imaging and brain registration in zebrafish and found embryonic exposure to low-dose ethanol (0.5% v/v) in the water during hypothalamic development (22-24 hours post-fertilization) to increase the dispersion and alter the migration and anatomical distribution of Hcrt neurons, causing more than 25% to be abnormally located. We then laser ablated these Hcrt neurons and found this to block behaviors induced by embryonic ethanol, including increased voluntary ethanol-gelatin consumption, suggesting that these neurons play a role in mediating ethanol-induced behavioral disturbances. We next sought to determine if PAE in rats similarly affects the anatomical distribution of these peptide neurons and also possibly their specific characteristics. We used iDISCO brain clearing with Hcrt and MCH immunolabeling that allows one to examine the total population of these neurons. We found that PAE, at a low-moderate dose (2 g/kg/day) from embryonic day 10 (E10) to E15, increases the density of Hcrt and MCH neurons in hypothalamic areas where they are normally most concentrated, specifically the perifornical lateral hypothalamus, zona incerta and dorsomedial area, but has little effect on the size, shape and number of processes of these peptide neurons. However, analyses of other hypothalamic areas, such as the mammillary nuclei where Hcrt and MCH are not normally detected, revealed populations of both Hcrt and MCH neurons in PAE rat offspring. Moreover, the majority of these abnormally located neurons exhibited altered features, including unusually round somas, small size, and fewer processes, relative to their standard phenotype in areas where they naturally exist. These findings suggest that, in addition to increasing their density in brain areas where they are normally located, PAE alters the anatomical distribution of these peptide neurons and causes some to migrate outside their normal location and have abnormal phenotypes that may contribute to the behavioral disturbances produced in the offspring.

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Poster

P736. Alcohol Intake and Preference: Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P736.04

Topic: G.09. Drugs of Abuse and Addiction

Support: NIAAA Grant R21AA026356

Title: The effects of perinatal sex hormone manipulation on alcohol response following a juvenile traumatic brain injury

Authors: *R. OLIVERIO[1], K. KARELINA[2], R. VELAZQUEZ-CRUZ[2], B. WHITE[2], B. WHITEHEAD[2], Z. M. WEIL[2];
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Abstract: Approximately 2.8 million traumatic brain injuries (TBIs) occur each year in the United States, and a large portion of the population affected by these injuries is composed of children. Notably, individuals who suffer from a TBI early in life experience a myriad of adverse risks. For example, women injured in adolescence are more likely to develop an alcohol use disorder in adulthood than women injured at any other developmental stage. We reported similar findings in a preclinical model, wherein female mice injured early in life drank significantly more than uninjured females. However, we did not see this effect in males. Given these sex differences, we aimed to investigate the role of sex hormones in the response to alcohol following a juvenile TBI. We hypothesized that male sex hormones reduced drinking following TBI. Therefore, we manipulated sex hormones in mice perinatally through exposure to androgens in females and gonadectomies in males. We then performed a mild traumatic brain injury at postnatal day 21 using a standard closed-head injury model. In adulthood, we employed additional sex hormone manipulations via gonadectomies in male and female mice. Alcohol preference was then assessed through a conditioned place preference paradigm. Injured female mice for which no sex hormone manipulations were performed developed a conditioned place preference to ethanol, whereas injured females which were provided testosterone perinatally or gonadectomized in adulthood did not. This indicates that sex hormones are involved in the response to alcohol following a TBI early in life.

Disclosures: R. Oliverio: None. **K. Karelina:** None. **R. Velazquez-Cruz:** None. **B. White:** None. **B. Whitehead:** None. **Z.M. Weil:** None.

Poster

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Program #/Poster #: P736.05

Topic: G.09. Drugs of Abuse and Addiction

Support: National Plan on Drug Abuse, Ministerio de Sanidad of Spain (PNSD015I2019)

Title: Receptor Protein Tyrosine Phosphatase β/ζ modulates ethanol effects on hippocampal neurogenesis in male and female adolescent mice but only regulates ethanol intake in male mice

Authors: M. GALÁN LLARIO, M. RODRÍGUEZ-ZAPATA, T. FONTÁN, E. GRAMAGE, M. VICENTE-RODRÍGUEZ, C. PÉREZ-GARCÍA, M. C. OVEJERO-BENITO, J. M. ZAPICO, A. RAMOS, B. DE PASCUAL-TERESA, *G. HERRADON;
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Abstract: Binge drinking causes neuroinflammation and neurotoxicity in the adolescent brain and is highly related to the development of alcohol use disorder (AUD). There is an urgent need to discover new pharmacological targets not only to limit ethanol consumption but to prevent brain damage caused by ethanol in adolescents. Pleiotrophin (PTN) is a cytokine upregulated in the brain after ethanol administration. PTN binds Receptor Protein Tyrosine Phosphatase (RPTP) β/ζ and acts as an endogenous inhibitor of its phosphatase activity. Both, PTN and MY10, a small-molecule inhibitor of RPTPβ/ζ, decrease binge-like ethanol consumption and reward in mice and rats. We hypothesize that RPTPβ/ζ also plays a role in chronic ethanol consumption and in the neural alterations associated with ethanol. To test this hypothesis, male and female adolescent C57BL/6J mice were used in an intermittent access to ethanol (IAE) model using a 2-bottle choice protocol. Before each drinking session, mice received an administration of MY10 (60 mg/kg, i.g.), or vehicle as control, and ethanol consumption was measured. At the end of the 4-weeks IAE protocol, brains were removed for immunohistochemistry analysis of Iba-1 and GFAP in prefrontal cortex and doublecortin (DCX) in dentate gyrus. In acute ethanol experiments, mice were treated with MY10 one hour before the administration of 6 g/kg ethanol (i.p.). Eighteen hours after ethanol administration, brains were dissected and subjected to the same immunohistochemistry analysis. Adolescent male mice treated with MY10 drank less ethanol than controls and showed a reduced preference for the ethanol solution in the IAE model. In contrast, MY10 did not seem to have relevant effects on ethanol intake in female mice. This effect of MY10 was not accompanied by significant alterations in glial responses. In acute ethanol experiments, we observed an overall increased activation of microglia in female adolescent mice, especially in those treated with MY10 and ethanol, suggesting gender differences in RPTPβ/ζ function. Acute ethanol induced a significant decrease on hippocampal neurogenesis in both male and female adolescent mice, which was completely prevented by treatment with MY10 in both sexes. In summary, inhibition of RPTPβ/ζ significantly reduced ethanol consumption in the IAE model only in male mice. RPTPβ/ζ critically modulates ethanol-induced decreases on hippocampal neurogenesis in both male and female adolescent mice.

Disclosures: M. Galán Llario: None. **M. Rodríguez-Zapata:** None. **T. Fontán:** None. **E. Gramage:** None. **M. Vicente-Rodríguez:** None. **C. Pérez-García:** None. **M.C. Ovejero-Benito:** None. **J.M. Zapico:** None. **A. Ramos:** None. **B. de Pascual-Teresa:** None. **G. Herradon:** None.

Poster

P736. Alcohol Intake and Preference: Mechanisms I

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Program #/Poster #: P736.06

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant AA027301

Title: Withdrawal from oxycodone dependence in mice promotes sex specific increases in alcohol drinking

Authors: A. BOISVERT[1], S. HUITRON-RESENDIZ[2], A. J. ROBERTS[2], *A. KIMBROUGH[1]; [1]Basic Med. Sci., Purdue Univ., West Lafayette, IN; [2]Animal Models Core, The Scripps Res. Inst., La Jolla, CA

Abstract: Substance use disorder (SUD) is a major societal problem throughout the world. In humans, one substance of abuse is commonly used to self-medicate withdrawal symptoms from dependence of another substance of abuse. As such, withdrawal from a specific drug may modulate the intake of another drug to promote excessive use. Preclinical models of self-medication and are not readily available, thus the present study sought to design a preclinical model of self-medication and determine if withdrawal from opioid dependence would modulate alcohol drinking behavior. Mice (n=40) were given daily two-bottle choice (2BC) access for two hours to 15% alcohol and water over two weeks (weeks 1-2) to determine baseline of alcohol drinking behavior. After baseline testing, for one week (week 3) half of the mice (n=20; 10 female, 10 male) were given twice daily injections of oxycodone (12 hours apart; 5 m/kg initially and ramping up to 20 mg/kg in later weeks), while the other half (n=20; 10 female, 10 male) were injected with saline. The following week (week 4), mice underwent 2BC drinking, without daily oxycodone injections. This two-week cycle of oxycodone injections for 1 week, followed by 2BC drinking for 1 week, was repeated for a total of 5 cycles. After the 5th week of 2BC post-oxycodone injections, mice were tested initially after 72 hours of withdrawal for thermal pain sensitivity using the tail-flick test. The mice were tested again after 96 hours of withdrawal, but immediately after a 2BC alcohol session to determine if alcohol could alleviate the presence of heightened pain sensitivity.In females, mice in withdrawal from oxycodone showed a significant increase in alcohol intake compared to mice injected with saline on post oxycodone weeks 3-5. Male mice did not show an effect of oxycodone on alcohol drinking. When tested for thermal pain sensitivity, female mice in withdrawal from oxycodone showed a significantly faster latency to withdrawal their tail compared to saline injected controls. Interestingly, this difference was eliminated after the mice had a 2BC alcohol session prior to testing. Males did not show differences in thermal pain sensitivity across groups. The present study suggests that oxycodone withdrawal promotes alcohol intake in a sex dependent manner. Similarly,

oxycodone withdrawal appears to modulate pain sensitivity in a sex dependent manner that can be alleviated by alcohol drinking. These results suggest females may be more susceptible to excessive alcohol drinking if already dependent on oxycodone. Future studies will explore brain mechanisms involved in the co-use of alcohol and opioids.

Disclosures: A. Boisvert: None. **S. Huitron-Resendiz:** None. **A.J. Roberts:** None. **A. Kimbrough:** None.

Poster

P736. Alcohol Intake and Preference: Mechanisms I

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Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant AA017823
NIH Grant DA048336

Title: Brief-access to ethanol induces an increase in ethanol taste detection threshold without affecting its salient taste quality: a potential role for the insula in escalation of ethanol intake

Authors: *A. MUKHERJEE, M. PALADINO, S. HONEYCUTT, G. LONEY;
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Abstract: Alcohol use disorder (AUD) is a critical public health concern, characterized by escalating drinking despite adverse outcomes. Preclinical studies demonstrate that animals given intermittent 24-h access to ethanol (EtOH) escalate their intake of EtOH and display blunted sensitivity to quinine-adulteration of EtOH. Previously, we have shown that rats undergoing intermittent brief-access EtOH drinking (10s presentations across 30 min) rapidly escalate EtOH intake and display decreased sensitivity to a quinine (QHCl)-adulteration of EtOH but not sucrose. Here, we examined whether the escalation of EtOH intake following brief-access exposure, and subsequent QHCl-insensitivity, was associated with changes in the psychophysical perception of the taste of EtOH. Briefly, mildly water-restricted EtOH-exposed and naïve rats were trained in a taste-detection paradigm such that they made one operant response when given a taste stimulus (0.4 mM QHCl or 200 mM sucrose) and a separate, distinct operant response when given water. Next, these rats were tasked with detecting various concentrations of EtOH (0.469% to 5%) amongst water trials. Prior brief-access EtOH significantly increased EtOH taste detection thresholds, such that a higher ethanol concentration was required in order for exposed rats, relative to naïve, to reliably distinguish between EtOH and water. In a separate cohort of rats, we examined whether brief-access exposure altered the salient taste quality (i.e. ‘bitterness’ or ‘sweetness’) of EtOH. Here, rats were trained to make one operant response when presented quinine and a separate operant response when presented sucrose. Next, the rats were randomly

given various EtOH concentrations (1.25% to 40%) and their responding on the QHCl ('bitter') or sucrose ('sweet') operandum was measured. Generally, rats reported that the taste of EtOH became more sucrose-like as concentration was increased. Importantly, there were no differences between exposed and naive animals in the perceived taste quality of EtOH. In other words, rats that had escalated their EtOH intake did not find EtOH to taste less 'bitter' or more 'sweet'. As such, the reduced sensitivity to QHCl-adulteration following escalation of EtOH intake is likely not a sensory phenomenon, but a motivational one. Currently we are examining the necessity of the insular cortex (IC) to this escalation of EtOH drinking and change in taste perception with pharmacological and chemogenetic inactivation studies. Furthermore, we are probing the relevant IC-afferents to this escalation, particularly connections with the nucleus of the solitary tract and parabrachial nucleus.

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Poster

P736. Alcohol Intake and Preference: Mechanisms I

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Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant M00234

Title: Alcohol intake and preference are regulated by striatal expression of Bmal1 and Per2 in male and female mice

Authors: *N. DE ZAVALIA, S. FERRARO, K. SHÖTTNER, P. SOLIS, G. PARENT, S. AMIR;
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Abstract: Alcohol use disorders (AUDs) are among the most severe public health problems worldwide and have dramatic consequences for individuals' health and productivity, their families, and society. Drastic differences have been identified between men and women in the propensity for alcohol abuse and their biological response to alcohol. A growing body of literature connects perturbations in circadian rhythms and the genes that control the molecular clock to the development and progression of AUDs. The striatum plays a crucial role in alcohol's reinforcing effects, as well as in the development and maintenance of AUDs. The aim of this work was to study ethanol intake in conditional knock-out mice that lack the *Bmal1* or *Per2* clock gene specifically in the striatum. We created a conditional *Bmal1* or *Per2* knock-out mouse using the Cre-loxP system. *Bmal1* or *Per2 Floxed* mice were crossed to *Gpr88-Cre* mice to specifically knock out *Bmal1* or *Per2* in Gpr88 expressing striatal medium spiny projection neurons (MSN). Twelve-18 week old WT, Knock-out (KO) and Heterozygote (HET) male and

female mice were tested in a 24 h intermittent ethanol (15% v/v) access protocol or sucrose (0.25 or 2% m/v) access protocol. Selective deletion of *Bmal1* or *Per2* mRNA and protein was observed in the striatum of KO mice compared with WT. Deletion of *Bmal1* from MSN disrupted the striatal expression of *Per2* and the clock-controlled gene *Dbp*. The lack of striatal *Bmal1* or *Per2* expression did not affect circadian locomotor activity rhythms and sucrose intake, but it significantly altered voluntary alcohol intake and preference. Strikingly, *Bmal1 and Per2* KO male mice drink significantly more alcohol and have a higher alcohol preference than WT mice (n=10/group, $P < 0.05$). *Bmal1* KO female mice drink significantly less alcohol and have a lower alcohol preference than WT mice (n=10/group, $P < 0.05$). No significant differences in alcohol intake and alcohol preference between *Per2* KO and WT female mice were observed. Interestingly, in both males and females, deletion of one allele of *Bmal1* or *Per2* did not affect alcohol intake and preference. Alcohol behaviour in Gpr88Cre/+ male and female mice was similar to Gpr88+/+ controls. These results demonstrate that a *Bmal1*/*Per2*-dependent mechanism in MSN of the striatum plays a sexually dimorphic role in the control of alcohol intake in mice, conferring protection from excess drinking in males and risk in females. Further studies are being carried out to determine the mechanisms that explain how clock genes exert their protective function and to understand the sex differences observed in this study.

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Poster

P736. Alcohol Intake and Preference: Mechanisms I

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Program #/Poster #: P736.09

Topic: G.09. Drugs of Abuse and Addiction

Support: R01 AA027682 (Ron)

Title: Alcohol activates Rac1 signaling in the dorsomedial striatum and contributes to alcohol drinking behavior in female and male mice

Authors: *Z. HOISINGTON[1], K. PHAMLUONG[1], Y. EHINGER[2], D. RON[3];
[2]Neurol., [1]Univ. of California San Francisco, San Francisco, CA; [3]Dept. of Neurol., UCSF, San Francisco, CA

Abstract: Alcohol activates Rac1 signaling in the dorsomedial striatum and contributes to alcohol drinking behavior in female and male mice

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The small G-protein Rac1 is a signaling molecule that promotes cytoskeletal rearrangement. We

and others previously showed that alcohol produces alterations in neuronal structure in the striatum [1-3]. We therefore examined the contribution of Rac1 to alcohol-dependent neuroadaptations in the striatum. To do so, mice underwent 7 weeks of intermittent access to 20% alcohol using a two bottle choice paradigm (IA20%2BC) and Rac1 activity was measured. We found that chronic excessive alcohol intake activates Rac1 in the dorsomedial striatum (DMS) and that the activation of Rac1 was maintained after 24 hours of alcohol withdrawal. In contrast, IA20%2BC did not activate Rac1 in the neighboring dorsolateral striatum (DLS). Consumption of 10% alcohol or sucrose did not alter Rac1 activity in the DMS, and the furthermore activity of a closely related small G-protein, RhoB, was unaltered by alcohol in the DMS. We further show that the consequence of alcohol-dependent Rac1 activation in the DMS is the stimulation of LIM kinase (LIMK), and the phosphorylation of its substrate, Cofilin. Next, to examine if Rac1 contributes to alcohol intake, we systemically administered Rac1 inhibitor NSC23766 (10mg/kg) to female and male mice that had undergone 7 weeks of IA20% 2BC paradigm 3 hours before a drinking session. We found that that systemic administration of NSC23766 reduces alcohol but not sucrose intake in female and male mice without altering water intake locomotion, anxiety, or blood alcohol concentration. Together, our data suggest that Rac1 plays a role in cellular and behavioral adaptations that drive and/or maintain alcohol use.

1. Wang, J., et al., *Alcohol Elicits Functional and Structural Plasticity Selectively in Dopamine D1 Receptor-Expressing Neurons of the Dorsomedial Striatum.* J Neurosci, 2015. **35**(33): p. 11634-43.
2. Laguesse, S., et al., *mTORC2 in the dorsomedial striatum of mice contributes to alcohol-dependent F-Actin polymerization, structural modifications, and consumption.* Neuropsychopharmacology, 2018. **43**(7): p. 1539-1547.
3. Laguesse, S., et al., *Prosapip1-Dependent Synaptic Adaptations in the Nucleus Accumbens Drive Alcohol Intake, Seeking, and Reward.* Neuron, 2017. **96**(1): p. 145-159.
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Disclosures: Z. Hoisington: None. **K. Phamluong:** None. **Y. Ehinger:** None. **D. Ron:** None.

Poster

P736. Alcohol Intake and Preference: Mechanisms I

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Support: NIH Grant R01AA026347
NIH Grant P50AA022537

Title: The effects of adolescent social isolation stress on binge drinking and cognitive behaviors in C57BL/6J mice

Authors: *J. LODHA, A. MORGAN, A. B. PAIS, A. C. PAIS, G. ELMISURATI, M. F. MILES, J. T. WOLSTENHOLME;
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Abstract: Rationale: Adolescence is characterized by high levels of social interaction and increased risk-taking behavior. Ethanol is one of the most commonly abused substances among adolescents. Due to their increased susceptibility to the rewarding effects and decreased susceptibility to its negative effects, adolescents tend to drink more than adults, often drinking in binges. Disruptions of cortical development by social isolation and/or binge ethanol during this critical period can lead to attention, social, and cognitive deficits later in life, and increase risk for alcohol use disorders. An individual's response to ethanol can be affected by early life stress, social interaction, and sex.
Objectives: Identify long-term behavioral and neurobiological consequences of adolescent social isolation stress and binge drinking in adult male and female C57BL/6J mice.
Methods: Social isolation was used as a social stressor during adolescence. C57BL/6J mice were single or neighbor housed with or without intermittent access to ethanol (2-BC 15% w/v EtOH) from P28-58. In another set of experiments, we used an adolescent (P27-44) drinking in the dark (DID) model (20% v/v EtOH) to test the effects of isolation stress on drinking behavior in single and group housed mice. In a third DID experiment, we modified the neighbor cages by removing the ports so animals could roam freely among the cages. Following DID, mice underwent novel object recognition as adults. BECs were correlated to intake for the DID mice.
Results: Although neighbor housing did not alter drinking behaviors in adolescence or adulthood, it did reduce cognitive deficits compared to single housed mice. Neighbor housing facilitated sharing of nesting material among female mice. During adolescence, neighbor housed ethanol drinkers built their nests significantly further from shared ports compared to water drinkers. In the DID study, socially isolated males and females drank significantly more ethanol than their group housed counterparts when drinking occurred under social conditions.
Conclusions: Social isolation during adolescence leads to cognitive deficits in adult animals, indicative of impairments to the prefrontal cortex. Neighbor housing ameliorated cognitive deficits but did not alter drinking behavior in adolescence or adulthood. In the DID model, social isolation increased adolescent drinking in males and females. Females drank more than males in both the 2-BC and DID models. Further investigation is needed into cognitive deficits following adolescent DID and social isolation, their effects on adult drinking, and underlying morphological and genomic changes in the prefrontal cortex.

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Poster

P759. Opioids: Reinforcement, Seeking, and Reinstatement VI

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P759.01

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH grant ZIA000069
NIDA IRP

Title: The Rewarding Effects of Ketamine are Mediated through Opioid Receptors

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Abstract: In recent years the use of ketamine enantiomers for the treatment of depression has gained traction. Ketamine is a racemic mixture consisting of the R-ketamine and S-ketamine enantiomers. Despite its promising efficacy, ketamine's antidepressant mechanism of action is still under debate. Ketamine's antagonism at N-methyl D-aspartate receptors (NMDAR) cannot fully explain ketamine's antidepressant effects and other putative targets have been characterized (including opioid receptors). Previous experiments from our lab showed that S-ketamine has higher affinity for NMDAR, mu-opioid receptors (MOR) and kappa-opioid receptors (KOR) than R-ketamine. We also showed that rats self-administered S-ketamine but not R-ketamine, and that the opioid antagonist naltrexone partially blocked S-ketamine induced locomotion in mice. To further elucidate the rewarding properties of S-ketamine in a rat self-administration model, we trained Sprague Dawley rats (n=10, 5 male, 5 female) to self-administer S-ketamine on a fixed-ratio schedule (FR1) (0.5mg/kg/infusion, I.V., 1-hour sessions 6 days/week) for a 16-day training phase. We used selective drugs to probe the putative receptor targets involved in S-ketamine seeking behavior. Once the rats were trained and responded steadily for S-ketamine we pretreated them with multiple doses for each of the following drugs: TRV130 (MOR partial agonist, S.C.), buprenorphine (competitive opioid receptor agonist, S.C.), and MK-801 (non-competitive NMDAR antagonist, S.C.) . Our previous PET studies with [^{18}F]-fallypride also indicated a divergence between the dopaminergic actions of S-ketamine and R-ketamine, since R-ketamine was not self-administered, we wanted to test the involvement of dopamine in the self-administration behavior. Hence, we pre-treated rats with the D2-like antagonist Haloperidol (0.1mg/kg, S.C.) and found rats did not change their S-ketamine intake. Instead, rats responded lower when pretreated with partial opioid receptor agonists and MK-801 at doses that did not significantly decrease food intake. These results suggest that binding to both opioid MOR receptors and NMDAR are necessary for the rewarding effects of S-ketamine. This work was supported by NIDA/NIH.

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Poster

P759. Opioids: Reinforcement, Seeking, and Reinstatement VI

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Program #/Poster #: P759.02

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant ZIA000069

Title: The abuse liability of methadone's enantiomers

Authors: *M. LEVINSTEIN, M. MICHAELIDES;
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Abstract: Methadone is a synthetic opioid used for chronic pain management and for opioid maintenance therapy; this racemic mixture is comprised of a 50/50 mixture of *d*- and *l*-methadone. Methadone and its enantiomers have nanomolar affinity for the mu opioid receptor; however, these compounds also interact with other receptors, specifically the N-methyl-D-aspartate receptor (NMDAR). Recently, *d*-methadone has gained interest as a potential rapid-acting antidepressant due to its activity at the NMDAR and it is assumed to be less active as an opioid. Here we test the abuse liability of *d*-, and *l*- methadone utilizing pharmacological and behavioral assays. In order to understand the pharmacological effects of these compounds more fully, we first screen them for competitive binding or inhibition against a selected panel of receptors and enzymes, including known targets for addictive drugs and medications. We also investigate the drug abuse related behavior in mice. We first test psychomotor sensitization by repeatedly injecting mice with vehicle, *d*-methadone, or *l*-methadone and measuring locomotor activity for three days. Three days later, we inject all mice with escalating doses of *d*-methadone in a single test session and assessed locomotor activity. Additionally, we test the rewarding effects of *d*- or *l*-methadone using the conditioned place preference (CPP) procedure.

Disclosures: M. Levinstein: None. **M. Michaelides:** None.

Poster

P759. Opioids: Reinforcement, Seeking, and Reinstatement VI

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Program #/Poster #: P759.03

Topic: G.09. Drugs of Abuse and Addiction

Support: Swedish Research Council, VR project number 2018-02320

Title: Sex differences in choosing oxycodone over healthy rewards

Authors: V. SCHWABL[1], M. ATUDOREI[1], G. AUGIER[1], *E. AUGIER[2];
[1]Lïnköping Univ., Linköping, Sweden; [2]Dept. of Clin. and Exptl. Med., Linköping, Sweden

Abstract: Use of addictive substances accounts for almost 10% of global disease burden. Opioid misuse in particular has recently become a major concern and a rapidly evolving public health crisis that requires innovative scientific approaches. Oxycodone is one of the most abused prescription opioids, but despite its prevalent misuse, the number of preclinical studies investigating oxycodone-seeking behaviors is relatively limited. Furthermore, opioid use disorder is characterized by a progressive shift of decision making, in which opioids are increasingly chosen over healthy non-drug rewards. Only a subset of people transition from recreational to pathological use. By contrast, in commonly used animal models, nearly all rats learn to self-administer addictive drugs, including opioids and animals have no alternative to drug use.
The primary goal of the present study was therefore to investigate potential sex differences in oxycodone self-administration and choice of oxycodone at the expense of healthy rewards. Using operant conditioning, we first trained male and female rats to self-administer oxycodone orally. We found that males earned more oxycodone reinforcers and showed a higher motivation for oxycodone. Blood oxycodone metabolites concentrations were in addition strongly correlated with oxycodone reinforcers earned during self-administration. Finally, we used an exclusive choice-based method to identify both male and female rats that continue to self-administer oxycodone at the expense of a high-value natural reward, a sweet solution, and found that, in agreement with our previous work with alcohol, only a minority of outbred rats (10% or less) choose oxycodone over an alternative high-value reward.
Together, our results indicate profound sex differences in oxycodone-related behaviors. They also confirm and extend to oxycodone previous data obtained with preclinical choice models.

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Poster

P759. Opioids: Reinforcement, Seeking, and Reinstatement VI

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Topic: G.09. Drugs of Abuse and Addiction

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NIDA Grant T32 DA035200

Title: Peer-induced remifentanil seeking in male and female rats

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Abstract: Re-association with drug-associated peers contributes to relapse in humans, and we have recently found that exposure to cocaine-associated peers triggers reinstatement in rats. Here we extend these findings to opioids by pairing one peer with the fast-acting opioid remifentanil and a different peer with saline. In two experiments, male (n=10) and female (n=11) Sprague Dawley rats (8-9 weeks of age) received 28 days of twice-daily self-administration on a terminal FR5 schedule. Each day, remifentanil (6 µg/kg/infusion) was available during one session in the presence of one same-sex peer (S+) and saline was available during another session in the presence of a different peer (S-). Following extinction, we tested reinstatement to the S+, S-, or no peer; Experiment 2 also included a novel peer. Following pre-training, rats in Experiments 1 and 2 increased active lever pressing for remifentanil but not saline across sessions. Rats also decreased their active lever pressing across extinction. Compared to the no peer condition, rats in Experiment 2 increased active lever pressing in response to the S+, S-, and novel peers; there was no difference between S+, S-, and novel partner-induced active lever pressing. When combining the results from both experiments to power for potential sex differences in peer-induced reinstatement, we found that females consistently pressed the active lever more than males. Furthermore, with both experiments combined, rats increased active lever pressing in response to the S+ and S- partners, with the S+ peer eliciting significantly greater responding compared to the S- peer regardless of sex. Thus, similar to cocaine, these experiments show that re-exposure to a remifentanil-associated peer after extinction elicits greater drug seeking relative to a saline-associated peer in both males and females. However, drug seeking was also triggered by the S- peer and a novel peer, indicative of either stimulus generalization (associative) and/or social facilitation (non-associative).

Disclosures: S. Malone: None. **L. Hammerslag:** None. **M. Carbone:** None. **S. DiMeo:** None. **M. Bardo:** None.

Poster

P759. Opioids: Reinforcement, Seeking, and Reinstatement VI

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P759.06

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant DA042792

Title: Examining the effect of neuropathic pain on drug-seeking ensembles in the dmPFC

Authors: *B. C. SARKA[1], Q.-S. LIU[2], C. L. STUCKY[3], S. LIU[1], C. M. OLSEN[4]; [1]Pharmacol. and Toxicology, [2]Dept. of Pharmacol. and Toxicology, [3]Cell Biology, Neurobio. & Anat., [4]Neurosci. Res. Ctr., Med. Col. of Wisconsin, Milwaukee, WI

Abstract: Approximately 50 million Americans suffer from chronic pain, and opioids are commonly prescribed for such individuals. While short term use can still pose a risk, those who take opioids for over a year are at an increased risk for long term opioid use. It is not uncommon for patients to require dose increases to maintain analgesic affects because tolerance builds up. Unfortunately, nearly a quarter of chronic pain patients have reported misusing their prescription at some point. We are currently investigating the effect that chronic pain has on drug-seeking behavior at the neuronal level. Drug-seeking behavior is associated with reactivation of an ensemble of neurons sparsely scattered throughout the dorsomedial prefrontal cortex (dmPFC). Prior research has demonstrated that chronic pain leads to an increase in intrinsic excitability of neurons in the dmPFC. We tested the hypothesis that chronic pain would increase oxycodone seeking behavior, and that the pain state would differentially increase intrinsic excitability in dmPFC drug seeking ensemble neurons. TetTag mice were trained to self-administer oxycodone (via intravenous infusion) for 10 days. After 7 days forced abstinence, a drug seeking session (extinction conditions) was performed and the ensemble was tagged. Mice received spared nerve injury (SNI) to induce chronic pain during the period between a first and second seeking session. Shortly thereafter we performed electrophysiology on individual neurons within the dmPFC to assess intrinsic excitability of the drug-seeking ensemble and non-ensemble neurons. Additionally, we compared the two seeking sessions to see if the SNI exacerbates seeking behavior. We have seen mild, insignificant trends in seeking behavior comparing SNI mice compared to Sham. However, the intrinsic excitability of SNI dmPFC neurons differs from Sham dmPFC neurons, and significant differences have been detected between distinct drug-seeking ensemble neurons versus non-ensemble neurons. We are also beginning to look at sex differences, and early data suggests that females may show an increase in neuronal intrinsic excitability compared to males.

Disclosures: B.C. Sarka: None. **Q. Liu:** None. **C.L. Stucky:** None. **S. Liu:** None. **C.M. Olsen:** None.

Poster

P759. Opioids: Reinforcement, Seeking, and Reinstatement VI

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P759.07

Topic: G.09. Drugs of Abuse and Addiction

Support: U01DA050243
U01DA044399

Title: Effect of Oxycodone behaviors in Cacna1h knockout mice

Authors: *W. B. LYNCH[1,2], J. A. BEIERLE[1,2,3], R. H. COLE[1], R. BHANDARI[1], A. ARSLAN[4], G. PELTZ[4], C. D. BRYANT[1];

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Abstract: The genetic basis of Opioid use disorder (OUD) is still largely unknown. Quantitative trait locus mapping in rodent models offers several advantages to human GWAS (controlled sample sizes, controlled environment, within-species gene validation) and can elucidate genetic causes of addiction-relevant model phenotypes. To identify genetic variants of OUD-relevant behaviors in mice, we surveyed 29 strains of mice for strain-specific differences in behavioral responses to the semi-synthetic opioid oxycodone (OXY). Using an OXY conditioned place preference (CPP) experimental design, we found robust strain differences in OXY-induced locomotion, place preference, and state-dependent learning, indicative of causal genetics for these phenotypes. Subsequent haplotype association mapping identified a region on chromosome 17 associated with OXY-induced locomotion containing the candidate gene *Cacna1h*, encoding a voltage-gated calcium channel subunit. We subsequently tested *Cacna1h* knockout (KO) mice on a mixed 129/C57BL/6N background backcrossed to C57BL/6N to the N2 generation (and then subsequently intercrossed) in OXY CPP, in addition to hot plate analgesia, elevated plus maze, and light-dark conflict to assess differential responses to OXY tolerance and withdrawal between genotypes. We did not observe any significant genotypic differences in distance traveled in response to OXY. However, *Cacna1h* KOs with previous OXY history travel shorter total distances during the place preference session. The effect of *Cacna1h* KO was specific to locomotion associated with OXY CPP, as there were no genotypic differences in OXY CPP or state-dependent learning, nor were there any significant differences OXY antinociceptive tolerance or spontaneous withdrawal in the elevated plus maze or the light/dark box. Our negative findings fail to support a role for Cacna1h in opioid addiction model behaviors, with the notable limitation that we have only tested a single genetic background.

Disclosures: W.B. Lynch: None. **J.A. Beierle:** None. **R.H. Cole:** None. **R. Bhandari:** None. **A. Arslan:** None. **G. Peltz:** None. **C.D. Bryant:** None.

Poster

P759. Opioids: Reinforcement, Seeking, and Reinstatement VI

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Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P759.08

Topic: G.09. Drugs of Abuse and Addiction

Support: R01 DA 019958
R21 A 049565

Title: Engagement of Beta-Arrestin 2 at the μ-opioid receptor reduces the abuse liability of opioids in mice during operant self-administration

Authors: *L. FELTH, Z. ROSA, S. W. GOODING, J. GIPOOR, A. GAUR, J. WHISTLER; Univ. of California, Davis, Davis, CA

Abstract: In 2019, 500,000 people died as a result of opioid overdose. About 10% of persons prescribed opioids will develop an opioid use disorder (OUD). The explanation for why some individuals transition to misuse of opioids, placing themselves at higher risk for overdose, remains unclear. Despite their significant risks, opioid analgesics, including morphine, codeine, Vicodin and Oxycontin, are still regarded as the gold standard for alleviating pain clinically. All of these drugs exert their analgesic effects through activation of a G protein coupled receptor (GPCR), specifically the G_i coupled μ- Opioid receptor (MOR). When MOR is activated by its endogenous ligand, a regulatory molecule called βeta-Arrestin-2 (βAR2) is recruited to the receptor, causing titration of signal transduction and subsequent internalization and recycling of the receptor. Conversely, when MOR is activated by many clinically relevant opioid drugs, such as morphine, it does not effectively recruit βAR2. The lack of βAR2 recruitment results in receptors remaining present on the membrane without being internalized and recycled back to the membrane. In short, in the presence of endogenous ligand, G-protein signal transduction follows an on/off pattern, whereas in the presence of exogenous drugs signaling remains 'on' and causes homeostatic adaptions, such as cAMP superactivation. This cAMP superactivation has been implicated in neuronal and behavioral plasticity associated with correlates of OUDs seen in mice, including compulsive drug seeking. We hypothesize that effective recruitment of βAR2 to activated MORs will reduce compulsive drug seeking in mice. To investigate this hypothesis, this study utilizes four different genotypes of mice, with varying βAR2 recruitment profiles in response to morphine: WT(poorly recruits βAR2), RMOR (strongly recruits BAR), βAR2-KO (does not recruit βAR2), βAR2-KO/RMOR (does not recruit βAR2). We utilized a novel oral operant self-administration paradigm for 16 weeks to model the transition from impulsive to compulsive drug use. At the end of 16 weeks, operant self-administration was extinguished and reinstatement was assessed to model OUDs. Our results suggest that βAR2 recruitment influences OUDs and that drugs that recruit βAR2 will have a reduced abuse liability.

Disclosures: L. Felth: None. **Z. Rosa:** None. **S.W. Gooding:** None. **J. Gipoor:** None. **A. Gaur:** None. **J. Whistler:** None.

Poster

P759. Opioids: Reinforcement, Seeking, and Reinstatement VI

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P759.09

Topic: G.09. Drugs of Abuse and Addiction

Support: St. Michael's Hospital Foundation
Lung Health Foundation
J. P. Bickel Medical Grant

Title: Identification of novel pain therapies without the side-effect of respiratory depression using phenotype-based drug discovery in larval zebrafish

Authors: S. ZAIG[1], D. SRIKRISHNARAJ[1], C. DA SILVEIRA SCARPELLINI[1], ***G. MONTANDON**[2,1];
[1]Keenan Res. Ctr. for Biomed. Sciences, St. Michael's Hosp., Toronto, ON, Canada; [2]Univ. of Toronto, Toronto, ON, Canada

Abstract: Introduction: Opioid drugs produce pain relief as well as respiratory depression which can be lethal with overdose. Opioid overdoses are equally due to the abuse of drugs such as fentanyl or pain killers like oxycodone. During the COVID19 pandemic, the opioid epidemic reached almost 100,000 deaths, highlighting the urgent need to identify pain killers with reduced liability so they can be safely prescribed. However, most pharmaceutical approaches have so far failed due to the complexity of the nervous system and the various phenotypes associated with opioid drugs. Using our newly-developed drug discovery platform in larval zebrafish, we propose to identify new drugs with potent analgesic properties and reduced respiratory liability. **Methods:** Using phenotype-based approaches in larval AB zebrafish *in vivo,* we can quickly and effectively screen new drug compounds to either target respiratory depression or analgesia (*Zaig et al., 2021, eLife*). We first screened customized drug libraries targeting voltage-gated calcium channels and the adenylyl cyclase pathway, which are both known to mediate neuronal inhibition by opioid drugs. **Results:** To test the role of calcium channels, we combined fentanyl and the calcium channel activators nefiracetam (1µM) or FLP-64176 (1µM) with both preventing respiratory depression by fentanyl (1µM). Analgesia by fentanyl, quantified by the escape swimming response to the noxious stimulus formalin, was prevented by FLP-6476. To target the adenylyl cyclase pathway, we combined fentanyl and the phosphodiesterase inhibitors pentoxifylline (50µM), roflumilast (1µM), or dipyridamole (100µM). Pentoxifylline prevented respiratory depression by fentanyl but not roflumilast and dipyridamole. We are also genetically deleting calcium channels and phosphodiesterase molecules using CRISPR-cas9 and are developing opioid withdrawal assays to determine the impact of new drug compounds on opioid addiction. **Discussion:** The successful identification of new drug combinations that can prevent respiratory depression while preserving analgesia suggests that our drug discovery platform in larval zebrafish will substantially accelerate the identification of new opioid drugs with reduced morbidity and mortality. **Acknowledgements:** Research was funded by the St. Michael's Hospital Foundation, the Lung Health Foundation and the J.P. Bickell Foundation. SZ was supported by the Canadian Institutes of Health Research Canada Graduate Scholarship and the St. Michael's Hospital Research Training Centre Scholarship.

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Poster

P815. Encoding-Retrieval Interactions in Human Long-Term Memory

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P815.01

Topic: H.07. Long-Term Memory

Support: NSF 1633873

Title: Retrieval-related anterior shift is moderated by age and relates to memory performance

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Abstract: Cognitive aging is associated with a decline in memory for contextual details of previously experienced episodes. Recent research has shown an increasing interest in a phenomenon termed the retrieval-related 'anterior shift'. The term refers to systematic differences in the localization of neural activity observed at encoding and retrieval, such that retrieval of a perceptual image (e.g., a scene) is associated with a response in category-selective cortex that is localized more anterior than the response elicited when the image is experienced directly. However, both the moderating effects of age on anterior shift and its relationship with memory performance remain unexplored. In the present study, younger (N = 24) and older adults (N = 24) underwent functional magnetic resonance imaging (fMRI) as they completed encoding and retrieval tasks. At encoding, participants viewed words paired with images of faces and scenes. At retrieval, participants determined whether test words were new or old and, if judged old, whether the word had been studied with an image of a face or a scene. Source memory performance - the ability to correctly judge whether a word had been paired with a scene or a face - was lower in the older age group. Turning to the fMRI data, we localized peak scene- and face-selectivity for each individual participant within the face-selective precuneus (PCU) and in three scene-selective (parahippocampal place area [PPA], medial place area [MPA], and occipital place area [OPA]) regions of interest (ROIs). In line with recent research with scene stimuli, we found strong evidence for an anterior shift in the PPA, MPA and OPA. We extended these findings to demonstrate an anterior shift in the precuneus (PCU) for face stimuli, although this effect was observed in older adults only. An age group x ROI ANOVA revealed that anterior shift was larger in older relative to younger adults, and that this age effect did not differ according to ROI. Lastly, the magnitude of the anterior shift in the PPA exhibited a robust negative correlation with source memory performance (after controlling for age), indicating that greater encoding-retrieval displacement of scene-selectivity within the PPA was associated with poorer memory performance. These results shed light on the functional significance of anterior shift in relation to memory success and provide an increased understanding of sources of memory decline in older age.

Disclosures: S. Srokova: None. **P.F. Hill:** None. **M.D. Rugg:** None.

Poster

P815. Encoding-Retrieval Interactions in Human Long-Term Memory

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P815.02

Topic: H.07. Long-Term Memory

Support: Multi-University Research Initiative Grant (ONR/DoDN00014-17-1-2961)
NWO Rubicon grant (446-17-009)

Title: Effect of context-dependent temporal structure on episodic memory

Authors: *S. COLLIN[1], S. SRIVATSAN[2], A. BEUKERS[2], U. HASSON[2], K. NORMAN[2];
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Abstract: One of the key objectives of learning is to infer the latent causes of our sensory observations. This concept of latent causes is closely tied to schemas: You make sense of your current sensory observations by inferring that the current event is an instance of a particular kind of schema, which allows you to draw on prior knowledge relating to that schema to make predictions about what will happen next. Theories of schema-dependent memory posit that having a representation of the temporal structure of an event can boost memory for details from that event. However, in the real world, the temporal structure of events is often complex and context dependent. In this study, we investigated how context-dependent temporal structure is represented in the brain during narrative perception, and if and how this relates to memory. We ran a 2-day fMRI-study (N = 40) in which we exposed participants to many unique animated videos of wedding ceremonies with sequences of rituals originating from a novel (fictional) culture, where each video was accompanied by a unique narrative. Each ceremony, despite the regularities in the sequence of rituals, was unique. Participants learned the context-dependent temporal structure of the wedding ceremonies on day 1 and came back on day 2 to watch and recall novel ceremonies. The results, analyzed with multivariate pattern analysis, showed history-independent coding (i.e., only coding for the current event) in lateral occipital and superior parietal regions and history-dependent coding (i.e., coding for the event that is currently shown as well as the preceding event) in medial occipital regions that were both relevant to memory for episodic details of the ceremonies.

Disclosures: S. Collin: None. **S. Srivatsan:** None. **A. Beukers:** None. **U. Hasson:** None. **K. Norman:** None.

Poster

P815. Encoding-Retrieval Interactions in Human Long-Term Memory

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P815.03

Topic: H.07. Long-Term Memory

Support: NSF Grant 1756417

Title: Stress-testing the memory system: Inter-item distraction disrupts temporal and semantic organization in free recall

Authors: *R. A. CUTLER[1], N. W. MORTON[2], S. M. POLYN[1];
[1]Psychological Sci., Vanderbilt Univ., Nashville, TN; [2]The Ctr. for Learning & Memory, The Univ. of Texas at Austin, Austin, TX

Abstract: Temporal information situates episodic memories along a personal timeline, while semantic information draws from an existing knowledge store to imbue those episodes with meaning. We present a set of fMRI and behavioral experiments that examine the interactions of temporal and semantic information in memory search. Participants studied lists of 27 items from 3 categories that elicit distinct patterns of brain activity (celebrities, landmarks, objects). Same-category items were presented in blocks of 3 to capture the combined effect of temporal proximity and semantic similarity. During the study period participants were challenged with an inter-item distraction task; *light distraction* was a simple vigilance task while *heavy distraction* was mental arithmetic. We found that heavy distraction disrupted the temporal and semantic organization of recall sequences, suggesting that a demanding task during encoding interferes with a cognitive process that takes advantage of patterns within the list structure. This drop in recall performance was associated with decreased category representation in brain regions that facilitate memory search. Our neural analyses show that increased inter-item distraction can be captured as a decrease in a classifier's ability to decode the category-membership of an item. Distraction also decreases the correspondence between neural patterns and formal models that capture semantic structure. This suggests that increasing the intensity of an inter-item task disrupts a search process that relies on the relational structure of the list to retrieve items from memory. We interpret the behavioral and neural consequences of distraction in terms of a retrieved context model of memory search.

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Poster

P815. Encoding-Retrieval Interactions in Human Long-Term Memory

Location: Virtual Only

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Program #/Poster #: P815.04

Topic: H.07. Long-Term Memory

Support: NIH R01 NS102201

Title: Relating the strength of conjunctive representations to episodic retrieval effects in task-switching

Authors: *B. RANGEL[1], E. HAZELTINE[2], J. R. WESSEL[2];
[2]Psychology, [1]Univ. of Iowa, Iowa City, IA

Abstract: Goal-directed behavior necessitates the combination of external stimuli with appropriate actions and their associated intended outcomes. For example, during the goal-directed behavior of unlocking a door, an external stimulus (door) needs to be paired with an appropriate action (retrieving/inserting the key) and an intended outcome (the opening). Prominent cognitive theories such as the Theory of Event Coding propose that the temporal association of stimulus, action, and outcome representations leads to their integration into a joint episodic memory trace, also known as an "event-file" (Hommel 2009). While the retrieval of such event-files from memory allows for more efficient reactivation of their contained representations when necessary, this retrieval process is automatic, and may not always be contextually appropriate – especially when the environmental contingencies changed. For instance, one may find oneself trying to retrieve the key for a door despite having lost it the day before. In the laboratory, such scenarios are simulated using task-switching paradigms in which contextual task stimuli first cue a task with a preexisting event file, but are then followed by a task conditions that are incommensurate with the action contained in said event file – resulting in a prominent "repetition cost" to reaction time (Hommel et al. 2014; Neill 1997). There is currently ongoing debate about whether this repetition cost is attributable to inhibitory effects (e.g., Mayr & Keele, 2000; Mayr 2002) or indeed due to the retrieval of the event file (e.g., Grange & Kowalczyk, 2017; Grange 2018). Recently, Kikumoto & Mayr (2020) used Linear Discriminant Analysis and Representational Similarity Analysis of Electroencephalography data to produce the first direct neurophysiological evidence for the existence of conjunctive representations (i.e., event-files) in the human brain. Utilizing these methods, we successfully estimated the representational strength of each individual task component (cue/target/response) and their combined conjunctive representation (i.e. the event-file), on a trial-by-trial basis in a sample of N=35 subjects performing a task switching paradigm. A mixed-model analysis demonstrated that event-file strength in a previous trial predicts RT in the current trial when retrieval is prompted by a repeated cue. These results provide the first neurophysiological evidence that the Repetition Cost incurred by incongruent event files can be partially attributed to the retrieval of conjunctive representations - i.e., to the neural representation of past event-files.

Disclosures: B. Rangel: None. **E. Hazeltine:** None. **J.R. Wessel:** None.

Poster

P815. Encoding-Retrieval Interactions in Human Long-Term Memory

Location: Virtual Only

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Program #/Poster #: P815.05

Topic: H.07. Long-Term Memory

Support: NSF CRCNS grants IIS-1822683 (to K.K.)
NSF CRCNS grants IIS-1822929 (to T.N.)

Title: Repetition-related memory signals in parietal cortex integrate information about stimulus content

Authors: *Y. ZHAO[1], E. J. ALLEN[4], Y. WU[6], T. NASELARIS[7], K. N. KAY[5], B. A. KUHL[2], J. HUTCHINSON[3];
[2]Psychology Dept., [3]Dept. of Psychology, [1]Univ. of Oregon, Eugene, OR; [4]Psychology, [5]Radiology, Univ. of Minnesota Twin Cities, Minneapolis, MN; [6]Univ. of Minnesota, Minneapolis, MN; [7]Neurosciences, Med. Univ. of South Carolina, Charleston, SC

Abstract: When a stimulus from the past is re-encountered, this can elicit increased neural activation relative to a novel stimulus (repetition enhancement) or decreased activation (repetition attenuation). Studies of episodic memory have consistently found that repetition enhancement occurs within parietal cortex and that these enhancement effects are related to behavioral expressions of successful episodic remembering. Repetition attenuation, in contrast, is more typically observed in regions of occipitotemporal cortex and is less consistently related to behavioral expressions of episodic memory. Separately, pattern-based fMRI studies have found that information about the content of stimuli is reflected in both parietal cortex and ventral temporal cortex, but it is less clear how or whether these content representations are integrated with repetition-related memory signals. In the current 7T fMRI study, eight participants performed a continuous recognition task spanning 30-40 fMRI scan sessions and up to 10,000 unique, naturalistic images. We calculated the difference in multivoxel fMRI activations between the first presentation (initial encoding) and second presentation (retrieval) for each stimulus and tested whether these repetition-related differences in activity patterns carried information about the visual content of the image. To quantify the content of each image, we passed the images through VGG16, a convolutional neural network designed for image content classification and detection. We then reduced features from the output layer to 10 PCA dimensions and used ridge regression to test whether repetition-related changes in fMRI activity patterns predicted the PCA content scores. We found that repetition-related differences in medial and lateral parietal cortex predicted image content. Moreover, these predictions were significantly better for hits (correct recognition) compared to misses (failed recognition) indicating that the presence of content information was directly related to successful recognition. Interestingly, repetition-related differences in occipitotemporal cortex also predicted image content, but the success of these predictions were less dependent on successful behavioral recognition. Collectively, our results indicate that repetition-related enhancements which have consistently been observed in parietal cortex directly integrate information about the content of what is being remembered.

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Poster

P815. Encoding-Retrieval Interactions in Human Long-Term Memory

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P815.06

Topic: H.07. Long-Term Memory

Title: Retrieval practice in a recognition memory word pair paradigm

Authors: *A. T. J. CAWLEY-BENNETT[1], D. MINONDO[2], J. R. MANNS[1];
[1]Psychology, [2]Neurosci. and Behavioral Biol., Emory Univ., Atlanta, GA

Abstract: The retrieval practice effect (also termed the testing effect) is a widely-replicated and robust phenomenon demonstrating that retrieving information from memory after recent learning leads to better retention than restudying the information. For example, cued recall (table - _____) of a studied word pair (table - window) leads to better subsequent memory for the target word (window) as compared to restudying the word pair. Several hypotheses have been proposed to explain this effect, yet disagreement overshadows consensus. We sought to address this debate by examining how cued recall retrieval practice affected subsequent memory for both cue words and target words in a study in which the final test consisted of a yes-no recognition memory test of either the cue word or target word contrasted against novel words. Participants were also asked to make confidence ratings for each yes-no judgment. Performance on the final recognition memory test in the retrieval practice condition was compared to performance in a restudy control condition. Existing hypotheses regarding the retrieval practice effect disagree as to whether the subsequent memory benefit should be observed for cue words, for target words, or for both. Preliminary results indicate that cued recall retrieval practice benefitted subsequent memory for both the cue words and target words, although the effect size was smaller for cue words. Planned analyses of receiver operating characteristic (ROC) curves will address for both cue words and target words whether retrieval practice positively impacted the shape of hit rates as a function of false alarm rates as compared to the restudy control condition. The results of the present study have implications for basic memory research on the retrieval practice effect as well as classroom best practices.

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Poster

P815. Encoding-Retrieval Interactions in Human Long-Term Memory

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Program #/Poster #: P815.07

Topic: H.07. Long-Term Memory

Support: A83470
RGPIN-2016-05523
NSERC CGS-D Scholarship

Title: Implicit memory for target location facilitates explicit memory-guided attention in real-world sound-clips

Authors: *M. FISCHER[1,2], M. MOSCOVITCH[1,2], C. ALAIN[2,1];
[1]Univ. of Toronto, Toronto, ON, Canada; [2]Rotman Res., Baycrest Ctr. for Geriatric Care, Toronto, ON, Canada

Abstract: What is the relation between awareness, attention, and memory? Long-term memory (LTM) of learned target locations can facilitate target detection. However, it remains unclear what mechanisms enable this perceptual facilitation. We first created long-term associations between audio-clips and the location of an embedded tone. Then, we measured behavioural and neuroelectric activity during a cue-target detection task. An a priori sample size calculation suggested testing a minimum of thirty-two participants in order to achieve an effect size of d = .90.

At learning, thirty-five participants (22 female, M = 22.6 years) were presented with 80 different audio-clips (half included a lateralized pure tone target, referred to as memory trials), and indicated whether the tone was low or high. While the location of the tone remained constant throughout exposure, the frequency of the tone ($500_{Hz}/1_{kHz}$) and the position (i.e., time) of the tone within the audio-clip (1300_{ms}, 1800_{ms}, 2300_{ms}) varied randomly. A surprise memory test followed in which participants were told to press a button as quickly as possible when they heard a *faint* lateralized tone (target), which was present in *all* audio-clips. They also indicated if the clip was (i) old/new; and (ii) if the tone was on left/right/not present when they heard the clip at exposure.

Participants showed good explicit memory for the clip and for the presence/absence of the tone, but *not* for the *specific* location of the tone. They were also faster in responding to the target on memory trials than on trials in which the target was presented for the first time. This benefit in response times coincided with changes in the stimulus-locked sustained potential *prior* to target onset at approximately $800\text{-}1000_{ms}$ and $1800\text{-}2300_{ms}$ over frontal sites.

These findings show that LTM can facilitate signal detection even when there are variations in time and frequency of the to-be-attended material. Furthermore, *explicit* memory for both the clip and the presence or absence of the tone facilitated target detection, an effect that was accompanied by an *implicit* memory of the specific spatial location of the tone.

Disclosures: M. Fischer: None. **M. Moscovitch:** None. **C. Alain:** None.

Poster

P815. Encoding-Retrieval Interactions in Human Long-Term Memory

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Program #/Poster #: P815.08

Topic: H.07. Long-Term Memory

Support: ISF grant 1960/19 to SYG
Ariane de Rotschild Women Doctoral scholarship to KT

Title: The role of visual and motor aspects of eye movements in memory retrieval

Authors: *K. TAUB, Y. GOSHEN-GOTTSTEIN, S. YUVAL-GREENBERG;
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Abstract: Eye movements during memory encoding act as retrieval cues: evidence shows that memory retrieval is improved when eyes are shifted to the same position where they were located during retrieval. However, it is yet unknown which component of eye movements is involved in this effect: the motor component, which is the specific muscle contraction that characterizes each movement performed by the eye, or the sensory component - which is the visual image that falls on the retina. In this research we examined the role of visual and motor aspects of eye movements during memory retrieval, and investigated individual differences in the ability to benefit from these cues. In our experiment (N=72), we dissociated the visual and motor aspects of eye movements during memory encoding and retrieval. In each encoding trial, participants were asked to fixate at one of the four corners of the screen, and then followed a fixation cross to perform two sequential saccades to the left or right, towards the horizontal center. They were subsequently presented with a target word at the final landing position. The trials varied in saccade direction, left or right, and in vertical location, as the horizontal saccades were performed either at the top or bottom part of the screen. During retrieval, participants performed the same task but were also asked to indicate whether the target word was studied or unstudied. Trials were divided into four conditions, with each retrieval trial matching its accompanying encoding trial in: the direction of the saccades (motor-only cue), the vertical location (visual-only cue), both (motor-visual cue) or neither (no-cue). Results showed that while cues were beneficial to memory retrieval compared with the no-cue condition, there was no difference in performance following the appearance of a single cue (either motor-only or visual-only) and the appearance of both cues together (motor-visual cue). Findings also revealed that participants varied in their ability to benefit from eye movements as retrieval cues: while some participants profited from motor and visual cues, others did not. Finally, we found that recognition performance in the motor-only condition was positively correlated across participants with performance in the visual-only condition, even after controlling for memory capabilities. Thus, individuals who tend to benefit from eye movements as retrieval cues, tend to gain from both aspects of these cues (motor and perceptual) to a similar extent. These findings

suggest that the ability to benefit from the encoding-retrieval compatibility of eye movements is an individual ability, which is independent of memory abilities.

Disclosures: K. Taub: None. **Y. Goshen-Gottstein:** None. **S. Yuval-Greenberg:** None.

Poster

P815. Encoding-Retrieval Interactions in Human Long-Term Memory

Location: Virtual Only

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Program #/Poster #: P815.09

Topic: H.07. Long-Term Memory

Support: DGAPA-PAPIIT IN215218
DGAPA-PAPIIT IA205218
DGAPA-PAPIIT N217918

Title: Association of the ability to inhibit irrelevant information with alpha activity in the encoding of spatio-temporal information in episodic memory

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Abstract: Episodic memory is the ability for encoding, storing, and recalling memories that have been personally experienced and that contains a space-time structure. When an event is encoded, decreased alpha-band frequency has been associated with better performance in episodic memory. On the other hand, in attention tasks, decreased amplitude of alpha-band has been associated with selection of information; in contrast, an increase in alpha-band amplitude is associated with the inhibition of irrelevant information. The participation of the selection sub-process of attention within the episodic memory has been reported; however, it is unknown whether the inhibition sub-process ability of attention is associated with the encoding of target information and its spatio-temporal context. Here, we evaluated the potential relationship between alpha activity in the encoding phase of spatio-temporal context of target information in episodic memory with the ability to inhibit irrelevant information. Healthy 22 young volunteers answered an episodic memory task in which target stimuli appeared in specific spatial (up or down) and time (first or second list) context during encoding; this information had to be remembered during the recovery phase. Electroencephalographic activity was recorded during the encoding phase. Complementary, participants answered Eriksen's flanker task. In this task, subjects had to inhibit flanker distractors to answer to target stimulus. Direct correlations were found between the mean amplitude of alpha on the parietal and frontal electrodes during the encoding phase of the episodic memory task, and the percentage of correct responses in the

Eriksen task. Also, inverse correlations were found between the amplitude of alpha and reaction times of type of trial in the Eriksen task. These results, in conjunction with reports in the literature, allow us to interpret that the increase in the amplitude registered during the encoding phase of the episodic memory seems to correspond to an alpha activity associated with the inhibition sub-process of irrelevant information.

Disclosures: A. Polo-Romero: None. **U. Caballero-Sánchez:** None. **D. Zenteno-Morales:** None. **M. Méndez-Díaz:** None. **O.E. Prospéro-García:** None. **A.E. Ruiz-Contreras:** None.

Poster

P870. Place Cells: Learning and Encoding of Goals

Location: Virtual Only

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Program #/Poster #: P870.01

Topic: H.09. Spatial Navigation

Support: NIH 1K99AG068602
AFOSR FA9550-18-1-0469
NIH DP1EB025835
Harrison Gardner Jr. Award

Title: Chronic electrophysiology of hippocampal neurons with neuron-like electronics

Authors: *T. J. ZWANG[1], A. ZHANG[2], N. L. PETTIT[3], C. D. HARVEY[3], C. M. LIEBER[4], B. T. HYMAN, MD,PhD[1];
[1]Massachusetts Gen. Hosp., Charlestown, MA; [2]Chem. Engin., Stanford Univ., Palo Alto, CA; [3]Harvard Med. Sch., Boston, MA; [4]Dept. of Chem. and Chem. Biol., Harvard Univ., Cambridge, MA

Abstract: The risk of Alzheimer's disease increases with age and, after onset, pathology appears to spread or worsen with time. It is not clear why some neural cell populations become vulnerable with age, yet others are resistant. Technological limitations have prevented chronic cellular-level tracking of interconnected brain regions during the many months or years before onset of age-related cognitive defects. To overcome these limitations, we have developed techniques to track electrophysiology of individual neurons in behaving mice, monitor their interactions across brain regions, and determine how they change over long periods of time. We utilized neuron-like electronics enable stable electrophysiological recordings on the scale of one year. We implanted these electronics in the hippocampus of male and female C57BL/6J mice (n=6) and monitored head-fixed running on a ball in virtual reality. We observed the presence of place fields that lasted multiple months. Changing the virtual environment caused the place fields of these cells to remap. A second cohort of THY-Tau22 mice (n = 8) had electronics implanted in the hippocampus and entorhinal cortex and show the ability to record correlated activity of

individual neurons across brain regions. These experiments demonstrate that the use of flexible electronics that do not induce astrogliosis in nearby tissue allows for recording activity of the same neurons over periods longer than months and does not require re-positioning of electrodes or the use of microdrives. Furthermore, these data show the stability of place fields and correlated activity between brain regions for mice running through a virtual maze.

Disclosures: T.J. Zwang: None. **A. Zhang:** None. **N.L. Pettit:** None. **C.D. Harvey:** None. **C.M. Lieber:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Owner of patents regarding mesh electronics. **B.T. Hyman:** None.

Poster

P870. Place Cells: Learning and Encoding of Goals

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Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P870.02

Topic: H.09. Spatial Navigation

Support: Postdoctoral NRSA 5 F32 NS077840-03
NIH NIMH R01 2R01MH083686-06

Title: Long-term cell tracking using volumetric registration to compensate for brain deformation

Authors: K. FINKE[1], **Z. CHEN**[1], J. T. BOYD-MEREDITH[3], D. W. TANK[3], J. L. GAUTHIER[2];
[2]Biol. Dept, [1]Swarthmore Col., Swarthmore, PA; [3]Princeton Univ., Princeton, NJ

Abstract: The hippocampus plays a vital role in navigation. It is unclear, however, how the hippocampus supports spatial learning over long timescales in complex tasks. One limitation has been the ability to track neural activity for more than a few days or weeks. Even with two-photon imaging, which allows precise anatomical localization of recorded cells, brain deformations make it difficult or impossible to repeatedly image the same plane. Here, we introduce an analysis technique to track calcium activity in large populations at cellular resolution over months of training despite substantial deformations. This technique is composed of two stages: 1) aligning each imaging session to a reference volume, and 2) identifying which cells were recorded on each day, using the reference volume to distinguish unique neurons. Though these processes can be largely automated, manual verification is required to ensure accuracy. To facilitate user inspection of the alignment and cell identification, we have developed a graphical user interface for each stage that allows for rapid verification and correction of the automated procedures. This analysis pipeline has allowed us to measure the activity of ~800 unique CA1 pyramidal neurons across 40 behavioral sessions spanning 90 calendar days. The results of this

analysis are described in a companion poster. We anticipate our technique for tracking large populations could allow for recording longer timescales of activity in other brain areas as well.

Disclosures: K. Finke: None. **Z. Chen:** None. **J.T. Boyd-Meredith:** None. **D.W. Tank:** None. **J.L. Gauthier:** None.

Poster

P870. Place Cells: Learning and Encoding of Goals

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Program #/Poster #: P870.03

Topic: H.09. Spatial Navigation

Support: Postdoctoral NRSA 5 F32 NS077840-03
NIH NIMH R01 2R01MH083686-06

Title: Hippocampal maps might not be one-size-fits-all

Authors: K. FINKE[1], ***E. KAROLCZAK**[1], G. STRAUS[1], E. HAPGOOD[1], A. METOYER[1], C. RICHMOND[1], C. ALVARADO[1], T. BOYD-MEREDITH[2], D. W. TANK[2], J. L. GAUTHIER[1]; [1]Swarthmore Col., Swarthmore, PA; [2]Princeton Univ., Princeton, NJ

Abstract: The hippocampus is critical for navigation and learning goal-directed tasks. Hippocampal place maps form almost immediately in a new environment, but it is unclear whether these maps are sufficient to support behavior, or if instead they must evolve beyond their initial representations. In other words, does the hippocampal map itself need to "learn" something about the environment? To interrogate this possibility, we trained mice on two virtual reality tasks and imaged ~1,000 CA1 neurons during every behavioral session, from first exploration through expert performance. Because the hippocampal tissue gradually changed shape over time, it was impossible to return to the same imaging plane every day. To account for these deformations, we developed a novel analytical technique to track the activity of individual neurons, as described in a companion poster. In the behavioral task, mice learned to associate a water reward with several locations along the track, and their reward anticipation strategies revealed gradual refinement across sessions. Though place cells and reward-associated cells changed in many ways throughout learning, a preliminary analysis revealed that the development of expert behavior coincided with predictive encoding; in both tasks, spatial fields shifted earlier at the precise time when mice began to accurately anticipate reward. This raises the possibility that initial hippocampal place maps are not able to support goal-directed behavior, but rather they must first develop representations that anticipate upcoming environmental features. We further interrogated expert performance using clustering analyses, and we identified discrete reward anticipation strategies that were interleaved throughout behavioral sessions. Preliminarily analyses comparing neural activity across different strategies found that reward-associated cells,

unlike place cells, behaved differently. Reward cell fields tended to shift earlier on traversals with more accurate anticipatory behavior, whereas place cell fields did not. This unexpected distinction implies that reward cells may be more strongly related to prediction of the goal than the general population of place cells. Taken together, our observations are consistent with hippocampal place maps needing to evolve beyond their initial representations in specific ways to facilitate goal-directed behaviors.

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Poster

P870. Place Cells: Learning and Encoding of Goals

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Program #/Poster #: P870.04

Topic: H.08. Learning and Memory

Support: Office of Naval Research (N000141612829)
the National Science Foundation (IIS-1735004)
DARPA (W911NF1820)

Title: Hippocampus as a generative circuit for predictive coding of future sequences

Authors: *Y. CHEN[1], H. ZHANG[1], T. J. SEJNOWSKI[2];
[1]UCSD, La Jolla, CA; [2]Salk Inst., La Jolla, CA

Abstract: Rationale and theory. Neurons in the hippocampus (HPC) CA1 region receive both direct and indirect inputs from entorhinal cortex (EC). Due to synaptic transmission delay, information flow through the indirect pathway via dentate gyrus and CA3 is delayed. The computational principle of this seemingly redundant and asynchronous pathway remains unclear. We propose that the EC-HPC circuit is anatomically wired for training a generative model where the recurrent CA3 network is constantly outputting predicted future states to CA1, a downstream region which then compares that with the actual input from EC and generates prediction errors. In so doing, the transmission delay in the circuit could be canceled out and the internal representation stored in the CA3 recurrent network gets updated. This hypothesis extends the predictive coding regime to the time dimension and can be compared with state space models such as the Kalman filter. Methods and results. We trained a traditional recurrent neural network (RNN) with input and output layers mimicking EC inputs and CA3 output predictions, respectively. The network was trained through back propagation through time in a self-supervised manner such that the CA3 prediction of any future state should match the EC input at those corresponding time points. We used sequential input currents to simulate a rat receiving

location- specific inputs in an environment and random spontaneous currents to mimic the animal's sleeping behavior. In our simulations, the error computing CA1 neurons in the network showed both one-shot activation upon remapping to a new environment and replay of multiple sequences during sleep. When a more realistic grid-cell like EC input was given to the network, the recurrently connected CA3 neurons also showed place fields. In addition, by minimizing future prediction errors, the trained network was more robust to noise interference compared to a conventional loss function that suppresses current prediction errors. Conclusions. These results suggest that the proposed generative model may be a unifying framework to explain how HPC and EC interact to participate in memory formation and spatial localization. Furthermore, this bio-inspired architecture points to a more robust and generalizable RNN training algorithm.

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Poster

P870. Place Cells: Learning and Encoding of Goals

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NIH NRSA Grant 5F31AG066410

Title: Goal discrimination in hippocampal non-place cells when place information is ambiguous

Authors: *L. ZHANG[1], S. M. PRINCE[3], A. L. PAULSON[4], A. C. SINGER[2];
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Abstract: Memory-guided navigation relies on hippocampal neurons, like place cells, that encode features of the environment. However, little is known about hippocampal place codes when spatial information, like cues, provide ambiguous information about finding goals. Non-place cell activity, pyramidal cells that fire without strong spatial modulation in an environment that are typically ignored, may be well-suited to carry task-relevant non-spatial information. We find that when spatial cues and goal information are conflicting, non-place cell firing distinguishes between ambiguous spatial cues. On correct trials non-place cells had higher firing rates and altered gamma phase modulation at task-relevant cues compared to incorrect trials while place cells showed no such differences. Finally, this goal discrimination in non-place cells

is absent in a mouse model of Alzheimer's disease that has memory impairment. Our findings show that non-place cells differentiate ambiguous goal information that place cells do not and reveal "non-coding" cells have special coding contributions.

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Poster

P870. Place Cells: Learning and Encoding of Goals

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Topic: H.09. Spatial Navigation

Support: Packard Fellowship for Science and Engineering
NIH-NINDS R01 NS109226

Title: Spatially specific, learning-dependent interneuron activity encodes goals

Authors: *N. JEONG, V. P. NGUYEN, S. R. THOMAS, A. C. SINGER;
The Wallace H. Coulter Dept. of Biomed. Engin., Georgia Inst. of Technol. and Emory Univ., Atlanta, GA

Abstract: Remembering precise locations of importance in distinct contexts is a key component of spatial memory. Over the past few decades, the hippocampal literature has shown extensive evidence that excitatory pyramidal cells code for various spatially relevant cues. In contrast, inhibitory interneurons have been overlooked and ascribed functions such as balancing global network activity. Evidence linking inhibitory activity to spatial learning has been scarce because interneurons are generally thought to fire action potentials in a spatially uniform manner and are therefore not thought to carry significant spatial information. However, the firing activity of excitatory neurons is under the control of inhibitory interneurons, and therefore interneurons could gate enhanced excitation and information transfer. Such gating would most likely occur in important locations such as rewarded locations during behavior. To test this hypothesis, we recorded from single units in hippocampal area CA3, a region known to be important for developing new spatial memories, in head-fixed mice during spatial learning. Mice learned to lick in specific locations of familiar and novel virtual environments to trigger reward delivery. We found a selective reduction in firing rates of most interneurons as mice reached learned reward zones. The magnitude of this reduction depended on the relative familiarity of the environment and correlated with learning performance as animals learned reward locations in a novel environment. These findings could not be explained by position-related changes in speed or licking behavior. Furthermore, these reductions in interneuron firing did not represent a simple balancing of changes in excitatory activity. Our results demonstrate spatially specific,

learning-dependent interneuron activity and suggest that inhibition plays a significant role in gating spatial information for correct behavioral performance.

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Poster

P870. Place Cells: Learning and Encoding of Goals

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Program #/Poster #: P870.07

Topic: H.09. Spatial Navigation

Support: Sainsbury Wellcome Centre Core Grant from the Gatsby Charitable Foundation and Wellcome Trust (090843/F/09/Z)
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Title: Hippocampal place cells use vector computations to navigate.

Authors: *J. ORMOND, J. O'KEEFE;
Sainsbury Wellcome Ctr., London, United Kingdom

Abstract: The hippocampal cognitive map supports navigation towards, or away from, salient locations in familiar environments. One strong candidate for underpinning navigation are the CA1 place cells which provide information about the animal's current location. In open-field foraging tasks lacking a specific goal, place cells provide a non-directional measure of current position. When a goal is introduced, however, place cells become directional and their fields move in the direction of the goal when it is moved. Previous work has shown that cells in the hippocampal formation encode heading towards a goal and other locations in the bat, mouse, primate, and human, but an important caveat about such studies is that the subject usually moves towards the goal whenever possible, precluding full assessment of the neuronal activity in non-goalward directions. We recorded CA1 place cells while rats navigated to a goal on the honeycomb maze. The maze tests navigation via both direct and indirect paths to the goal and allows the directionality of place cells to be assessed at each choice point. A large proportion of place fields (~30%) showed strong directional polarization characterized by vector fields which converged to unique locations, which we termed convergence sinks (ConSinks), distributed throughout the environment. The distribution of these ConSinks was centred near the goal location, and the population vector field also converged there, providing a strong navigational signal. Changing the goal location led to the movement of ConSinks and vector fields towards the new goal. ConSink cells also encoded distance and allocentric direction to their sink positions, indicating that the necessary information for precise navigation to the sinks is encoded by these cells. Directional and distance tuning was broadly distributed, indicating that the ConSink cell population has the information necessary to calculate the goal direction vector from

any location. The directional tuning of the population forms a peak in the goalward direction, and its distribution falls off smoothly with deviation from this direction, except on error trials, when the distribution is disrupted. Animals broadly sample direction relative to the goal, particularly on correct trials, suggesting that they solve the task by identifying the available direction which produces the highest firing rate within the ConSink cell population. ConSink cells, therefore, appear to be an important component of a system allowing flexible navigation towards goal locations.

Disclosures: J. Ormond: None. **J. O'Keefe:** None.

Poster

P870. Place Cells: Learning and Encoding of Goals

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Program #/Poster #: P870.08

Topic: H.09. Spatial Navigation

Support: ONR MURI grant: N000141310672
NSF IIS grant #1703340

Title: Multifield and Multiscale Coding of Megaspace by Dorsal and Ventral Hippocampal Place Cells.

Authors: Y. QIN, ***M. CONTRERAS**, C. STERZINAR, B. HARLAND, J.-M. FELLOUS; Univ. of Arizona, Tucson, AZ

Abstract: Animals navigate in familiar and new environments across a range of spatial scales. It is unclear, however, how the mammalian brain forms multiscale representations of complex natural environments, and how these representations are used for implementing successful spatial navigation. We previously recorded wirelessly from dorsal hippocampal place cells while rats performed a goal-oriented spatial task in a large open environment (the megaspace; 5.3 × 3.5 m, 18.6 m2). Rats were trained to follow a small food-baited robot to facilitate high-resolution place cell recordings in the megaspace. We found that place cells within the dorsal CA1 exhibited multiple subfields of different sizes in the megaspace supporting a multi-scale representation of space. Our results demonstrate that each dorsal place cell is active in several sections of the same environment and show that each location of the environment is represented by a unique combination of subfields of different sizes. These results suggest that dorsal hippocampal place cells use a coding scheme that challenges our present understanding of how the hippocampus represents an animal's position in space.
We extended our work by conducting simultaneous wireless electrophysiological recordings from dorsal and ventral place cells while rats perform a rodent version of the traveling salesperson problem (TSP). This allowed us to study whether ventral cells exhibit the same

multifield and multiscale coding scheme as dorsal cells, and to investigate how this dorsoventral coding mechanism may contribute to spatial optimization. The TSP is a spatial optimization task that requires that a subject find the shortest possible route to visit a fixed set of identical reward cups without re-visits. Experiments were also conducted in conditions where the reward locations were differentiated by the presence of small objects (local cues). In this goal directed task, we show that the multiscale and multifield nature of coding is largely conserved in ventral CA1. Additional work is underway to determine if place field locations and place cell firing dynamics are related to the ability of the animal to optimize its trajectory.

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Poster

P870. Place Cells: Learning and Encoding of Goals

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Topic: H.09. Spatial Navigation

Support: NIH Grant U01NS090541
NIH Grant U19NS104648
NIH Grant F32MH119749
Simons Collaboration on the Global Brain

Title: Low-dimensional neural manifolds in the hippocampus contain geometric representations of learned knowledge

Authors: *E. H. NIEH[1], M. SCHOTTDORF[1], N. W. FREEMAN[1], R. J. LOW[1], S. LEWALLEN[1], S. KOAY[1], L. PINTO[1], S. C. VENDITTO[1], J. L. GAUTHIER[1], C. D. BRODY[1,2,3], D. W. TANK[1,3,4];
[1]Princeton Neurosci. Inst., [2]Howard Hughes Med. Inst., [3]Dept. of Mol. Biol., [4]Bezos Ctr. for Neural Dynamics, Princeton Univ., Princeton, NJ

Abstract: Hippocampal neurons encode physical variables such as space or auditory frequency in cognitive maps. In addition, human fMRI studies have shown that the hippocampus can also encode more abstract, learned variables. However, their integration into existing neural representations of physical variables is unknown. Here, we show the integration of physical and learned task variables as smooth gradients on task-specific, low-dimensional neural manifolds, instantiated by the coordinated activity of single neurons. Male and female adult mice (n=7 mice) learned a task traversing the stem of an immersive virtual reality T-maze, while visual cues were presented randomly on the left and right walls. Mice were rewarded for turning to the side that had more cues at the end of the maze. Using 2-photon calcium imaging, we show that

individual dorsal CA1 neurons jointly encode accumulated evidence with spatial position in mice performing this task (n=917 of 3144 total neurons). Nonlinear dimensionality reduction estimated the neural manifold to be approximately 4-6 dimensions. Within this low-dimensional space, both physical and abstract variables were jointly mapped in an orderly fashion, creating a geometric representation that we demonstrate to be similar across animals. Furthermore, using a method combining generalized linear models (GLMs) with hidden Markov models (HMMs), different trials in the task could be clustered into distinct behavioral states. We show that manifolds fit to some clusters of trials better organize behavioral variables, such as accumulated evidence, compared to manifolds fit to all trials in a session. Other clusters have little representation of accumulated evidence, consist mostly of left or right trials, and coincide with poor task performance. These results demonstrate that CA1 neurons can have distinct patterns of neural activity representing different behavioral strategies, i.e. remapping, in animals performing complex decision-making tasks. The existence of multiple behavioral states represented by different neural manifolds suggests that errors independent of trial difficulty seen in various perceptual decision-making tasks, i.e. “lapses”, may correspond to alternate suboptimal strategies, instead of mistakes due to noise or motor errors.

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Poster

P890. Cognitive Natural Aging: Neural Mechanisms I

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Program #/Poster #: P890.01

Topic: H.12. Aging and Development

Support: NIH-NIA R01-AG059028-01

Title: Multiplexed immunofluorescence reveals distinct neuron and astrocyte clusters in the rhesus monkey prefrontal cortex that are differentially altered in normal aging

Authors: W. CHANG[1], M. MEDALLA[1], P. CASTRO-MENDOZA[1], E. MCDONOUGH[2], L. LOWERY[2], M. VARGHESE[3], P. HOF[3], D. E. MEYER[2], ***J. LUEBKE**[1];
[1]Boston Univ., Boston, MA; [2]GE Res., NIskayuna, NY; [3]Neurosci., Icahn Sch. of Med. At Mount Sinai, New York, NY

Abstract: Multiplexed immunofluorescence was used to sequentially stain and image 18 biomarkers in tissue microarrays containing PFC sections from young (6.5, 11.7 y.o.), middle-aged (15.3, 17.3, 17.6, 19.2 y.o.) and aged (24.8, 28.4 y.o.) behaviorally characterized rhesus monkeys. Tissue from two additional middle-aged subjects (17.7, 19.7 y.o.), that received daily

dietary curcumin over 3 years, was included to assess whether this compound prevents age-related changes. Cells were precisely localized within gray matter laminae and white matter using machine learning-based segmentation of DAPI images, and classified as neurons or astrocytes using cell type-specific biomarkers.In separate analyses of neurons and of astrocytes, we performed dimensionality reduction and cluster analysis on the cellular intensities of biomarkers known to be closely associated with the aging process. This approach allowed us to identify different subpopulations (clusters) of neurons and of astrocytes that are characterized by distinctive biomarker intensity profiles. Eight out of a total 25 neuronal clusters and 12 out of a total 18 astrocytic clusters incorporate proportions of cells that are either increased or decreased with age (Fisher's exact test, $p<0.05$). For example, "Neuron Cluster 1", characterized by low intensities of 3-nitrotyrosine and amyloid β (Aβ), and high intensities of cleaved caspase-3, TDP43 and TSPO (Mann Whitney U, $p<2.5e-17$), contains significantly higher percentages of the total number of neurons in middle-aged (17.7%) and aged (16%) relative to young monkeys (10%; $p<9e-09$). The percentage of total neurons in "Neuron Cluster 1" (7.2%) of curcumin-treated monkeys is significantly lower than that of middle-aged controls ($p=5.62e-35$). Interestingly, neurons within "Neuron Cluster 1" are preferentially located in the supragranular layers of the cortex ($p=1.36e-24$). As another example, "Astrocyte Cluster 3" characterized by low intensity of Aβ and high intensity of vimentin ($p<0.008$), differs significantly between age groups, with 14.5%, 7.7% and 2.7% of the total number of astrocytes in young, middle-aged and aged respectively ($p<5.12e-5$). "Astrocyte Cluster 3" cells are preferentially localized in the white matter ($p=1.87e-68$). As with "Neuron Cluster 1" curcumin treatment ameliorates the age-related changes to "Astrocyte Cluster 3" ($p=0.006$).These two examples -"Neuron Cluster 1" and "Astrocyte Cluster 3"- are just two of the 43 clusters identified with 6 (of 18) biomarkers. Ongoing analyses will reveal many additional interactions between biomarkers in different cell types, as well as alterations to specific clusters during aging.

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Poster

P890. Cognitive Natural Aging: Neural Mechanisms I

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Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P890.02

Topic: H.12. Aging and Development

Support: Generalitat Valenciana APOST2019

Title: Age-dependent changes in the neural coding of the cholinergic basal forebrain during a probabilistic Pavlovian task

Authors: *S. MARTÍNEZ-BELLVER[1,2], A. VELENCEI[1], N. SOLARI[1], C.-H. DE BEVAL[1,3], P. HEGEDÜS[1,4], B. HANGYA[1];
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Abstract: Acetylcholine mediates multitude of cognitive functions including arousal, attention, sensory processing, reinforcement expectation, reward and addiction. The basal forebrain cholinergic input to the cortical mantle plays a central role in the modulation of these cognitive processes. Age-related changes in the morphology of cholinergic neurons has been observed across species, characterized by dendritic, synaptic, and axonal degeneration. However, the link between cholinergic activity during learning and the normal or pathological age-related neurodegeneration is still missing. To clarify the role of the cholinergic basal forebrain in learning during the normal aging process, we performed neuronal recordings in the nucleus of the horizontal limb of the diagonal band (HDB), in head-fixed ChAT-cre mice during a probabilistic auditory cued-outcome task. Animals from 3 to 18+ months of age were injected with an adenoassociated virus carrying channelrhodopsin-2, transfecting cholinergic neurons, allowing us to optically tag them. Animals showed decreased anticipatory licking correlated with aging, being old animals less likely to lick during reward-predicting trials, compared to young ones. Surprisingly, tagged cholinergic neurons in the young animals seemed to respond to the reward but not punish-related cue, whereas in the old animals no response to cue was observed in contrast to the young ones. Additionally, both punishment and reward activated part of the recorded neurons in all the animal groups. We additionally performed fiber photometry experiments with an acetylcholine fluorescent sensor in order to determine the cholinergic influx over the prefrontal cortex and basolateral amygdala, the main cholinergic projections of HDB. Results showed a similar pattern to that observed in the basal forebrain cholinergic neurons. Finally, we managed to modify the associative learning of the Pavlovian task contingencies of 3 months old animals by specifically inhibiting the cholinergic neurons of HDB during the CS presentation. With this paradigm, young animals showed learning deficits similar to those observed by aged animals. Overall, our study reveal new information on the role of the basal forebrain in associative learning, and how this system is affected by ageing.

Disclosures: S. Martínez-Bellver: None. **A. Velencei:** None. **N. Solari:** None. **C. de Beval:** None. **P. Hegedüs:** None. **B. Hangya:** None.

Poster

P890. Cognitive Natural Aging: Neural Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P890.03

Topic: H.12. Aging and Development

Support: NIH Grant AG065819

Title: Reward, sociability, affective behavioral and functional neuroimaging differences in young, middle, and aged C57BL/6J male and female mice

Authors: M. POMPILUS, J. BURAIMA, *M. FEBO;
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Abstract: According to the WHO, mental health conditions affect 15% of adults ages 60 and older and accounts for 6.6% of total disability. These conditions are often underdiagnosed because they occur along with other primary systemic and neurological conditions. More research on the neurobiological regulation of mood and affective states in aging subjects is thus needed. We investigated reward preferences, sociability, and anxiety-like behavior in male and female C57BL/6J mice tested after arrival at 10, 30, and 60 weeks of age. Mice were behavioral assessed using protocols for sucrose preference, social interaction/recognition, open field activity, and contextual fear condition (CFC). Mice were then imaged at 4.7 Tesla under combined dexmedetomidone/isoflurane anesthesia to measure resting state functional connectivity across cortical, striatal, hippocampal, and amygdala nodes. Initial analyses of general locomotor activity in an open field environment indicates that 10-week-old mice had higher levels of horizontal activity than 30- and 60-week-old mice. The total distanced moved during testing showed a similar difference, however, stereotyped activity was not different between the 3 age groups. Interestingly, 10-week-old mice ambulated at much higher levels in the center of the test arena than 30- and 60-week-old mice. We used a 3-day CFC protocol that included a conditioning day, a retrieval day, and a 2nd retrieval day with changes to context. Mice in all age groups acquired tone-shock paired freezing behavior at the same rate and extent on day 1. All mice showed robust conditioned fear to tone alone on day 2, with 60-week-old mice trending towards higher levels of conditioned freezing relative to 10- and 30-week-old mice. On day 3, 10-week-old mice had lowest levels of conditioned freezing in a modified context. Our data suggest that aged (60-week-old) C57BL/6J mice are less active, exhibit anxiety like behavior in an open field, and express higher levels of fear conditioning and more difficulty in suppressing conditioning in a modified context when compared to younger 10-week-old mice. Ongoing analyses will investigate reward preferences, sociability, and functional connectivity in these mice. The present preliminary results show initial evidence of differential expression of affective and cognitive behaviors in aged C57BL/6J mice.

Disclosures: M. Pompilus: None. **J. Buraima:** None. **M. Febo:** None.

Poster

P890. Cognitive Natural Aging: Neural Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P890.04

Topic: H.12. Aging and Development

Support: NIH Grant AG058748
NIH Grant AG034570
NIH Grant AG062542
NIH Grant AG044292
Alzheimer's Association Award AARF-17-530186

Title: The role of dopamine in age-related changes in regional homogeneity

Authors: *J. H. PARENT[1], C. J. CIAMPA[1], R. M. GARRETT[1], T. M. HARRISON[2], C. N. GORDON[1], W. J. JAGUST[3], A. S. BERRY[1];
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Abstract: Regional homogeneity (ReHo) is an fMRI measure of the nonparametric concordance of neighboring voxel timeseries that has been interpreted to reflect local functional connectivity. Aging is associated with alterations in ReHo, which previous research has associated with accumulation of AD-related pathology and reduced glucose metabolism. However, aging is also associated with changes in the dopamine (DA) system. Given DA's neuromodulatory function and broad innervation of most cortical and subcortical regions, we hypothesized that ReHo would be related to individual differences in DA function in aging. Here, we used [^{18}F]Fluoro-m-tyrosine ([^{18}F]FMT) PET, a measure of DA synthesis capacity, and 3T resting state fMRI to establish relations between ReHo, DA function, and cognition in cognitively normal older adults (OA) (n = 34; 21 females, mean education: 17 mean age: 77 (range = 62-85 years). To limit the possible impact of AD-related pathology, OA were defined as β-amyloid negative via [^{11}C]Pittsburgh Compound B PET. To describe the effect of older age on ReHo, cross-sectional group-level analyses were performed using 29 young adults (12 females, mean education: 16 years, mean age: 25 years (range = 20-35 years)). Analyses relating ReHo to DA and cognition were limited to OA. We found that OA tended to have significantly lower ReHo in cortical regions and higher ReHo in subcortical regions compared to young adults. Next, we tested the relation between ReHo and [^{18}F]FMT net tracer influx (Ki) in a midbrain region of interest (ventral tegmental area/substantia nigra). We found that OA with higher [^{18}F]FMT Ki were those with higher ReHo in prefrontal cortex regions including dorsal anterior cingulate cortex (dACC, r = .379, p=.035). Thus, higher midbrain DA synthesis capacity in older age was associated with "youth like" dACC ReHo patterns. To establish the relevance of relations between [^{18}F]FMT Ki and ReHo to cognition, we tested their independent and combined association with performance on the Stroop task, which is consistently linked with both DA and dACC function. Higher [^{18}F]FMT Ki was associated with better Stroop performance (r=.540, p=.001). However, we did not find a significant relation between dACC ReHo, and Stroop performance (r= .112, p= .534). Lastly, we tested whether [^{18}F]FMT Ki mediated the relation between dACC ReHo and Stroop performance. We found a significant indirect effect of [^{18}F]FMT Ki (mean boostrapped unstandardized β= 2.03, p = .006). Together, this study describes a distributed pattern of ReHo changes in aging and reveals a potential neuromodulatory pathway through which dACC ReHo is related to executive control in OA.

Disclosures: J.H. Parent: None. **C.J. Ciampa:** None. **R.M. Garrett:** None. **T.M. Harrison:** None. **C.N. Gordon:** None. **W.J. Jagust:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Optoceutics. F. Consulting Fees (e.g., advisory boards); Biogen, Bioclinica, Roche/Genentech, Grifols. **A.S. Berry:** None.

Poster

P890. Cognitive Natural Aging: Neural Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P890.05

Topic: H.12. Aging and Development

Support: NIH Grant R01 AG039684
NIH Grant R56 AG052576
NSF GRF 1644868

Title: Cortical iron mediates age-related decline in fluid cognition

Authors: *C. M. HOWARD[1,2], S. JAIN[2], A. D. COOK[2], L. E. PACKARD[2], H. A. MULLIN[2], N.-K. CHEN[2], C. LIU[2], A. W. SONG[2], D. J. MADDEN[2,1,3];
[1]Ctr. for Cognitive Neurosci., [2]Brain Imaging and Analysis Ctr., Duke Univ., Durham, NC; [3]Dept. of Psychiatry and Behavioral Sci., Duke Univ. Med. Ctr., Durham, NC

Abstract: Brain iron dyshomeostasis disrupts various critical cellular functions and age-related iron accumulation in deep gray matter and sensorimotor regions may contribute to oxidative stress, deficient neurotransmission, and cell death. While recent studies have linked excessive brain iron to cognitive function in neurodegenerative pathology contexts, the role of brain iron accumulation in age-related decline in fluid cognition (encompassing speed-dependent cognitive abilities) remains unknown. We validated age-related accumulation of iron in deep gray matter regions, measured by quantitative susceptibility mapping (QSM), in 67 healthy participants (18-78 years, 35 female), in relation to known histology for these regions. Voxelwise QSM analyses defined cortical regions for which susceptibility was associated with both age and fluid cognition. QSM susceptibility values were negatively associated with fluid cognition in the right inferior temporal gyrus, bilateral putamen, posterior cingulate gyrus, motor, and premotor cortices. These results align with previous literature suggesting that voxelwise QSM methods are valid measures for the detection of age-related differences in brain iron in both cortical and subcortical regions. A mediation analysis indicated that susceptibility in the right inferior temporal gyrus was a significant mediator of the relation between age and fluid cognition. Additionally, age and right inferior temporal gyrus susceptibility interacted to predict fluid cognition, such that brain iron was negatively associated with cognitive decline for adults over 45 years. This novel finding suggests that iron has a mediating role in cognitive decline, is a

critical source of variance in cognitive aging studies, and may be an early biomarker of neurodegenerative disease.

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Poster

P890. Cognitive Natural Aging: Neural Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P890.06

Topic: H.12. Aging and Development

Title: Cognitive reserve modulates the neurophysiological complexity of the aging brain

Authors: C.-M. HUANG[1,2], Y.-C. SU[1,2,3], C. LIN[4,5], O. J.-L. TZENG[1,2,6,7,8], S.-C. WU[10], ***H.-W. HUANG**[9];
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Abstract: Normal aging experiences a general decrement in physiological function and cognitive abilities, and the age-related neurocognitive declines is highly variable within the aged population. Previous studies have suggested that Cognitive Reserve (CR) factor such as education could cope with the neurocognitive impairments emerging as a consequence of age-associated brain damage or pathology. Given that age-related and individual differences in neurocognitive function are associated with dynamic processes of the functional brain networks, we hypothesized that CR may involve facilitating age-related changes in flexibility and efficiency of neural networks and individual variations in educational level would be associated with altered functional brain complexity of the aging brain. In this resting-state functional magnetic resonance imaging (rs-fMRI) study, we applied multiscale entropy, cross-sample entropy, and neuropsychological approaches to examine whether and how educational level impacts the complexity of brain function, intrinsic networks, and cognitive ability in healthy aging. The rs-fMRI results showed that, compared with the healthy elder with lower CR, the healthy elder with higher CR presented better performance of executive function, youthful brain entropy and increased efficiency of networks in fronto-parietal (FPN), cingulo-opercular (CON),

and default mode network (DMN). These findings provide neurobiological evidence of CR and suggest that CR may facilitate and scaffold neural networks of the aging brain to act as a protective mechanism.

Disclosures: C. Huang: None. **Y. Su:** None. **C. Lin:** None. **O.J. Tzeng:** None. **S. Wu:** None. **H. Huang:** None.

Poster

P890. Cognitive Natural Aging: Neural Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P890.07

Topic: H.12. Aging and Development

Support: CSUN Grant

Title: Cognitive effects of environmental enrichment on aged obese male mice

Authors: *C. C. BAKER[1], L. R. BANNER[2], T. UHLENDORF[3];
[1]Biol., California State University, Northridge, Northridge, CA; [2]Biol., California State University, Northridge, Simi Valley, CA; [3]Cal State Univ. Northridge, Northridge, CA

Abstract: Humans are living longer than ever before. Cognitive impairments are one of the most devastating health challenges that occur with increasing lifespan. Normal aging is associated with atrophy and alterations in hippocampal synapses, plasticity, and spatial memory. Coupled with the increased aging population, approximately 40% of Americans aged 65-74 are obese. Many long-term complications arise in patients with obesity and type-2 diabetes including dementia and Alzheimer's disease. These cognitive effects begin early as they are seen in children and adolescents with type-2 diabetes and a lifetime exposure to obesity could have an additive effect on age-related cognitive decline as has been seen with late life cardiovascular disease. Environmental enrichment has been shown to improve the cognitive decline in in normal aging and Alzheimer's disease. In this study we sought to determine whether environmental enrichment would improve cognition in long-term obese aged C57Bl6/J mice to the same extent it does in control aged mice. Two-month old animals were randomly assigned to standard laboratory mouse cages with either regular chow (RC) or a high-fat diet (HFD). Animals on a HFD developed obesity and elevated blood glucose levels. After 18 months, mice were further randomly assigned into HFD-Control (n=9), HFD-Enrichment (n=8), RC-Control (n=9), and RC-Enrichment (n=11) for two months. Control groups were housed in standard mouse cages while enriched cages were larger and contained a variety of toys rotated on a weekly basis. After two months of enrichment, mice were tested using the Morris Water Maze, Novel Object Recognition, and Elevated Plus Maze. Each trial was filmed and analyzed with tracking software. A decrease in body weigh was seen in both groups in the enriched environment with

the RC animals losing the most weight. Blood glucose levels were also slightly lower in both enrichment groups compared to controls. Although both enriched groups (HFD and RC) showed cognitive improvement over their respective controls, the greatest improvements were seen in the mice on regular chow in the enriched environment. The RC-Enrichment improved their time to the platform in the Morris Water Maze and had a significantly greater exploratory ratio in the Novel Object Recognition compared to all other groups. In the Elevated Plus Maze, both enrichment groups showed a substantial reduction in anxiety (greater number of crossings and more time in open arms), although the RC-Enrichment group showed the greatest changes. Taken together these results may indicate that aging in the context of long-term obesity has a greater cognitive impact than aging alone.

Disclosures: C.C. baker: None. **L.R. Banner:** None. **T. Uhlendorf:** None.

Poster

P890. Cognitive Natural Aging: Neural Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P890.08

Topic: H.12. Aging and Development

Title: The balance of hippocampal-caudate functional connectivity is altered in older adults

Authors: *M. FREEDBERG[1], R. FOROUGHI[2];
[1]Kinesiology and Hlth. Educ., Univ. of Texas, Austin, TX; [2]Kinesiology and Hlth. Educ., The Univ. of Texas at Austin, Austin, TX

Abstract: The hippocampus and caudate are hubs in the episodic and procedural memory networks, respectively. Aging disproportionately affects episodic memory (memory for personally experienced facts and events) more than procedural memory (memory for skills and habits). This suggests that the hippocampal network is more vulnerable to aging than the caudate one. However, to our knowledge, a direct comparison of age-related changes in hippocampal and caudate network connectivity has not been made. Here, we compared the magnitude and valence of hippocampal and caudate connectivity changes between younger and older adults. We expected to see relatively greater hippocampal network decoupling (i.e., decreases in network connectivity) compared to the caudate network. Single sessions of resting-state fMRI data acquired from younger (18-45 yrs; n = 103) and older (55-75 yrs; n = 125) adults were obtained from the Nathan Kline Institute-Rockland Sample (NKI-RS) repository. Resting-state functional connectivity (rsFC) was measured from the left anterior hippocampus and the head of the right caudate based on their specific roles in episodic and non-motor procedural memory, respectively. Whole-brain hippocampal and caudate rsFC were compared between groups. Statistical significance was set to $p < 0.001$ (FDR-corrected, $q < 0.05$). Examining hippocampal and caudate rsFC separately, we observed that the hippocampus showed significant age-related

decoupling from the medial prefrontal cortex and striatum, while the caudate became disconnected from several subcortical regions, including the striatum and sensorimotor regions. Contrary to our expectations, the caudate showed significantly greater network decoupling compared to the hippocampus, in general. Finally, we also noted a dynamic shift in rsFC between networks: While the hippocampus increased in rsFC with the thalamus, the opposite was true for the caudate. In contrast, we noted that while the caudate significantly increased in rsFC with several frontal areas, such as the dorsal and ventral medial prefrontal cortex, the opposite was true for the hippocampus. Our results challenge the notion that the hippocampus undergoes stronger age-related network decoupling than the caudate and indicate that both networks undergo compensatory shifts: While the hippocampus becomes more associated with the thalamus and less associated with the prefrontal cortex, the opposite is true of the caudate. These results suggest that aging causes a shift in the configuration of each network, including an exchange of network resources.

Disclosures: M. Freedberg: None. **R. Foroughi:** None.

Poster

P890. Cognitive Natural Aging: Neural Mechanisms I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P890.09

Topic: H.12. Aging and Development

Title: Norepinephrine possibly mediates the effect of chronic amphetamine on memory performance in aged rats.

Authors: *S. SCOGNAMIGLIO, G. DEZFULI, K. J. KELLAR;
Georgetown Univ., Washington, DC

Abstract: Understanding the neural mechanisms of normal and pathological aging will become increasingly important as the average lifespan continues to rise. Memory impairment is attributed to normal aging processes with the hippocampus and the cerebral cortex playing key role in the encoding and retrieval of memories. Norepinephrine (NE) is an important modulator of memory, especially emotional memory (Tully *et al.*, 2010). Previously, we have demonstrated that acute treatment with amphetamine (0.5mg/kg) enhances memory performance on the novel object recognition (NOR) task in aged Fischer 344 rats, with a reversible and short-term effect on memory performance (Scognamiglio *et al.*, 2021). The purpose of this study was to investigate whether chronic amphetamine treatment has a more robust and possible long-term effect on memory performance in aged rats and whether the effect of amphetamine on object recognition memory is NE dependent. To this end, in the first set of experiments we administered amphetamine for 2 weeks via drinking water at doses of 0.25 mg/kg (week 1) and 0.5 mg/kg (week 2) and tested aged Fischer 344 rats on the NOR task. To determine the extent to which NE

per se influences the cognitive effects of amphetamine, we examined the effects of systemic administration of β and α adrenergic antagonists propranolol, metoprolol, and prazosin prior to the NOR test phase in aged rats chronically treated with amphetamine. Results from the present studies demonstrate: **(a)** aged rats chronically treated with amphetamine display improved object recognition memory compared to acute administration, **(b)** memory performance goes back to baseline 2 months after chronic amphetamine treatment, **(c)** both propranolol and prazosin attenuate the amphetamine effect on cognition while metoprolol completely abolishes amphetamine effects on cognition. An additional goal of this study was to assess the outcome of chronic amphetamine treatment on a cellular level by analyzing dendritic spines in the cerebral cortex and hippocampus. The results will be reported.

Disclosures: S. Scognamiglio: None. **G. Dezfuli:** None. **K.J. Kellar:** None.

Poster

P898. Cognitive Disorders: Pharmacology II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P898.01

Topic: B.02. Ligand-Gated Ion Channels

Title: Ropanicant (SUVN-911): An orally active neuronal nicotinic acetylcholine α4β2 receptor antagonist for the management of depression

Authors: *R. ABRAHAM, N. GANUGA, R. SUBRAMANIAN, S. PETLU, S. RAVELLA, N. BOGARAJU, V. MIDDEKADI, V. CHOWDARY PALACHARLA, A. MANOHARAN, P. JAYARAJAN, R. NIROGI;
Suven Life Sci., Hyderabad, India

Abstract: Depression is a common and severe psychological condition with change in mood affecting approximately 300 million people worldwide. It is characterized by loss of interest, pleasure, feelings of guilt, low self-worth, low confidence, disturbed sleep or appetite, poor concentration, and is associated with suicidal thoughts. Reports from literature suggest that neuronal nicotinic acetylcholine α4β2 receptor antagonist has antidepressant like activity. Ropanicant is a clinical candidate for the treatment of depression. The pharmacokinetics of ropanicant was assessed. Efficacy of ropanicant in depression was assessed using an olfactory bulbectomized (OBX) Swiss albino mice. OBX mice were evaluated in the tail suspension test. Similarly ropanicant was evaluated in a forced swimming test and brain microdialysis studies. Ropanicant was found to have favorable pharmacokinetics with adequate brain penetration and good oral bioavailability. A reduction in the duration of immobility in the tail suspension test as well as forced swimming test was observed. SUVN-911 produced a significant increase in cortical levels of norepinephrine and altered the expressions of brain-derived neurotrophic factor, indicating the possible neurochemical basis for the effects observed in animal models of

depression. Changes were observed in calcium binding adaptor molecules (Iba1) as well. From the above studies, we conclude that ropanicant may have potential in addressing depression.

Disclosures: R. Abraham: A. Employment/Salary (full or part-time):; employement. **N. Ganuga:** A. Employment/Salary (full or part-time):; employement. **R. Subramanian:** A. Employment/Salary (full or part-time):; employement. **S. Petlu:** A. Employment/Salary (full or part-time):; employement. **S. Ravella:** A. Employment/Salary (full or part-time):; employement. **N. Bogaraju:** A. Employment/Salary (full or part-time):; employement. **V. Middekadi:** A. Employment/Salary (full or part-time):; employement. **V. Chowdary Palacharla:** A. Employment/Salary (full or part-time):; employement. **A. Manoharan:** A. Employment/Salary (full or part-time):; employement. **P. Jayarajan:** A. Employment/Salary (full or part-time):; employement. **R. Nirogi:** A. Employment/Salary (full or part-time):; employement.

Poster

P898. Cognitive Disorders: Pharmacology II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P898.02

Topic: H.13. Schizophrenia

Title: Establishing Signature Profile of Muscarinic M4 Selective Molecules by Evaluating their Orthosteric/Allosteric Binding Properties and Biased Signaling

Authors: S. S. PIN, M. TOH, H. N. NGUYEN, P. A. IREDALE;
Cerevel Therapeut., Cambridge, MA

Abstract: The muscarinic M4 receptor is a Gi-coupled G-protein receptor frequently targeted by the pharmaceutical industry for psychiatric disorders such as schizophrenia. The aim of this study is to understand the behavior of M4 receptor in response to the binding and activation by various agonists with distinct chemical structures. Combining classical radioligand binding and modern receptor pharmacology, we have done a comprehensive analysis of known M4 selective molecules at the binding and functional level to determine their preferences for allosteric/orthosteric binding sites, as well as their preferred signaling pathway. Equilibrium binding and full kinetics profiles were determined. Molecules have a broad range of binding affinity and functional potency. At the kinetics level, they have similar k_{off}; the differences in K_d is driven by the various level of k_{on}. Functional profiling was done at the G-protein activating, β-arrestin recruitment, and at the cAMP level. Our results show that M4 agonists have a variety of mode of actions. All have a varying degree of binding affinity to the orthosteric and/or allosteric binding site. Signal transduction can be mediated through the orthosteric as well as the allosteric sites. At the functional level, we evaluated activation of Gαi1, Gαi2, Gαi3, GαoA, GαoB, GαZ, β-arrestin and cAMP accumulation. Agonists have distinct signaling signature profile. Overall, M4 agonists appear to have a variety of unique identities that can be distinguished at the binding

and functional levels. Additional studies are needed to fully understand the clinical implications for each profile.

Disclosures: S.S. Pin: None. **M. Toh:** None. **H.N. Nguyen:** None. **P.A. Iredale:** None.

Poster

P898. Cognitive Disorders: Pharmacology II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P898.03

Topic: H.08. Learning and Memory

Title: Cholinergic receptor blockade impairs spatial memory retrieval and minimizes retrieval-induced alterations in matrix metalloproteinase-9

Authors: *M. L. OLSON, B. BADENOCH, M. BLATTI;
Concordia Col., Moorhead, MN

Abstract: Alzheimer's Disease (AD) is associated with a loss of cholinergic function, and the basal forebrain cholinergic system remains the most common target of pharmaceutical treatments for AD. Recently, some have argued that AD may be best characterized as a deficit of memory retrieval; however, research reports investigating the role of the cholinergic system in memory retrieval are often confounded by the fact that they utilize experimental methodologies that fail to fully isolate the retrieval process from acquisition and consolidation processes. Utilizing a methodology which isolates spatial memory retrieval in the circular water maze, we show that the central administration of the cholinergic antagonist scopolamine reliably impairs the retrieval of previously consolidated spatial memories. Matrix metalloproteinase-9 (MMP-9) is a zinc-dependent endopeptidase that regulates the extracellular matrix as well as several other substrates implicated in memory. Using immunoblotting and gelatin zymography, we show that modifications in hippocampal MMP-9 expression are associated with spatial memory retrieval. Further, we provide evidence that the cholinergic system is an important regulator of some of these retrieval-induced changes in MMP-9. These results further elucidate the well-established and important role of MMP-9 in learning and memory and indicate a possible connection between the cholinergic dysfunction and MMP-9 dysregulation that is commonly seen in AD.

Disclosures: M.L. Olson: None. **B. Badenoch:** None. **M. Blatti:** None.

Poster

P898. Cognitive Disorders: Pharmacology II

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Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P898.04

Topic: H.08. Learning and Memory

Support: CPRIT RP180055
NCI R01 CA224672
The Quincy and Estine Lee Endownment Fund
CPRIT Research Training Award RP170345

Title: Plasticity-related signaling pathways in the medial prefrontal cortex are potential targets of the antidepressant, Vortioxetine, which reverses cognitive impairments associated with androgen deprivation therapy for prostate cancer.

Authors: *A. DORNBUSCH[1,2], A. VAIANA[1,2,3], J. GELFOND[4], T. L. JOHNSON-PAIS[3,5], R. LEACH[3,6], C. RAMAMURTHY[3,7], I. THOMPSON[5,9], D. A. MORILAK[1,2,3,8];
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Abstract: Prostate cancer is one of the most common types of cancer in men. Androgen Deprivation Therapy (ADT) is a first-line treatment for late-stage, metastatic, androgen-responsive prostate cancers. Despite ADT's success to slow cancer growth, there are a number of unwelcome side effects. These include profound cognitive impairment in areas such as executive function, memory, and attention. Further, patients receiving ADT have reduced functional connectivity within the medial prefrontal cortex (mPFC). There are currently no treatment options available to patients that experience cognitive decline. Vortioxetine is a multimodal drug that enhances cognition in patients with major depressive disorder. We hypothesized that vortioxetine might also be beneficial to cognitive decline associated with ADT. We have found that vortioxetine reverses deficits in a rodent model of ADT using surgical castration. Androgen depletion impaired attentional set-shifting, which is mediated in the mPFC. Chronic treatment with vortioxetine (28 mg/kg/day) administered through the diet rescued these deficits (Sharp et al., 2019). Because the androgen receptor is a transcription factor, the impairments seen after ADT are likely related to changes in gene expression. Vortioxetine has also been shown to induce changes in neuroplasticity-related gene expression in healthy rats. It is well documented that castration reduces spine density within the mPFC. Given this, we hypothesized that vortioxetine might reverse cognitive deficits by rescuing plasticity-related gene expression altered by castration. Castrated or intact male Sprague-Dawley rats were treated with vortioxetine or control diet for 17 days. Gene expression in the mPFC was analyzed using a whole rat genome microarray (Agilent). Castration alone significantly altered 1,221 genes within the mPFC (adjusted $p<0.05$). Of the genes altered by castration, many are key for activity-dependent plasticity which may underlie cognition, including Shank1, Akt, MAPK, and CaMKII. Vortioxetine had no significant effect on individual gene expression. However, Ingenuity Pathway Analysis revealed that vortioxetine significantly altered gene clusters involved in

synaptic plasticity (q value<0.05). Preliminary studies are underway to examine changes in the activation state of relevant proteins in the mPFC after castration, as well as chronic vortioxetine treatment. These experiments will provide insight into the mechanisms by which vortioxetine improves cognition after ADT in a rodent model. In conclusion, vortioxetine may be an effective treatment in prostate cancer patients that experience cognitive decline after ADT.

Disclosures: A. Dornbusch: None. **A. Vaiana:** None. **J. Gelfond:** None. **T.L. Johnson-Pais:** None. **R. Leach:** None. **C. Ramamurthy:** None. **I. Thompson:** None. **D.A. Morilak:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); H. Lundbeck, Copenhagen, Denmark.

Poster

P898. Cognitive Disorders: Pharmacology II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P898.05

Topic: H.13. Schizophrenia

Support: Conte Center grant (P50 MH103222)

Title: Enduring adverse effects of adolescent cannabis abuse involve kynurenic acid: relevance for the etiology and treatment of cognitive dysfunctions.

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Abstract: Cannabis abuse during adolescence is a risk factor for cognitive impairments in psychiatric disorders later in life (Renard et al., 2016). Specifically, early exposure to Δ9-tetrahydrocannabinol (THC; i.e. the main psychotropic component of cannabis) causes enduring cognitive deficits, which critically involve impaired glutamatergic function in the prefrontal cortex (PFC; Zamberletti et al., 2014). Remarkably, these adverse effects are qualitatively very similar to those caused in developing rodents by experimental increases in brain kynurenic acid (KYNA), a neuroactive metabolite of tryptophan degradation (Pocivavsek et al., 2014).To date, the possible causal relationship between cannabinoids, KYNA and cognition has not been investigated in adolescence. We therefore examined the effect of chronic adolescent THC exposure on KYNA levels in the rat brain. Male Wistar rats were chronically treated with vehicle or ascending intraperitoneal (i.p.) doses of THC starting on postnatal day (PND) 35 until PND 45, as described (Zamberletti et al., 2014). In adulthood (PND 75), cognitive assessment (Y-maze) and extracellular KYNA/glutamate levels were measured in the PFC by *in vivo* microdialysis, before and after an acute challenge with L-kynurenine (L-KYN), the biological precursor of KYNA (5 mg/kg; i.p.). By using the selective, brain-penetrable KAT II inhibitor PF-

04859989 (30 mg/kg, i.p., 1 hr prior the Y-maze; Kozak et al. 2014), we then examined whether blockade of KYNA neosynthesis prevents the cognitive impairment. Compared to vehicle-treated controls, extracellular basal KYNA levels were higher in the PFC of adult rats chronically exposed to THC in adolescence ($p<0.01$). No changes were observed in extracellular glutamate levels. Following a challenge with L-KYN, extracellular KYNA levels increased similarly in both groups (i.e. vehicle- and THC-treated; $p<0.001$ and $p<0.01$ respectively). Chronic adolescent THC exposure negatively affected short-term memory (reduced spontaneous alternation) in adult animals ($p<0.001$), while pre-treatment with PF-04859989, which prevented the THC-related KYNA increase ($p<0.01$), partially restored the impairment ($p<0.05$). We conclude that the observed alterations in PFC KYNA signalling might be involved in the cognitive dysfunction induced by the exposure to THC during the adolescence. In the translational realm, these experiments raise the prospect of prevention of KYNA neosynthesis as a promising novel approach to combat some of the detrimental long-term effects of adolescence cannabis use.

Disclosures: S. Beggiato: None. **L. Ferraro:** None. **R. Schwarcz:** None.

Poster

P898. Cognitive Disorders: Pharmacology II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P898.06

Topic: H.08. Learning and Memory

Support: NRPU-3766

Title: Modulation of brain neurotransmitters and BDNF levels by curcumin improves depression and associated pseudodementia in rats

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Abstract: Rationale Depressive state adversely affects the memory functions, especially in the geriatric population. The initial stage of memory deficits associated with depression is particularly called as pseudodementia. It is the starting point of memory disturbance before dementia. The purpose of this research was to study depression and its consequent pseudodementia. **Material and methods** For this purpose 24 male albino Wistar rats were divided into four groups. Depression was induced by 14 days of chronic restraint stress (CRS) daily for 4 h. After developing a depression model, pattern separation test was conducted to monitor pseudodementia in rats. Morris water maze test (MWM) was also performed to observe spatial memory. It was observed that model animals displayed impaired pattern separation and spatial memory. Treatment was started after the development of pseudodementia in rats.

Curcumin at a dose of 200 mg/kg was given to model rats for one week along with the stress procedure. Following the treatment with curcumin, rats were again subjected to the aforementioned behavioral tests before decapitation. Corticosterone levels, brain derived neurotrophic factor (BDNF) and neurochemical analysis were conducted. **Results** Model rats showed depressogenic behavior and impaired memory performance. In addition to this, high corticosterone levels and decreased hippocampal BDNF, 5-HT, dopamine (DA), and acetylcholine (ACh) levels were also observed in depressed animals. These behavioral biochemical and neurochemical changes were effectively restored following treatment with curcumin. Hence, it is suggested from this study that pseudodementia can be reversed unlike true dementia by controlling the factors such as depression which induce memory impairment.
Funding Higher education commission of Pakistan and University of Karachi.

Disclosures: S. Haider: None. **A. Afzal:** None. **Z. Batool:** None.

Poster

P898. Cognitive Disorders: Pharmacology II

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Program #/Poster #: P898.07

Topic: H.08. Learning and Memory

Support: CPRIT RP180055
NCI R01 CA224672

Title: Cognitive impairments and changes in inflammatory cytokines in a prostate cancer rodent model treated with docetaxel

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Abstract: Prostate cancer (PC) is the most common cancer to affect men, with more than 3.1 million men in the United States having been diagnosed with PC at some point in their lifetime. In fact, metastatic PC is the second leading cause of cancer-related deaths. Mainstay treatment of metastatic PC consists of androgen deprivation therapy and chemotherapy, both of which are associated with cognitive impairments. Microtubule stabilizers, like docetaxel, are often used as chemotherapy for PC. Patients who receive docetaxel as therapy experience difficulties in working memory, cognitive flexibility, and attention. These cognitive changes are accompanied by dysfunction in the prefrontal cortex and hippocampus, but mechanisms underlying these impairments remain unknown. To study possible mechanisms for chemotherapy-induced cognitive impairments, a syngeneic rat model of PC was utilized. Male Copenhagen rats were

implanted subcutaneously with syngeneic Dunning R-3327 G cell tumors or subjected to sham implants, then treated with either docetaxel (3 i.p. injections over 5 days, 6 mg/kg each) or vehicle. Cognitive impairments, specifically in visuospatial memory and cognitive flexibility were assessed using the novel object task and the attentional set-shifting test, respectively. Additionally, an array of inflammatory cytokines were measured in plasma and in medial prefrontal cortex and hippocampus. Preliminary data show that docetaxel reduced discrimination in the novel object task primarily in tumor-bearing rats, with less effect in sham controls. Significant increases in the pro-inflammatory cytokines interleukin-6 and tumor necrosis factor alpha were observed in the plasma of tumor-bearing rats. Moreover, interleukin-6 mRNA was increased in the dorsal hippocampus of tumor-bearing rats compared to sham controls. Ongoing experiments will assess potential disruption of the blood-brain barrier in rats with prostate cancer. Results from this study suggest that peripheral and central inflammatory signaling associated with prostate cancer may increase brain vulnerability to the potentially detrimental effects of docetaxel, contributing to cancer therapy-induced cognitive impairment.

Disclosures: A. Asher: None. **M. Girotti:** None. **A. Sharp:** None. **D.A. Morilak:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); H. Lundbeck, Copenhagen, Denmark.

Poster

P898. Cognitive Disorders: Pharmacology II

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Program #/Poster #: P898.08

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Title: Blockade of the adenosine A2A receptor protects against cisplatin-induced cognitive impairments

Authors: *A. OLIVEROS, M. ABDUR RASHID, K.-H. YOO, A. CORUJO-RAMIREZ, J. TANG, J. HAWSE, D.-S. CHOI, M.-H. JANG;
Mayo Clin. Col. of Med., Rochester, MN

Abstract: Cisplatin, a platinum-based chemotherapy, has been clinically reported to potentiate cognitive impairments, known as chemobrain, and affects approximately 14 million cancer survivors in the United States without a known cure. These cognitive impairments are reported to

occur well-after chemotherapy cessation, thus it is imperative that novel therapeutics against chemobrain are investigated to improve quality of life in cancer survivors. To elucidate the pathological molecular mechanisms by which chemobrain impairs cognition, we have developed a mouse model resembling intermittent cisplatin chemotherapy in humans and performed RNA-seq analysis in the hippocampus, a brain structure critical for learning and memory. Our RNA-seq analysis revealed robust and selective elevations in the adenosine A2A receptor (A2AR) gene in this region. Given that A2AR is known to play a critical role in learning, memory, and neurodegenerative conditions, we hypothesize that A2AR inhibition can prevent cisplatin-induced cognitive impairments. Consistent with our prior results, we observed that cisplatin impairs adult neurogenesis, neuronal morphogenesis of newborn neurons and dendrite spine formation in the adult mouse hippocampus, leading to learning and memory dysfunction as well as increased anxiety. Remarkably, we found that pretreatment with the specific A2AR antagonist Istradefylline significantly prevents the aberrant neuronal and cognitive deficits induced by cisplatin, without promoting tumor growth or interfering with the anti-tumor activity of cisplatin. Mechanistically, we found that cisplatin's neurotoxic effects are cAMP-CREB and ERK signaling dependent in both the mouse hippocampus as well as human cortical neurons derived from iPSCs. Collectively, our results suggest that robust A2AR induction may be a key contributor to cisplatin-induced memory impairments via cAMP-CREB and ERK signaling pathways. Therefore, targeting the A2AR as a novel translational therapeutic target can be efficacious in protecting against cisplatin-induced chemobrain and consequently, have far-reaching synergistic effects on improved quality of life for cancer survivors.

Disclosures: A. Oliveros: None. **M. Abdur Rashid:** None. **K. Yoo:** None. **A. Corujo-Ramirez:** None. **J. Tang:** None. **J. Hawse:** None. **D. Choi:** None. **M. Jang:** None.

Poster

P898. Cognitive Disorders: Pharmacology II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P898.09

Topic: H.05. Working Memory

Title: Assessing the effects of different doses of hericium erinaceus dietary supplementation on C57BL/6J mice and its role in cognitive behavior

Authors: *M. N. RODRIGUEZ[1], S. L. LIPPI[2];
[1]Angelo State Univ., Mission, TX; [2]Angelo State Univ., San Angelo, TX

Abstract: Hericium Erinaceus, also known as lion's mane, has shown beneficial neurocognitive and neurobiological effects in both healthy and transgenic mice. It has also shown to lessen anxiety and depression after 4 week consumption in human subjects. Not enough research has been conducted on the effects of lion's mane on healthy mice. It is important to assess the

benefits that Hericium Erinaceus may provide in order to prevent health complications and/or pathological disease. This study served as a baseline for the effects of Hericium Erinaceus Mushroom Mycelium Powder (Host Defense Mushrooms) on Wild-Type mice. Moreover, it was a pilot to an upcoming study assessing the effects of Hericium Erinaceus Mushroom Mycelium Powder (Host Defense Mushrooms) on the tauopathy mouse model. It was hypothesized that mice who received the highest dose of powder (2000 mg) would perform better than those receiving 1000 mg and the control. Twenty-four adolescent C57BL/6J male mice were randomized into 3 groups of 8: One control group and 2 experimental. For 9 weeks, all groups were administered 150 g of wet food every 4 days, while the experimental groups' wet food contained doses of Hericium Erinaceus Mushroom Mycelium Powder (Host Defense Mushrooms). Each Orirat semi-self-contained caging system (Animal Care Systems), housed with 2-3 mice received this. The first experimental group received 1/3 teaspoons of powder containing 1000 mg of Hericium Erinaceus and 825 mg of mycelium polysaccharides, while the second group received 2/3 teaspoons containing 2000 mg of Hericium Erinaceus and 1650 mg of mycelium polysaccharides. All mice were also provided 200 g of dry Teklad pellets ad libitum. Throughout the duration of the study the mice, dry food, and remainder of the wet food were weighed every 4 days to ensure the consumption of the wet food in comparison to the dry food, and to assess weight gain of the mice as a result of maturation along with dietary consumption. Behavior as assessed at 8 weeks through Open Field (OF), Elevated Zero Maze (EZM), and Morris Water Maze (MWM) tests. Trending significance of time spent in the surround ($p = .059$) indicated that mice who consumed 1000 mg of Hericium Erinaceus spent more time in the surround ($M = 152.65$) than mice who consumed 2000 mg ($M = 122.27$). This signifies that those who took a higher dose of Hericium Erinaceus experienced less anxiety. Future analyses of EZM and MWM will be further indicative of the effects of Hericum Erinaceus on the cognitive behavior of Wild-Type mice.

Disclosures: M.N. Rodriguez: None. **S.L. Lippi:** None.

Poster

P898. Cognitive Disorders: Pharmacology II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P898.10

Topic: H.05. Working Memory

Support: AG042178
AG047812
NS105473
AG060767
AG069333
AG066347

R41 AG060836

Title: Diethyl (3,4-dihydroxyphenethylamino) (quinolin-4-yl) methyl phosphonate protects against amyloid beta induced defective mitophagy, synaptic damage and cognitive dysfunction

Authors: *C. BOSE[1], M. VIJAYAN[2], P. REDDY[3];
[2]Intrnl. Med., [1]Texas Tech. Univ. Hlth. Sci. Ctr., Lubbock, TX; [3]Intrnl. medicine, Texas Tech. Univ. Hlth. sciences Ctr., lubbock, TX

Abstract: ABSTRACT: The purpose of our study is to determine the protective effects of the newly discovered molecule DDQ (diethyl (3,4-dihydroxyphenethylamino) (quinolin-4-yl) methyl phosphonate) against mutant APP and amyloid-beta (Aβ) in Alzheimer's disease (AD). To achieve our objective, we used a well characterized amyloid-beta precursor protein (APP) transgenic mouse model (Tg2576 strain). We administered DDQ, a 20 mg/kg body weight (previously determined in our laboratory) intra-peritoneally 3-times per week for 2 months, starting at the beginning of the 12th month, until the end of the 14th month. Further, using biochemical and molecular methods, we measured the levels of DDQ in the blood, skeletal muscle, and brain. Using Morris Water Maze, Y-maze, open field, and rotarod tests, we assessed cognitive behavior after DDQ treatment. Using q-RT-PCR, immunoblotting, transmission electron microscopy, and Golgi-cox staining methods, we studied mRNA and protein levels of longevity genes SIRTUINS, mitochondrial number & length, and dendritic spine number and length in DDQtreated APP mice. Our extensive pharmacodynamics analysis revealed high peak levels of DDQ in the skeletal muscle, followed by serum and brain. Our behavioral analysis of rotarod, open field, Y-maze, and Morris Water Maze tests revealed that DDQ ameliorated cognitive decline (Morris Water Maze), improved working memory (YMaze), exploratory behavior (open field), and motor coordination (rotarod) in DDQ-treated APP mice. Interestingly, longevity genes SIRTUINS, mitochondrial biogenesis, fusion, mitophagy, autophagy and synaptic genes were upregulated in DDQ-treated APP mice relative to untreated APP mice. Dendritic spines and the quality mitochondria were significantly increased in DDQ treated APP mice. Current study findings, together with our previous study observations, strongly suggest that DDQ has anti-aging, and anti-amyloid-beta effects and a promising molecule to reduce age- and amyloid-beta-induced toxicities in AD.

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Poster

P930. Advances in Methods for Imaging Neural Activity

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P930.01

Topic: I.04. Physiological Methods

Title: Simultaneous optical imaging of neuronal activity from multiple brain regions in songbirds with a Multiscope.

Authors: J. PENG[1], W. GONZALEZ[1], *Y. SOUDAGAR[2], A. PAPANICOLAOU[2], R. CHRISTO[2], C. LOIS[1];
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Abstract: Fluorescent imaging of neuronal activity is a powerful method to monitor single-neuron spikes from large numbers of cells over time. Two-photon microscopes (2PM) have high resolution and can image cells deep in the brain. However, 2PM requires the animal to be head fixed, and many animals, such as songbirds, do not perform certain behaviors while head fixed. Miniature implantable microscopes (miniscopes) can be used to image neuronal activity in freely behaving animals. However, miniscopes are relatively heavy (around 2 grams) and bulky, which makes it very challenging to image in young or small animals, or to image simultaneously from 2 or more brain regions in the same animal. In an attempt to overcome the limitations of the miniscopes due to their size and weight, we applied a recently developed multiscope system based on imaging fiber optics, which is much smaller and lighter (less than 0.5 grams) than miniscopes, to study song circuits in songbirds. We have carried out pilot experiments to measure neuron activity in freely behaving zebra finches with calcium sensitive dyes (Oregon green and Cal-520) or GCaMP delivered by viral vectors. The minimal size and weight of the implant has made it possible to simultaneously image both left and right hemispheres in the same animal and image in very young animals (~30 days of age), neither of which has been achieved in songbirds previously. Using the fiber optic multiscope system, we have investigated the activity of neurons in the premotor nucleus HVC in two instances: (a) coordinated activity between the left and right premotor nuclei HVC in freely singing adults, and (b) the responses of neurons in juveniles as they form a memory of the song from their tutors. These preliminary experiments indicate that the fiber optic based mutiscope has the potential to overcome some critical limitations of 2PM and miniscopes to investigate the neuronal basis of behavior in freely moving animals.

Disclosures: J. Peng: None. **W. Gonzalez:** None. **Y. Soudagar:** None. **A. Papanicolaou:** None. **R. Christo:** None. **C. Lois:** None.

Poster

P930. Advances in Methods for Imaging Neural Activity

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Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P930.02

Topic: I.04. Physiological Methods

Support: T32-AG020506
R37AG008796
R01 NS113804

Title: In vivo multi-day calcium imaging of hippocampus with miniscopes in freely moving rats

Authors: *H. S. WIRTSHAFTER, J. F. DISTERHOFT;
Northwestern Univ., Chicago, IL

Abstract: Calcium imaging using GCaMP calcium indicators and miniature microscopes has been used to image cellular populations during long timescales and in different task phases, as well as to determine neuronal circuit topology and organization. The hippocampus (HPC) has long been studied with cellular resolution using *in vivo* electrophysiology, but this technique does not allow the same cells to be definitively followed over days, or the visualization of cellular organization within the structure. Because the HPC is essential for many tasks of memory, spatial navigation, and learning, calcium imaging of large populations of HPC can shed new insight on cell changes and organization over time during these tasks.To our knowledge, all reported uses of hippocampal *in vivo* calcium imaging have been done in mouse. However, rats have many basic experimental advantages over mice: they able to do more complex behaviors, more closely mirror human physiology, and are less stressed by handling. Rats are also an advantageous organism to use for calcium imaging: Due to their larger size, rats are able to support larger implants, including a wider diameter imaging lens and a larger camera, thereby enabling the recording of a greater number of cells. Additionally, rats are less prone to post-operative seizures than mice and may be at a decreased risk of seizures after prolonged GCaMP expression as compared to mice. In this poster, we will present the first *in vivo* calcium imaging from CA1 hippocampus in freely moving rats. Using GCaMP7c and the UCLA Miniscope, we will demonstrate that hundreds of cells (mean 226+-90 cells per session) can reliably be visualized and held across weeks using this technique. We will additionally show proof of method by showing that a large number of place cells (>70% of cells) can be recorded on a navigational task involving retrieving food reward on a linear track, and that these place cells are sufficient for decoding animal position.

Disclosures: H.S. Wirtshafter: None. **J.F. Disterhoft:** None.

Poster

P930. Advances in Methods for Imaging Neural Activity

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Topic: I.04. Physiological Methods

Support: HHMI (M.J.S.)

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Title: Long-term, in vivo voltage imaging reveals learning-induced, distributed neural depression in a Drosophila memory center

Authors: *C. HUANG[1], J. LUO[3], L. A. ROITMAN[2], S. WOO[2], M. KANNAN[4], G. VASAN[6], V. A. PIERIBONE[5], M. J. SCHNITZER[7];
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Abstract: Time-lapse studies of neural dynamics in live animals are crucial toward understanding brain function and plasticity. However, in *Drosophila melanogaster*, long-term recordings of neural spiking have been infeasible, and many cellular electrophysiological properties remain unexplored. To address this obstacle, we used laser microsurgery to create a chronic fly preparation for longitudinal voltage-imaging studies. In combination with recently developed FRET-opsin voltage indicators, this preparation allowed us to perform imaging studies of neural voltage dynamics with single-spike resolution for up to 50 days in adult flies. Using this approach, we systematically and quantitatively characterized spontaneous spiking, stimuli-induced spiking, and learning-induced plasticity over multiple days in 6 subtypes of mushroom body output neurons (MBONs), which are crucial for aversive olfactory conditioning. The MBONs showed diverse spontaneous firing rates and bursting dynamics, revealing heterogeneities across the different MBON subtypes. Further, we observed MBON dendritic spikes, which manifested as a temporally broadened action potentials that backpropagated from the axonal region. Notably, all the MBONs had elevated firing activities during odor presentation, but this odor-induced excitation gradually converted into inhibitory responses after repeated odor exposures, revealing fast neural plasticity during sensory adaptation. After olfactory classical conditioning, some MBONs also selectively decreased their spiking responses to the punishment-predicting odor. This cue-specific depression lasted from 1-48 hr in different subtypes of MBONs as a form of learning-induced distributed neural plasticity in the fly memory system. Overall, our approach for long-term voltage imaging facilitates investigations of neurophysiology and neuroplasticity that were previously challenging in awake behaving flies. Our approach is compatible with a variety of recent optical imaging and optogenetic tools and opens new opportunities to study how individual neurons respond over time to environmental influences and change across the lifespan of adult flies.

Disclosures: C. Huang: None. **J. Luo:** None. **L.A. Roitman:** None. **S. Woo:** None. **M. Kannan:** None. **G. Vasan:** None. **V.A. Pieribone:** None. **M.J. Schnitzer:** None.

Poster

P930. Advances in Methods for Imaging Neural Activity

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Topic: I.04. Physiological Methods

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European Research Council Grant (714609)
SNSF Project Grant (175667)
SNSF Eccellenza grant (181239)

Title: Long-term imaging of the ventral nervous system in behaving adult Drosophila melanogaster.

Authors: *L. HERMANS, M. KAYNAK, J. BRAUN, V. LOBATO RIOS, S. GÜNEL, F. AYMANNS, M. SAKAR, P. RAMDYA;
Swiss Federal Inst. of Technol. Lausanne, Lausanne, Switzerland

Abstract: Studying long time-course biological phenomena requires the ability to measure the same tissue repeatedly. In neuroscience, such tools would make it possible to study, for example, long-term memory, brain aging, and neurodegeneration. Here, I will describe a suite of microfabricated technologies that enable long-term, minimally invasive optical recordings of the adult *Drosophila* melanogaster ventral nervous system (VNS)—neural tissues that are functionally equivalent to the vertebrate spinal cord. This consists of a two-component system - a transparent window into the thorax, and an implant that pushes thoracic tissues away from the imaging region of interest. Additionally, we have developed a flexible tethering approach for repeatedly mounting and remounting animals under the 2-photon microscope. Taken together, these tools now open up the possibility of monitoring neural activity, and tissue dynamics in flies over the course of their lifetime. Preliminary results will show the impact of the technology on *Drosophila's* survival and behavior and the loss of sensory neurons following a leg injury.

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Poster

P930. Advances in Methods for Imaging Neural Activity

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Program #/Poster #: P930.05

Topic: I.04. Physiological Methods

Support: R43HL150989

Title: Calibration of optical measurement of membrane potential in eukaryotic cells

Authors: A. COSTANTINO[1], B. K. PANAMA[2], *M. W. NOWAK[1], R. L. RASMUSSON[2], G. C. L. BETT[2];
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Abstract: We developed a method for calibration of optical signals to determine resting membrane potential and action potential (AP) amplitude in mV, for eukaryotic cells. Voltage sensitive fluorescent dyes enable non-invasive observation of APs, but need to be calibrated to provide information about absolute resting potential and amplitude. We used Cytovolt1 (di-4-ANBDQBS) (Cytocybernetics, NY), a voltage sensitive dye that has a much longer lasting stable signal than older dyes such as di-4-ANNEPS. This allows adequate time to permeabilize the membrane and change extracellular $[K^+]_o$ without rundown. We validated a method using a K^+ ionophore and ionic gradients to calibrate optical signals with voltage clamp in HEK293 cells. Cytovolt1 operates via the Stark effect, shifting the emission spectrum in response to membrane voltage (V_m). The emission spectrum is bell-shaped, so we used voltage clamp to verify that the dye response was linear over the physiological range (-80 to +40 mV). We measured changes in fluorescence intensity as a function of the voltage clamp potential, using a 720 nm long pass emission filter and excitation at 660 nm. This relationship was highly linear with deviation from linearity of less than 2% over the range -80 to +40 mV. The linearity of response greatly simplifies our calibration method. The V_m of cells which are dominated by a K^+ conductance will track the K^+ Nernst potential closely. We made HEK293 cell membranes highly permeable and selective for potassium by application of the K^+ ionophore, valinomycin. We then exposed the cells to 10, 25, 60 and 150 mM $[K^+]$. Using the Nernst Equation, these correspond to a V_m of -68, -45, -23 and 0 mV. We calculated the resting V_m of HEK cells as -76 ± 4.9 mV (n=11 cells). The relationship between calculated Nernst potential in the presence of valinomycin showed a linear correlation with changes in fluorescence intensity. To validate these fluorescence-voltage changes in valinomycin treated cells with actual membrane voltages, we correlated the K^+-induced fluorescence changes to fluorescence changes under voltage clamp in single-dye loaded cells in the whole cell patch configuration. Fluorescence measurements were recorded in response to membrane potentials from -80 to +40 mV. The slopes under the two conditions were highly correlated (-0.0051 V/mV (n=6, R^2=0.99) vs. -0.0060 V/mV (n=11, R^2=0.94) for voltage clamp and ionophore experiments, respectively). The close agreement of the ionophore method of calibration vs. voltage clamp validates permeabilization as a method to determine absolute membrane voltage parameters, such as AP amplitude and resting potential, with high accuracy.

Disclosures: A. Costantino: None. **B.K. Panama:** A. Employment/Salary (full or part-time):; Cytocybernetics, Inc.. **M.W. Nowak:** None. **R.L. Rasmusson:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Cytocybernetics, Inc. **G.C.L. Bett:** E. Ownership Interest (stock, stock options,

royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Cytocybernetics, Inc..

Poster

P930. Advances in Methods for Imaging Neural Activity

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P930.06

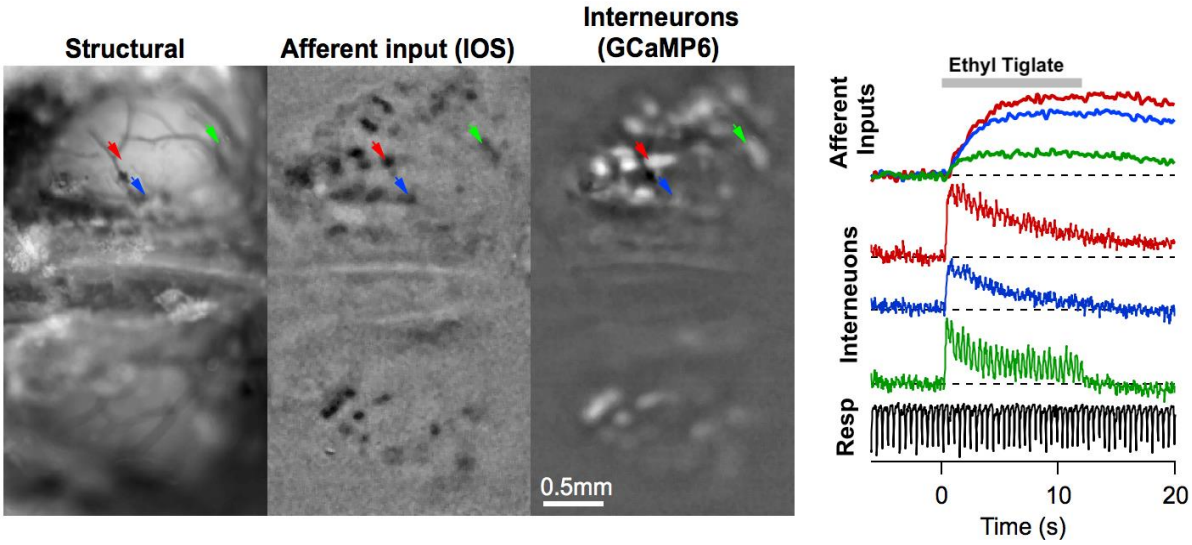
Topic: I.04. Physiological Methods

Support: The Royal Society
The Academy of Medical Sciences grant: SBF002\1033
The Physiological Society

Title: Low cost optical multimodal imaging to study neural circuit function

Authors: C. STEFENS[1], L. VANDE PERRE[2], ***J. JOHNSTON**[1];
[1]Univ. of Leeds, Leeds, United Kingdom; [2]Univ. Libre De Bruxelles, Brussels, Belgium

Abstract: We have developed a low-cost solution for wide-field brain imaging that enables simultaneous measurement of activity from different components of a neural circuit along with other experimental or physiological signals. We obtain a field of view of >4.5 mm using optics from a consumer SLR camera. Using open source LED drivers (optostim) we illuminate alternate frames with different wavelengths to achieve imaging rates of 50 frames per second (fps) for 2 channels or 100/n-channels fps for any combination of channels. We demonstrate its utility by combining optical imaging of intrinsic signals with fluorescent imaging of interneurons expressing GCaMP6 driven by the VGAT promoter to image odour evoked activity in the olfactory bulb afferent inputs and periglomerular neurons.



The image shows structural details of the olfactory bulb (green illumination) alongside the response images for afferent input (IOS, far red illumination) and interneuron activity (GCaMP6, blue illumination). The time course of these optical changes are plotted on the right for 3 glomeruli along side the simultaneously recorded respiration signal. We show that in vivo pharmacological manipulations combined with simultaneous measurement of afferent input and interneuron activity provides a powerful means to study feedback circuits within the olfactory bulb.

Disclosures: C. Stefens: None. **L. Vande Perre:** None. **J. Johnston:** None.

Poster

P930. Advances in Methods for Imaging Neural Activity

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P930.07

Topic: I.04. Physiological Methods

Support: SBIR Grant 2R44MH088176-03

Title: Cross-species wildtype swimming pattern recognition by Remanent Imaging and machine learning

Authors: *L. C. LEUNG[1], P. MOURRAIN[2], P. R. MARTINEAU[1];
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Abstract: We present Remanent Imaging (RI): a novel imaging paradigm for next-generation behavioral assays. RI sets the exposure time as the inverse of the frame rate and, when combined with darkfield illumination, allows data-efficient capture and analysis. Our analytics simultaneously reveal individual and population measures from short timescales to sleep/wake daily cycles and potentially to entire organismal lifetimes. Here we present new data from the recording of multiple fish species that demonstrate how RI can be leveraged to identify species-specific locomotion for the automated classification of aquatic research animal species. We will also explore how RI can be combined with computer vision/machine learning techniques to augment performance of this classification.

Disclosures: L.C. Leung: None. **P. Mourrain:** None. **P.R. Martineau:** None.

Poster

P962. Computational and Analysis Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P962.01

Topic: I.06. Computation, Modeling, and Simulation

Support: NIMH R01MH122752
NIH P20GM103653

Title: Unsupervised mouse behavior analysis with self-organizing maps

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[2]Psychological and Brain Sci., [1]Univ. of Delaware, Newark, DE; [3]Emory Univ., Atlanta, GA

Abstract: A major objective of neuroscience is understanding how brain activity gives rise to complex behavior. Animal models are instrumental for investigating this relationship, as we can manipulate and quantify their behavior in controlled settings. Previously, studying behavior involved laborious manual annotation, and was subject to user bias. Newer supervised machine learning techniques have been instrumental, but still rely on human input for training. Therefore, unsupervised machine learning approaches are needed. To address this issue, we applied a computational model to recordings of freely interacting mice (B6.CAST-Cdh23Ahl+/Kjn) in mixed-sex groups (n = 11, 2 males and 2 females per group). To improve training times, we selected an hour-long period for training, after validating that this block contained a representative sampling of behaviors. A supervised approach for automated behavior annotation, JAABA (Kabra et al., 2013), allowed us to first extract discrete instances of social and non-social behaviors. For each frame of video, JAABA calculates a suite of features for each animal, including information about position and movement. We then calculated additional features to capture characteristics of group interactions. With these features and a subset of those from JAABA, we applied a Self-Organizing Map (SOM) (Kohonen, 2001), creating a two-

dimensional representation of the data. To visualize potential clustering, we projected discrete instances of the behaviors extracted by JAABA onto the trained SOM network. Specifically, we separated subsets of our data containing positively identified examples of 12 types of behaviors (e.g., female approaching either male or female, female fleeing male, female investigating either male or female, males fighting, male fleeing male, male approaching either male or female, male chasing female, male chasing male, male investigating either male or female, male mounting female, and walking) and applied hierarchical agglomerative clustering to their projections. We evaluated the clustering using silhouette scores. Scores closer to 1 indicate high-density, well-separated clusters, while scores closer to 0 indicate greater overlap between clusters. When using 12 clusters (one per behavior classified by JAABA), the mean silhouette score was 0.36. However, after tuning the clustering parameters, we found that removing the constraint of using 12 clusters allowed the silhouette score to improve. Our approach may lead to new techniques for unsupervised behavior annotation, thus leading to a potentially more robust quantification of social behavior.

Disclosures: J. Turk: None. **M.R. Warren:** None. **J.P. Neunuebel:** None.

Poster

P962. Computational and Analysis Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P962.02

Topic: I.06. Computation, Modeling, and Simulation

Support: NSF Robust Intelligence 1704436
NSF NeuroNex 2015317

Title: Using Synthetic Nervous Systems to Model the Multifunctional and Adaptive Feeding Behavior of Aplysia californica

Authors: *Y. LI[1], J. P. GILL[2], P. J. THOMAS[3], V. A. WEBSTER-WOOD[4], H. J. CHIEL[2], R. D. QUINN[1];
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Abstract: Multifunctionality and adaptability are critical for animals to successfully interact with the environment. How do the brain and the body coordinate such multifunctional behaviors? To address this question in the marine mollusk *Aplysia californica*, we are modeling multifunctional feeding behavior, which adapts in response to external sensory cues. Our group previously created a hybrid Boolean/continuous neuromechanical model that could qualitatively reproduce multifunctional *Aplysia* feeding behavior [1]. This previous model was

computationally efficient but lacked many physiological details of the individual neurons and synapses. To assess the effect of neuron dynamics on the simulated behavior, we have translated our Boolean network model into a Synthetic Nervous System (SNS). An SNS is a computational neuroscience model that contains components whose parameters can be set based on actual measurement from neurons.

The SNS network is composed of 3 command-like neurons, 9 interneurons, and 3 motor neurons, similar to the Boolean model [1], but also incorporated intrinsic properties based on a more detailed conductance-based model that was previously published [2]. When simulated without sensory feedback, the model can generate distinct motor patterns somewhat similar to those previously reported in isolated *Aplysia* buccal ganglion [2]. By incorporating feedback from a biomechanical model, the complete simulation with the SNS control can qualitatively reproduce biting (attempts to grasp the food), swallowing (moving the grasped food into the esophagus), and rejection (pushing inedible material out of the buccal mass). Additionally, the SNS can capture post inhibitory rebound and bistability dynamics similar to those observed in the *Aplysia* feeding circuit [2].

The SNS model of *Aplysia* feeding behavior balances biological detail and computational efficiency. The SNS model has a 10-fold simulation time reduction compared with the more complex spiking models of the same circuitry [2]. In the future, we will expand our SNS model to include additional identified neurons and compare model results with animal data. In addition, we are developing optimization and machine learning techniques to tune unknown model parameters. We will also assess the ability of this model to predict animal behaviors and as a robotic controller.

[1] Webster-Wood, et al. Biol Cybern 114, 557–588 (2020). [2] Costa et al. Learn Memory 27, 236–249 (2020).

Disclosures: Y. Li: None. **J.P. Gill:** None. **P.J. Thomas:** None. **V.A. Webster-Wood:** None. **H.J. Chiel:** None. **R.D. Quinn:** None.

Poster

P962. Computational and Analysis Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P962.03

Topic: I.06. Computation, Modeling, and Simulation

Support: NIH Grant 5R01EB029271

Title: Computational modeling of electrical stimulation of peripheral nerves: A new adaptive meshing technique reduces complexity of realistic large-scale models built from CNN segmentation

Authors: *J. DU[1], A. MORALES[2], J.-M. BOUTEILLER[2], G. LAZZI[2];
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Abstract: Among different kinds of electrical stimulation to nerves, electrical stimulation of peripheral nerves has long been used and proven effective in many applications. Although numerous modeling studies have emerged in past years to help guide the design in neural stimulation devices and provide safety criteria, most of the studies use either simplified nerve model with homogeneous fascicles or heterogeneous fascicles populated by axons with artificial radii and locations. No peripheral nerve model has yet been capable of capturing all the morphological and structural details of a complex nerve structure.
With the help of high-resolution imaging, realistic computational modeling, machine learning methods and adaptive meshing technique, we developed a multiscale computational model of the sciatic nerve capable of capturing the model's computational complexity within a reasonable range. Utilization of such model aims to help the development of more effective stimulating devices but also more accurate safety criteria that would ensure that stimulation does not induce nerve injury. Model construction involves first performing image segmentation of high-resolution nerve cross-sectional images using a convolutional neural network method and then building realistic peripheral nerve models from the segmentation results. Then in order to keep the 3D heterogeneous nerve model size within a reasonable range, in addition to the multiscale meshing previously developed by our group which can selectively maintain high resolutions at material boundaries and merge inside, we propose another adaptive meshing technique. With our proposed adaptive meshing, a high level of details and fine resolution is maintained near the nerve periphery, in locations proximal to the contact electrode, and the voxel size is increased further away from nerve periphery where fine resolution is unnecessary. Using this methodology, the number of nodes and edges are decreased resulting in decreased computational complexity and reduced simulation runtimes - while maintaining reasonable accuracy.
Results to be presented include the realistic peripheral nerve model construction results, new adaptive meshing technique and its results validation.

Disclosures: J. Du: None. **A. Morales:** None. **J. Bouteiller:** None. **G. Lazzi:** None.

Poster

P962. Computational and Analysis Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P962.04

Topic: I.06. Computation, Modeling, and Simulation

Title: Automatic segmentation of corticospinal tract track in patients with brain pathology

Authors: Y. H. MOSHE[1], M. ARTZI[2], *M. TEICHER[1];
[1]Gonda Brain Res. Ctr., Ramat-Gan, Israel; [2]The Sagol Brain Inst., Tel-Aviv, Israel

Abstract: Preoperative white matter (WM) fibers visualization and quantitative assessment can assist in the selection of surgical approach, and setting maximum boundaries for the extent of lesion resection while preserving function. Diffusion tensor imaging (DTI) can be used for WM fibers visualization and quantitative assessment. Currently, reconstruction of white matter tract is heavily rely on manual user intervention and requires prior anatomical knowledge, predefined thresholds Etc. The main limitations of DTI tractography methods, which depend on user intervention, are that they are time consuming, and are heavily prone to Inter and intra rater reliability errors. Recently, TractSeg was proposed as a novel, fully automatic approach for direct WM tract segmentation. TractSeg is based on fully convolutional neural network (FCNN) that directly segments WM tracts in fields of fiber orientation distribution function (fODF) peaks. Although showing promising results for the task of automatic fiber tractography, the implementation of TractSeg for the clinical task as of pre-operative fibers visualization in patients with brain lesion is not straight forward. TractSeg predictive model was trained on healthy subjects, and to relative homogeneous dataset in terms of study population and acquisition parameters.This does not represent the clinical scenario, where there are substantial brain morphological changes due to cerebral pathology, as well as high heterogeneity in terms of data acquisition parameters and data and quality.The aim of this study was to implement and optimize TractSeg tool on extremely large cohort of 675 DTI scans, focusing on the corticospinal tract (CST) segmentation, while tackling with uncertainty in the manually CST labels used for model training. Automatic tool computer aided diagnosis (CAD) tool for identification of the lesion area (target area for resection), mapping WM tracts and identifying the fibers in risk (area for preservation) may promote the approach of personalized medicine in patient's candidate for brain surgery

Disclosures: Y.H. Moshe: None. **M. artzi:** None. **M. Teicher:** None.

Poster

P962. Computational and Analysis Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P962.05

Topic: I.06. Computation, Modeling, and Simulation

Support: NIH Grant F32 NS095580
NIH Grant MH112729
NIH Grant DA035913

Title: Benchmarking and scaling of an accurate method for fitting multicompartment models to data

Authors: *A. LADD[1], K. E. BOUCHARD[2], J. BALEWSKI[3], R. BEN-SHALOM[4];
[1]Univ. of California Berkeley, Berkeley, CA; [2]Univ. of California Berkeley, Lawrence Berkeley

Natl. Lab., Berkeley, CA; [3]Lawrence Berkeley Natl. Lab., Berkeley, CA; [4]Univ. of California Davis, Davis, CA

Abstract: Effective algorithms for modeling the biophysical properties of a neuron can lead to an overall improved understanding of how ion channel function and dysfunction affects neuronal activity. A challenge in creating these models is determining realistic channel conductances for a wide spectrum of possible neuronal morphological constraints. Here we present an efficient algorithm for the simulation and evaluation of neuronal models and benchmark this algorithm's scaling performance on high performance computing systems.
A common algorithmic approach in generating neuron models are genetic algorithms. The procedure in which the genetic algorithm constructs realistic neuronal models can be broken down into three phases that repeat until a stopping criteria: (1) initialization of a new generation of parameters for the model, (2) simulation of the models with those parameters, and (3) evaluation of those models. The goal of our work was to minimize the amount of time it takes to initialize, simulate, and evaluate one generation. To accomplish this goal, we have developed a genetic algorithm that uses high performance computing (HPC) nodes to maximize the computational efficiency and minimize the run time of the algorithm. To evaluate the benefits of our redesigned genetic algorithm we determined critical aspects of what defines an efficient simulation/evaluation based algorithm for constructing neuronal models. The results show that both simulation of neuronal models and evaluation of resulting voltage traces are computationally costly and that both tasks can potentially act as bottlenecks. We demonstrate how to balance the use of computational resources with simulation problem size. Furthermore, we show that our GPU/CPU based design enables simulating multiple neuronal responses to multiple stimuli with more efficient resource utilization with nominal cost for run time. This analysis demonstrates an opportunity for researchers to advantage using more experimental stimuli in simulation-based neuronal modeling, enabling the development of richer biophysical neuronal models. The analysis also shows how the endeavor of neuronal modeling can benefit from the development of well-designed and efficient software for simulating and evaluating electrophysiological spike traces.
In conclusion, this analysis shows a methodology for benchmarking the performance of simulation based neuronal modeling and provides future directions and considerations for improving algorithmic efficiency and scalability in simulation based neuroscience.

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Poster

P962. Computational and Analysis Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P962.06

Topic: I.06. Computation, Modeling, and Simulation

Title: Phase-phase coupling network analysis performed on sEEG recordings

Authors: *M. PARRA[1], A. TEKRIWAL[3], D. R. KRAMER[2], J. A. THOMPSON[4]; [2]Neurosurg., [1]Univ. of Colorado Anschutz Med. Campus, Aurora, CO; [3]Dept. of Physiol. and Biophysics; Dept. of Neurosurg., [4]Neurosurg., Univ. of Colorado Sch. of Med., Aurora, CO

Abstract: Epilepsy is a neurological condition characterized by recurrent seizures that may induce a spectrum of semiologies, caused by excessive, synchronous hyperexcitability in neural activity. Approximately 1/3 of people with epilepsy fail to respond to first line medications and may be diagnosed as treatment refractory. Medical management of treatment refractory epilepsy (TRE) often requires surgical intervention. Recent advances in surgery include novel neuromodulatory interventions for TRE which are predicated on a pathophysiological framework that considers epilepsy as a product of network dysfunction. In this study, we propose to use a novel method, phase-phase coupling analysis, for assessing the network properties of seizure generating brain wide circuits as they shift over time and in response to significant events such as seizures. We introduce a novel exploratory method to analyze phase-phase coupling statistics (PPCS) over a long period of time, using the Hilbert transform, that can be used to generate directed graphs that show the flow of information between areas. We define nodes in this process as each sEEG electrode contact and use a thresholded version of statistically significant PPCS to generate the connections in our network. This process is repeated over discrete timesteps, of 30 second lengths. The result of this analysis is a time varying graph that shows how connections are established and broken over time; revealing whole brain LFP mechanics (<200 Hz). After generating these graphs, we then analyze time periods corresponding to significant events using graph theoretic methods. We analyze the small worldness and centrality measures of the networks generated as a measure to determine deviation from baseline brain activity. Through tracking these larger time-scale network changes, events such as response to medication or shifts in seizure frequency may be able to be monitored. In this study, we propose to use this method to monitor and analyze whole brain activity taken from human epilepsy patients undergoing invasive monitoring for seizure localization, and analyze several days worth of continuous data. Preliminary analysis reveals strong intraregion beta:beta coupling (13-30 Hz), followed by a decrease in this beta:beta coupling in the left orbitofrontal region, over a three hour period. This pattern occurs simultaneously with a gradual increase in high gamma:gamma (>100 Hz) coupling strength in the latter half of this three hour period.

Disclosures: M. Parra: None. **A. Tekriwal:** None. **D.R. Kramer:** None. **J.A. Thompson:** None.

Poster

P962. Computational and Analysis Tools I

Location: Virtual Only

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Program #/Poster #: P962.07

Topic: I.06. Computation, Modeling, and Simulation

Support: ANR-17-RHUS-0004, EPINOV (https://anr.fr)
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PHRC-I 2013 EPISODIUM (grant number 2014-27)
Fondation pour la Recherche Médicale (DIC20161236442)

Title: Amortized Bayesian inference of SEEG recordings

Authors: *M. HASHEMI, A. VATTIKONDA1, V. SIP, J. JHA, M. GUYE, F. BARTOLOMEI, M. WOODMAN, V. JIRSA;
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Abstract: Adjusting the stimulation parameters is a key challenge to infer pathological brain networksfrom a large amount of data. Although an efficient Bayesian inversion requires painstakingparameter tuning, the novel methods in deep learning (DL) can efficiently provide theposterior distribution and the relation between model parameters for different observations ofa patient without having to re-run inference, the so-called amortized inference. In this work,we constructed an efficient and accurate method to perform amortized inference for a whole-brain model of epilepsy spread in the context of estimating the brain regional epileptogenicityfrom SEEG recordings. The aim of employing simulation-based-inference (SBI) methodtailored to Bayesian rule, is to perform flexible and efficient inference for complex modelswhen standard methodologies cannot be applied, due to analytic or computational difficulties.The core of the methodology only requires forward-simulations from the computer model,rather than model-specific analytic calculation or exact evaluation of likelihood functionusing MCMC algorithms. Of specific interest is to produce inference for state-space modelsgovern by coupled nonlinear differential equations. One of the goals is to automatically returnthe required (low-dimensional) summary statistics of the observation via ad-hoc DLalgorithms based on a large number of forward-simulations. We demonstrate the efficiencyand accuracy of the training DL such as normalizing flows to automatically infer thedistribution of unknown parameters (the posteriors) of coupled Epileptor model at the whole-brain level given only observed SEEG and prior information. Our results indicated 100%accuracy for the in-silico SEEG recording as generated by virtual brain modeling ofindividual patients. We also investigate the performance of the proposed method againstretrospective data (N=50). The amortized Bayesian framework used in this work proposes anappropriate patient-specific strategy for estimating the epileptogenicity of the brain regions toimprove outcomes after epilepsy surgery.

Disclosures: M. hashemi: None. **A. Vattikonda1:** None. **V. Sip:** None. **J. Jha:** None. **M. Guye:** None. **F. Bartolomei:** None. **M. Woodman:** None. **V. Jirsa:** None.

Poster

P962. Computational and Analysis Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P962.08

Topic: I.06. Computation, Modeling, and Simulation

Support: NIH Grant MH121119

Title: An eLearning Approach to USV Hand Scorer Training with Acoustilytix: A Web-Based Automated USV Scoring Platform

Authors: *C. DUVAUCHELLE[1], C. B. ASHLEY[2], C. CERVANTES[3], E. C. MACASLAN[2], N. MITTAL[4], S. M. FLEMING[5], J. BAILEY[3], M. D. NIEVERA[3], S. I. SOULEIMANOVA[3], B. NYAOGA[3], L. LICHTENFELD[3], W. MADDOX[6];
[1]Pharmacol. and Toxicology, Univ. of Texas At Austin, Austin, TX; [2]Cornerstone Res. Corp, Dayton, OH; [3]Univ. of Texas, Austin, TX; [4]ZS Mgmt. Consulting, San Francisco, CA; [5]Pharmaceut. Sci., Northeast Ohio Med. Univ., Rootstown, OH; [6]CD&SC, Austin, TX

Abstract: Ultrasonic vocalizations (USVs) are known to reflect neurotransmitter activity in the ascending limb of the mesolimbic dopaminergic and cholinergic neurotransmitter systems and serve as noninvasive, real-time biomarkers of dopaminergic and cholinergic neurotransmission. In addition to reflecting emotional processing in the limbic system, recent studies have shown USV utility in differentiating between sex and rat strain, transgenerational effects of substance exposure and prediction of future alcohol consumption. Although a valuable experimental tool, collecting and processing USV data is time-intensive and costly, creating a significant bottleneck by limiting researchers' ability to employ fully effective experimental designs, and serves as a barrier to entry for other researchers. We are currently developing Acoustilytix, a web-based automated USV detection and scoring algorithm that is environment- and animal-agnostic. Building upon the fact that some level of hand scoring competency is needed in any USV lab, we describe a new feature that leverages learning science principles to optimize hand scoring training. In brief, trainees are presented, one by one, with a large number of calls (e.g., 1000) which they may examine visually with a spectrogram or auditorily at a reduced rate. For each call, the trainee selects a call category and is given immediate corrective feedback (based on the expert scorer's classification). If in error, the trainee is allowed to make another classification judgement. This process continues until the trainee provides the correct classification for each call. To test the new feature, we asked five individuals who had been give traditional hand scoring training and had been hand scoring USVs for months (6000-31,000 calls classified by each hand scorer) to score a single test file with 1000 calls that had been hand scored by an

expert. We used a kappa statistic to compare each hand scorer with the expert hand scorer and found moderate inter-rater reliability ranging from .30 - .55 with a mean of .42. Next, we asked two individuals with no prior USV hand scoring experience to complete the same training and test and found kappa values of .42 and .47 which are comparable to those of the 5 experienced scorers. Next, we asked the 5 experienced scorers to complete Acoustilytix training and a subsequent test, and found kappa values ranged from .30 - .75 (average kappa = .60). These findings suggest that hand scoring training in which the learner is given immediate call-by-call feedback, and where the teaching signal is based on the call classification from an expert hand scorer leads to more accurate and consistent hand scoring.

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Poster

P962. Computational and Analysis Tools I

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Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P962.09

Topic: I.06. Computation, Modeling, and Simulation

Support: NS100531-01A1

Title: A machine learning approach to analysis of skilled reaching behavior in mice identifies behavioral compensation after spinal cord injury

Authors: *N. O'NEILL[1], K. MAH[1], M. MURRY[2], J. L. BIXBY[3], V. LEMMON[4]; [2]Neurosurgery/Miami Project to Cure Paralysis, [1]Univ. of Miami Miller Sch. of Med., Miami, FL; [3]Miami Proj to Cure Paralysis, Univ. Miami, Miller Sch. Med., Miami, FL; [4]Neurolog. Surgery, Univ. of Miami, Miami, FL

Abstract: Injuries to the cervical spinal cord represent around 60% of human spinal cord injuries (SCIs). For these patients, any recovery of arm or hand function is a major priority. Similarities between human and rodent “reach-to-eat movements” mean that analysis of mouse forelimb reaching behavior has potential clinical relevance. While recovery in the single pellet reaching assay is normally measured by successful pellet retrieval, this outcome measure fails to account for compensatory movements that may be used in the task. Our goal was to establish a skilled forelimb reaching task that can be used to distinguish restoration of function from compensation after SCI. We used a machine learning approach to automate the quantitative assessment of videos. Computer vision (DeepLabCut) was used to track individual paw features, allowing for a more refined assessment of reaching behavior after SCI. Kinematic analysis of movements pre- and post-injury illuminated deficits in specific features of the reaching motor pattern (pronation)

that persisted independent of the ability to successfully retrieve pellets. A C5 cervical SCI causes specific deficits in pronation and grasping. To continue retrieving pellets after a loss in those movements, animals can compensate by approaching the pellet with a shallower angle and sweeping across rather than turning the paw over. The change in distance between two points on the paw in the X plane provides a quantitative measure of paw angle (and thus pronation). By plotting this relative distance over the course of an attempt, we can measure the extent of pronation that animals exhibit during a reach. We compared animals from opposite ends of the spectrum with respect to post-injury success at 4 WPI (>35%, or pre-injury levels, vs. complete loss of skill, or <5%). Comparing pronation capabilities between these groups suggests that, despite large differences in retrieval success, the two sets of animals displayed nearly identical deficits in pronation capabilities compared to pre-injury baseline. Our results suggest that, even for an assay requiring fine motor skills, measurements of recovery on a binary (success/fail) scale largely measure an animal's ability to compensate rather than a restoration of normal function in the injured tracts and pathways.

Disclosures: N. O'Neill: None. **K. Mah:** None. **M. Murry:** None. **J.L. Bixby:** None. **V. Lemmon:** None.

Poster

P962. Computational and Analysis Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P962.10

Topic: I.06. Computation, Modeling, and Simulation

Title: N2A: a language and software tool for large-scale modeling

Authors: *F. ROTHGANGER, *F. ROTHGANGER;
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Abstract: Sharing models and data is a prerequisite for progress toward a full understanding of brain function. The Neuroscience Information Framework (NIF) does this for many forms of descriptive data. Interchange languages such as NeuroML/LEMS provide a simulator-agnostic description of models, and repositories such as NeuroML-DB and ModelDB make them searchable and accessible.
However, exchanging models and data is not sufficient. It is also necessary to assemble those shared models into larger functional units, ultimately reaching the level of an entire nervous system. To create abstract descriptions of function, the modeling system must be capable of crossing all the scale levels, and component models should be expressed in a form suitable for automated analysis.
N2A is an effort toward these challenging goals. It treats models as data rather than code. This declarative approach describes a model as a set of attributes and equations, without specifying a

step-by-step procedure for simulation. It emphasizes the relationships between values within a model and the relationships between models in a larger functional unit.
The declarative approach allows one model to directly extend and modify another, simply by referencing the parent model and declaring new values for specific attributes and equations. A model may also incorporate other models as components, allowing the assembly of arbitrarily deep systems. We will demonstrate an open-source implementation of N2A.

Disclosures: F. Rothganger: None.

Poster

P982. New Developments in Transcranial Stimulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P982.01

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Brain Initiative Grant 1RF1MH124909

Title: Induced electric fields in the brain exhibit orientation-specific effects on neocortical neurons in awake non-human primates

Authors: *H. TRAN[1], I. ALEKSEICHUK[1], S. SHIRINPOUR[1], B. VAIL[2], D. MAISSON[2], B. VOLOH[2], J. ZIMMERMANN[2], A. OPITZ[1];
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Abstract: Transcranial alternating current stimulation (tACS) is a non-invasive method to modulate ongoing neural activity in the brain. Previous work shows that external oscillations entrain neural firing activity in a phase- and dose- dependent manner. Thus, tACS holds promise as a new treatment option for neuropsychiatric disorders characterized by pathological brain oscillations. However, neural responses to oscillating electric fields exhibit large variability. We hypothesize that the neural responses to the external field largely depend on the neocortical column orientations relative to the induced electric fields.
Here, we first perform a computational study to investigate the relationship between 10 Hz tACS and neural entrainment in morphologically realistic neocortical neurons exhibiting ongoing spiking activity (set of 25 cells – 5 cells for each cortical layer). The orientation of the electric field is in the y-axis orientation corresponding to the somato-dendritic axis of pyramidal cells and intensities were between 0.1 mV/mm and 3 mV/mm. Neural entrainment was quantified by computing the phase-locking value (PLV) of spikes times across neurons and field strengths. We found that 1) pyramidal neurons are more entrained than interneurons resulting in a higher phase-locked spiking to the applied oscillation (pyramidal cells n=15, 0.0788 +/- 0.0821 and interneurons n=10, 0.0495 +/-0.0414 at 1mV/mm) and 2) pyramidal cells are more preferentially entrained when parallel to the electric field direction.

Second, we perform in-vivo recordings of single-unit activity in awake non-human primates while applying tACS in 3 orientations (Anterior-Posterior, Left-Right and diagonal orientations) and 2 intensities for each of them (0.4mA and 0.8mA). We record neuronal activities using a 128-channel amplifier at 30 kHz sampling rate with glass coated electrodes. Preliminary findings in one macaque (8 y.o., male) show that neurons (N=159) exhibit significant entrainment to tACS with strong orientation preference (Rayleigh test $p \leq 0.01$).
Overall, these findings advance our mechanistic understanding of tACS effects and highlight the importance of neuronal architecture on the susceptibility to the external electric fields. Highlighting neural directional sensitivity represents a milestone for understanding brain networks interactions.

Disclosures: H. Tran: None. **I. Alekseichuk:** None. **S. Shirinpour:** None. **B. Vail:** None. **D. Maisson:** None. **B. Voloh:** None. **J. Zimmermann:** None. **A. Opitz:** None.

Poster

P982. New Developments in Transcranial Stimulation

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Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P982.02

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Grant R01NS109498

Title: Investigating early neural responses during transcranial magnetic stimulation in non-human primates

Authors: *N. D. PERERA[1], S. SHIRINPOUR[1], I. ALEKSEICHUK[1], M. WISCHNEWSKI[1], G. LINN[2], C. E. SCHROEDER[3], A. Y. FALCHIER[2], A. OPITZ[1];
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Abstract: Transcranial Magnetic Stimulation (TMS) is a non-invasive neuromodulation technique commonly utilized in brain research and clinical applications in neuropsychiatric disorders. Despite its widespread use, the underlying neural mechanisms of TMS and how it modulates brain activity is still poorly understood. It has been debated that some of the TMS evoked responses could be the result of peripheral effects rather than direct neural activation. To this end, we recorded brain activity from an anesthetized monkey with implanted electrodes projecting from visual area (V2) to frontal eye field (FEF), secondary auditory cortical (AUD) and temporal (TE) regions in the left hemisphere. The exact locations of electrode contacts were determined by co-registering post- and pre-surgery MR images. Biphasic single-pulse TMS was

delivered over the left hemisphere to 4 different locations. A control stimulation location over the right hemisphere was used to demonstrate spatial specificity. A sham protocol was executed by tilting the coil away from the head to mimic the auditory response. Stimulation intensities were set to 10, 25, 50, 70 and 90% of the maximum stimulator output. We preprocessed the neural data by removing channels contaminated with large noise through visual inspection, removing the TMS and the muscle artifacts (calculated from external EMG data), interpolating, down-sampling to 1 kHz and bandpass filtering at 1-50 Hz. For analysis, we performed time-locked averaging across trials. Cluster based ANOVA test indicated time windows with significantly different amplitudes from 25 - 100 ms ($p < 0.006$) for anterolateral stimulation location and from 25 - 136 ms ($p < 0.001$) for lateral stimulation location. Post-hoc multiple comparisons of the N50 peak of TMS evoked potentials revealed intensity-dependent clusters. Linear regression of N50 with intensity showed a maximum correlation of 0.52 ($p = 5.5\times10^{-12}$) for lateral coil location and 0.37 ($p = 4.0\times10^{-27}$) for anterolateral coil location. The activation region, quantified by the normalized N50 amplitudes per electrode showed movement with the coil location. Furthermore, contacts of AUD electrode in the auditory belt revealed consistent low frequency oscillations in both TMS and sham conditions, which is attributed to the auditory response to the 'click' sound. Our study provides preliminary evidence that the TMS effects are dose dependent, localized and can be separated from peripheral stimulation effects.

Disclosures: N.D. Perera: None. **S. Shirinpour:** None. **I. Alekseichuk:** None. **M. Wischnewski:** None. **G. Linn:** None. **C.E. Schroeder:** None. **A.Y. Falchier:** None. **A. Opitz:** None.

Poster

P982. New Developments in Transcranial Stimulation

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Program #/Poster #: P982.03

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Brain Initiative Grant 1RF1MH124909
University of Minnesota MnDRIVE Initiative

Title: Boosting beta oscillations enhances working memory performance in surgical epilepsy patients

Authors: *I. ALEKSEICHUK[1], C. SAIOTE[2,4], S. D. KOENIG[2], M. WISCHNEWSKI[1], T. A. BERGER[1], J. A. PEBBLES[2], R. A. MCGOVERN, III[2], M. C. PARK[2], A. B. HERMAN[3], D. P. DARROW[2], A. OPITZ[1];
[1]Dept of Biomed. Engin., [2]Dept of Neurosurg., [3]Dept of Psychiatry, Univ. of Minnesota, Minneapolis, MN; [4]Waisman Ctr., Univ. of Wisconsin-Madison, Madison, WI

Abstract: Neural oscillations reflect and organize brain functions. Transcranial alternating current stimulation (TACS) can non-invasively modulate brain oscillations by entraining neural ensembles to the phase and frequency of the applied current. Thus, TACS presents a versatile tool for basic research and therapeutic applications for "oscillopathies." However, the mechanisms of TACS and their effect on cognition are still incompletely understood. Research in surgical epilepsy patients, who undergo invasive brain recordings, presents a unique opportunity to explore a causal mechanistic link between the modulation of brain oscillations, changes in neurophysiology, and cognitive functioning. Here, we aimed to investigate the causal role of beta oscillations in supporting human cognitive functions. Patients who met clinical guidelines and provided informed consent were implanted with intracranial subdural grids and depth electrodes (100-200 contacts) according to medical needs. The study is conducted in accordance with the Declaration of Helsinki and IRB regulations (University of Minnesota, MN). Intracranial recordings were acquired at a high sampling rate (16 kHz) and dynamic range (24 bits). Participants performed a classical 2-back working memory task while receiving either sham stimulation (passive control), beta TACS at 23 Hz (main condition) or theta TACS at 4 Hz (active control) using bilateral temporal montage. Altogether, five participants performed 45 experimental sessions with different conditions, totaling over 5000 task trials. Generalized linear mixed effect model shows significant effect of stimulation on memory performance: $F(2,5178) = 8.9$, $p = 0.0001$. Post-hoc t-tests reveal significant improvement ($p < 0.05$) following beta TACS in comparison to sham or theta TACS, while theta TACS induced no changes. Bayesian hierarchical diffusion drift modeling of cognitive performance demonstrates an improvement in the quality of the memory process (drift rate *v*, Bayes factor = 3.8). In contrast, there are no changes in the speed-accuracy tradeoff (threshold *a*). Beta TACS improves the memory process in all studied patients with an effect size of Hedges' $g = 0.78$. In invasive brain recordings, we observe a significant and frequency-specific increase in beta oscillations following beta TACS. This significantly correlates with improved working memory performance ($p < 0.05$), providing a mechanistic link from stimulation to behavior. The present findings demonstrate a causal role of beta oscillations in working memory performance and the feasibility of supporting cognitive functions in humans via modulation of beta activity.

Disclosures: I. Alekseichuk: None. **C. Saiote:** None. **S.D. Koenig:** None. **M. Wischnewski:** None. **T.A. Berger:** None. **J.A. Pebbles:** None. **R.A. McGovern:** None. **M.C. Park:** None. **A.B. Herman:** None. **D.P. Darrow:** None. **A. Opitz:** None.

Poster

P982. New Developments in Transcranial Stimulation

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Program #/Poster #: P982.04

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH grant RF1MH117428
Minnesota's Discovery, Research, and Innovation Economy (MnDRIVE)

Title: Phase-specific modulation of cortical excitability using closed-loop TMS-EEG

Authors: *M. WISCHNEWSKI, Z. J. HAIGH, S. SHIRINPOUR, I. ALEKSEICHUK, A. OPITZ;
Biomed. Engin., Univ. of Minnesota, Minneapolis, MN

Abstract: Transcranial magnetic stimulation (TMS) is able to activate neurons in the motor cortex non-invasively. As such, TMS can be a useful method to promote recovery of motor disorders, for example after stroke. However, due to neurophysiological variability TMS effects differ between and within participants. On source of variability is the phase of motor cortex-related neural oscillations. In this within-subjects, double-blind study, we investigate the role of brain oscillation phase of the motor cortex Mu (8-13 Hz) and Beta (14-30 Hz) rhythm. For this we used a novel closed-loop TMS-electroencephalogram (EEG) system. Phase-specific TMS with our closed-loop system is achieved in two steps. First, during the training phase, specific individual Mu and Beta frequency is determined in a three-minute resting-state EEG. In the second step phase-specific TMS-EEG is applied concurrently. For this, EEG oscillation are read out and the phase is analyzed in real-time. The phase information is forwarded to the TMS and phase-specific pulses are applied. Cortical excitability is tested by measuring the motor-evoked potential (MEP) of the first dorsal interossei (FDI) muscle, at different phases of the Mu and Beta oscillation. For analysis MEPs are normalized to the grand average. Four phases are being explored: the peak, trough, rising-phase, and falling phase. We hypothesized phase-dependent MEP amplitude modulations for both Mu and Beta rhythms. Preliminary results (n = 4) showed that normalized MEPs were largest at the Mu rising phase (1.024 ± 0.018 mV) and smallest at the Mu falling phase (0.938 ± 0.030 mV). In contrast, analyzing MEPs during Beta oscillations revealed that MEPs were largest at the falling phase (1.032 ± 0.016 mV), and smallest at the trough (0.964 ± 0.015 mV). From these data the tentative conclusion can be reached that MEP amplitude modulation is both phase- and frequency-specific, with opposing results for the Mu and Beta rhythm. Our results imply modulation of motor cortex activity may be optimal when phase is considered. Therefore, our results provide valuable insights for clinical application of TMS in rehabilitation of motor-related disorders, such as stroke.

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Poster

P982. New Developments in Transcranial Stimulation

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Program #/Poster #: P982.05

Topic: I.08. Methods to Modulate Neural Activity

Support: RF1MH124909

Title: Transcranial alternating current stimulation induces neural entrainment in neuronal network models

Authors: *Z. ZHAO, S. SHIRINPOUR, H. TRAN, A. OPITZ;
Biomed. Engin., Univ. of Minnesota Twin Cities, Minneapolis, MN

Abstract: Transcranial alternating current stimulation (tACS) is a non-invasive brain stimulation technique that can modulate ongoing neural activity in the brain by affecting single-neuron spike timing and causing neural entrainment. Thus, tACS may be a promising treatment for neuropsychiatric disorders as well as a useful tool for studying human cognition. However, how the external oscillating electric fields interact with neuronal spiking activity in a neuronal network is still insufficiently understood. Experiments using invasive or non-invasive methods are crucial to address the fundamental questions about the neural responses to stimulation. In addition, computational modeling helps explain the neuronal dynamics observed from recordings by exploring the model parameters and testing hypotheses. We conduct a computational modeling study to investigate the relationship between the electric field and neural entrainment in the coupled population of neurons. Here, we employ a neuron model that consists of two compartments (soma and apical dendrite), which involves the cable equation as well as multiple ionic currents such as Hodgkin-Huxley kinetics. Therefore, we are able to model the effect of the extracellular field on a neuron in a straightforward way. Furthermore, we spatially arrange and connect excitatory and inhibitory neurons to match the realistic neuron distribution in cortical layers. Then, we explore different parameters in this neuronal network, such as types of synapses and synaptic strength to probe how a single neuron and neuronal population respond to the extracellular electric field. We measure the neural entrainment by computing the phase-locking value (PLV) of spike times across neurons with different intrinsic firing rates and electric field strengths. Preliminary results suggest that the excitatory synaptic connection amplifies the entrainment effect on the post-synaptic neurons while the inhibitory synapse reduces the entrainment effect. Investigating the network dynamics will provide additional insights on how tACS affect neuron spiking patterns on the network level. A better understanding of tACS mechanisms of action allows the design of more effective tACS treatment modalities for neuropsychiatric disorders.

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Poster

P982. New Developments in Transcranial Stimulation

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Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Grant R01MH118930
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BMBF Grant 01GQ1804B
von Behring Röntgen Foundation Grant

Title: Neuron Modeling for Transcranial Magnetic Stimulation (NeMo-TMS) Toolbox

Authors: *S. SHIRINPOUR[1], N. HANANEIA[2], J. ROSADO[3], H. TRAN[1], C. GALANIS[4], A. VLACHOS[4], P. JEDLICKA[2], G. QUEISSER[3], A. OPITZ[1];
[1]Univ. of Minnesota Twin Cities, Minneapolis, MN; [2]Justus-Liebig-University, Giessen, Germany; [3]Temple Univ., Philadelphia, PA; [4]Univ. of Freiburg, Freiburg, Germany

Abstract: Transcranial Magnetic Stimulation (TMS) of the dorsolateral prefrontal cortex is currently utilized as a non-invasive neuromodulation tool for the treatment of depression. Nevertheless, the basic mechanisms underlying TMS effects at the neuronal level are largely unknown. Since the recording of single-unit activity *in vivo* during TMS is challenging in animals, and typically not possible for humans, computational modeling is an important tool to investigate the neuronal response to TMS. Here, we develop a new multi-scale pipeline (NeMo-TMS) for modeling TMS effects across spatial scales. On the macro-scale, we generate a realistic head model from MRI that includes multiple tissue types (white matter, grey matter, cerebrospinal fluid, skull, and scalp) and use the SimNIBS software to simulate the electric field induced in the brain for a given TMS coil position and orientation using Finite Element Method (FEM). Afterward, simulated electric fields from the previous step are coupled with morphologically realistic neuronal models. These models are solved by discretizing the neuronal branches into small compartments and by numerically solving the cable equation and channel dynamics in the NEURON environment. These neuron-scale simulations allow investigating membrane voltage (depolarization/hyperpolarization), action potential initiation and propagation, field intensity, and orientation necessary for modulating neuron response, etc. Then, we incorporate the membrane voltage data to simulate the calcium concentration induced by voltage-gated calcium-channels at the subcellular scale by solving the calcium dynamics equations. This step is important for understanding neural plasticity due to rTMS protocols. In this study, we also provide results to demonstrate how different neuron types behave distinctly for the same external electric field and therefore make a case for the importance of such multi-scale realistic modeling of neurons. Our pipeline can facilitate research as a tool for hypothesis testing and prediction technique for experiments. Additionally, modeling has the potential to be used for calculating the dose needed for efficient treatment in clinical applications.

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Poster

P982. New Developments in Transcranial Stimulation

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Program #/Poster #: P982.07

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Grant RF1MH117428

Title: Extending SimNIBS to integrate non-invasive brain stimulation with functional imaging data and primate head segmentation

Authors: *K. MANTELL[1], S. SHIRINPOUR[1], X. LI[3], O. PUONTI[4,5], K. MADSEN[4,6], Z. HAIGH[1], E. CRUZ CASILLO[1], I. ALEKSEICHUK[1], T. HENDRICKSON[2], T. XU[3], A. THIELSCHER[4,5], A. OPITZ[1];
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Abstract: Non-invasive brain stimulation (NIBS) is currently being used as a treatment method for psychiatric disorders such as major depression, obsessive-compulsive disorder, and substance use disorder. NIBS is also increasingly utilized as a research tool for causal manipulations of brain activity. Two of its main methods, Transcranial Magnetic Stimulation (TMS) and Transcranial Electric Stimulation (TES) create electric fields in the brain to modulate neural activity. Precise knowledge of these electric fields is crucial for experimental design and choice of stimulation parameters for precise dosing. Computational modeling has been used extensively to predict the induced electric fields. Due to the growing clinical and research applications of NIBS, there is increasing demand in the accessibility of such tools. SimNIBS is one of the leading platforms for NIBS electric field modeling (simnibs.org). In this project, we are increasing the functionality and accessibility of SimNIBS. This includes: 1) Developing automatic non-human primate head segmentation. This will open the SimNIBS tool more researchers interested in translational research. 2) Extending the visualization and graphical interfaces of SimNIBS. This will make the software more user-friendly and bring additional features such as intuitive selection of regions of interest. 3) Improving human MRI segmentation to increase accuracy, speed, and robustness of head model generation. 4) Utilizing software containers to provide simple cross-platform and high-performance access to SimNIBS. 5) Adding functionality to integrate neuronavigation systems with SimNIBS to assist in accurate targeting of the brain and to accurately model test conditions. 6) Generating automated recommendations for optimal coil positions and electrode locations. This will help users plan tailored experimental and clinical setups.Thus far, progress on these updates have resulted in several publications including (Wang et al., NeuroImage, 2021), (Puonti et al., NeuroImage,

2020), and (Saturnino et al., J. Neural Eng., 2021) for items 1), 3), and 6), respectively.Our continued work in these six areas of developments will widen the pool of users such as clinicians, human, and animal researchers to incorporate modeling platforms in their work. Modeling is a crucial part of efforts to understand the NIBS mechanism of action, to translate the experimental findings between animals and humans, and to find the optimal targeting and dosing of brain stimulation. Altogether, these functions will help researchers and clinicians increase non-invasive brain stimulation treatment efficacy.

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Poster

P982. New Developments in Transcranial Stimulation

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Program #/Poster #: P982.08

Topic: I.08. Methods to Modulate Neural Activity

Title: Effects of repetitive transcranial magnetic stimulation on somatosensory evoked potentials via a miniaturized coil in rats

Authors: *W. JIANG, R. ISENHART, Z. LU, H. XU, J. PACE, D. LEE, C. LIU, D. SONG; USC, Los Angeles, CA

Abstract: While repetitive transcranial magnetic stimulation (rTMS) is a widely-used non-invasive technique for neuromodulation, its underlying neurobiological mechanisms remain unclear. Rodent models are useful tools for elucidating plasticity-like mechanisms of rTMS since they allow simultaneous invasive recording of the neural circuits to monitor the electrophysiological effects of rTMS. However, the lack of small coils suitable for small animal stimulation hinders the rTMS studies in rodent models; Most rodent rTMS studies have used commercially available coils designed for human brain stimulation, which lack stimulation focality. To address this, we have designed, fabricated, and tested a miniaturized TMS coil for rodent experiments. The TMS coil (5 mm outer diameter) was constructed with a C-shaped iron powder core. Insulated copper wires were wound (30 turns) around the core and evenly distributed over its circumference. The magnetic field generated by the TMS coil was measured with a search-coil magnetometer. A maximum magnetic field strength of 473 mT was achieved. The induced electric field was measured with a dipole probe in saline. Maximum electric field (5.2 V/m) was focally created at the center of the coil. To evaluate the effect of rTMS on cortical activities, Platinum-Iridium microelectrodes were implanted in the somatosensory cortices of anesthetized rats (n = 3) to record somatosensory evoked potentials (SSEPs). SSEPs were evoked by electrically stimulating the median nerve of the contralateral forelimb using

subdermal needle electrodes. High frequency (10 Hz) rTMSs were delivered for three minutes above the recording site (1800 pulses). SSEPs were monitored before and after rTMS. Results show that 10 Hz rTMS suppresses the SSEP amplitudes by 33±7% compared to the baseline. SSEPs in the first 5 minutes following rTMS show a slightly decreased latency (91±4%). SSEP amplitudes gradually recover to 85±12% of baseline 10 minutes after rTMS, and 84±14% of baseline 15 minutes after rTMS. The mean latency increases to 107±11% of baseline 10 minutes after rTMS, and 95±3% 15 minutes after rTMS. These results indicate that rTMS (3 minutes, 10 Hz) can effectively modulate SSEPs in rats. The miniaturized TMS coil and the stimulation-recording paradigm described here provide a valuable experimental platform for studying the effect of TMS on brain circuits in rodents. It has the potential to assist the development of novel therapeutic strategies for treating various neurological disorders in humans.

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Poster

P982. New Developments in Transcranial Stimulation

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Topic: I.08. Methods to Modulate Neural Activity

Support: French ANR 19-CE37-0021

Title: Long-lasting inhibitory effects of low-intensity rTMS on activity and excitability of neocortical pyramidal neurons in vivo.

Authors: M. BOYER[1], P. BAUDIN[2], C. STENGEL[2], A. VALERO-CABRE[2], A. LOHOF[1], S. CHARPIER[2], *R. SHERRARD[1], S. MAHON[2];
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Abstract: Repetitive Transcranial Magnetic Stimulation (rTMS), either at classically-used high intensity (in Teslas, T) or sub-excitation low intensity (μT-mT; LI-rTMS), is used to increase neural circuit plasticity and shows promise in treating human neurological dysfunction. However, the processes activated by magnetic fields at the single neuron level remain largely unknown, preventing the optimization of potentially therapeutic rTMS protocols.
We investigated the impact of continuous focal 10-Hz LI-rTMS on neuronal excitability with in vivo intracellular and electrocorticographic (ECoG) recordings from the primary somatosensory cortex in sufentanil-sedated Sprague-Dawley rats of either sex. Neuronal electrical properties were compared before and after 10 minutes LI-rTMS, so that each neuron was its own internal control. All experiments were approved (APAFIS no. 18003-2019051019017280) and were in accord with the European Union directive (2010/603/EU).

In pyramidal cortical neurons (n=16), this LI-rTMS protocol produced: (1) a large hyperpolarization of membrane potential, (2) a near complete abolition of spontaneous firing and (3) a decreased amplitude of ongoing synaptic events. This alteration of spontaneous activity correlated with significant changes in neuronal membrane excitability, notably a decrease in the input resistance and gradual and persistent increase in the current threshold required to fire cortical neurons.
These findings indicate that a short period of continuous 10-Hz LI-rTMS can strongly, and durably, reduce the membrane and synaptic properties of somatosensory cortical neurons. We are currently further investigating the functional outcome of this time-dependent modulation of cortical excitability by analyzing the ECoG patterns and whisker-evoked sensory responses before and after LI-rTMS.

Disclosures: M. Boyer: None. **P. Baudin:** None. **C. Stengel:** None. **A. Valero-Cabre:** None. **A. Lohof:** None. **S. Charpier:** None. **R. Sherrard:** None. **S. Mahon:** None.

Poster

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Time: Tuesday, November 9, 2021, 10:15 AM - 11:15 AM

Program #/Poster #: P982.10

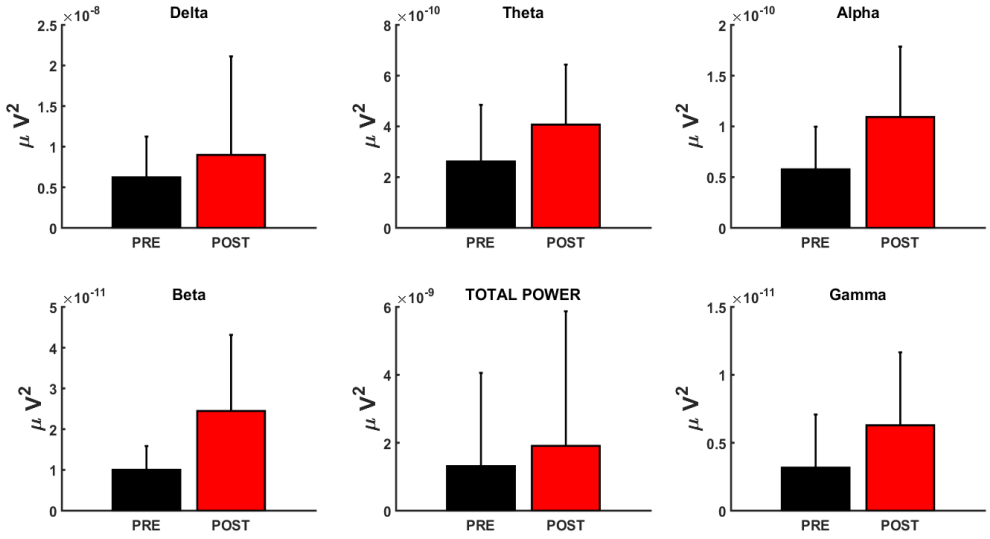
Topic: I.08. Methods to Modulate Neural Activity

Title: Transcranial pulse stimulation in Alzheimer´s disease: A global power boost on EEG-oscillations?

Authors: *L. WOJTECKI[1], C. A. TRENADO COLIN[2], C. CONT[3], K. M. ZIMMERMANN[4], N. STUTE[4], A. SCHNITZLER[5];
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Abstract: Background: Transcranial Pulse Stimulation (TPS) is a new therapy that uses shockwaves for the treatment of Alzheimer´s Disease (AD). In humans, there is first evidence for beneficial clinical effects after a series of six TPS sessions. Magnetic resonance imaging studies suggest up-regulation of memory networks as well as increased cortical thickness after stimulation. Effects on oscillatory electrical brain activity measured with resting state electroencephalography (EEG) have not been reported so far. **Methods:** A consecutive number of four pilot TPS-treated AD-patients was examined. TPS-naïve resting EEGs were recorded before treatment and compared with EEG-recordings directly after first stimulation. All patients received 4Hz TPS of about 6000 pulses of 0.2 mJ/mm2 (navigated bifrontally, biparietally, bitemporally and praecuneus) using the Neurolith System (Storz Medical). Frontal, parietal, central and temporal channel clusters were analyzed with regard to their alpha, theta delta and

gamma power. **Results:** The immediate effect after TPS on grand-average EEG resting state showed an increase of power in all frequency bands and for total power (Figure), although a significant difference between pre- and post-stimulation was not revealed at each specific band across four patients due to power variability. However, the average power increase was reflected by a power change in each individual patient and by consistent changes across EEG-channels. **Discussion / Conclusion:** Although limited due to a high variability (and possibly due to underpower) we find these pilot measurements remarkable as we see consistent oscillatory changes after one single session of TPS across frequency domains, EEG-channels and patients. More measurements should be added including longitudinal recordings over the treatment course of six sessions with correlations of clinical treatment effects.



Disclosures: **L. Wojtecki:** F. Consulting Fees (e.g., advisory boards); Consulting fees from Storz Medical. **C.A. Trenado Colin:** None. **C. Cont:** None. **K.M. Zimmermann:** None. **N. Stute:** None. **A. Schnitzler:** None.

Poster

P008. Glial Development and Neuronal Interactions

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P008.01

Topic: A.01. Neurogenesis and Gliogenesis

Support: R01 NS088529

Title: Investigating a role for Tcf3/12 during development in the ventral telencephalon and oligodendrogenesis

Authors: *M. TALLEY[1,2], D. NARDINI[1], L. EHRMAN[1], R. R. WACLAW[1];
[1]Cincinnati Children's Hosp. Med. Ctr., Cincinnati, OH; [2]Univ. of Cincinnati, Cincinnati, OH

Abstract: Class II pro-neural and pro-glial basic helix-loop-helix (bHLH) transcription factors (TFs), such as Olig2 and Ascl1, are known to be required and sufficient for stem cell differentiation into distinct cell types in the central nervous system. bHLH TFs dimerize in order to bind to DNA and promote those cell fates. Class II bHLH TFs preferentially heterodimerize with class I bHLH transcription factors, also known as E-proteins. E-proteins are thought to be widely expressed, however, their unique function in the developing telencephalon remains largely unknown. In this study, we characterize the expression of all three E-proteins throughout forebrain development, including their expression within the oligodendrocyte (OL) lineage in the postnatal brain. In addition, we generated *Tcf3/12* conditional double knockouts (cdKOs) using *Olig2*$^{Cre/+}$ to understand the requirement of *Tcf3* and *Tcf12* in the ventral telencephalon and oligodendrogenesis. Double knockouts show an expansion of progenitor regions in the embryonic ventral telencephalon resulting in abnormal morphology of the medial ganglionic eminence. Also, *Tcf3/12* cKOs exhibit a reduction of oligodendrocyte progenitor cells (OPCs) in mid and late embryonic stages. Interestingly, OPC numbers recover at postnatal stage, but a decrease in mature OLs remains at 1 and 2 months of age. Our preliminary data indicates that cortical interneurons from the Olig2+ progenitor lineage are less affected by the loss of *Tcf3/12* than OLs. Combined, our results suggest that *Tcf3* and *Tcf12* play a key role in ventral progenitors of the embryonic telencephalon and in multiple stages of the oligodendrocyte lineage.

Disclosures: M. Talley: None. **D. Nardini:** None. **L. Ehrman:** None. **R.R. Waclaw:** None.

Poster

P008. Glial Development and Neuronal Interactions

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P008.02

Topic: A.01. Neurogenesis and Gliogenesis

Support: CIHR Doctoral Award

Title: Changes in internodal length during development.

Authors: *A. BEAUDRY-RICHARD[1,2], N. JABASSINI[1], C. CORDANO[1], R. KOTHARY[2], J. R. CHAN[1], A. J. GREEN[1];
[1]Univ. of California San Francisco, San Francisco, CA; [2]Univ. of Ottawa, Ottawa, ON, Canada

Abstract: The pattern of myelin plays a crucial role in determining the speed of conduction of electrical signals in the nervous system. The principal determinants of internodal length during

development remain to be elucidated. Consequently, we don't yet understand the dynamic responsiveness of internodes and their capacity to tune their length. This project will characterize primary myelination dynamics and assess how internodal length is tuned over time. The mouse model of stochastically sparse labeled oligodendrocytes PLP-CreER^{T};STARS allows for precise quantification of internodal length. We use this model to study oligodendroglial myelination in the corpus callosum and the optic nerve. Preliminary results from these experiments reveal that each oligodendrocyte has a wide variety of internodal lengths during early development (postnatal day [P]14): internodes (reported as median [min-max]) are 33 μm long [12 to 89 μm] in corpus callosum of mice (N=3). We observed that early in development most internodes measured between 20 and 40 μm, and that the distribution of internodal length is skewed to shorter internodes (Skewness: 1.22). In the optic nerve, internodes measured at P18 were 77 μm long [51 to 144 μm], while at 7 months old (mo) internodes measured 120 μm long [50 to 192 μm]. We noted a shift in the distribution of internodal length between early (P18) and late (7mo) development: most internodes in the optic nerve at P18 measure between 60-80 μm and the general distribution was skewed to shorter internodes (Skewness = 0.74). However, internodes at 7mo mostly measured 100-120 μm and the distribution was Gaussian (Skewness = 0.07). In addition, we utilize the NG2-CreER^{T};Tau^{mgfp} model to characterize newly formed internodes during early, middle, and late development to learn how myelin wraps axons under normal conditions. Further experiments to study the impact of extrinsic cues on internodal length are ongoing. These approaches will shed light on how internodal length patterning is structured for optimal neuronal function. Not only do we establish that myelination during development is a dynamic process, but we also provide a model for internodal length determination.

Disclosures: A. Beaudry-Richard: None. **N. Jabassini:** None. **C. Cordano:** None. **R. Kothary:** None. **J.R. Chan:** None. **A.J. Green:** None.

Poster

P008. Glial Development and Neuronal Interactions

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P008.03

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH/NIDCD Grant DC007695

Title: Ultrastructural analysis and 3D cellular reconstruction of developing astrocytes in the medial nucleus of the trapezoid body from volume electron microscopy

Authors: *E. AMICK, D. HELLER, V. MOSES, M. ATTALLA, L. DE VRIES, E. FRAHM, A. LYNN, A. GABBARD, K. CSEH, G. SPIROU;
Med. Engin., Univ. of South Florida, Tampa, FL

Abstract: Mature astrocytes display myriad morphological features, including blood vessel contacting endfeet, finger-like projections at tripartite synapses, and vellus processes ensheathing neuronal cell bodies, and diverse physiological characteristics. While the relationship between morphological diversity within cell types and functionality has been studied in multiple brain regions, the exact role of this heterogeneity remains elusive. The medial nucleus of the trapezoid body (MNTB) represents an excellent neurodevelopmental model due to the rapid maturation of a highly homogenous neuronal population that is monoinnervated by the largest terminal in the mammalian brain, the calyx of Held (CH). The CH terminal also provides a system for studying astrocytic regulation of synaptogenesis. Here, we utilized a developmental series of serial block-face scanning electron microscopy volumes to visualize astrocytic morphology at an ultrastructural level during CH development. In addition to the characteristic features above, we observed several novel morphological features, including large mitochondrial widths, prevalent stacks of endoplasmic reticulum, and axonal ensheathment. A comparison of all cell types identified these unique features as being exclusive to astrocytes across developmental ages, providing additional morphological characteristics for cell type identification. Along with these ultrastructural observations, we completed the first full astrocyte reconstructions at postnatal day (P)2, P4 and P6 to fully observe and analyze time-dependent morphological variations. Initial comparisons reveal a preservation of characteristic features across developmental ages, although some refinement is observed in the older astrocytes. A notable difference is the increased volume and degree of ramification of the P6 cell relative to younger ages. Endfeet were observed to be formed by all three reconstructed cells. Axonal wrapping was not observed at P2, while the P6 astrocyte formed more wraps than the P4. All three cells were observed to form vellus processes on MNTB neuronal cell bodies, with the P6 astrocyte contacting the largest number of cells. However, the dynamic morphology of astrocytes during CH maturation needs to undergo further analysis and quantification to elucidate their developmental roles. This study highlights the complex morphology of astrocytes and their diverse heterogeneity by identifying novel ultrastructure morphological features. Furthermore, our analysis of 3D cellular structures can be implemented for future cell-type classifications and computational models for astrocytic biophysical properties.

Disclosures: E. Amick: None. **D. Heller:** None. **V. Moses:** None. **M. Attalla:** None. **L. de Vries:** None. **E. Frahm:** None. **A. Lynn:** None. **A. Gabbard:** None. **K. Cseh:** None. **G. Spirou:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); syGlass.

Poster

P008. Glial Development and Neuronal Interactions

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P008.04

Topic: A.01. Neurogenesis and Gliogenesis

Support: NIH Grant NS082203

Title: Actin regulation impacting myelination in the central nervous system.

Authors: *K. DAHL[1], W. B. MACKLIN[2];
[1]Univ. of Colorado, Aurora, CO; [2]Dept Cell & Dev Biol., Univ. Colorado Med. Sch., Aurora, CO

Abstract: In the central nervous system (CNS), myelin is a specialized membrane elaborated by oligodendroglia, which facilitates rapid conduction of action potentials. Myelination is a critical element of normal vertebrate neurotransmission and CNS myelin pathophysiologies, such as multiple sclerosis or leukodystrophies, are associated with profound neurological disfunction. Therefore, it is essential to understand the molecular mechanisms that drive myelin formation. During myelination, oligodendrocytes undergo dramatic expansion and modification of cell processes and membranes. These complex morphological transitions are driven by various signaling pathways, including mTOR, which enhance filamentous actin (F-actin) polymerization during early remodeling of membranes committed to myelin sheath formation. However, the extent to which mTOR regulates actin dynamics and the downstream pathways recruited during this process have not been fully determined. Current studies focus on signaling from the rapamycin-insensitive mTOR complex 2 (mTORC2, defined by the subunit RICTOR) through the GTPase Rac1 to regulate actin dynamics and myelination. Rac1 activation requires the interaction of guanine nucleotide exchange factors (GEFs), at least one of which, TIAM1, mediates Rac1 activation via mTORC2 signaling. Branched F-actin, polymerized through Rac1, is necessary for early process extension in cultured oligodendrocytes. Here, we examine how mTORC2 impacts actin dynamics in oligodendrocytes both in vivo and in vitro. Driving loss of mTORC2 in oligodendrocyte progenitor cells (OPCs) using PDGFRa-Cre;RICTOR$^{fl/fl}$ mice altered myelination and reduced the complexity of premyelinating oligodendrocytes during early development. In vitro, pharmacological inhibition of mTOR decreased both F-actin and Rac1 activity. Adenovirus-mediated genetic ablation of mTORC2 impeded progression of OPC differentiation past the early A2B5 expressing stage, while Sholl analysis indicated reduced membrane complexity in these cells. In addition, OPCs overexpressing shRNA to knockdown TIAM1 reduced F-actin and process extension. These data demonstrate that Rac1-mediated mTORC2 signaling is essential for regulating actin dynamics during oligodendrocyte differentiation.

Disclosures: K. Dahl: None. **W.B. Macklin:** None.

Poster

P008. Glial Development and Neuronal Interactions

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P008.05

Topic: A.01. Neurogenesis and Gliogenesis

Support: NSERC RGPIN-2020-06512

Title: Cortical layer distribution of SOX9+ astrocytes from V-SVZ neural/stem progenitor cells depends on time of birth

Authors: *I. KORTEBI[1,2], T. SHARMA[2], D. LOZANO-CASASBUENAS[2], A. OLFAT[2], E. DANIELE[1,2], M. FAIZ[1,2];
[1]Inst. of Med. Sci., [2]Div. of Anatomy, Dept. of Surgery, Univ. of Toronto, Toronto, ON, Canada

Abstract: Cortical astrocytes are highly heterogeneous cells with distinct gene expression profiles, phenotypes, and functions. They are born from neural stem/progenitor cells (NSPCs) during embryonic development starting at E15 and undergo a period of rapid expansion in the first postnatal week (P4-P7). Whether time of astrocyte birth from NSPCs determines their position within the murine cerebral cortex is not known. To determine whether time of birth influences astrocyte distribution in the upper (UL) vs. lower (LL) cortical layers, we permanently labelled ventricular-subventricular zone (V-SVZ) NSPCs and their progeny via *in utero* electroporation with PiggyBac (PB)-CAG-EGFP at E13, E15, 16, and 17 and sacrificed animals at P21. Immunohistochemistry (IHC) for the astrocyte lineage marker SOX9 was performed and EGFP+SOX9+ astrocytes were quantified and compared between UL and LL cortical layers within each timepoint. PB-based labelling of EGFP+SOX9+ astrocytes showed that E15- and E16-born astrocytes were primarily found in the UL at P21. As an alternative birth dating method, 5-ethynyl-2'deoxyuridine (EdU) was administered to pregnant mice at E13 and E16 and tissue was collected at P3 prior to astrocyte expansion. EdU labelling showed that SOX9+EdU+ astrocytes born at E13 were primarily found in the LL at P3, while those born at E16 were primarily found in the UL at P3. Altogether, these results suggest that the distribution of SOX9+ cortical astrocytes is influenced by time of birth from V-SVZ NSPCs.

Disclosures: I. Kortebi: None. **T. Sharma:** None. **D. Lozano-Casasbuenas:** None. **A. Olfat:** None. **E. Daniele:** None. **M. Faiz:** None.

Poster

P008. Glial Development and Neuronal Interactions

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P008.06

Topic: A.01. Neurogenesis and Gliogenesis

Support: The Glaucoma Foundation (SK)
Dr. John P. and Therese E. Mulcahy Endowed Professorship in Ophthalmology (SK)

Illinois Society for Prevention of Blindness (HNH, AKG)
"Smart FDI" (project no. 01.2.1-LVPA-T-848-01-005) under the Promotion of Scientific Research, Experimental Development and Innovations Priority of the Operational Programme of the European Union Structural Funds for 2014-2020 (GK)

Title: Functional effects of astrocyte-derived exosomes on neurite outgrowth

Authors: *S. KAJA[1], H. N. HARIANI[2], A. K. GHOSH[3], G. KALESNYKAS[5], W. K. JONES[4]; [1]Ophthalmology and Mol. Pharmacol. & Neurosci., [2]Grad. Program in Neurosci., [3]Grad. Program in Biochem. and Mol. Biol., [4]Mol. Pharmacol. & Neurosci., Loyola Univ. Chicago, Maywood, IL; [5]Experimentica Ltd., Vilnius, Lithuania

Abstract: Exfoliation glaucoma is the most common identifiable secondary form of open-angle glaucoma and associated with deposition of extracellular fibrillar material in ocular tissues. Genetic polymorphisms in lysyl oxidase-like 1 (Loxl1) are the most common cause of exfoliation glaucoma. The objective of this study was to determine the functional effects of extracellular vesicles (exosomes) derived from Loxl1-deficient primary optic nerve head astrocytes (ONHAs) on neuronal differentiation and viability. We generated an ONHA culture with ~50% Loxl1 knockdown by transducing ONHAs with lentivirus harboring Loxl1-targeting shRNA. Exosomes from primary cultures of rat ONHAs (naïve, scramble- and Loxl1 shRNA-transduced) were prepared by ultracentrifugation. Exosomes were characterized by nanoparticle tracking analysis and immunoblotting. Uptake of exosomes into rat E18 primary cortical neurons was assessed by quantitative microscopy. The functional effects of exosomes on primary neurons *in vitro* were determined by quantification of cell viability and neurite outgrowth after addition of exosomes. ONHA-derived exosomes expressed the prototypical markers, Alix and CD63, but were devoid of CD81. Exosomes derived from naïve as well as scramble- and Loxl1 shRNA-transduced ONHAs were similar in size (119.8 ± 4.3, 121.9 ± 2.8 and 121.1 ± 2.5 nm, respectively, n = 3, $P = 0.89$). Uptake of exosomes into primary neurons was time-dependent, peaking at 4h after addition (n = 3, $P < 0.05$). Viability of rat primary E18 cortical neuronal cultures did not differ in the presence of exosomes. However, naïve and scramble-transduced ONHA exosomes increased neurite outgrowth of primary cortical neurons by 20.1 ± 3.3% and 18.7 ± 2.0%, respectively (n=5, $P < 0.05$ compared with media alone). In contrast, exosomes from Loxl1 shRNA-transduced ONHAs prevented any increase in neurite outgrowth (2.2 ± 4.0% change; n = 5, $P = 0.98$).These data suggest ONHA-derived exosomes can exert functional effects on primary neurons *in vitro*. Altered exosome-mediated neuron-glia signaling in exfoliation glaucoma characterized by a chronic reduction of LOXL1 expression may contribute to the ocular manifestations of the disease.

Disclosures: S. Kaja: B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Acuisee, Inc.. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Experimentica Ltd., K&P Scientific LLC. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds);

Experimentica Ltd., eyeNOS, Inc., K&P Scientific LLC. F. Consulting Fees (e.g., advisory boards); Experimentica Ltd.. **H.N. Hariani:** None. **A.K. Ghosh:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Experimentica Ltd., K&P Scientific LLC. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); eyeNOS, Inc.. F. Consulting Fees (e.g., advisory boards); Experimentica Ltd., K&P Scientific LLC. **G. Kalesnykas:** A. Employment/Salary (full or part-time):; Experimentica Ltd.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Experimentica Ltd.. **W.K. Jones:** None.

Poster

P008. Glial Development and Neuronal Interactions

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P008.07

Topic: A.01. Neurogenesis and Gliogenesis

Support: PID2019-109858RB-I00
RD16-0015/0019
2019AEP033
LINKA20268

Title: Genetic approach to study the contribution of Sonic Hedgehog in the CNS physiological myelination and the remyelination process after damage in the adult

Authors: S. NOCERA[1], M. A. MARCHENA[2], Y. LAÓ[3], L. SOBRINO[3], E. BURGOS[4], Ó. GÓMEZ-TORRES[4], R. LUJÁN[4], ***F. DE CASTRO**[5];
[1]GNDe, Cajal Institute, CSIC, Madrid, Spain; [2]Univ. Europea de Madrid, Villaviciosa de Odón (Madrid), Spain; [3]Inst. Cajal-CSIC, Madrid, Spain; [4]Univ. de Castilla-La Mancha, Toledo, Spain; [5]Inst. Cajal (CSIC), Madrid, Spain

Abstract: Sonic Hedgehog (Shh) is the best-studied ligand of the Hedgehog pathway. Shh regulates vertebrate organogenesis and it is one of the main gradient-forming morphogens organizing the CNS. Shh gradients continues to act in the adult CNS modulating specification and self-renewal of neural stem cells, and is upregulated after acute brain injury. Recently, it has been shown that cell progenitors in the postnatal forebrain V-SVZ require Shh signaling to generate the oligodendrocytes of the corpus callosum. Oligodendrocytes are the glial cells whose myelin sheaths envolve myelinated axons axons, a critical step in development and the normal functioning of the CNS. These myelin-forming cells derive from oligodendrocyte precursor cells (OPCs) during pre- and postnatal CNS development, and a relevant population of OPCs physiologically remains during adulthood. These adult OPCs support myelin maintenance and physiological plasticity, as well as after demyelinating damage (Multiple Sclerosis -MS-, others)

they react in a restorative assay. Shh has been shown over-expressed in MS lesions, potentially regulating OPC responses driving to remyelination. However, the direct effect of Shh pathway on OPCs during postnatal development and in pathological conditions is still an open question, with substantial impacto n the role of Shh in MS.In our current work we show that the up or down-regulation of Shh signaling pathway affects OPC differentiation into myelin-forming oligodendrocytes both postnatally and in the adult, as well as modulate remyelination after damage. Furthermore, implanting OPCs in CNS areas that do not present myelin in physiological conditions (the retina), we show that the activation or the inhibition of Shh signaling cascade depends on the molecular environment, which is dynamic depending on the stage of development.

Disclosures: S. Nocera: None. **M.A. Marchena:** None. **Y. Laó:** None. **L. Sobrino:** None. **E. Burgos:** None. **Ó. Gómez-Torres:** None. **R. Luján:** None. **F. de Castro:** None.

Poster

P008. Glial Development and Neuronal Interactions

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P008.08

Topic: A.03. Stem Cells and Reprogramming

Support: SCRMC Postdoctoral Fellowship
Nancy & Jean Pierre Boespflug Foundation for Myopathic Research

Title: Tgfbeta1-gpnmb signaling inhibits oligodendrogenesis from adult neural stem cells following demyelination

Authors: *D. RADECKI, J. SAMANTA;
Univ. of Wisconsin, Madison, WI

Abstract: Demyelinating diseases, such as multiple sclerosis (MS), are debilitating diseases where the underlying degenerative and potential regenerative mechanisms are incompletely understood. Since regeneration of oligodendrocytes (OLs) and myelin can prevent neurodegeneration, understanding the molecular mechanisms of remyelination is essential to design therapeutic strategies for repair of demyelinating lesions. Adult neural stem cells (aNSCs) lining the lateral ventricles in the mammalian brain, including humans and mice, have the potential to regenerate myelin. In the healthy brain, a subset of these aNSCs, expressing the transcription factor Gli1, migrate to the olfactory bulb and differentiate into interneurons and astrocytes, but do not generate OLs. Following demyelination, these aNSCs can be recruited to lesions where they can differentiate into myelinating OLs. However, the pathways regulating aNSC recruitment, and their ability to generate OLs in adults is incompletely understood. Furthermore, remyelination is enhanced when Gli1 is inhibited in these aNSCs. By analyzing the

transcriptome of these cells following demyelination, we identified GlycoProtein Non-metastatic Melanoma B (Gpnmb) as the most significantly downregulated gene in aNSCs lacking Gli1 expression, suggesting its role as a negative regulator of remyelination. Indeed, global loss of Gpnmb in Gpnmb-LacZ knock-in mice, resulted in enhanced generation of OLs from Gli1 aNSCs following demyelination. Loss of Gpnmb also downregulated the expression of TGFβR2, a TGFβ1 receptor subunit, which suggested that Gpnmb acts downstream or in tandem with TGFβ1 which is highly expressed in demyelinating lesions. Remarkably, *in vitro* treatment of aNSCs by TGFβ1 lead to upregulation of Gpnmb. Overexpressing Gpnmb in aNSCs further lead to upregulation of TGFβR2 along with inhibition of mature OL generation *in vitro*, suggesting a positive feedback regulation of the pathway leading to amplification of TGFβ1 signaling in aNSCs following demyelination. Consistently, genetic loss of TGFβR2 specifically in Gli1 aNSCs, resulted in enhanced recruitment and generation of mature OLs from these cells in the demyelinating lesion. Thus, TGFβ1 released from demyelinating lesions, induces Gpnmb expression, which further upregulates TGFβR2 to amplify the pathway in Gli1 aNSCs, thereby inhibiting their recruitment and differentiation into mature OLs.

Disclosures: D. Radecki: None. **J. Samanta:** None.

Poster

P008. Glial Development and Neuronal Interactions

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P008.09

Topic: A.03. Stem Cells and Reprogramming

Support: CIHR Project Grant PJT-175254
Medicine by Design Cycle 2 Funding
Temerty Foundation Pathway Grant

Title: Generation of unique oligodendrocyte lineage cells following direct lineage reprogramming of reactive astrocytes

Authors: *J. BAJOHR, A. OLFAT, K. LEE, H. TAHA, D. LOZANO CASASBUENAS, M. FAIZ;
Univ. of Toronto, Toronto, ON, Canada

Abstract: Direct lineage reprogramming is a novel strategy which converts one cell type to another without the need for a pluripotent intermediate. Oligodendrocyte (OL) loss is characteristic of many neurological conditions, and thus astrocyte to OL reprogramming is of interest for central nervous system repair. Given the differential loss of OLs and their progenitors following disease, it is important to be able to generate OL lineage cells at different stages of development. Here we show that astrocytes can be converted to various cells of the OL lineage

in vitro when transduced with different transcription factors (TFs) involved in OL fate determination. Using GFAP driven lentiviruses containing TFs of interest, postnatal (P1-P5) cortical astrocytes were transduced and assessed for markers of the OL lineage at varying time points. Unexpectedly, our data showed that ectopic expression of a TF involved later in OL development resulted in the generation of O4+ newly forming OLs (nfOLs) relative to controls 14 days post transduction (DPT), while ectopic expression of an early OL fate specification TF did not. Although this early OL fate specification TF led to the increased production of PDGFRA+ oligodendrocyte progenitor cells (OPCs) by 14 DPT, longer culture (22 DPT) did not lead to an increase in O4+ nfOLs for this TF. Ectopic expression of a TF important for OL myelination showed a trend towards increased generation of MBP+ myelinating OLs. This lends support to the idea that different TFs can be used to produce different OL cell types. Live cell imaging of the reprogramming process confirmed that iOPCs and iOLs originate from S100β expressing astrocytes. Altogether, these findings demonstrate reprogramming of astrocytes to cells of the OL lineage, and that this technology can be tailored to generate cells at different stages of maturity. These studies lay the groundwork for novel, reprogramming-based strategies for diseases involving OL lineage cell loss.

Disclosures: J. Bajohr: None. **A. Olfat:** None. **K. Lee:** None. **H. Taha:** None. **D. Lozano Casasbuenas:** None. **M. Faiz:** None.

Poster

P066. Animal Models of Developmental Disorders Other Than Autism I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P066.01

Topic: A.07. Developmental Disorders

Support: (P01 HD083157)
(NIH U54HD090257)

Title: Phenotypic alterations in the hypoglossal nucleus of a mouse model of 22q11.2DS-LgDel

Authors: C. CLARKSON-PAREDES[1], X. WANG[4], P. MISTRY[2], L. MATSIYEVSKIY[1], D. S. MENDELOWITZ[1], S. A. MOODY[3], A. S. LAMANTIA[5], *A. POPRATILOFF[1];
[1]George Washington Univ., Washington, DC; [2]George Washington Univ., washington, DC; [3]Anat. and Regen Biol, George Washington Univ., Washington, DC; [4]Dept. of Pharmacol. and Physiol., The George Washington University, SMHS, Washington, DC; [5]Virginia Tech., Roanoke, VA

Abstract: DiGeorge/22q11 Deletion Syndrome (22q11DS) is a complex disorder characterized by cardiovascular and craniofacial abnormalities, intellectual disability, schizophrenia, autistic

spectrum, or attention-deficit/hyperactivity, among other pathologies. 22q11DS is caused by a microdeletion of 30-50 genes of chromosome 22, while clinical manifestations are linked to early perinatal developmental feeding/swallowing disorders (dysphagia) whose origins remain poorly understood. Previously, we have shown that the LgDel mouse model of 22q11.2 DS mimics the human phenotypic spectrum, including perinatal dysphagia, and exhibits changes in neuronal excitability in the hypoglossal nucleus (nXII) motor neurons, that controls the tongue movement and play a key role in the swallowing process. Therefore, we asked whether nXII circuits abnormalities are present and associated with LgDel feeding/swallowing difficulties. We analyze whether alteration in neuronal excitability is induced by changes in neurotransmitters content or distribution in the nXII. We used a ChAT:GFP reporter to identify nXII motor neurons in young and adult animals. Confocal 3D reconstructions of motor neurons demonstrate dendritic and axonal abnormalities, including degenerating dendrites. Also, backscatter scanning electron microscopy allowed us to analyze nXII location, neuronal identity, and ultrastructural characteristics in motoneurons affected by the genetic lesion. nXII motoneurons of one-week-old LgDel showed dilated mitochondrial cristae, paler mitochondrial matrix, and swollen Golgi and endoplasmic reticulum compared to age-matched wild type. At later ages, we found that LgDel nXII motor neurons and interneurons displayed cytoskeletal sparse, organelle depleted, paler cytoplasm, as well as reduced synaptic frequency. Also, we found hypertrophy of perineuronal, perisomatic, perivascular, and perisynaptic astrocytic processes, indicating glial changes in the modulation of the LgDel synaptic neurotransmission. To explore alterations in the inhibitory/excitatory afferents in the LgDel group, we classified/quantified the number of presynaptic terminals contacting the soma of nXII motor neurons at P7 using 2D-SEM tile-acquisitions. In addition, we quantified and compared the GABA content in presynaptic terminals of nXII during development, finding a significant decrease in the GABA presynaptic content in nXII of LgDel group, supporting the idea of imbalance neurotransmission levels that could directly affect the functionality of this nucleus in the feeding and swallowing circuits during development.

Disclosures: C. Clarkson-Paredes: None. **X. Wang:** None. **P. Mistry:** None. **L. Matsiyevskiy:** None. **D.S. Mendelowitz:** None. **S.A. Moody:** None. **A.S. LaMantia:** None. **A. Popratiloff:** None.

Poster

P066. Animal Models of Developmental Disorders Other Than Autism I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P066.02

Topic: A.07. Developmental Disorders

Title: Partial penetrance phenotype in the 16p11.2 deletion syndrome mouse model

Authors: *J. TARANDA[1,2], J. VELINSKOVA[3], S. MUZUMDAR[1], K. PRADHAN[4], K. UMADEVI VENKATARAJU[1], R. PALANISWAMY[1], J. SUN[1], S. TURCAN[2], J. GILLIS[1], L. VELISEK[3], P. OSTEN[1];
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Abstract: 16p11.2 deletion is a recurrent copy number variation of 600 kb chromosomal region that includes 28 genes and 1/200 in patients diagnosed with autism spectrum disorder (ASD). Here, we describe several new phenotypes with clinical relevance in the 16p11.2 deletion (df/+) mouse model. First, we show that in response to a convulsant drug pentylenetetrazol (PTZ), some, but not all, df/+ mice are more prone to seizures at lower doses of the drug than wild type littermates. Second, we further probe this phenotype in the df/+ mouse model using electrocorticography (ECoG) day/night recording, which revealed 1) changes in the awake and non-REM sleep time with significant alterations in low-frequency oscillations, 2) abnormal spontaneous cortical activity, and 3) increased susceptibility of approximately 50% df/+ mice to PTZ. Third, we provide evidence for higher excitability in thalamocortical pathways in about 50% of df/+ mice injected with PTZ, using our method of mapping whole-brain activation by serial two-photon (STP) tomography in transgenic c-fos-GFP reporter mice. Fourth, employing our STPT images analysis pipeline, we discover that 16p11.2 animals with more PTZ susceptibility present a significant reduction in some areas of the cortex, for example, the somatosensory sensory or motor cortex. Fifth, we ask whether the increased PTZ susceptibility in some df/+ mice represents a partially penetrant phenotype and, if so, whether high-susceptibility (HS) df/+ mice show different behaviors from their siblings low-susceptibility (LS) df/+ mice. This experiment, which comprised monitoring of home-cage behaviors (three 24-hour sessions over one week), followed by 3 PTZ injections (once a week, repeated three times), demonstrated that the df/+ mice indeed fall into two distinct HS and LS PTZ-dependent phenotypes and the HS df/+ show selective disruptions in sleep and increased grooming behavior. Finally, we analyze the whole-transcriptome RNA sequence from bulk samples from the cortex of P2 animals and the somatosensory cortex of adult male animals. Additionally, we included animals that presented HS and LS seizure phenotype and their wild-type littermates after to performed biopsies from the same brain area employed in the adult without any manipulation. Overall, our results reveal a seizure-like phenotype of partial penetrance that can be linked to sleep deficits and repetitive behaviors in isogenic 16p11.2 df/+ mice. The 16p11.2 df/+ mouse model thus recapitulates several important clinical features of autism, providing us with a unique opportunity to study the underlying brain circuit changes and test novel therapeutic approaches.

Disclosures: J. Taranda: None. **J. Velinskova:** None. **S. Muzumdar:** None. **K. Pradhan:** None. **K. Umadevi Venkataraju:** None. **R. Palaniswamy:** None. **J. Sun:** None. **S. Turcan:** None. **J. Gillis:** None. **L. Velisek:** None. **P. Osten:** None.

Poster

P066. Animal Models of Developmental Disorders Other Than Autism I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P066.03

Topic: A.07. Developmental Disorders

Support: MRC Grant No. MR/R014736/1
PhD Scholarship Emma and Leslie Reid

Title: Altered behaviour, metabolism and neural activity in a mouse model of intellectual disability with a mutation in PDZD8 gene

Authors: *A. PANTIRU, S. CLAPCOTE, J. JOHNSTON;
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Abstract: Intellectual disability often results from neurodevelopmental disorders that begin in childhood and are characterised by impaired cognitive functioning and an intelligence quotient below 70. Although intellectual disability is caused by both environmental and genetic causes, severe forms are often associated with defects in single genes. Three siblings from a consanguineous family were diagnosed with intellectual disability, developmental delay and autistic features. This phenotype cosegregated with a homozygous frameshift mutation in the PDZD8 (PDZ domain containing protein 8) gene. PDZD8 is an ER-mitochondria tethering protein which regulates calcium dynamics in mammalian neurons and was not previously associated with human disease. The aim of this project is to characterise a mouse model with a frameshift mutation in PDZD8. PDZD8 disruption in mice leads to intellectual disability, altered spatial memory and hyperactivity compared to WT controls. Molecular characterization of PDZD8 mutation further displays changes in gene expression, with gene related to mitochondria, cellular stress, neurogenesis and brain growth increased. In addition, preliminary in vivo imaging displayed changed in spontaneous cortical activity in PDZD8 mice containing a calcium indicator GCaMP6 compared to WT. Currently, changes in neuronal dynamics in PDZD8 mice are further investigated. Overall, PDZD8 disruption in mice was liked to behavioural, molecular and neuronal activity changes.

Disclosures: A. Pantiru: None. **S. Clapcote:** None. **J. Johnston:** None.

Poster

P066. Animal Models of Developmental Disorders Other Than Autism I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P066.04

Topic: A.07. Developmental Disorders

Title: C. elegans as a model for gain and loss of function in GRIN1 and GRIN2A-D variants

Authors: *S. OKAFOR[1], P. J. ROY[2], A. J. RAMSEY[1];
[1]Pharmacol. and Toxicology, [2]Mol. Genet., Univ. of Toronto St. George Campus, Toronto, ON, Canada

Abstract: GRIN disorder is caused by rare mutations within one of the seven GRIN genes that code for N-methyl-D-aspartate receptor (NMDAR) subunits. Pathogenic variants have been identified across the GRIN sequence and their functional impact can be either gain- or loss-of-function. We are developing mutant *c. elegans* as a model system to determine the impact of pathogenic variants on NMDA receptor function. The *c. elegans* genes *nmr-1* and *nmr-2* are orthologous to human GRIN1 and GRIN2 genes. *nmr-1* and *-2* form heterodimeric complexes that function like human NMDARs and are co-expressed on motor interneurons: AVE, AVA, PVC, AVD, and RIM. To characterize gain (GoF) and loss of function (LoF) in variants of GRIN1 and GRIN2A disorder we will co-inject human GRIN1 and GRIN2A genes into wildtype nematodes and into *nmr-1;nmr-2* deletion nematodes then develop mutants carrying documented patient variants. Next we will characterize locomotory phenotypes with our custom-built worm tracking system. We hypothesize that our overexpression model and our ectopic expression model will experience abnormal levels of reversals, forward movement, foraging behavior, omega turns, and locomotory speed. GoF variants should experience an increase in the aforementioned locomotory behaviors while LoF variants should experience a decrease. In addition we expect that ectopically expressing GRIN1 and GRIN2A will result in partial rescue of *nmr-1;nmr-2* deletion locomotor phenotypes. We will then use our data and methods to screen for FDA approved drugs that will mitigate or rescue the effects of GoF or LoF phenotypes.

Disclosures: S. Okafor: None. **P.J. Roy:** None. **A.J. Ramsey:** None.

Poster

P066. Animal Models of Developmental Disorders Other Than Autism I

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Program #/Poster #: P066.05

Topic: A.07. Developmental Disorders

Support: Startup Fund by School of Molecular and Cellular Biology, University of Illinois at Urbana-Champaign
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NARSAD Young Inverstigator Grant 27018 to N-P.T.

Title: Infantile spasms-linked Nedd4-2 mediates hippocampal plasticity and learning via cofilin signaling

Authors: *K. LEE[1], J. ZHU[1], C. A. CURIA[2], C. A. CHRISTIAN-HINMAN[1,2,3], J. S. RHODES[2,3,4], N.-P. TSAI[1,2,3];
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Abstract: Individuals affected by infantile spasms (IS), such as those carrying mutations in an IS linked gene, neural precursor cell expressed developmentally downregulated gene 4-like (Nedd4-2), exhibit developmental delays and learning disabilities, but the underlying mechanism is unknown. Using conditional Nedd4-2 knockout mice, we uncovered that Nedd4-2 functions to maintain the excitatory synapses in hippocampal neurons and allows for late-phase long-term synaptic potentiation (L-LTP) at Schaffer-collateral synapses in the hippocampus. We also found that Nedd4-2 is required for multiple forms of hippocampus-dependent learning and memory. Mechanistically, we showed that a loss of Nedd4-2 leads to a decrease in actin polymerization caused by reduced phosphorylation of the actin-depolymerizing protein cofilin. Using a cell permeable peptide to promote phosphorylation of endogenous cofilin in Nedd4-2 knockout neurons, we could restore the number of hippocampal excitatory synapses and hippocampal L-LTP and partially restore hippocampus-dependent learning in mice. Together, our results reveal a novel mechanism underlying IS-associated learning disabilities and may provide information for future therapeutic strategies for IS.

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Poster

P066. Animal Models of Developmental Disorders Other Than Autism I

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Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P066.06

Topic: A.07. Developmental Disorders

Support: SUNY-OW FDG
FUN-ELP
RAM-FITW
C-STEP/LSAMP

Title: Developmental lead (Pb^{2+})-exposure causes altered brain excitability in the adult rat: in-vivo pilocarpine induced seizure pathophysiology

Authors: G. CRUZ[1], M. VASQUEZ[2], E. CABAÑAS[1], J. JOSEPH[1], M. MIAN[1], S. K. V. MADHIRA[1], L. S. NEUWIRTH[3];
[1]Biol. Sci., [2]Chem. & Physics, [3]Psychology & SUNY Neurosci. Res. Inst., SUNY Old Westbury, Old Westbury, NY

Abstract: Lead (Pb2+) is neurotoxicant, that causes early neurodevelopmental pathologies in the offspring of Pb2+-exposed dams. Studies show that maternal Pb2+-exposure disrupts the GABA-shift contributing to altered brain excitability and potentially epilepsy. Unlike high-level Pb2+-exposure that produces encephalopathy and seizure, low-level Pb2+exposure does not produce such overt symptoms. However, subtle neurotoxic influences of Pb2+ on altering the CNS excitation:inhibition balance can cause a range of neurodevelopmental disorders at subthreshold seizure levels. Thus, in order to assess how Pb2+-exposure might cause neurodevelopmental aberrations in brain excitability, we exposed Long Evans Hooded Rats to in-vivo pilocarpine-induced (380 mg/kg) seizures. Control rats (0 ppm) were compared against Perinatal Pb2+-exposed rats (150 ppm & 1,000 ppm) to determine if Pb2+-exposure dose-dependently exacerbated the developmental neuropathological conditions. Rats were tested 1-2 months following the cessation of Pb2+-exposure at PND 22. Rats were anesthetized and in-vivo recording electrodes were stereotaxically implanted into the hippocampal CA1 region of the hippocampus and recorded for 1 hr post-pilocarpine seizure induction. The seizure data were subjected to a Fourier Transform using a Power Spectrum Density analysis to convert all raw brain seizure activity into isolated brain waves (i.e., alpha, beta, delta, gamma, theta rhythms) in addition to slow and fast ripples to assess pathophysiological states that persisted into adulthood from the early neurodevelopmental Pb2+-exposures. The results show that pilocarpine-induced seizures in a sex-dependent and Pb2+-dependent manner producing aberrant brain excitability. The findings from this preliminary study suggest that Pb2+-exposure may cause sex-dependent vulnerabilities as a function of dose, and the time-period of development that emanate from a disrupted GABA-shift in early neurodevelopment. These findings are consistent with the subtleties of Pb2+-exposure causing neurodevelopmental pathologies of prefrontal cortical damage with concurrent learning and memory hippocampal deficits as a model for human fronto-executive dysfunction and intellectual disabilities; albeit, emerging from aberrant neurodevelopmental programming of GABAergic excitation with persistent elevations of brain activity across the lifespan.

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Poster

P066. Animal Models of Developmental Disorders Other Than Autism I

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Topic: A.07. Developmental Disorders

Support: Student Blugold Commitment Differential Tuition funds through the University of Wisconsin-Eau Claire Office of Research Sponsored Programs
Freshwater Collaborative of Wisconsin (FCW) Freshwater Research Experience for Undergraduate Award

Title: Investigating gene-environment interactions of developmental methylmercury exposure in zebrafish

Authors: *A. LUTZKE, C. DICKEY, E. JOHNSON, B. S. CARTER;
Univ. of Wisconsin Eau Claire, Eau Claire, WI

Abstract: Methylmercury (MeHg) is a common environmental contaminant, and prenatal or early environmental exposure to MeHg is known to cause developmental abnormalities. MeHg is cleared from the body by oxidative stress metabolism pathways involving glutathione (GSH). The amount of time MeHg remains in the body varies greatly across individuals, and this variation may be due in part to genetic polymorphisms in genes involved in MeHg metabolism. Certain alleles of GSH-related genes are correlated with elevated blood MeHg levels in adult humans; how these alleles affect MeHg metabolism during development is not well known. Our objective is to produce and validate mutant zebrafish lines with loss-of-function mutations in GSH-related genes (*gstp1, gstp2, gclm*) using CRISPR-Cas9 reagents and high-resolution melt analysis genotyping (HRMA). CRISPR targeting and genotyping primers were designed using CHOPCHOP web tools. Embryos were microinjected at 1-cell stage with Cas9 protein complexed with specific guide RNA per gene target. HRMA primers were confirmed using synthetic DNA templates matching the targeted gDNA sequence. Methods were implemented and confirmed using reagents targeting tyrosinase (*tyr,* pigment gene). HRMA was able to distinguish between PCR products from synthetic DNA templates differing by 10 base pairs for all genes tested, and it was able to distinguish mutation in *tyr* mutant fish compared to vehicle-injected fish. These results enable future experiments defining genetic influence on developmental toxicology of methylmercury.

Disclosures: A. Lutzke: None. **C. Dickey:** None. **E. Johnson:** None. **B.S. Carter:** None.

Poster

P066. Animal Models of Developmental Disorders Other Than Autism I

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Program #/Poster #: P066.08

Topic: A.07. Developmental Disorders

Support: Canadian Institutes of Health Research

Scottish rite charitable Foundation of Canada

Title: Anxiety-related behaviors are impaired in adult mice exposed to perinatal cerebellar injuries

Authors: *E. GUARNIERI, M. MENGUS, R. IMANE, S. TREMBLAY;
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Abstract: Developmental cerebellar cognitive affective syndrome affects growing preterm infants diagnosed with perinatal cerebellar injuries during their neonatal intensive care unit stay. To better characterize motor, social and behavioral long-term outcomes, and their cellular mechanisms, we experimentally induced right-sided cerebellar hemorrhages (CBH) and/or early inflammation state (EIS) in 2 days-old transgenic mice (B6.129P2(Cg)-C*x3Cr1*$^{\text{CreERT2-EYFP/iDTR}}$) with or without transient right-sided cerebellar microglial cell depletion. Sensorimotor and behavioral tests were blindly performed at P4-P14 (Grasping reflex acquisition) and at P60-P85 (Marbles burying, Open field, Y maze, Elevated Plus Maze, Social recognition test, Morris water maze, Rotarod and Footprint analysis). Our preliminary cohort results suggested that in presence of microglial cells, male mice exposed to CBH+EIS spent significantly (*P=0.01*) less time in the periphery area of the Open field (78.2 ± 3.5 %, *n*=8) compared to control males (92.9 ± 1.4 %, *n*=7). They also tended to spend less time in the closed arms of the Elevated Plus maze (74.6 ± 4.7 %, *n*=8) and increase their locomotor activity (5559 ± 314 cm, *n*=8) while performing these tests compared to control males (77.3 ± 1.8 %; 4020 ± 221 cm, *n*=7). In male mice, microglial cell depletion prior to insult seems to attenuate these anxious behaviors suggesting a deleterious role of microglia on brain development. Interestingly, in absence of microglial cells, female mice exposed to CBH+EIS tended to have better short-term memory on the Morris Water Maze test (7.3 ± 1.9 sec, n=3) compared to non-depleted females (22.2 ± 6.0 sec, *n*=5) also suggesting a deleterious impact of microglia on cognitive abilities acquisition. These results revealed long-term impairments induced by perinatal cerebellar injuries with sex differences and impact of microglial cell activation on brain development. Increasing number of tested animals is required to confirm these preliminary results.

Disclosures: E. Guarnieri: None. **M. Mengus:** None. **R. Imane:** None. **S. Tremblay:** None.

Poster

P066. Animal Models of Developmental Disorders Other Than Autism I

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Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P066.09

Topic: A.07. Developmental Disorders

Support: NIH CHRCDA-K12
Children's National Board of Visitors

Title: Maternal Immune Activation and Hypoxia Induces Cerebellar Injury in Mice

Authors: *P. KRATIMENOS[1], I. KOUTROULIS[2], A. SATHYANESAN[5], E. GOLDSTEIN[3], M. GRANA[6], A. VIJ[7], B. JABLONSKA[5], V. GALLO[4];
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Abstract: Background: Preterm birth is often associated with maternal immune activation (MIA), and premature infants are also exposed to prolonged periods of hypoxia (Hx) and inflammation. Preclinical studies of perinatal hypoxia in mice have identified developmental alterations in the cerebellum; however, the cellular and molecular mechanisms underlying the effects of combined MIA and hypoxia on cerebellar development in are unknown. **Hypothesis**: We hypothesize that MIA and chronic hypoxia results in cerebellar injury and disruption of Purkinje cells (PCs) circuitry, leading to adaptive learning deficits. **Methods**: We utilized the "double-hit" model of prematurity (MIA+Hx). We injected lipopolysaccharide (LPS) to the dams during mid- to late gestation, embryonic days 15 and 16 (E15, E16). Then we exposed the pups to sub-lethal chronic Hx from postnatal day 3 to 11 (P3-P11). We included 4 groups: Double hit (DH), saline-Hx, MIA-normoxia, saline-normoxia. We included 6-8 mice/group for morphological and behavioral analysis and 3 mice/group for transcriptomic analysis. We performed morphological assessment and transcriptomic analysis of the cerebellar cortex at two timepoints (P15 and P35). We also tested the DH mice at P35 using the Erasmus Ladder, a fully automated system for real-time monitoring of cerebellar-mediated motor learning and behavior. **Results**: The cerebellum of the DH mouse model revealed alterations in the external and internal granule cell layer and phenotypically immature PC with poor dendritic arborization, as compared to controls. Interestingly, although there were 1,529 differentially expressed genes (DEGs) between DH and controls (1188 up-regulated, 341 down-regulated), there were only 19 DEGs between Controls and Hx (9 up-regulated and 10 down-regulated) and only 13 DEGs (9 up-regulated and 4 down-regulated) between the DH and MIA-only group, indicated that MIA is a significant contributor to cerebellar injury and acts synergistically with Hx (second hit) on the cerebellum. In addition, a cerebellum-specific behavioral assessment (Erasmus ladder) in the DH mice showed an increased percentage of missteps compared to the controls. In addition, associative cerebellar learning, pairing an obstacle and a tone during the challenge phase, revealed a significant cerebellar motor learning deficit in the DH mice. **Conclusions**: In "double-hit" mice, a combination of prenatal maternal immune activation and postnatal chronic Hx (the two main insults in premature neonates), induced specific patterns of cerebellar injury, similar to premature humans, and compromised adaptive cerebellar learning, a hallmark of autism spectrum disorder.

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Poster

P089. Ionotropic GABA Receptors and Glycine Receptors I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P089.01

Topic: B.02. Ligand-Gated Ion Channels

Support: NIH Grant 1R01MH114908-01
NIH Grant T32NS086749

Title: α5 $GABA_A$ receptor negative allosteric modulation - effects on inhibitory and excitatory hippocampal neurotransmission

Authors: *J. L. NUWER[1], M. L. BRADY[1], N. V. POVYSHEVA[2], A. COYNE[1], T. C. JACOB[1];
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Abstract: In the central nervous system, GABA type A receptors ($GABA_A$Rs) generate fast inhibitory signals to dampen and control neuronal activity. $GABA_A$Rs are heteropentameric ion channels assembled from a pool of 19 subunit genes (α1-6, β1-3, γ1-3, δ, π, ε, θ, and ρ1-3), each with distinct spatial, temporal, and subcellular expression patterns. The specific subunit composition and arrangement confers distinct functional and pharmacological properties. α5 subunit-containing $GABA_A$Rs are enriched in the hippocampus and play critical roles in development, synaptic plasticity, cognition, and memory. Negative allosteric modulators (NAMs) that target α5 $GABA_A$Rs enhance cognition and have shown promise in preclinical studies to mitigate cognitive impairment in Down syndrome, schizophrenia, and post-anesthesia. Despite *in vivo* efficacy in both rodents and humans, no study has examined the effects of sustained α5 NAM treatment on inhibitory and excitatory synapse plasticity to identify mechanisms of action. Here we used L-655,708, an imidazobenzodiazepine that acts as a highly selective but weak α5 NAM, in conjunction with microscopy, biochemical, and electrophysiological techniques to better understand the functional effects of reducing α5 $GABA_A$R signaling in both developing and mature neurons. We show that 2-day pharmacological reduction of α5 $GABA_A$R signaling in immature neurons delays dendritic spine maturation and the NMDA receptor (NMDAR) GluN2B/GluN2A developmental shift, while 2-day treatment in mature neurons enhances surface synaptic GluN2A and reduces GluN2B. Despite these changes, total levels of key excitatory synapse proteins were largely unchanged, and mEPSCs were unaltered. Additionally, 2-day α5 NAM treatment does not alter the total surface levels or distribution of α5 $GABA_A$Rs, reduce the gephyrin inhibitory synaptic scaffold, or impair phasic or tonic inhibition. Furthermore, mature neurons remained sensitive to α5 NAM-mediated inhibition of the $GABA_A$R tonic current after 2-day α5 NAM treatment.

Together, these results provide initial mechanistic insight into α5 $GABA_AR$ regulation of dendritic spine and circuit maturation via an NMDAR-dependent mechanism. Importantly, α5 NAMs may have sustained efficacy with reduced tolerance liability than non-specific $GABA_AR$-targeting drugs, such as benzodiazepines. We are currently investigating the effects of long-term α5 NAM treatment and the regulatory mechanisms governing α5 $GABA_ARs$ and α5 NAM-induced plasticity. The results of these studies will be invaluable in determining therapeutic application of α5 $GABA_AR$-preferring drugs.

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Poster

P089. Ionotropic GABA Receptors and Glycine Receptors I

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Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P089.02

Topic: B.02. Ligand-Gated Ion Channels

Title: Influence of GABA-A Receptor Intracellular Domain on Channel Activation and Desensitization

Authors: *A. TUNG[1], L. BLECKER[2], C. M. CZAJKOWSKI[2];
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Abstract: Rapid opening and closing of GABA-A receptor (GABAR) channels regulate signaling in the brain, but the mechanisms underlying the channel's open, closed, and desensitized transitions remain unclear. Recent GABAR cryo-EM structures provide details about its extracellular and transmembrane domains, but little information exists about its intracellular domain (ICD) and its role in regulating channel gating. Interestingly, the intracellular region of the GABAR α subunit following the C-terminal end of the M3 transmembrane helix is lysine rich. We hypothesize that mechanisms underlying GABAR gating transitions may involve charged interactions between these lysine residues and other regions of the GABAR ICD. Here, we individually mutated three lysines in this region to alanine (α1K327A, α1K328A, and α1K336A). The mutations' effects on GABA EC50 and macroscopic current desensitization were measured using two-electrode voltage clamping of oocytes expressing wild-type and mutant GABARs. α1K327A increased GABA EC50 6- fold (212.8 ± 49.9 μM, n = 6) compared to α1β2γ2 WT (35.98 ± 1.03 μM, n=9) and significantly reduced GABAR desensitization. The extent of current desensitization after 60 seconds in saturating GABA (10mM) was 30.16 ± 6.72% (n=9) versus WT (63.0 ± 15.0%, n=13). α1K328A decreased GABA EC50 2- fold (18.4 ± 6.09 μM, n=3) and also significantly reduced the extent of current desensitization at 60 sec (18.2 ± 3.65%, n=5) compared to WT. α1K336A decreased GABA

EC50 2-fold (17.1 ± 2.45 μM, n=2) and had little effect on the extent of desensitization at 60 sec (59.3 ± 4.50%, n=5) compared to WT. Based on a GABAR ICD homology model, a1N386 is predicted to interact with a1K327. We found that alanine substitution of α1N386 also increased GABA EC50 4- fold compared to WT and had no effect on the extent of GABAR desensitization at 60 sec (69.8 ± 6.24%, n=3). These data indicate structural perturbations of residues in the GABAR ICD can influence macroscopic GABAR current desensitization and suggest that charged residues in the ICD play a role in regulating GABAR gating transitions.

Disclosures: A. Tung: None. **L. Blecker:** None. **C.M. Czajkowski:** None.

Poster

P089. Ionotropic GABA Receptors and Glycine Receptors I

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Program #/Poster #: P089.03

Topic: B.02. Ligand-Gated Ion Channels

Support: NIH Grant R01NS108378
NIH Grant F31MH126542

Title: Beta3 subunit phosphorylation determines the assembly of synaptic and extrasynaptic $gaba_ars$

Authors: *C. CHOI[1], J. L. SMALLEY[1], S. J. MOSS[1,2], P. A. DAVIES[1];
[1]Neurosci., Tufts Univ. Sch. of Med., Boston, MA; [2]Neuroscience, Physiol. and Pharmacol., Univ. Col. London, London, United Kingdom

Abstract: Impaired inhibitory signaling via γ-aminobutyric acid type A receptors ($GABA_ARs$) underlies the pathophysiology of many neuropsychiatric disorders such as autism and epilepsy. Neuronal inhibition is regulated by two major $GABA_AR$ populations: α1-containing synaptic and α4-containing extrasynaptic subtypes that mediate phasic and tonic currents, respectively. Having distinct α subunits ensures proper receptor localization within the plasma membrane by controlling interactions with the inhibitory synapse scaffold, and is thus critical to the fidelity of GABAergic inhibition. While both subtypes are required for inhibitory transmission, the mechanism underlying the assembly of the two $GABA_AR$ subtypes remains unknown. The membrane trafficking of $GABA_ARs$ is subject to modulation by phosphorylation of the serine 408 and 409 residues (S408/9) within the β3 subunits, which are common components of the two $GABA_AR$ subtypes. Substituting serines with alanines (S408/9A) compromises GABAergic inhibition and leads to autism-like behaviors. However, the role of S408/9 phosphorylation in mediating $GABA_AR$ assembly has not been explored. To this end, we examined the subunit compositions and protein interactors of endogenous α1- and α4-containing $GABA_ARs$ in 8-12-week-old male and female C57Bl/6 and S408/9A animals using immunoprecipitation coupled

with quantitative mass spectrometry, western blots, and bioinformatics analyses. We also performed confocal microscopy on cultured neurons from C57Bl/6 and S408/9A pups. Lastly, to understand the mechanism involved in $GABA_AR$ assembly, we examined the expression patterns of $GABA_AR$ subunits across different cellular compartments. We found that, in C57Bl/6 animals, the α1 and α4 subunits form distinct populations of $GABA_ARs$ and interact with distinct sets of proteins. In addition, the two subtypes are associated with different levels of phosphorylated S408/9 and have different expression patterns across cellular compartments. In S408/9A animals, however, the α1 and α4 subunits co-assemble, resulting in the formation of α1α4-containing $GABA_ARs$ and mis-trafficking of α4 subunits into synapses via interactions with synapse-targeting proteins. These results suggest that the phospho-regulation of S408/9 plays a critical role in the assembly of α1-containing synaptic and α4-containing extrasynaptic $GABA_AR$ subtypes. Mutating S408/9 compromises $GABA_AR$ assembly and thus inhibition, by altering the proteome and localization of α4-containing $GABA_ARs$. Collectively, these results suggest that S408/9 may be a novel therapeutic target for disease conditions with impaired GABAergic inhibition.

Disclosures: C. Choi: None. **J.L. Smalley:** None. **S.J. Moss:** None. **P.A. Davies:** None.

Poster

P089. Ionotropic GABA Receptors and Glycine Receptors I

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Program #/Poster #: P089.04

Topic: B.02. Ligand-Gated Ion Channels

Support: CONICET PIP11220130100266

Title: Diazepam-induced down-regulation of the $GABA_A$ receptor alpha 1 subunit expression mediated by a calcium signaling pathway

Authors: N. B. MEDINA, M. V. PIETKO, ***M. C. GRAVIELLE**;
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Abstract: Benzodiazepines are widely prescribed drugs for the treatment of anxiety, sleep disorders, and epilepsy; however, their long-term use is limited by the development of tolerance. In previous studies we reported that prolonged exposure of rat cortical neurons to diazepam, a classic benzodiazepine, induced transcriptional repression of the $GABA_A$ receptor α1 subunit gene. This regulatory process was prevented in the presence of nifedipine, an inhibitor of L-type voltage-gated calcium channels. The aim of this work was to investigate the signaling cascade triggered by the persistent treatment with diazepam that leads to the regulation of $GABA_A$ receptor expression. Results from this study indicated that a 48 h treatment of rat cortical neurons

with diazepam (1 μM) produced an increase in the intracellular calcium concentration measured in a multi-well plate reader using the calcium-sensitive fluorescent dye Fura 2-AM and a high potassium concentration (60 mM KCl) as a stimulus. This increase was abolished in the presence of nifedipine, suggesting that diazepam exposure induced an increase in the calcium influx through L-type voltage-gated calcium channels. Quantitative real-time PCR and western blots experiments showed that the diazepam-induced down-regulation of the α1 subunit expression was blocked by picrotoxin, a $GABA_A$ receptor channel blocker, and H-89, a protein kinase A inhibitor. Taken together, our results suggest that sustained stimulation of $GABA_A$ receptors by benzodiazepines activates an intracellular signaling cascade that involves the stimulation of the calcium influx through L-type voltage-gated calcium channels which results in the activation of a protein kinase A cascade, finally leading to the transcriptional repression of α1 subunit gene. This regulatory mechanism depends on the activation of $GABA_A$ receptors. Elucidation of the signaling pathway activated by the sustained exposure to benzodiazepines will contribute to understand the molecular bases of tolerance.

Disclosures: N.B. Medina: None. **M.V. Pietko:** None. **M.C. Gravielle:** None.

Poster

P089. Ionotropic GABA Receptors and Glycine Receptors I

Location: Virtual Only

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Program #/Poster #: P089.05

Topic: B.02. Ligand-Gated Ion Channels

Support: NIH Grant 1R01MH123748
NIH Grant 1F30MH126548

Title: Allopregnanolone effects on inhibition in hippocampal parvalbumin interneurons

Authors: *X. LU, P. LAMBERT, A. BENZ, C. F. ZORUMSKI, S. J. MENNERICK; Psychiatry, Washington Univ. In St. Louis, Saint Louis, MO

Abstract: Allopregnanolone (AlloP) is an endogenous neurosteroid that potentiates ionotropic GABAergic inhibition in the CNS. AlloP has a rapid antidepressant effect and recently been approved for treating postpartum depression in women. However, the antidepressant mechanism of AlloP is largely unknown but could involve selective action at $GABA_A$ receptors containing the δ subunit. Previously, we and others showed that AlloP potentiates both phasic and tonic inhibition in the dentate granule cells (DGCs), which express α4δ-containing receptors. Using pharmacoresistant receptor subunit knock-in mice, we in fact found little evidence for selectivity at δ subunit-containing receptors despite previous evidence. In contrast to DGCs, hippocampal fast-spiking parvalbumin (PV) interneurons express an unusual variant partnership of δ subunits with α1 subunits. Here we hypothesized that native α1δ receptors may provide a strong substrate

for AlloP, resulting in potentiation of tonic disinhibition to DGCs. Contrary to the hypothesis, electrophysiology from genetically tagged PV interneurons in hippocampal slices showed that 100 nM AlloP promoted phasic inhibition by increasing the sIPSC decay, but tonic inhibition was not detectably altered. Co-application of 100 nM AlloP with 5 µM GABA did result in tonic current augmentation. Furthermore, 100 nM AlloP decreased the membrane resistance and the number of action potentials of DGCs but not PV interneurons. Taken together, our results indicate that PV interneurons possess low sensitivity to AlloP, which makes it unlikely that PV tonic current is a substrate for AlloP-induced tonic disinhibition.

Disclosures: X. Lu: None. **P. Lambert:** None. **A. Benz:** None. **C.F. Zorumski:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Sage Therapeutics. F. Consulting Fees (e.g., advisory boards); Sage Therapeutics. **S.J. Mennerick:** None.

Poster

P089. Ionotropic GABA Receptors and Glycine Receptors I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P089.06

Topic: B.02. Ligand-Gated Ion Channels

Support: Austrian Science Fund (FWF) P33130

Title: Selective targeting of the etomidate site of α6-containing $GABA_A$receptors

Authors: *P. SCHOLZE[1], K. SENDOREK[1], M. ERNST[1], X. SIMEONE[2];
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Abstract: $GABA_A$ receptors containing the α6 subunit are highly expressed in cerebellar granule cells, and less abundantly in many other neuronal and peripheral tissues. Animal models of several neuropsychiatric conditions suggest that compounds selectively activating or positively modulating cerebellar α6-containing $GABA_A$ receptors can alleviate essential tremor and motor disturbances in Angelman and Down´s syndrome, as well as impaired prepulse inhibition in neuropsychiatric disorders. Moreover, they have been suggested to reduce migraine and trigeminal-related pain by acting on α6-containing $GABA_A$ receptors on trigeminal ganglia. Within the scope of this study, we aim to develop novel α6-selective ligands as therapeutic prospects in neuropsychiatric conditions involving central and peripheral α6-containing $GABA_A$ receptors. The α6 subunit features unique amino acids contributing to ligand pockets in the ECD near “loop” C as well as in the TM1 contributing to the binding sites for etomidate and loreclezole. Not only the already known influence on furosemide is exerted by this upper TM1 segment, but several other scaffolds including some loreclezole derivatives target the etomidate pocket with some degree of selectivity that is conferred by these two amino acids. The two

unique amino acids in TM1 were subjected to mutational analysis and their impact on various ligands thought to bind at or near the etomidate site was investigated using two electrode voltage clamp electrophysiology on GABA receptors expressed in *Xenopus laevis* oocytes. We show that both mutants influence modulatory potency and efficacy of several known α6-preferring compounds.

Disclosures: P. Scholze: None. **K. Sendorek:** None. **M. Ernst:** None. **X. Simeone:** None.

Poster

P089. Ionotropic GABA Receptors and Glycine Receptors I

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Program #/Poster #: P089.07

Topic: B.02. Ligand-Gated Ion Channels

Support: NIH Grant R01NS081986
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Title: Glutamine synthetase and its role in astrogliosis and neurotransmission.

Authors: *S. F. J. NG[1], S. J. MOSS[1,2,3];
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Abstract: Glutamine synthetase (GS) is an enzyme that plays a central role in the regulation of nitrogen metabolism, through catalyzing the ATP-dependent condensation of glutamine and ammonia to glutamine: Glutamate + ATP + NH_4^+ → Glutamine + ADP + phosphate. It is predominantly found in astrocytes in the brain and contributes to detoxifying NH_4 and limiting glutamate accumulation to prevent excitotoxicity. It is the only enzyme capable of synthesizing glutamine, which is the principal metabolic precursor for the inhibitory neurotransmitter γ-aminobutyric acid (GABA), indicating its importance in in maintaining neurotransmitter supply and supporting neuronal inhibition. The essential role GS plays in the brain is revealed by the rare congenital mutations in humans, contributing to brain malformations, resulting in neonatal death. Constitutive inactivation of GS in mouse brain causes deaths 1-3 days after birth. Consistent with its role in supporting GABAergic inhibition, deficits in GS expression are found in hippocampi from human patients with temporal lobe epilepsy (TLE). However, whether these deficits directly contribute to the pathophysiology of epilepsy remains elusive. To address this, we have used stereotaxic injection of *Cre* recombinase-expressing adeno-associated viruses to locally ablate GS expression within the hippocampus of adult GS-FLOXED (GS^{flox}) mice. The impact of this manipulation on glia and neuronal structure, inhibitory neurotransmission, and seizure sensitivity were examined. Our results suggests that locally ablating GS expression in

hippocampal subdomains is sufficient to induce reactive astrogliosis, a hallmark of TLE, suggesting that agents which augment GS activity may be a useful therapeutic strategy to limit epileptogenesis.

Disclosures: S.F.J. Ng: None. **S.J. Moss:** None.

Poster

P131. Signal Propagation in Neural Networks

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Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P131.01

Topic: B.09. Network interactions

Support: 1R01NS104911-01

Title: Subthreshold synaptic inputs to space-specific neurons

Authors: *P. HARRAST[1], W. M. DEBELLO[1], D. FIORAVANTE[2];
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Abstract: Neuronal computations performed by space-specific neurons (SSNs) in the barn owl inferior colliculus are well-studied via extracellular recordings, but there is very little data regarding subthreshold synaptic events. We used whole-cell voltage-clamp recordings and concentric bipolar stimulation in 35-50d old animals to map the topography of synaptic inputs preserved in ex vivo slices, and begin to determine the rules of integration for pairs of input locations. Recordings were visually targeted to large somas in the external nucleus of the inferior colliculus (ICX). Retrospective imaging revealed that approximately half of the cells contained toric-like spines, signature features of Type I SSNs. To map input topography, the electrode with narrow field of excitation was placed in the adjacent input nucleus, the lateral shell of the inferior colliculus (ICCls), and brief current pulses were applied in ascending steps from ~10 - 1000 microamps. The threshold current intensity for evoking an EPSC was determined. EPSCs were shown to be glutamatergic by pharmacology and manipulation of holding potentials. The bipolar electrode was then moved to a new location and the stimuli repeated. In this horizontal plane of section, different locations represent different values of interaural time difference (ITD). As predicted, the largest EPSCs were elicited when the bipolar electrode was placed at an iso-ITD contour within ICCls, and the EPSC magnitudes decreased as the sites of stimulation were moved to locations representing more ipsi or more contra ITDs. To measure EPSC integration, a second bipolar electrode was deployed. Once two effective sites were found, stimuli were presented individually or synchronously. Preliminary results indicate that the EPSCs elicited by synchronous stimulation were often larger than predicted by the sum of individual stimulations (i.e. supralinear summation). This effect appeared to be limited to nearby stimulation sites. Finally, at some single stimulation sites we observed paired-pulse facilitation. In total, these

experiments demonstrate the feasibility of measuring the topography, summation rules and capacity for short-term plasticity of synaptic inputs to SSNs. In addition to probing integration across the ITD axis, future experiments will use orthogonal slices which expose the sound frequency axis and a different set of neuronal computations.

Disclosures: P. Harrast: None. **W.M. DeBello:** None. **D. Fioravante:** None.

Poster

P131. Signal Propagation in Neural Networks

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Program #/Poster #: P131.02

Topic: B.09. Network interactions

Support: NIMH R37MH087027
ONR MURI N00014-16-1-2832
The JPB Foundation

Title: Propofol anesthesia changes dynamic stability in cortex

Authors: *L. KOZACHKOV[1], A. EISEN[1], A. BASTOS[1,2], J. A. DONOGHUE[1], M. K. MAHNKE[1], S. L. BRINCAT[1], E. N. BROWN[1,3], E. K. MILLER[1];
[1]Picower Inst. Learning Memory, MIT, Cambridge, MA; [2]Dept. of Psychology and Vanderbilt Brain Inst., Vanderbilt Univ., Nashville, TN; [3]The Dept. of Anesthesia, Critical Care and Pain Med., Massachusetts Gen. Hospital/Harvard Med. Sch., Boston, MA

Abstract: Understanding the mechanisms by which anesthetics induces loss of consciousness (LOC) is critical for making anesthesia care safer and for understanding consciousness. Recent work suggests an optimal range of “Dynamic Stability” is critical for normal cortical function[1]. Dynamic Stability is the ability of network activity to “stay in its lanes”: be dynamic yet consistent, performing the same computations regardless of initial state and in the face of being perturbed by other activity, noise, and distractions. Too much stability and networks cannot process and integrate new inputs; too little stability and network activity spirals off into “chaos”. We tested how propofol, a widely used anesthetic, affects Dynamic Stability. We used local field potentials (LFPs) from the prefrontal cortex in non-human primates (NHPs) anesthetized with propofol [2]. We applied techniques in machine learning [3] to produce a time series with statistical properties approximating those of the neural data. We then computed the spectrum of Lyapunov exponents – a quantification of the degree of (in)stability present in the system. Preliminary results indicates that after propofol-induced LOC, there is increased stability in cortical networks relative to the awake state. This excess stability may contribute to LOC in part by rendering cortical networks impervious to inputs thereby disrupting cortical communication.
1. Kozachkov L, Lundqvist M, Slotine J-J, Miller EK. Achieving stable dynamics in neural

circuits. Haith AM, editor. PLOS Comput Biol. 2020;16: e1007659. doi:10.1371/journal.pcbi.10076592. Bastos AM, Donoghue JA, Brincat SL, Mahnke M, Yanar J, Correa J, et al. Neural effects of propofol-induced unconsciousness and its reversal using thalamic stimulation. Elife. 2021;10: 1–28. doi:10.7554/ELIFE.608243. Pathak J, Lu Z, Hunt BR, Girvan M, Ott E. Using machine learning to replicate chaotic attractors and calculate Lyapunov exponents from data. Chaos. 2017;27: 121102. doi:10.1063/1.5010300

Disclosures: L. Kozachkov: None. **A. Eisen:** None. **A. Bastos:** None. **J.A. Donoghue:** None. **M.K. Mahnke:** None. **S.L. Brincat:** None. **E.N. Brown:** None. **E.K. Miller:** None.

Poster

P131. Signal Propagation in Neural Networks

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Program #/Poster #: P131.03

Topic: B.09. Network interactions

Support: NIH/NINDS 5R01NS114120-02

Title: Subthreshold oscillations can propagate as a wave by volume conduction

Authors: *C.-C. CHIANG[1], D. M. DURAND[2];
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Abstract: Rhythmic oscillations at different frequency bands have been observed in several regions of the brain. Some of these oscillations are classified as subthreshold membrane potentials (STO) and can influence neural spikes' timing. We reproduced the oscillating activity at different frequency ranges (5-50 Hz) in the longitudinal hippocampal slice expressing optopatch proteins by optical stimulation with blue laser pulse trains. We found that the optogenetically-induced neural oscillations could propagate across the hippocampal slice and go through a complete transection of the tissue. The propagation speed of these neural oscillations is about ~0.1m/s and is independent of the oscillating frequency. The speed increases with increasing amplitudes of the wave. The electric fields generated by these waves are 0.5~0.9 mV/mm between somatic and dendritic regions and ~0.4 mV/mm along the cell layer. We also observed that these subthreshold oscillations could interfere with each other. Optical stimulation applied simultaneously at each end of the slice generated a larger oscillation in the middle of the tissue (constructive effect). Similarly, when two stimulation waveforms with 180° phase delay are generated, an oscillation with a smaller amplitude was measured in the middle of the slice (destructive effect). Finally, we conducted pharmacological experiments to characterize these oscillations and found that these induced oscillations are not affected by the NMDA blocker, APV, but the speed increases when the HCN channels are inhibited. These results indicate that hippocampal subthreshold oscillations can propagate by volume conduction and generate

interference patterns. The role of these subthreshold waves in neural function is unknown, but the results suggest that at low voltage amplitude, the lack of refractory period could allow a novel analog form of preprocessing of neural activity.

Disclosures: C. Chiang: None. **D.M. Durand:** None.

Poster

P131. Signal Propagation in Neural Networks

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Program #/Poster #: P131.04

Topic: B.09. Network interactions

Support: NS102871
NS116724
NS121850

Title: Control of sympathetic preganglionic output strength by modulation of spike propogation in branching axons along paravertebral sympathetic chain ganglia

Authors: *M. HALDER[1], Y. LI[1], A. J. SOKOLOFF[2], M. SAWCHUK[1], S. HOCHMAN[3]; [2]Dept Physiol, [1]Emory Univ., Atlanta, GA; [3]Dept Physiol, Emory Univ. Sch. Med., Atlanta, GA

Abstract: Spinal sympathetic preganglionic neurons (**SPNs**) issue divergent axonal projections onto ganglionic neurons (**GNs**) across multiple paravertebral chain ganglia. SPN axons labelled from ventral roots (**VRs**) issue branching axons which travel rostro-caudally in the interganglionic nerve (**IGN**) to innervate GNs across many segments. Neurobiotin labeling of adult mouse SPN axons have begun to identify the complexity of SPN branching. For 165 SPN axons originating from T7 VR, projections were seen across at least 10 segmental ganglia (mean of 140 synapses/axon in T5-T7 ganglia alone). Here we explore whether spike propagation failures across ganglia represent a physiologically modifiable feature of divergence gain control. Adult mouse thoracic sympathetic chain ganglia were dissected to remain in continuity *in situ* with connections from VRs. Synaptic transmission was blocked with hexamethonium. SPN axons were recruited at 2X suprathreshold intensities in thoracic VRs. Population conduction across branching (in IGN) and unbranching (in splanchnic) axons were recorded with suction electrodes. Recordings at 22º over 2hrs showed individual SPN axons undergoing spontaneous conduction block. After scaling for temperature dependent changes in spike height and width, increasing temperature from 22º progressively reduced rectified-integral responses in both unbranching (35% at 28º; 46% at 36º; n=2) and branching SPNs (27% and 35.6%; n=2). At 31-36º, spontaneous conduction block was always seen across ≥ 3 intervening ganglia (n=6/6) but not ≤2 segments (n=0/2). To assess activation history of spike propagation, we compared evoked responses before and 2s after 10 pulses of 5 & 10 Hz stimulus trains and observed facilitation of

conduction across ≥ 3 ganglia following 10Hz (n=3/3) and 5Hz (n=4/7) stimulation. SPNs express α_5-$GABA_A$ receptors and may be constitutively active (10.1523/jneurosci.2951-08.2008). We used $GABA_A$ receptor pharmacology to explore actions on spike conduction. The $GABA_A$ antagonist bicuculline (10μM) significantly reduced rectified integral values of the population volley in branching (34%; $p < 0.05$; n=6) but not unbranching SPNs (29%; $p < 0.1$; n=2). L655,708 (20μM), a negative allosteric modulator for α_5-GABAARs also reduced conduction in branching SPNs (33% $p < 0.06$; n=4). In sum, we demonstrate that conduction block of spikes in SPN axonal projections is prevalent and modifiable in a distance-, temperature-, activity- and neuromodulation-dependent manner. Further studies on $GABA_A$ receptor pharmacology are planned to better understand their subunit composition and mechanisms of endogenous modulation.

Disclosures: M. Halder: None. **Y. Li:** None. **A.J. Sokoloff:** None. **M. Sawchuk:** None. **S. Hochman:** None.

Poster

P131. Signal Propagation in Neural Networks

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Topic: B.09. Network interactions

Support: NIH grant RO1 MH111439

Title: Fluctuation of inter-regional delays in the human cerebral cortico-cortical communication

Authors: *J.-Y. MOON[1], *K. MÜSCH[1], C. E. SCHROEDER[2], C. J. HONEY[1];
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Abstract: In electric signals such as field-potentials measured across regions of the human brain, parietal signals frequently phase-lead the signals in temporal and frontal cortex, while waves of activity propagate along parieto-temporal pathways. In order to better understand the functional properties of such large-scale pattern of signal flow, we asked: (i) are inter-regional delays stable or variable over time? (ii) do the patterns of signal propagation co-vary with endogenous cortical rhythms? (iii) do the patterns of signal flow vary with external stimulus properties? We recorded electrocorticographic signals from the lateral cortical surface of 10 human participants as they listened to a 7-minute auditory narrative. In sliding 2-second windows, we identified inter-regional delays by computing the cross-correlation of voltage signals between nearby electrode pairs. For each time window, and for each electrode-pairs, we identified the time delay of maximal inter-electrode correlation from raw voltage signals. *Variable Latencies*: We found that

the auditory pathway exhibited a gradient of delays, with posterior temporal regions leading anterior temporal regions on average. However, the latencies between stages of auditory processing were not stable and fluctuated over time. Two distinct electrophysiological states were evident: one characterized by longer inter-channel latencies (propagating state), and the other shorter latencies (synchronized state). We also observed that latency changes were not confined to local region but global patterns change between these two states. *Low-Frequency Power*: Latencies were longer during bursts of 6-16 Hz power (propagating state) and were shorter during bursts of broadband power (synchronized state), consistent with models in which low-frequency oscillations regulate corticocortical interactions. The inter-regional delays were mostly endogenous, as the correlation between responses under repeated stimulus was weak. *Computational Model*: We designed a computational model for the inter-regional flows using a Stuart-Landau coupled oscillator model. Employing a network topology derived from human neocortical anatomy, we reproduced the inter-regional correlation and delay pattern. Our model indicates that inter-regional latencies increase when the strength of inter-regional influence are increased. Altogether, the data and models suggest that the couplings between human cortical regions are not stable in their delays, and that instead they transition between synchronized (associated with higher broadband power) and propagating states (associated with higher lower-frequency 6-16 Hz power).

Disclosures: J. Moon: None. **K. Müsch:** None. **C.E. Schroeder:** None. **C.J. Honey:** None.

Poster

P131. Signal Propagation in Neural Networks

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Topic: B.09. Network interactions

Support: EU Grant No. 945539 (Human Brain Project SGA3)
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Title: An enriched E-GLIF model for hippocampal CA1 pyramidal neurons and interneurons

Authors: *M. MIGLIORE[1], **A. IUORIO**[1,2], E. SPERA[1], V. DE FALCO[1,3], R. MIGLIORE[1], A. GEMINIANI[4], C. CASELLATO[4], A. MARASCO[3];
[1]Natl. Res. Council, Palermo, Italy; [2]Fac. of Mathematics, Univ. of Vienna, Vienna, Austria; [3]Dept. of Mathematics and Applications "R. Caccioppoli,", Univ. of Naples "Federico II", Naples, Italy; [4]Dept. of Brain and Behavioral Sci., Univ. of Pavia, Pavia, Italy

Abstract: Hippocampal CA1 pyramidal neurons and interneurons exhibit complex and highly variable firing dynamics that play a key role in modulating the dynamics of the network to which they belong. These patterns can be successfully described by morphologically and biophysically realistic conductance-based models [1], but they cannot be easily reproduced by point neuron models. This can be fundamental when dealing with very large-scale networks including multiple brain areas/regions, because of the current technical limitations of supercomputer systems. Building up on a previous work [2], we have implemented an enriched Extended generalized leaky integrate-and-fire (E-GLIF) model for hippocampal CA1 pyramidal neurons and interneurons including more flexible reset rules for the membrane potential and intrinsic currents at each spike event. The aim is to reproduce the rich set of electrophysiological behaviours of hippocampal CA1 neurons as a functions of the incoming current (injected or synaptic). The linear nature of the equations (in contrast with other implementations), and an ad-hoc nondimensionalization procedure, allowed us to sensibly reduce the number of parameters and compute more tractable analytic solutions. This approach gave us the ability to perform a rigorous analysis of the equilibria stability as well as the monotonicity properties of the membrane potential as a function of the model's parameters. Furthermore, it was possible to determine general constraints on the model parameters, which were optimized against experimental findings. Such constraints, linked to the type of firing, lead to different functions for the reset rules depending on the pattern considered. Differently from the linear G-LIF models proposed in a recent work, which require a special set of experimental protocols and features, the optimization process for our model is based on basic experimental data such as spike numbers and spike-times.
The models have been validated against electrophysiological data recorded from hippocampal neurons activated by external (constant) stimuli. However, the models can reproduce typical firing behaviour of hippocampal CA1 pyramidal neurons and interneurons also in the presence of synaptic inputs. Finally, these reset rules can contribute in creating infinite copies of neurons statistically indistinguishable within the experimental variability ranges, therefore increasing the ability to better reproduce the full range of realistic firing scenarios, to build realistic circuits. 1. Migliore R. et al. PLoS computational biology 14.9 (2018): e1006423. 2. Geminiani A. et al. Frontiers in neuroinformatics 12 (2018): 88.

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Poster

P131. Signal Propagation in Neural Networks

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Grant from the Swiss National Supercomputing Centre (CSCS) under project ID ich002 and ich011

Title: Full-scale point-neuron model of mouse hippocampal CA1 microcircuit

Authors: *D. GANDOLFI[1,2], J. MAPELLI[2], S. M. G. SOLINAS[3], A. MARASCO[4], A. IUORIO[5,6], E. SPERA[5], V. DE FALCO[5,4], R. G. DE SCHEPPER[7], A. GEMINIANI[7], C. CASELLATO[7], E. U. D'ANGELO[7,8], M. MIGLIORE[5];
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Abstract: The coupling of a bottom-up modeling approach with simplified neuron models represents a valuable strategy to simulate large-scale brain networks dynamics and high-level behaviors. In the present work, a data-driven and computationally efficient point-neuron representation is at the basis of a full-scale spiking network model of activity propagation within a mouse CA1 region. The network building computational effort was greatly reduced by exploiting a recently developed boosting modelling framework for Python, the Brain Scaffold Builder module (BSB), which provides multiple placement and connectivity strategies enabling simulations of arbitrary experimental conditions. Excitatory and inhibitory neurons (10% excitatory-inhibitory ratio) were placed in the simulation volume according to the Blue Brain Cell Atlas. Neuron pairs specific connectivity was obtained by modeling the geometrical probability volumes associated to pre-synaptic axons and post-synaptic dendrites. The neuronal morphological features (axonal and dendritic arborization shape and extension) were taken from literature and from Neuromorpho.org and MouseLight databases. Similarly to the formalism adopted in DTI tractography, for the majority of cell types the axonal probability volume was modelled as an ellipsoid and it was parametrized as a covariance matrix (eigenvalues corresponded to the semiaxes lengths while eigenvectors corresponded to 3D orientations). Dendritic arborizations were parametrized either as conical volumes with different heights, radii and orientations or as a combination of ellipsoids. This parametric description of axonic and dendritic volumetric extent allowed to consider the experimentally observed preferential orientation of pyramidal cell axons along the direction of the minimum distance between CA3 and subiculum, thus implementing the strong directionality in Pyramidal-to-Pyramidal activity propagation along transversal hippocampal slices that has been observed experimentally. Cell pairs were connected through a short-term dependent plasticity synapse model showing facilitation and depression as in Ecker et al 2020. Single neuron electrophysiological properties were implemented following a new G-lif implementation of hippocampal pyramidal neurons and interneurons.The initial benchmarks and validations reported in this work, show that by exploiting the BSB modeling framework and NEST simulator it is possible to build, with a

relatively easy workflow, a full-scale data driven network with biological plausible features able to reproduce the essential functional activity of a large scale circuit.

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Poster

P131. Signal Propagation in Neural Networks

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P131.08

Topic: B.09. Network interactions

Support: EU Grant Agreement No. 945539 (Human Brain Project SGA3)
Centro Fermi project “Local Neuronal Microcircuits”
Fenix Infrastructure resources, which are partially funded from the European Union's Horizon 2020 research and innovation programme through the ICEI project under the grant agreement No. 800858

Title: The Brain Scaffold Builder: a new neuroinformatic framework for realistic brain modelling, a cerebellar use case

Authors: R. DE SCHEPPER[1], A. GEMINIANI[1], S. MASOLI[1], M. RIZZA[1], A. ANTONIETTI[1], E. D'ANGELO[1,2], ***C. CASELLATO**[1];
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Abstract: Data-driven modelling of the brain requires a neuroinformatic framework implementing a general strategy to accommodate experimental data at different scales. To this end, we have developed the Brain Scaffold Builder (BSB), a tool for structural and functional microcircuit modelling. The BSB provides an organized staged workflow, multiple strategies for cell placement and connectivity, a configuration system capable of including detailed neuronal and synaptic models and the support for multiple simulators with transparent parallel processing. It is provided as an open-source package, applicable for multi-scale modelling of different brain areas. The interfaces with several simulators (NEURON, NEST, Arbor) allow to investigate the same brain region at different levels of resolution, depending on the scientific question about structure-function relationship. BSB effectiveness was tested on the cerebellar network, which has a complex geometry and raises a broad set of modelling challenges. For the first time, an entire module of mouse cerebellar cortex was reconstructed using morphologically realistic conductance-based multi-compartmental neuron models of granule, Golgi, Purkinje, stellate and basket cells. Different connection rules, such as convergence-divergence, touch detection and

voxel intersection, were used to generate the connectome, unifying a collection of scattered experimental data into a coherent construct. Baseline and sensory-burst stimulation were used for functional validation against *in vivo* recordings, monitoring the impact of subcellular and cellular mechanisms on signal propagation and spatio-temporal processing in the cerebellum. The integration of structural and functional properties through the model provides a new "ground truth" about cerebellar circuit organization capable of predicting neural dynamics *in vivo*.

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Poster

P135. Oscillations and Synchrony: EEG Studies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P135.01

Topic: B.09. Network interactions

Support: Whitehall Foundation grant 2017-12-73
NIH National Institute of General Medical Sciences R01GM134363-01
UC San Diego Halıcıoğlu Data Science Institute Fellowship

Title: The shape of spectral peaks of EEG alpha-rhythms: Gaussian or not?

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Abstract: The alpha-rhythm is the most prominent oscillation in the human brain, characterized in the spectral domain by a peak in the 8-13 Hz frequency range. This spectral peak has been modelled frequently using the Gaussian distribution (Donoghue, Haller et al., 2020; Chiang et al., 2011; Blankertz et al., 2010), using a central frequency and a bandwidth parameter. We examined the Gaussian assumption using a large developmental EEG dataset using resting data in eyes open and eyes closed conditions of approximately five minutes in length. Data was available for 1205 participants, with ages ranging from 5 to 21 years.
We first computed the EEG power spectrum using Welch's method. Then the 1/f-component of the spectrum was fitted and subtracted. Recordings with a minimum spectral peak height of 10 dB (1/f-corrected) in the alpha-range were selected, retaining only datasets with a very high signal-to-noise ratio. The spectral peak was modelled using different types of probability distributions, centered around the individual alpha-peak: Gaussian, Laplace, Gamma as well as a kernel density estimation procedure. We find that spectral alpha-peaks for high signal-to-noise ratio exhibit a shape that is not well captured by a single Gaussian, underestimating the peakedness of the distribution, resulting in a reduced height of the spectral peak when modelled

by a Gaussian. In several instances, the spectral peak exhibited a non-symmetric skew and was not uni-modal. We illustrate how spatial-spectral decomposition can be applied to recover uni-modal spectral peaks to reduce the effects of rhythms with different peak frequencies overlapping in space and time.
An improved characterization of the shape of oscillatory peaks is informative for different purposes. First, this will lead to improved spectral parametrization procedures using for instance more appropriate basis functions for the spectral peaks. Second, a better spectral peak characterization may be helpful in deriving functional models of the alpha-rhythm, for instance, for disambiguating between dampening and desynchronization models (Liley & Muthukamaraswamy, 2020). The spectrum is a summary measure of neural oscillations, and the temporal structure of the alpha-rhythm exhibits more complexity than is commonly appreciated. But as spectra are easily computable, it would be of interest to exploit information present in spectra to the fullest extent to the most degree, for instance regarding long-range temporal correlations or waveform shape. Understanding the mechanisms underlying oscillatory activity in the human brain and their link to perception and cognition is central to understanding how our brain works.

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Poster

P135. Oscillations and Synchrony: EEG Studies

Location: Virtual Only

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Program #/Poster #: P135.02

Topic: B.09. Network interactions

Support: NSERC

Title: Resting state metabolite concentrations in the anterior cingulate cortex and their influence in inter-subject variability in Laser-Evoked-Potentials

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Abstract: Inter-subject variability is a major limiting factor for the adoption of Laser evoked potentials (LEPs) in clinical settings for the diagnosis of pain and pain conditions such as neuropathy. LEPs show clear deflections at latencies compatible with the conduction velocity of A δ-fibers, and the largest deflection is a negative-positive N2/P2 waveform measured at the scalp vertex. The anterior cingulate cortex (ACC) has been identified as one of the main cortical

generators of this waveform, and recently, an increase of glutamate (Glu) and glutamate + glutamine (Glx) concentrations in this region have been reported during the onset of pain. Here, we performed a correlational study using proton magnetic resonance spectroscopy (^{1}H-MRS) and surface electroencephalography (sEEG) to determine if resting state concentrations of Glu and Glx in the ACC can account for some of the inter-subject variability in the amplitude and latency of the N2/P2 components across a common level of pain perception. Four different levels of laser stimulation were presented, and inter-subject pain perception was controlled by analyzing a subset of LEPs rated to feel the same (i.e. same rating in the numeric rating scale) across all subjects. ^{1}H-MRS data were collected in an 8 mL voxel using a 3T Philips scanner. Food intake was standardized before data collection and females were tested only during their follicular phase. Preliminary results (n=4, 1f, mean age = 22.7 ± 5.5) show a negative inter-subject trend between then N2 amplitude and resting state concentrations of Glu and Glx (Glu, R^2=-0.71, p =0.29; Glx, R^2=-0.80, p=0.20) when controlling for perceived stimulus intensity. Furthermore, P2 amplitude also demonstrated a smaller negative trend with resting state concentrations of Glu and Glx (Glu, R^2=-0.51, p =0.49; Glx, R^2=-0.53, p=0.47). The latency of the N2 component also showed a positive trend with increasing levels of Glu and Glx (Glu, R^2=0.66, p =0.34; Glx, R^2=0.76, p=0.24). This relationship will be further examined in a larger cohort of healthy participants and other sEEG outcomes related to pain such as gamma-band oscillations will also be examined with relation to resting state concentrations of Glu and Glx. Understanding how the underlying biochemical environment of cortical generators affects inter-subject amplitude and latency of LEPs can help their implementation in clinical settings.

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Poster

P135. Oscillations and Synchrony: EEG Studies

Location: Virtual Only

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Program #/Poster #: P135.03

Topic: B.09. Network interactions

Title: Brain asynchrony following infection with SARS-CoV-2: Physiologic evidence based on electroencephalography and machine learning.

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Abstract: The high prevalence of symptoms consistent with central nervous system pathology following SARS-CoV-2 infection is contrasted by a lack of clinical tools to objectively assess

these symptoms and a poor understanding of their pathophysiology. Prior research based on the visual interpretation of electroencephalography (EEG) in patients with COVID-19 (C19) failed to identify distinct EEG signatures that discriminate between pre-existing illnesses and novel C19 pathology in the brain. Here, we performed quantitative analysis of EEG using a fully-automated EEG processing pipeline in large cohorts of patients at Cleveland Clinic, Ohio and Kings County Hospital Center, New York (n=156). First, compared to hospitalized patients prior to C19 in the same clinical environment, resting state global EEG power (averaged from 15 channels) in patients with confirmed C19 diagnosis is decreased in the 4-8 Hz theta band (0.19 $\mu V^2/Hz$ in C19+ vs. 0.22 $\mu V^2/Hz$ in C19-, $p = 0.011$), and increased in the 31-50 Hz gamma band (0.016 $\mu V^2/Hz$ in C19+ vs. 0.011 $\mu V^2/Hz$ in C19-, $p = 0.0064$). Moreover, phase amplitude coupling (PAC) between the theta phase and the gamma amplitude was lower in C19+ patients. Second, C19+ patients were distinguished from C19- patients with 71% accuracy using a binary neural net algorithm trained on PAC and spectral power features. These results demonstrate significant effects of SARS-CoV-2 on brain dynamics, particularly cross-frequency synchrony between bands known to have distinct biophysical mechanisms. This may potentially explain altered cognitive states in C19 'long-haulers' with neurological sequelae. Machine learning results, relative to visual analysis of EEG, further provide unbiased evidence for the impact of SARS-CoV-2 on brain activity irrespective of confirmed medical diagnosis or symptomatology, suggesting that a similar approach could be developed in the future to predict the trajectory of C19 disease.

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Poster

P135. Oscillations and Synchrony: EEG Studies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P135.04

Topic: B.09. Network interactions

Support: Center for Neurorestoration and Neurotechnology at the Providence VA Medical Center

Title: Transient neural oscillations reveal new biomarkers for therapeutic transcranial magnetic stimulation for MDD and PTSD

Authors: *A. MORRIS[1], S. TEMEREANCA[2], A. ZANDVAKILI[1], D. D. SLIVA[2], A. TYRKA[3], B. GREENBERG[1], L. CARPENTER[3], N. PHILIP[1], S. R. JONES[2];
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Abstract: Repetitive transcranial magnetic stimulation (TMS) is a therapy for major depressive disorder (MDD) and post-traumatic stress disorder (PTSD). TMS effectiveness varies across patients and optimal protocols are unknown. Electroencephalography (EEG) examines changes in brain dynamics associated with TMS (Zandvakili et al, 2019). Standard EEG analysis methods investigate oscillations in highly averaged data. Recent studies indicate that, in non-averaged signals, EEG oscillations often emerge as brief high power transients, coined "events" (Jones, 2016; Shin et al, 2017). We applied such methods to identify and analyze event features in an attempt to describe novel biomarkers of TMS and their relationship to clinical outcomes. We collected resting 8-channel EEG before and after therapeutic TMS applied to the left dorsolateral prefrontal cortex in 23 patients with comorbid MDD and PTSD. Using our open-source toolbox (https://github.com/jonescompneurolab/SpectralEvents) we tested if resting state oscillations can be quantified as events in non-averaged data using 2-second windows. We then assessed if event characteristics - including event number, power, duration, or frequency span - changed after TMS or correlated with symptom improvement. Averaged omnibus power spectral density in resting EEG data revealed distinct peaks in canonical frequency bands including delta/theta (1-6 Hz), alpha (7-14 Hz), and beta (15-29 Hz). Investigation of non-averaged signals showed that oscillations in each band emerged as transient high-power events before and after TMS. Across all subjects, the number of frontal delta/theta ($p = 0.023$) and alpha ($p = 0.023$) events and the duration of medial delta/theta ($p = 0.001$) events decreased after TMS. Pretreatment event characteristics correlated with subsequent clinical response. Frontal delta/theta event frequency span was negatively associated with MDD & PTSD improvement ($r^2 = 0.24, 0.22$); medial alpha event power and duration were positively associated with MDD improvement ($r^2 = 0.17, 0.19$) whereas medial alpha event rate and duration were positively associated with PTSD improvement ($r^2 = 0.18, 0.18$); and frontal beta duration was negatively associated with PTSD improvement ($r^2 = 0.18$). Novel methods to characterize spectral events in non-averaged signals successfully identified transient periods of high power activity in resting state EEG data and provided insight into changes in event features potentially predictive of TMS treatment response. With further effort, transient signatures in EEG oscillations may provide new biomarkers of response and a better mechanistic understanding of TMS.

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that research relationship even if those funds come to an institution.; Neuronetics, NeoSync, Janssen. F. Consulting Fees (e.g., advisory boards); Neuronetics. **S.R. Jones:** None.

Poster

P135. Oscillations and Synchrony: EEG Studies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P135.05

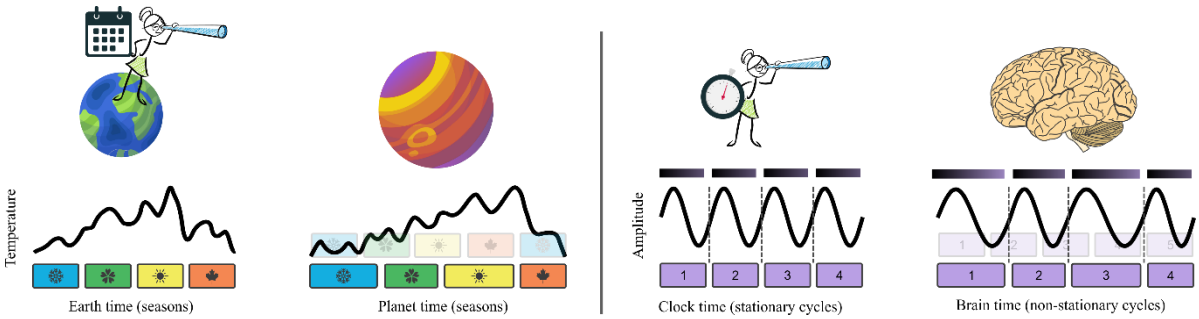
Topic: B.09. Network interactions

Support: ERC Grant 715714
ERC Grant 647954

Title: Clock time: a foreign measure to brain dynamics

Authors: *S. VAN BREE[1,2], M. MELCÓN[3], L. KOLIBIUS[1,2], C. KERRÉN[4], M. WIMBER[1,2], S. HANSLMAYR[1,2];
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Abstract: Human thought is highly flexible and dynamic, achieved by evolving patterns of brain activity across groups of cells. Neuroscience aims to understand cognition in the brain by analysing these intricate patterns. Here, we argue that this goal is impeded by the time format of our data - clock time. The brain is a system with its own dynamics and regime of time, with no intrinsic concern for the human-invented second. A more appropriate time format is cycles of brain oscillations, which coordinate neural firing and are widely implicated in cognition. These brain dynamics do not obey clock time - they start out of tune with clock time and drift apart even further as oscillations unpredictably slow down, speed up, and undergo abrupt changes. Since oscillations clock cognition, their dynamics should critically inform our analysis. We describe brain time warping as a new method to transform data in accordance with brain dynamics, which sets the time axis to cycles of clocking oscillations (a native unit) rather than milliseconds (a foreign unit). We also introduce the Brain Time Toolbox, a software library that implements brain time warping for electrophysiology data and tests whether it reveals information patterns in line with how the brain uses them.



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Poster

P135. Oscillations and Synchrony: EEG Studies

Location: Virtual Only

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Program #/Poster #: P135.06

Topic: B.09. Network interactions

Support: German Research Foundation
Leibniz Association
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Title: Individual slow wave events give rise to macroscopic fMRI signatures and drive the strength of the BOLD signal in human resting-state EEG-fMRI recordings

Authors: *M. ILHAN-BAYRAKCI[1], Y. CABRAL-CALDERIN[2], T. O. BERGMANN[3,1], O. TÜSCHER[1,3], A. STROH[3,1];
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Abstract: The slow wave state is a general state of quiescence that is interrupted by sudden bursts of activity or so-called slow wave events (SWE). Importantly, it is considered as the default activity of the cerebral cortex. Recently, the relationship between slow wave events and functional MRI (fMRI) blood oxygen level–dependent (BOLD) signals has been assessed in a rodent model which revealed a cortex-wide BOLD activation pattern. However, it remains unclear which macroscopic signature corresponds to these specific neurophysiological events in the human brain. Therefore, we analyzed well characterized simultaneous EEG-fMRI data during human Non-REM sleep. SWE were identified in the EEG data adapting a toolbox specific to slow wave detection. The individually detected SWE were then used as predictors in event-related fMRI analyses to examine the relationship between task-free, resting-state fMRI signals and slow wave events. For all 10 subjects we identified significant changes in BOLD activity in association with slow wave events covering substantial parts of the cortical and subcortical gray

matter. In line with the findings of the animal studies, we show a direct relation of a characteristic, non-oscillatory neurophysiological event to specific BOLD activation patterns. Notably, we found a correlation between the number of SWE and the spatial extent of these specific BOLD activation patterns, and we show that the strength of the BOLD response strongly depends on the amplitude of the SWE. Our study puts forward both a novel pipeline for analyzing resting state EEG - fMRI data based on well-defined events to be used as a regressor, and, provides direct evidence for a relation between the strength of a neuronal signal and its representation in BOLD fMRI. This is particularly important with regard to subsequent studies in early stages of neuropsychiatric disorders. As recent findings suggest hampered SWE propagation in neuropsychiatric diseases, it is critical to resolve the brain's slow wave activity state network architecture, which holds the potential for establishing an early imaging biomarker and a novel target for early treatment for a variety of diseases long before disease onset.

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Poster

P135. Oscillations and Synchrony: EEG Studies

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Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P135.07

Topic: B.09. Network interactions

Support: CIHR

Title: Sex-related Differences in Cortical EEG, Hippocampal LTD and De-depression Following Prolonged Exposure to Deep Coma Isoflurane Anesthesia

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Abstract: Cortical EEG activity beyond the isoelectric EEG of deep coma induced by inhalational anesthetics is poorly understood. Specific patterns of activity emerge in the EEG, as the dose of anesthetic is increased. In animal studies, when ISO % is increased, EEG activity changes from large-amplitude slow wave to burst suppression pattern, followed by an isoelectric EEG. Beyond the isoelectric EEG, randomly occurring fast, large-amplitude spikes (FLAS) appear. Since the transfer of information between the cortical and hippocampal networks is bi-directional, it is possible for activity in either of these networks to induce reciprocal changes in the other (PLoS One 2013; 8(9):e75257). However, the functional consequences within the hippocampal networks during this state are not known. The present study examined the effects of prolonged exposure to deep ISO (5%) on long-term depression (LTD) and de-depression (DD) of the excitatory post-synaptic potential (fEPSP) in the CA1 region of the hippocampus. We also

examined if changes to LTD and DD are sex-specific. Adult male or female Sprague Dawley rats (~ 600 g) were anesthetized and cortical EEG were recorded using screw electrodes positioned in S1 bilaterally. ISO % was gradually increased to 5%, representing a state of deep coma. This anesthetic plane was then maintained for over 4 h during which FLAS waves were continuously monitored, and the brains were removed subsequently for *in-vitro* experiments. Hippocampal slices were prepared from either naïve or ISO-exposed rats using standard procedures. CA1 field EPSPs were evoked by stimulating the Schaffer collaterals. LTD was induced using a low-frequency stimulation (1 Hz, 1200 pulses). After following LTD for 30 min, LTD was reversed using DD stimuli (4 X 100 Hz, 10 s inter-stimulus interval). When animals were exposed to ISO 5% for ~ 4 h, there was a significant reduction in the LTD of fEPSPs in males. Indeed, the DD stimuli, elicited a significant potentiation of the fEPSP. In females, however, LTD was not affected and DD stimuli reversed LTD to pre-tetanic baseline levels. At 1, 2, 3, and 4 h of deep ISO coma, the average number of FLAS waves recorded in 30 min epochs measured 12, 41, 88, and 119 in male rats and only 9, 14, 19, and 27 in female rats, respectively. LTD, among its other role(s) serves to balance the overall excitability of neural networks. Sustained deep ISO (5%) would be expected to "flood" the entire brain and thereby depress all neural activity. The greater FLAS wave activity that occurs during deep ISO coma and the accompanying marked suppression of LTD reversal in the male brain may reflect a novel sex-related shift toward excitatory processes requiring further study.

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Poster

P282. Movement Disorders Other Than Parkinson's: Clinical Trials

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Program #/Poster #: P282.01

Topic: C.04. Movement Disorders other than Parkinson's Disease

Support: FRIA (F.R.S.-FNRS)

Title: Fingerprint of essential tremor on motor adaptation

Authors: *F. BLONDIAUX[1,2], L. LEBRUN[2,3], B. J. HANSEEUW[2,3], F. CREVECOEUR[1,2]; [1]Inst. of Information and Communication Technologies, Electronics and Applied Mathematics, Univ. Catholique de Louvain, Louvain-la-Neuve, Belgium; [2]Inst. of Neurosci., Univ. Catholique de Louvain, Brussels, Belgium; [3]Dept. of Neurol., Cliniques Universitaires Saint-Luc, Brussels, Belgium

Abstract: Objectives : Essential Tremor (ET), is characterized by involuntary oscillations of the limbs, mainly during voluntary movements. Although its origin remains unclear, there is a consensus on a neurogenic cause originating from a dysfunction of the cerebello-thalamo-

cortical loop. Given that previous studies highlighted a role of cerebellum in motor adaptation of eye and arm movements to changes in the environment, we sought to investigate the ability of patients with ET to adapt their movements in two different paradigms: an upper-limb adaptation task and a saccadic adaptation task. We reasoned that a deficit of adaptation observed in this population would constitute functional evidence supporting the hypothesis that ET is linked to cerebellar impairment.
Methods: We enrolled 40 volunteers, 20 ET patients (58.57 +- 15.45 years) along with 20 aged-matched healthy volunteers (56.79 +- 12.44 years). A neurological assessment confirmed their inclusion in the study. The task of saccadic adaptation consisted in performing saccadic eye movements towards a visual target that was jumped at the onset of saccade, resulting in an endpoint error. The task of upper-limb control consisted in visually-guided reaching movements disturbed by an orthogonal velocity-dependent force fields.
Results: During the saccadic adaptation task, reduced and slower adaptation was observed for the ET group in comparison with controls' behaviour. For the upper limb adaptation task, a lower asymptote characterized the learning curve of ET patients but the learning rates were comparable. In both tasks, we were able to dissociate movement components linked to anticipation and to online execution. The purely anticipatory phase of the movement, when feedback related to the movement was not yet available, did not reveal any significant differences between groups.
Conclusion: Altogether, our results suggest that ET patients' reduced adaptation may originate from impairment in online monitoring of their movements within a trial. These results could therefore expose an inability to correctly combine internal and sensory feedback, which in combination with the effect of sensorimotor delays leads to the production of tremor. These results support the hypothesis of a cerebellar involvement in ET, and could pave the way to create new tests to develop a positive diagnostic strategy and complement current clinical evaluations.

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Poster

P282. Movement Disorders Other Than Parkinson's: Clinical Trials

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P282.02

Topic: C.04. Movement Disorders other than Parkinson's Disease

Support: NIDA U01DAo47713
NIA supplements
BRINM

Title: Ptprd phosphatase inhibitors and pams for rls, alzheimer's neurofibrillary pathology and reward from addictive substances

Authors: *G. UHL[1], I. HENDERSON[2], E. D. LEVIN[3], W. WANG[4], D. K. JOHNSON[5], T. PRISINZANO[6];
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Abstract: The receptor type protein tyrosine phosphatase PTPRD is highly expressed by neurons and concentrated in their presynaptic terminals. PTPRD's activation by extracellular binding partners is thought to change activity of its intracellular phosphatase in altering tyrosine phosphorylation of a number of synaptic proteins that include the tau kinases GSK3 beta and GSK3 alpha. Human PTPRD genomic variants are associated with Restless leg syndrome, densities of neurofibrillary tangles in Alzheimer's disease (AD) postmortem brains, levels of PTPRD expression in postmortem human brains and a number of addiction-associated phenotypes. Mice with reduced PTPRD expression display addiction, RLS and AD-related phenotypes. These and other results suggest that reduced PTPRD function would reduce reward from addictive drugs and that increased PTPRD function would reduce accumulation of neurofibrillary pathology and RLS symptoms. We now report that selected flavanols, especially quercetin, act as substrate-selective positive allosteric modulators of the ability of PTPRD's phosphatase to dephosphorylate phosphotyrosine (pY) GSK3 alpha/beta and thus to downregulate activity of this tau hyperphosphorylator. In silico docking studies provide a novel candidate mechanism for this action. We report that NHB1109, a novel analog of our lead compound PTPRD phosphatase inhibitor 7-BIA, displays good potency and selectivity in vitro, reasonable oral bioavailability and therapeutic index and ability to reduce mouse cocaine-conditioned place preference and rat remifentanil self administration. These results each support PTPRD as a novel target for new therapeutics that inhibit or enhance its activities in ways that should benefit these three common human disorders. Comparisons between quercetin effects on PTPRD's abilities to dephosphorylate pYGSK3 phosphopeptide *vs* its effects on dephosphorylation of other PTPRD substrates supports the novel concept of substrate-specific positive allosteric modulation.

Disclosures: G. Uhl: None. **I. Henderson:** None. **E.D. Levin:** None. **W. Wang:** None. **D.K. Johnson:** None. **T. Prisinzano:** None.

Poster

P282. Movement Disorders Other Than Parkinson's: Clinical Trials

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P282.03

Topic: C.04. Movement Disorders other than Parkinson's Disease

Title: Electroencephalographic endpoints as translational central pharmacodynamic biomarkers of the activity of PRAX-944, a novel T-type calcium channel blocker for the treatment of essential tremor

Authors: *L. SCOTT, C. B. PURYEAR, G. BELFORT, T. SNYDER, S. RAINES, Z. A. HUGHES, A. WEHR, M. HARD, B. RAVINA, M. WITTMANN;
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Abstract: Essential tremor (ET) is the most common movement disorder affecting approximately 7 million patients in the United States alone. Mounting evidence suggests that tremor is caused by increased neuronal burst firing and oscillations in cerebello-thalamo-cortical circuitry, which is thought to be driven by T-type Ca^{2+} channel activity. Furthermore, T-type Ca^{2+} channels regulate EEG power in the sigma band (σ-power, 11-15 Hz) during non-rapid eye movement (NREM) sleep. Therefore, we sought to use modulation of NREM σ-power as a translational biomarker for assessing whether we can achieve potentially therapeutic levels of central blockade of T-type Ca^{2+} channels in humans at doses that are well tolerated. We tested the effects of PRAX-944, a novel selective T-type Ca^{2+} channel blocker in clinical development for the treatment of ET, in the rat harmaline-induced tremor model. PRAX-944 dose-dependently reduced tremor by 50% and 72% with 1 mg/kg and 3 mg/kg doses, respectively. Both reductions were significantly different compared to vehicle ($n=15-45$, $p<0.05$). Effects in the rat spontaneous locomotor assay were assessed, and although doses of ≥10 mg/kg reduced total distance moved ($n=10-20$, $p<0.05$), activity was not reduced at doses ≤3 mg/kg. NREM σ-power was used as a mechanistic biomarker of T-type Ca^{2+} channel blockade in rat and human studies. PRAX-944 produced robust dose-dependent effects on NREM σ-power in rats across a similar dose range that decreased harmaline-induced tremor. Notably, 1 and 3 mg/kg doses of PRAX-944 reduced NREM σ-power by approximately 30 and 50% ($n=11$, $p<0.05$), respectively. Data from healthy participants, in a within-subject dose-escalation Phase 1 study, showed similar robust and dose-dependent reductions in NREM σ-power across a wide and well-tolerated dose range (5-120 mg) ($n=8-10$ per dose, $p<0.05$). Importantly, NREM σ-power was reduced by approximately 25-50% at doses of 20-80 mg, with no meaningful further reduction achieved above 80 mg. Additional exploratory analysis on awake EEG periods showed that gamma frequency power was also reduced in a dose-dependent manner. Administration of PRAX-944 in rats and humans produced consistent effects on NREM σ-power, which may represent a robust and translational biomarker of T-type Ca^{2+} channel blockade. Reduced NREM σ-power in rats was observed at doses that reduced harmaline-induced tremor. The comparable reduction in NREM σ-power in humans indicates that robust blockade of T-type Ca^{2+} channels was achieved at well-tolerated doses. These findings suggest that the doses of PRAX-944 tested here may hold promise for patients with ET.

Disclosures: L. Scott: A. Employment/Salary (full or part-time):; Praxis Precision Medicines. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Praxis Precision Medicines. **C.B. Puryear:** A. Employment/Salary (full or part-time):; Praxis Precision Medicines. E. Ownership Interest

(stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Praxis Precision Medicines. **G. Belfort:** A. Employment/Salary (full or part-time):; Praxis Precision Medicines. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Praxis Precision Medicines. **T. Snyder:** A. Employment/Salary (full or part-time):; Praxis Precision Medicines. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Praxis Precision Medicines. **S. Raines:** A. Employment/Salary (full or part-time):; Praxis Precision Medicines. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Praxis Precision Medicines. **Z.A. Hughes:** A. Employment/Salary (full or part-time):; Praxis Precision Medicines. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Praxis Precision Medicines. **A. Wehr:** A. Employment/Salary (full or part-time):; Praxis Precision Medicines. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Praxis Precision Medicines. **M. Hard:** A. Employment/Salary (full or part-time):; Praxis Precision Medicines. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Praxis Precision Medicines. **B. Ravina:** A. Employment/Salary (full or part-time):; Praxis Precision Medicines. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Praxis Precision Medicines. **M. Wittmann:** A. Employment/Salary (full or part-time):; Praxis Precision Medicines. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Praxis Precision Medicines.

Poster

P282. Movement Disorders Other Than Parkinson's: Clinical Trials

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P282.04

Topic: C.04. Movement Disorders other than Parkinson's Disease

Support: NBMI Science Ltd., EMERA006

Title: Effect of treatment with iron chelator NBMI on the expression of specific metabolic brain patterns in patients with multiple system atrophy and progressive supranuclear palsy: exploratory findings

Authors: *M. PEROVNIK[1,3], Ž. DRAKULIĆ[1], J. JAMŠEK[2], D. GEORGIEV[1,3], D. FLISAR[1,3], Z. PIRTOŠEK[1,3], M. TROŠT[1,2,3];

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Abstract: Objective: To explore the effect of N1, N3-bis (2-mercaptoethyl sophthalamide (NBMI)) treatment on brain metabolism. **Background:** Multiple system atrophy (MSA) and progressive supranuclear palsy (PSP) are progressive, incurable atypical parkinsonian syndromes, characterized by accumulation of α-synuclein and tau protein, respectively. Abnormal protein aggregation can be triggered by elevated ferric iron concentrations and abnormal brain iron accumulation has been observed before. NBMI is a safe and efficacious membrane and blood brain barrier passing metal chelator and antioxidant that strongly binds toxic metals. **Methods:** This was a pilot exploratory, randomised, placebo-controlled, double blinded, cross-over, Phase 2a study to explore efficacy and safety of NBMI treatment in 20 MSA or PSP patients (EMERA006, EudraCT: 2018-000506-34). 2 MSA and 3 PSP patients were additionally scanned with FDG PET twice, at baseline and at last visit. We performed TPR network analysis to measure the expression of MSA-related pattern (MSARP) and PSPRP in all FDG PET scans (1). **Results:** Clinical and imaging data are presented in Table 1. One MSA patient progressed slower than normal compared to normal progression of MSARP and Unified MSA Rating Scale (UMSARS) historical data (1, 2). The other MSA patient progressed similarly compared to the reference data. One PSP patient improved based on decreased PSPRP expression score and reduction in PSP Rating Scale (PSPRS) score. One PSP patient progressed slower than normal compared to normal progression of PSPRP and PSPRS historical data (1, 3). One PSP patient progressed similarly compared to the reference data based on PSPRP expression, but did not progress based on PSPRS score. **Conclusion:** Treatment with NBMI may slow down the progression of neurodegenerative changes in MSA and PSP at least in some patients. Larger groups and longer follow-up are needed to further assess its efficacy on group level. **References:** 1. Perovnik M et al. *MDS 2021* [in press]; 2. Foubert-Samier A et al. *Neurobiol Dis*. 2020; 3. Bang J et al. *Parkinsonism Relat Disord.* 2016.

Table 1. Expression of the corresponding metabolic pattern and clinical scale for each subject at each timepoint.

		MSARP/PSPRP [Z-score]			UMSARS/PSPRS				
	Dg	Baseline	Follow-up	Difference	Baseline	V1	V2	V3	Difference
S1	MSA	8.1	8.0	-0.1	61	63	59	60	-1
S2	MSA	0.5	1.8	+1.3	43	34	41	46	+3
S3	PSP	7.3	7.7	+0.4	39	41	42	46	+7
S4	PSP	7.7	9.1	+1.4	63	52	NA	63	0
S5	PSP	5.1	4.5	-0.6	37	27	37	34	-3

Disclosures: M. Perovnik: None. **Ž. Drakulić:** None. **J. Jamšek:** None. **D. Georgiev:** None. **D. Flisar:** None. **Z. Pirtošek:** None. **M. Trošt:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you

are a PI for a drug study, report that research relationship even if those funds come to an institution.; Principal investigator.

Poster

P282. Movement Disorders Other Than Parkinson's: Clinical Trials

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P282.05

Topic: C.04. Movement Disorders other than Parkinson's Disease

Support: NSF CBET grant 1806056

Title: The effect of afferent feedback on tremor propagation: a modeling study

Authors: *I. SYNDERGAARD[1], D. B. FREE[2], S. K. CHARLES[3], D. FARINA[4];
[2]Mechanical Engin., [3]Mechanical Engin. and Neurosci., [1]Brigham Young Univ., Provo, UT;
[4]Bioengineering, Imperial Col. London, London, United Kingdom

Abstract: Tremor is the most common movement disorder, affecting over 10 million individuals in the United States alone. A new field of tremor research is *tremor propagation*, which refers to the way tremor spreads throughout the upper limb because of mechanical coupling and neural feedback. Understanding tremor propagation is important because it may allow us to determine which muscles are most responsible for tremor, and thus where to intervene to suppress tremor optimally. So far, we have investigated tremor propagation using only open-loop multiple-input multiple-output (MIMO) models [1, 2]. While these studies yielded many significant insights into the mechanisms of tremor propagation, they did not include known neural feedback mechanisms.We expanded on prior models by including the effects of Golgi tendon organs (modeled as pure homonymous force feedback) and muscle spindles (type Ia and type II signals, modeled together as homonymous and heteronymous proportional/derivative feedback) to create a closed-loop MIMO model of the upper limb. Feedback gains were gleaned from the literature and stability analyses. The subsystem transformation from neural input to muscle force was analyzed with and without force feedback to simulate the effect of Golgi tendon organs on tremor. The full model was then analyzed by averaging magnitude ratios at tremor frequencies under the following conditions: without feedback, with force feedback alone, with proportional/derivative feedback alone, and with both force feedback and proportional/derivative feedback.We found that force feedback significantly increased the system’s response at tremor frequencies, especially as neural round-trip travel time increased (i.e. for more distal muscles). Consequently, force feedback increased the system’s bandwidth, amplifying the effect of tremorogenic neural signals. Compounding the problem, force feedback decreased the system’s response at low frequencies (such as in holding a posture), requiring greater neural drive to complete a desired movement, which may increase the tremorogenic signal as well. In contrast, proportional/derivative feedback had little effect on the average magnitude ratio of the system.

However, since it included heteronymous feedback, proportional/derivative feedback did spread tremor between the degrees of freedom of the arm.According to our simulations, neural feedback has a significant influence on tremor and how it spreads throughout the limb. Therefore, neural feedback should be included in models of tremor propagation. Understanding tremor propagation may eventually allow us to determine where to intervene to optimally suppress tremor.

Disclosures: I. Syndergaard: None. **D.B. Free:** None. **S.K. Charles:** None. **D. Farina:** None.

Poster

P282. Movement Disorders Other Than Parkinson's: Clinical Trials

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Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P282.06

Topic: C.04. Movement Disorders other than Parkinson's Disease

Support: NIH UH3NS095553

Title: Closing the loop on essential tremor in multiple subjects, using thalamic and cortical control signals

Authors: *B. PARKS, K. D. FOOTE, A. GUNDUZ;
Univ. of Florida, Gainesville, FL

Abstract: Essential tremor (ET) is a neurological disorder that is predominantly characterized by action tremor, is sometimes accompanied by rest tremor, and is one of the most common neurological disorders. Deep brain stimulation (DBS) of the ventral intermediate nucleus (Vim) region of the thalamus in an open loop setting is a common therapeutic strategy for tremor control. If tremor is not present during rest, continuous stimulation may lead to extraneous stimulation, potentially inducing side effects such as speech impairment, and may prematurely deplete the battery. Closed-loop DBS (CL-DBS) is a method of recording neurophysiology and triggering stimulation on only during specific epochs. As part of standard clinical care, a depth lead is placed within the Vim region; for the purposes of our study a cortical strip is placed over the primary motor cortex, specifically targeted at the hand knob region. Using an investigational bidirectional neurostimulator (Summit RC+S, Medtronic PLC), we recorded movement-related changes in spectral content from the cortical strip and the depth lead to implement cortical-based closed-loop (CL_C), and depth-based closed-loop (CL_D), respectively. To determine feasibility of both CL-DBS paradigms, movement tasks (goal-directed motions of the upper limbs), and holding tasks (maintaining position against gravity) are tested. The frequency band was independently chosen for each lead using a Fisher discriminant analysis, and was typically within the alpha/beta bands (8-30 Hz). The RC+S is equipped with a linear discriminant analysis, and utilizes the frequency band previously defined to check against a threshold for epoch determination. Here we show data collected from two patients, who both show consistent

performances using real-time power bands to drive closed-loop paradigms from either the cortex or the depth lead. Almost all trials, including both movement and holding tasks, performed above chance with $p < 0.05$ when using either the cortical or the depth lead to control stimulation. Clinical efficacy is simultaneously maintained, as determined by a tremor rating scale (TRS) score. All stimulation trials showed a lower score (less tremor) compared to a preoperative baseline score, and all closed-loop trials (CL_C and CL_D) were substantially equivalent to open-loop DBS. As proof of concept in two separate subjects, we have shown that there is potential to use the depth lead alone for recording neurological signals and subsequently triggering stimulation on/off. Translating these results to a broader population may decrease number of battery placement surgeries while maintaining clinical benefit.

Disclosures: B. Parks: None. **K.D. Foote:** None. **A. Gunduz:** None.

Poster

P337. Stroke Recovery: Non-Pharmacological Approaches to Therapy

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P337.01

Topic: E.04. Voluntary Movements

Support: Fondazione Baroni Grant 2020
Fondazione Roma grant (NCDS-2013-00000349)

Title: Bi-hemispheric transcranial direct current stimulation promotes neuroplasticity and anticipates motor recovery in a mouse model of ischemic stroke

Authors: M. BOLLA[1], S. A. BARBATI[1], V. LONGO[1], A. RE[1], F. PACIELLO[1], M. RINAUDO[1], F. VECCHIO[2], P. M. ROSSINI[2], C. GRASSI[3], *M. V. PODDA[3];
[1]Univ. Cattolica del Sacro Cuore, Rome, Italy; [2]IRCCS San Raffaele Roma, Rome, Italy; [3]Univ. Cattolica del Sacro Cuore; Fondazione Policlinico Universitario A. Gemelli IRCCS, Roma, Italy

Abstract: Introduction: Several studies have revealed that early treatment of ischemic stroke ameliorates stroke-related excess mortality and morbidity. However, stroke still represents one of the most common causes of death and long-term disability, so strategies promoting post-stroke functional recovery are necessary. For this purpose, we investigated the effect of bi-hemispheric transcranial direct current stimulation (tDCS, with the anode over the lesioned motor cortex and the cathode over the contralateral side) to promote motor function recovery and its mechanisms in a mouse model of motor cortex focal ischemia. **Methods:** Mice were subjected to Rose-Bengal photothrombosis in the forelimb area of the motor cortex. Seventy-two hours after stroke, mice underwent tDCS treatment (one session a day for 3 consecutive days). In order to evaluate motor function, grid-walking, single pellet reaching, and grip strength tests were performed. Local field potential recordings were used to assess brain connectivity. Western immunoblotting,

ELISA, qRT-PCR and Golgi-Cox staining were carried out to shed light on tDCS-promoted stroke recovery mechanisms. **Results:** Results showed an important role of tDCS to improve and anticipate motor recovery at the early subacute stage. In particular, tDCS was able to induce brain-derived neurotrophic factor (BDNF) expression and BDNF-dependent pathways, as well as to enhance structural plasticity and functional connectivity in motor and somatosensory cortices. The tDCS-promoted rescue was paralleled by enhanced plasma BDNF level, suggesting its potential role as circulating prognostic biomarker. Notably, in mice treated with BDNF TrKB receptor inhibitor (ANA-12) we did not observe the beneficial effects of tDCS on motor recovery and connectivity and the tDCS-induced increase of spine density, pERK, pCaMKII, pMEF and PSD-95. **Conclusions:** Our study points out that tDCS subacute treatment speeds up motor recovery after stroke. Here, enhanced BDNF expression following tDCS application appears fundamental to support the beneficial effects of tDCS. Therapies commonly adopted in stroke are based on physical rehabilitation, which is often delayed to a later stage when plastic responses are progressively lower. In this context, tDCS might be relevant to speed up and ameliorate post-stroke deficits in support of standard treatments.

Disclosures: M. Bolla: None. **S.A. Barbati:** None. **V. Longo:** None. **A. Re:** None. **F. Paciello:** None. **M. Rinaudo:** None. **F. Vecchio:** None. **P.M. Rossini:** None. **C. Grassi:** None. **M.V. Podda:** None.

Poster

P337. Stroke Recovery: Non-Pharmacological Approaches to Therapy

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P337.02

Topic: C.09.Stroke

Title: Role of astrocytic G6PD in oxidative stress and neuronal protection after ischemia

Authors: *Z. ZHANG, N. ZHANG, S. DING;
Univ. of Missouri-Columbia, COLUMBIA, MO

Abstract: Astrocytes are the predominant glia cell type in the central nervous system. Astrocytes are activated and experience notable changes in morphology, cell proliferation and gene expression after focal ischemic stroke (FIS). A growing body of evidence suggest that these reactive astrocytes play an important role in neuronal and brain protection after FIS. We found that glucose-6-phosphate dehydrogenase (G6PD), the rate limiting enzyme in pentose phosphate pathway (PPP), is highly unregulated in reactive astrocytes. PPP is a parallel pathway to glycolysis for glucose catabolism. In addition to providing essential metabolites for cell proliferation, PPP is vital for the production of NADPH which is essential for the production of reduced glutathione (GSH), the primary antioxidant for combating oxidative stress in disease condition. To study the effect of G6PD on metabolic reprogramming and oxidative stress after

ischemic stress, we overexpress G6PD in primary astrocytes through DNA transfection. We found G6PD overexpression significantly reduced extracellular acidification rate, an indirect measurement of glycolysis while did not affect oxygen consumption rate (OCR). Overexpression of G6PD increased the production of NADPH, but reduced NAD^+. Moreover, G6PD overexpression increased GSH with concurrent decrease in reactive oxygen species (ROS). Our results suggest G6PG overexpression causes metabolic shift from glycolysis to PPP without affecting mitochondrial function, and increases anti-oxidant capacity. Finally, G6PD overexpression can promote cell survival after oxygen-glucose deprivation (OGD) of astrocytes. Our results also show that the upregulated production of GSH dramatically decreases ROS production. In summary, our research suggests that reactive astrocytes can play an important role in neuronal and brain protection and recovery after ischemic stroke through reducing oxidative stress via increasing G6PD expression and promoting PPP might be a potential therapeutic approach for stroke.

Disclosures: Z. Zhang: None. **N. Zhang:** None. **S. Ding:** None.

Poster

P337. Stroke Recovery: Non-Pharmacological Approaches to Therapy

Location: Virtual Only

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Program #/Poster #: P337.03

Topic: C.09.Stroke

Support: Swedish Research Council (2017-02255, 2017-00991, 2017-00915 and 2018-02532)

Title: Elevated plasma neurofilament light chain levels predict improvement in the late phase after stroke

Authors: *M. PEKNA[1], A. STOKOWSKA[1], L. BUNKETORP KÄLL[1], C. BLOMSTRAND[1], J. SIMRÉN[1], M. NILSSON[2], H. ZETTERBERG[1], K. BLENNOW[1], M. PEKNY[1];
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Abstract: Background: Although functional recovery is most pronounced in the first 6 months after stroke, improvement is possible also in the late phase. We explored the value of plasma neurofilament light chain (NfL), a biomarker of axonal injury and secondary neurodegeneration, for the prediction of functional improvement in late phase after stroke.**Methods:** We measured baseline plasma NfL levels in 115 participants of a trial on the efficacy of multimodal rehabilitation in late phase after stroke. We determined the association between NfL levels, impairment in balance, gait and cognitive domains, and improvement 3 and 9 months later.**Results:** Plasma NfL levels were associated with the degree of impairment in all three domains. Individuals with meaningful improvement in balance and gait capacity had higher

plasma NfL levels compared with non-improvers (*P*=0.001 and *P*=0.018, respectively). Higher NfL levels were associated with improvement in balance (OR 2.34, CI: 1.35-4.27, *P*=0.004) and gait (OR 2.27, CI: 1.25-4.32, *P*=0.009). Elevated plasma NfL levels showed a positive predictive value for cognitive improvement, and this effect was specific for the intervention targeting the cognitive domain. The association of NfL levels with cognitive improvement withstood correction for baseline impairment, age and total years of schooling (OR 7.54, CI: 1.52-45.66, *P*=0.018).**Conclusions:** In addition to its established role as a biomarker in the acute phase, elevated circulating NfL levels may predict functional improvement in late phase after stroke. Our results should prompt further studies into the use of plasma NfL as a biomarker in late phase after stroke.

Disclosures: M. Pekna: None. **A. Stokowska:** None. **L. Bunketorp Käll:** None. **C. Blomstrand:** None. **J. Simrén:** None. **M. Nilsson:** None. **H. Zetterberg:** None. **K. Blennow:** None. **M. Pekny:** None.

Poster

P337. Stroke Recovery: Non-Pharmacological Approaches to Therapy

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Topic: C.09.Stroke

Support: NIH R01NS093057
NIH RF1MH11422701
NIH R25
NREF grant

Title: Circuit specific plasticity changes after post-stroke optogenetic stimulation

Authors: *T. CHIANG[1], S. VAHDAT[4], A. V. PENDHARKAR[1], S. HARVEY[2], H. UCHINO[2], Z. CAO[1], A. KIM[1], M. CHOY[2], H. CHEN[1], H. J. LEE[2], M. Y. CHENG[1], J. H. LEE[3], G. K. STEINBERG[1];
[1]Neurosurg., [3]Neurol. & Neurolog. Sciences, Bioengineering, Neurosurg., [2]Stanford Univ., Stanford, CA; [4]Applied Physiol. and Kinesiology, Univ. of Florida, Gainesville, FL

Abstract: Background: Functional recovery after stroke is attributed to rewiring of neural connections. We have demonstrated that optogenetic stimulations in the ipsilesional primary motor cortex (iM1) can promote functional recovery. Recently our brain imaging data indicate that iM1 stimulated mice can restore activation of the ipsilesional cortico-thalamic tract. To address whether iM1 stimulation enhanced post-stroke plasticity in the cortico-thalamic tract, we investigate the expression of plasticity markers in the ipsilesional thalamus (iTH). We also selectively stimulate the cortico-thalamic circuit and examine its effects on plasticity changes

and functional outcome. Methods: For iM1 stimulation, mice were injected with AAV1-CaMKIIa-hChR2 virus and implanted with optical fiber in iM1 (C57BL6 male mice, 6-7 weeks old). To selectively stimulate the cortico-thalamic circuit, mice were injected with AAV1-CaMKIIa-hChR2 virus in iM1 and an optical fiber was implanted in the ipsilesional thalamus (iTH). Six weeks were allowed for virus expression and mice underwent transient middle cerebral artery occlusion (tMCAo, 30min). Mice received optogenetic stimulations from PD5-14. Functional recovery was evaluated by rotating beam test at pre-stroke and post-stroke days (PD) 4, 7 and 14. Brain sections were immunostained with plasticity marker synaptophysin and imaged for co-localization of synaptophysin and ChR2. Western blot analysis of plasticity markers was performed for thalamic tissue. Results: Rotating beam test show significant functional improvement in iM1-stimulated mice (Stim group) when compared to non-stimulated mice (NoStim group). iM1-stimulated mice showed higher expression of axonal growth marker GAP43 in iTH. Immunostaining and co-localization analyses with pre-synaptic marker Synaptophysin indicate that iM1-stimulated mice exhibited higher co-localization of $ChR2^{+}$/$Synaptophysin^{+}$ terminals in thalamus. Furthermore, histology showed robust ChR2 is expression in the iM1-iTH motor cortico-thalamic circuit, and selective stimulation of this cortico-thalamic circuit is sufficient to improve recovery, Conclusion: Our results show that cortico-thalamic circuit is a key circuit involved in post-stroke recovery. Ongoing studies include investigating plasticity changes in the iTH as well as transcriptome profiling of the cortico-thalamic circuit.

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Poster

P337. Stroke Recovery: Non-Pharmacological Approaches to Therapy

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Program #/Poster #: P337.05

Topic: C.09.Stroke

Support: German Research Council, comprehensive research center 1451
Köln Fortune

Title: Analysis of miRNA patterns in peri-ischemia and secondary degenerating tissue after focal experimental stroke: beneficial impact by enriched environment

Authors: *J. ROMER[1], A. LADWIG[1], V. ZIMMERMANN[1], S. BLASCHKE[1], M. A. RÜGER[1], M. BERGAMI[2], G. R. FINK[1], M. SCHROETER[1];
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Abstract: Modifiable risk factors such as diet and physical inactivity have a large impact on the outcome of stroke. Specifically, hyperglycemia in stroke has been shown to lead to higher morbidity and mortality. Blood brain barrier disruptions and increased inflammation amongst others were postulated as possible contributing factors. On the contrary, exercise is known to have positive effects on the brain such as reduction of inflammation, increased synaptic plasticity and neuronal arborization. This study asks for the influence of miRNA signals on the functional modification of the behavioral outcome after focal cerebral ischemia in the cortical motor area. Male mice were housed in enriched environment (EE) or control conditions (CC) with a high fat (HFD) or normal chow diet (NCD). After 6 weeks mice were subjected to focal photothrombotic stroke (PT). At day 16 post PT, mice were sacrificed, RNA was extracted from the infarct tissue and ipsilateral subcortical tissue (thalamus; as secondary degenerating area) and their respective contralateral counterparts. Anti (miR-9, -22, -124, -181c)- and pro-inflammatory (miR-147, -155, -210) micro RNAs and downstream mRNAs (NF-kB, STAT3, GFAP, Iba1, Arg1 and iNOS) were analyzed by qPCR. Protein expression for Arg1 and iNOS were further examined by Western Blot. In the infarct tissue we found a diet dependent correlation between miRs, specifically miR-155 was found reduced in NCD compared to HFD. Subcortically, most miRs showed a positive correlation in CC, while in EE, miRs appeared to be more differentially regulated. We also found a negative correlation between functional recovery and miR expression in the infarct area in the CC groups, but no correlation in the EE groups. Further, NF-kB and STAT3 expression was found highest in the EE HFD group in all areas, correlating inversely with subcortical miR expression. Environmental and dietary changes differentially influence miRNA expression patterns in stroke tissue and distant secondary degenerating areas. In CC, inflammatory and non-inflammatory miRNAs and mRNAs reacted together while in EE those were found independent of each other suggesting an inhibition of inflammatory cascades. Changing miRNA patterns may pose a mechanism by which EE acts beneficially after stroke.

Disclosures: J. Romer: None. **A. Ladwig:** None. **V. Zimmermann:** None. **S. Blaschke:** None. **M.A. Rüger:** None. **M. Bergami:** None. **G.R. Fink:** None. **M. Schroeter:** None.

Poster

P337. Stroke Recovery: Non-Pharmacological Approaches to Therapy

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P337.06

Topic: C.09.Stroke

Support: CRC 1451, german Research Council
Koeln Fortune, University of Cologne

Title: Both functional and structural effects of an enriched environment (EE) and nutrition on the outcome after experimental stroke in mice

Authors: *M. SCHROETER[1], A. LADWIG[1], V. ZIMERMANN[1], J. ROMER[1], S. BLASCHKE[1], H. BACKES[2], A.-L. CREMER[2], M. ASWENDT[1], D. WIEDERMANN[2], S. VLACHAKIS[1], M. A. RUEGER[1], M. BERGAMI[1], G. R. FINK[1];
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Abstract: Background: Stroke is the second most common cause of death worldwide and the most common cause for disability in adulthood in Germany. Comorbidities like diabetes and obesity have been shown to worsen the outcome, while physical therapy and an active lifestyle can ameliorate it. Aims: We examined the functional and structural effects of an enriched environment (EE) and nutrition on the mice outcome after experimental stroke. Methods: Male mice were housed in EE or control conditions (CC) with a high fat (HFD) or normal chow diet (NCD). At week 6, mice were subjected to focal photothrombotic stroke (PT). T2-weighed MRI imaging at day 2 after stroke measured infarct size. A neuroscore, a grid walk test, and an open-field test were performed for functional neurological assessment. PET imaging was performed before the stroke and at days 1 and 14 post-stroke. A week before and after PT, the mice underwent glucose and insulin tolerance tests. Immunohistochemistry (IHC) assessed neuronal damage, infarct volume, neovascularisation, glial scar formation, and microglial activation and polarization. Atlas-based brain mapping was used to identify specific brain regions of interest for analysis and correlation with PET and MRI data. Results: While mice in EE did not show any significant weight gain under HFD, they exhibited an impaired glucose tolerance compared to the group in CC. Immediately after stroke and during recovery, the EE NCD group showed better scores on the functional tests, although there was no significant difference between infarct size on MRI or FDG uptake in PET at day 2. On day 14, however, the EE group showed improved FDG uptake. IHC revealed an increased Arg1/iNOS ratio in the infarct, a denser infarct scar, and reduced astrogliosis in non-infarct brain tissue. Specifically, the EE NCD group revealed an inverse correlation of Iba1 and GFAP+ cells in most brain regions. Conclusion: EE improves functional outcomes after stroke. Immediate effects are possibly due to whole-brain network effects between neuronal and glial cells due to exercise in EE, while long-term effects might be caused by a better containment and improved pro-/anti-inflammatory balance within and around the infarct tissue.

Disclosures: M. Schroeter: None. **A. Ladwig:** None. **V. Zimermann:** None. **J. Romer:** None. **S. Blaschke:** None. **H. Backes:** None. **A. Cremer:** None. **M. Aswendt:** None. **D. Wiedermann:** None. **S. Vlachakis:** None. **M.A. Rueger:** None. **M. Bergami:** None. **G.R. Fink:** None.

Poster

P337. Stroke Recovery: Non-Pharmacological Approaches to Therapy

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Program #/Poster #: P337.07

Topic: C.09.Stroke

Support: NIH Grant R01NS094384
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Title: Testing the effects of common medications on VNS-mediated plasticity

Authors: *T. DANAPHONGSE, R. MORRISON, S. ABE, K. ADCOCK, V. EZHIL, A. SOMANEY, M. KILGARD, S. HAYS;
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Abstract: Vagus nerve stimulation (VNS) paired with rehabilitative training enhances recovery of function in a range of neurological injuries such as stroke, post-traumatic stress disorder, and spinal cord injury. Previous studies suggest that VNS-dependent recovery of motor function is supported by synaptic plasticity within central networks via activation of cholinergic, noradrenergic, and serotonergic systems. Patients undergoing VNS-paired rehabilitation are commonly prescribed medications that influence these neuromodulatory systems for issues directly stemming from their specific neurological injuries, related comorbidities, or other unrelated conditions. Based on their mechanisms of action, it is possible that these medications may impact treatment in patients undergoing VNS therapy. Alternatively, the common clinical doses of these medications may not reach a level to substantially impair VNS-dependent effects on the central nervous system. Because these medications are often critical to maintaining quality of life for those who take them, a clear understanding of how clinically-relevant doses of these drugs is critical to the clinical implementation of VNS-based therapies. In this study, we tested the effects of commonly prescribed medications at clinically relevant doses on VNS-mediated plasticity. We trained rats to perform a motor task paired with VNS and daily injections of either oxybutynin, prazosin, duloxetine, or saline, and measured subsequent reorganization of the motor cortex, a robust marker for neural plasticity that underlies many VNS-dependent benefits.

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Poster

P337. Stroke Recovery: Non-Pharmacological Approaches to Therapy

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P337.08

Topic: C.09.Stroke

Title: Genomics of functional network recovery: Looking at the cerebellum after lesioning motor neocortex in mice

Authors: *V. ZIMMERMANN[1], A. LADWIG[3], J. ROMER[3], S. BLASCHKE[2], M. RÜGER[3], M. BERGAMI[4], G. R. FINK[5], M. SCHROETER[6];
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Abstract: Genomics of functional network recovery: Looking at the cerebellum after lesioning motor neocortex in mice
Valentina Zimmermann[1], Anne Ladwig[1], Johannes Romer[1], Stefan Blaschke[1], Maria A. Rüger[1], Matteo Bergami[3], Gereon R. Fink[1], Michael Schroeter[1]
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A focal cortical lesion such as a stroke results in brain network changes in distant areas. Described as crossed cerebellar diaschisis the change of cerebellar (functional) activity is connected to recovery from stroke. Factors that can influence recovery are diet and exercise. Specifically, hyperglycemia and immobility are known risk factors for a poor outcome both in human and under experimental stroke conditions. In the present study we aimed to identify specific transcriptomic patterns in cerebellar tissue after experimental stroke in mice under different diet and environmental influences, to assess genomics in good versus poor recoverees after stroke in a paradigmatic way. Male mice were housed in enriched environment (EE) or under control conditions (CC) with a high fat (HFD) or normal chow diet (NCD). After 6 weeks mice underwent cortical photothrombotic stroke (PT). Functional tests assessed neurological deficits. 16 days post-infarct mice were sacrificed and RNA from cerebellar tissue was extracted. The transcriptome was sequenced and underwent bioinformatics analysis.We found most differentially expressed genes to be grouped into either inflammatory, cellular stress/ apoptosis, metabolic, neuroprotective, and in blood-brain-barrier associated pathways. In particular, EE ameliorated gene expression for neurotransmission and oxidative phosphorylation and decreased inflammatory genes, while the diet influenced mostly inflammation- and blood brain barrier associated genes. In conclusion, the combination of a healthy diet and physical exercise resulted in the most favorable outcome after stroke possibly due to gene expression modification in synaptic, neuroprotective, metabolic and anti-inflammatory pathways. This exploratory study thereby reveals molecular targets for improving the outcome after stroke.

Disclosures: V. Zimmermann: None. **A. Ladwig:** None. **J. Romer:** None. **S. Blaschke:** None. **M. Rüger:** None. **M. Bergami:** None. **G.R. Fink:** None. **M. Schroeter:** None.

Poster

P337. Stroke Recovery: Non-Pharmacological Approaches to Therapy

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Topic: C.09.Stroke

Support: the Korea Centers for Disease Control and Prevention (2019E-320202)
the Korean government (NRF-2020R1A2C3010304)

Title: Clustering of Multifaceted Functional Recovery Pattern after Stroke: Analysis of the KOSCO Data

Authors: *S. SHIN[1], Y.-S. LEE[1], J. LEE[1], W. CHANG[1], D. KIM[2], J. LEE[3], M. SOHN[4], M.-K. SONG[5], Y.-I. SHIN[6], Y.-S. LEE[7], M. JOO[8], S. LEE[10], J. HAN[11], J. AHN[12], G.-J. OH[9], S. CHOI[13], S.-Y. SEO[13], S. LEE[13], Y.-H. KIM[1];
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Abstract: This study aimed to cluster multifaceted functional recovery patterns of first-ever stroke patients using the unsupervised learning algorithm of artificial intelligence (AI) and to examine the validity of clusters by generating the prediction model using the boosting algorithm. This study was an interim analysis of the Korean Stroke Cohort for Functioning and Rehabilitation (KOSCO) dataset, which is a prospective multicenter cohort. Multifaceted functional assessments were performed five or six times repeatedly from seven days to 24 months of stroke onset. Out of 7,858 patients enrolled, 4,389 ischemic stroke (IS) and 1,146 hemorrhagic stroke (HS) patients who completed the functional assessment at 24 months after onset were included in this analysis. In k-means clustering analysis, multifaceted functional assessment scores, demographic features, and clinical information were used as input variables. The optimal cluster k number was determined by finding the highest k with at least a silhouette score of 0.2. IS patients were clustered into 11 groups while HS patients were clustered into 13 groups. Using 70% of the dataset, the prediction model for clustering was generated using the boosting algorithm. The validity of the prediction model was examined using the rest 30% of the dataset. All statistical analysis was implemented using the R (version 4.0.3). In both IS and HS, each cluster showed distinct clinical characteristics and functional recovery pattern which included the overall high function group, the overall low function group, the gradual improvement group, the late declining group, etc. Each cluster also showed different mean age and initial stroke severity measured by NIHSS scores. In validation of prediction model using

boosting algorithm, both IS and HS clusters showed high prediction accuracy (IS, 0.805; HS, 0.881). In our knowledge, this is the first study performing successful clustering of long-term functional recovery pattern in ischemic and hemorrhagic stroke patients using AI. Early identification and accurate prediction of long-term functional outcome will be useful for developing customized treatment of these patients.

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Poster

P337. Stroke Recovery: Non-Pharmacological Approaches to Therapy

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Program #/Poster #: P337.10

Topic: C.09.Stroke

Support: Academy of Finland #309479/2017
Doctoral Programme of Biomedicine - Helsinki University

Title: Dorsal skull meningeal lymphatic vessels drain blood-solutes after intracerebral hemorrhage

Authors: *A. VIRENQUE[1], R. BALIN[1], S. ANTILA[2], K. ALITALO[2], F. M. NOE[1];
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Abstract: Early intracranial hematoma drainage is crucial for the treatment of intracerebral hemorrhage (ICH). Currently, there are no available non-invasive strategies, which have been clinically approved as an aid in ICH resolution. The identification of novel therapeutic targets is therefore essential to develop safe and effective therapeutic strategies. Here we investigated the possible function of the meningeal lymphatic vessels (mLVs) in ICH resolution. To better identify the role of the mLVs we used two preclinical mouse models of ICH, mimicking focal cortical ischemic hemorrhage and subcortical extended hemorrhage, and characterized the dynamics of blood drainage through the dorsal mLVs. We also analyzed the effect of mLV drainage on the functional recovery after subcortical ICH, in mice presenting an acquired mLV deficit. After ICH induction in adult male and female mice, we observed a specific flow of blood-derived components within the mLVs, suggesting that the meningeal lymphatics can play a role in facilitating the hemorrhage drainage (no sex-related differences were observed). We also found that local formation of new mLVs is directly correlated with ICH-related neuroinflammation levels. Mice presenting acquired mLV dysfunction showed a delayed

recovery after ICH and a drop of functional connectivity in the areas affected by the intracranial hematoma. These findings suggest that meningeal lymphatics are involved in the resolution of ICH and could provide a valuable therapeutic target.

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Poster

P398. Peripheral Mechanisms of Neuropathic Pain Including Chemotherapy Induced Peripheral Neuropathy

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P398.01

Topic: D.03. Somatosensation – Pain

Support: NS113991

Title: A Novel WW Domain-dependent Interaction between Magi-2 and GluA1 Underlies a Potential Role in Synaptic Transmission in the Spinal Cord Dorsal Horn Following Inflammation

Authors: *G. SHEEHAN[1], J. KRISHNAN[1], P. AHUJA[1], A. BHATTACHARJEE[2]; [1]SUNY at Buffalo, Buffalo, NY; [2]Univ. at Buffalo - The State Univ. of New York, Buffalo, NY

Abstract: Membrane-Associated Guanylate Kinase, WW and PDZ Domain Containing 2 (Magi-2, S-SCAM) is a synaptic scaffolding molecule containing 2 WW and 6 PDZ domains that has been shown to regulate glutamatergic synaptic transmission. Here we show for the first time a novel WW-domain dependent interaction with the AMPA receptor subunit GluA1. This interaction leads to protection from protein degradation promoted by cotransfection with the E3-ubiquitin ligase Nedd4L. Mutation of a previously unidentified WW-domain within the GluA1 C-terminus blocked this Nedd4L-depedent degradation and enhanced GluA1 protein expression in the presence of Nedd4L. In addition to the protective effect, simultaneous transient transfection of GluA1 and Magi-2 led to increased surface expression of GluA1 as measured by membrane biotinylation. This increase was recapitulated by a two-fold increase in glutamate-induced peak GluA1 current densities in transiently transfected HEK-293T cells. Owing to the importance of calcium-permeable GluA1 homomers and their enhanced contribution to EPSCs in the dorsal horn (DH) of the spinal cord following various insults to peripheral sensory afferents we sought to explore a physiological role for the interaction of Magi-2 and GluA1 in the spinal cord. Lumbar DH tissue was collected from WT C57BL/6J mice and co-immunoprecipitation of GluA1 was confirmed via Magi-2 pulldown. Confocal immunofluorescence further illustrated colocalization of GluA1 and Magi-2 along dendrites in the superficial DH. Overall, Magi-2 was shown to have somewhat restricted expression in Lamina 1 and 2o, as well as in deeper lamina 4/5 with a higher degree of colocalization with TacR1 expressing neurons in Lamina 1 and 2

compared to PKCy expressing neurons in lamina 2i and 3. To probe for a role for Magi-2 in pain behaviors, lumbar SC tissue was taken following peripheral administration of CFA. An increase in the Magi-2 alpha isoform was seen in the ipsilateral DH. Further experiments will explore the behavioral and *ex vivo* electrophysiological effect of spinal cord DH Magi-2 knockdown in basal and inflammatory pain conditions.

Disclosures: G. Sheehan: None. **J. Krishnan:** None. **P. Ahuja:** None. **A. Bhattacharjee:** None.

Poster

P398. Peripheral Mechanisms of Neuropathic Pain Including Chemotherapy Induced Peripheral Neuropathy

Location: Virtual Only

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Program #/Poster #: P398.02

Topic: D.03. Somatosensation – Pain

Support: NIH grant NS113991

Title: Magi-1 deficiency decreases thermal sensitivity and increases hindpaw weight bearing in a nerve entrapment mouse model of neuropathic pain

Authors: *M. MARTIN[1], A. BHATTACHARJEE[2];
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Abstract: Neuropathic chronic pain is a leading cause of disability that can be caused by nerve entrapment, nerve injury, diabetes, viral infection, and chemotherapy. Current treatments either lack efficacy or have adverse side effects including abuse liability. Application of local anesthetics, which blocks all sodium channels, is currently considered as a first line treatment strategy for neuropathic pain. Instead of blocking sodium channels, an alternative method to treating neuropathic pain could be to disrupt their trafficking. Recent studies have indicated that WW domain-containing ubiquitin ligases are downregulated during nerve injury causing an increase in membrane targeting of sodium channels. The scaffold proteins responsible for sodium channel targeting and membrane stabilization in DRG neurons have not been fully characterized but our recently published studies identified the WW domain containing scaffold protein called Magi1, as a potential candidate (PMID: 30860870). Using an *in vivo* sciatic nerve knockdown approach strategy, we tested the consequences of Magi1 knockdown after established neuropathic pain. We used the sciatic nerve cuff model, which models nerve entrapment injury, in male and female mice. The Hargreaves thermal assay and dynamic weight bearing assay was used to confirm unilateral neuropathic pain, and was then followed by Magi1 *in vivo* knockdown. Magi1 knockdown was confirmed by Western analyses. We found an

increase in the thermal withdrawal latency during Magi1 knock-down in neuropathic mice compared to the shRNA treated mice. We also saw an increase of the percent weight borne on the ipsilateral paw in the Magi-1 knock-down mice using dynamic weight-bearing measurements compared to scrambled shRNA treated mice. Future studies will examine the consequences of Magi-1 knockdown on the electrophysiological properties of DRG neurons. Our studies reveal Magi-1 as a potential analgesic target for analgesia for nerve entrapment-induced neuropathic pain.

Disclosures: M. Martin: None. **A. Bhattacharjee:** None.

Poster

P398. Peripheral Mechanisms of Neuropathic Pain Including Chemotherapy Induced Peripheral Neuropathy

Location: Virtual Only

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Program #/Poster #: P398.03

Topic: D.03. Somatosensation – Pain

Support: NIH Grant NS113991

Title: Dynamic Weight Bearing More Closely Resembles the Thermal Hyperalgesia Phenotype, rather than Mechanical Allodynia during Sciatic Nerve Cuff-induced Neuropathic Pain

Authors: G. SHEEHAN, M. K. MARTIN, ***A. BHATTACHARJEE**;
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Abstract: Chronic constriction injury (CCI) of the sciatic nerve is a widely used model of neuropathic pain in rodents and two common behavioral phenotypes observed following CCI are thermal hyperalgesia and mechanical allodynia, as measured by the Hargreaves and Von Frey tests respectively. In this study, we used the sciatic nerve cuff variant of CCI in mice, which models nerve entrapment neuropathic pain in humans. Typically, thermal hyperalgesia recovers within 28-30 days following nerve cuff, while mechanical allodynia persists for at least 60 days (PMID: 18006342). Reflexive mechanical allodynia during CCI and indeed other neuropathic pain models, have been extensively used to model “chronic pain” where circuitry changes that occur in the lumbar spinal cord can be studied. Here we propose that in contrast to these evoked measures of nociceptive hypersensitivity, Dynamic Weight Bearing, where an unrestrained animal is allowed to freely roam, provides a more clinically relevant behavioral measure of ongoing pain in mice. We found that the effect of sciatic nerve cuff on the ratio of weight bearing by the injured relative to uninjured hindlimbs more closely resembles that of thermal hyperalgesia, following the trend toward recovery by about 28-30 days. Thus, we posit that non-myelinated, peptidergic C-Fibers innervating lamina I are the primary source of ongoing pain in mice following sciatic nerve cuff creating an anticipatory pain state that underlies the

recoverable weight bearing phenotype. We therefore propose that thermal hyperalgesia, rather than mechanical allodynia in the nerve cuff/CCI model is a more relevant behavioral measure when considering pharmacological interventions for persistent ongoing neuropathic pain.

Disclosures: G. Sheehan: None. **M.K. Martin:** None. **A. Bhattacharjee:** None.

Poster

P398. Peripheral Mechanisms of Neuropathic Pain Including Chemotherapy Induced Peripheral Neuropathy

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Program #/Poster #: P398.04

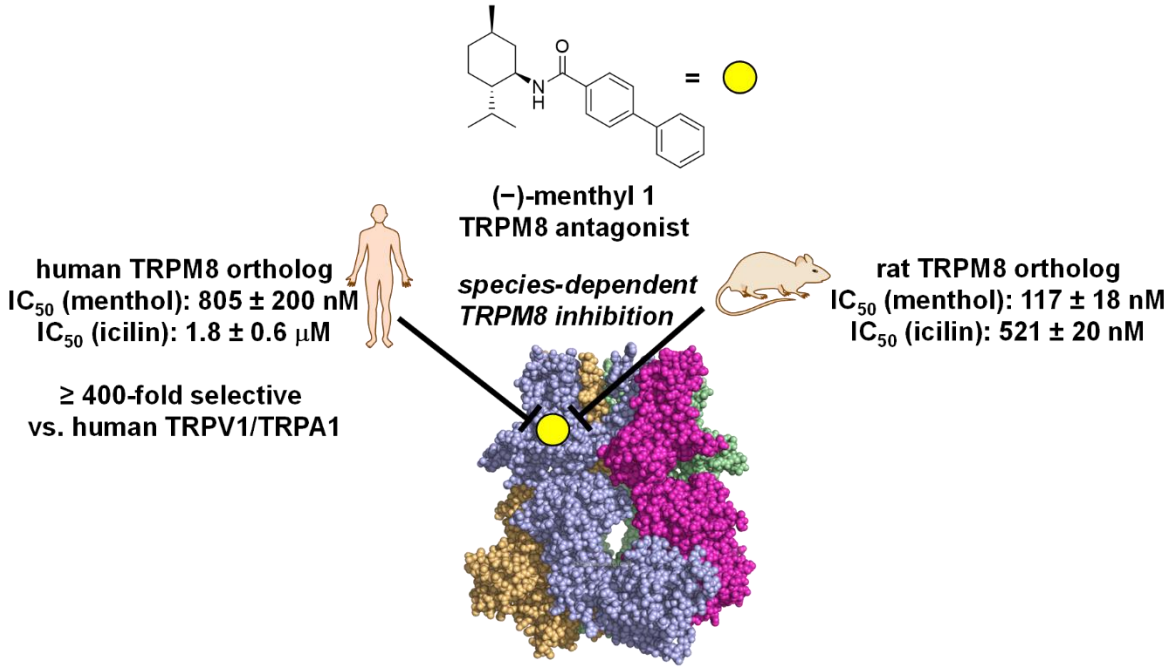
Topic: D.03. Somatosensation – Pain

Support: NIH grant U54GM104942−04
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Title: Structural and in vitro functional characterization of a menthyl TRPM8 antagonist indicates species-dependent regulation

Authors: *V. JOURNIGAN[1], D. ALARCÓN-ALARCÓN[2], Z. FENG[3], Y. WANG[3], T. LIANG[3], D. C. DAWLEY[1], A. AMIN[1], C. MONTANO[4], W. D. VAN HORN[4], X.-Q. XIE[3], A. FERRER-MONTIEL[2], A. FERNÁNDEZ-CARVAJAL[2];
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Abstract: TRPM8 antagonists derived from its cognate ligand, (−)-menthol, are underrepresented. We determine the absolute stereochemistry of a well-known TRPM8 antagonist, (−)-menthyl 1, using VCD and 2D NMR. We explore 1 for its antagonist effects of the human TRPM8 (hTRPM8) orthologue to uncover species-dependent inhibition versus rat channels. (−)-Menthyl 1 inhibits menthol- and icilin-evoked Ca^{2+} responses at hTRPM8 with IC_{50} values of 805 ± 200 nM and 1.8 ± 0.6 μM, respectively, while more potently inhibiting agonist responses at the rat orthologue (rTRPM8 IC_{50} (menthol) = 117 ± 18 nM, IC_{50} (icilin) = 521 ± 20 nM). Whole-cell patch-clamp recordings of hTRPM8 confirm the 1 inhibition of menthol-stimulated currents, with an IC_{50} of 700 ± 200 nM. We demonstrate that 1 possesses ≥400-fold selectivity for hTRPM8 versus hTRPA1/hTRPV1. (−)-menthyl 1 can be used as a novel chemical tool to study hTRPM8 pharmacology and differences in species commonly used in drug discovery.



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Poster

P398. Peripheral Mechanisms of Neuropathic Pain Including Chemotherapy Induced Peripheral Neuropathy

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Topic: D.03. Somatosensation – Pain

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NIH Grant AR069861
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NIH Grant DE027454

Title: IL-23/IL-17A/TRPV1 axis produces mechanical pain via macrophage-sensory neuron crosstalk in female mice

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Abstract: Sex dimorphism in chronic pain is a well-recognized clinical phenomenon, as females are more likely to suffer from chronic pain conditions. However, female-specific pain signaling is not well studied. Recently, we find that macrophages may use distinct pathways to modulate pain, and macrophage toll-like receptor 9 drives chronic pain only in males. In this study, we first investigate a female-dominant pathway which drives the crosstalk between macrophages and sensory neuron in chronic pain. Intraplantar (i.pl.) administration of interleukin 23 (IL-23) produces mechanical allodynia and conditioned place aversion in female but not male naïve mice. In the mouse model of chemotherapy-induced peripheral neuropathy (CIPN), *Il23* or *Il23r* deficiency or IL-23R antagonist P2305 exhibits anti-nociceptive effects only in female mice, indicating a female-specific role of IL-23/IL-23R axis in pain signaling. Notably, Estrogen deficiency after ovariectomy abrogates IL-23-induced pain and analgesic effects of P2305 against CIPN in females, and estrogen treatment enables IL-23-induced pain in males, implicating a regulative role of estrogen signaling. Female mice exhibit greater IL-23^{+} macrophage subset in DRG than males in CIPN *in vivo*, suggesting macrophages as the source of IL-23. Even IL-23 requires TRPV1^{+} C-fiber nociceptors to induce pain, IL-23 doesn't activate DRG sensory neuron directly. Macrophage ablation by clodronate abolishes IL-23-indued pain in females, whereas T cell deficiency by *Rag1*$^{-/-}$ or nude fails to do so, indicating an involvement of macrophage IL-23R signaling. Of note, IL-23 promotes macrophage release of IL-17A in females. Further, IL-17A induces mechanical allodynia, evokes calcium influx and potentiates action potential firing in DRG sensory neurons in a female-dominant manner, suggesting IL-17A as downstream effector of macrophage IL-23 signaling. Moreover, TRPV1 is also required for IL-23- and IL-17A-induced pain in females, and TRPV1 agonist capsaicin produces female-dominant mechanical allodynia in naïve mice. *In situ* hybridization evidence indicates female and male mice exhibit comparable *Il17ra*$^{+}$/*Trpv1*$^{+}$/*Erα*$^{+}$ DRG neurons, which provides neuronal components in IL-23/IL-17A/TRPV1 axis. Depletion of *Erα* in TRPV1^{+} C-fiber nociceptors reduces IL-23, IL-17A and capsaicin-induced pain in females, suggesting contributive role of neuronal estrogen signaling in this pain axis. In summary, this study demonstrates that the IL-23/IL-17A/TRPV1 axis regulates female-specific mechanical pain via neuro-immune interactions and suggests this axis as a novel target of precision therapy for women chronic pain patients.

Disclosures: X. Luo: None. **O. Chen:** None. **Z. Wang:** None. **S. Bang:** None. **J. Ji:** None. **S. Lee:** None. **Y. Huh:** None. **K. Furutani:** None. **Q. He:** None. **X. Tao:** None. **M. Ko:** None. **A. Bortsov:** None. **C. Donnelly:** None. **Y. Chen:** None. **A. Nackley:** None. **B. Temugin:** None. **R. Ji:** None.

Poster

P398. Peripheral Mechanisms of Neuropathic Pain Including Chemotherapy Induced Peripheral Neuropathy

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P398.06

Topic: D.03. Somatosensation – Pain

Support: NIH COBRE P20GM103643

Title: Phenotypic and functional characterization of CD4+ T cells in dorsal root ganglion of naïve and paclitaxel-treated male and female mice

Authors: *D. J. GOODE, N. E. MECUM;
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Abstract: Chemotherapeutic agents are often a life-saving cancer treatment, but the development of chemotherapy induced peripheral neuropathy (CIPN) is a major dose-limiting toxicity resulting in intractable pain that restricts the cancer survival rate. Recent reports demonstrate that mice lacking T cells have prolonged mechanical hypersensitivity after treatment with paclitaxel (PTX). The cytokines IL-10 and IL-4 have also been shown to be protective against CIPN, yet the sources of these cytokines in the dorsal root ganglion (DRG) after injury are unknown. The major IL-10 and IL-4 secreting immune cell is the CD4+ T cell; however, identification of CD4+ T cells in naïve and injured dorsal root ganglion (DRG) is still in its infancy. Here, we characterized the phenotype and functional properties of CD4+ T cells in acutely dissociated DRGs of naïve and PTX-treated female and male C57BL6/J mice using multi-color flow cytometry. Naïve females had a greater percentage of CD4+ T cells in the DRG compared to naïve male mice. The CD4+ T cells present in naïve male and female DRG had an effector/memory phenotype (CD44+) and were predominantly anti-inflammatory. However, the anti-inflammatory CD4+ T cell subpopulations were different in males and females. Females had a higher frequency of suppressive Foxp3+ T regulatory cells (Tregs) and IL-4 producing CD4+ T cells whereas males had a higher frequency of latency activated peptide+ (LAP; the pro-peptide of transforming growth factor-β) CD4+ T cells, another suppressive CD4+ T cell population. PTX increased anti-inflammatory Foxp3 Tregs and IL-4 producing CD4+ T cells in the DRG during the resolution of CIPN in both male and female mice; however, the frequencies were much higher in female DRG. IL-10 producing CD4+ T cells were also significantly greater in females than in males after PTX administration. Interestingly, PTX only increased pro-inflammatory IL-17A+ CD4+ T cells in the DRG of male mice, not female. The PTX-induced increase in CD4+ T cell populations in mouse DRG were transient as the frequencies returned to baseline levels when the mice were no longer mechanically hypersensitive. Our results demonstrate that PTX changes the phenotype and functional properties of CD4+ T cells in the DRG of male and female mice, indicating a potential target for treatment of CIPN.

Disclosures: D.J. Goode: None. **N.E. Mecum:** None.

Poster

P398. Peripheral Mechanisms of Neuropathic Pain Including Chemotherapy Induced Peripheral Neuropathy

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P398.07

Topic: D.03. Somatosensation – Pain

Title: Cannabichromene Effectively Reduces Cisplatin-Induced Neuropathic Pain

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Abstract: Cisplatin is a commonly used chemotherapy that has a dose limiting effect whereby 30-70% of patients develop peripheral neuropathy. Cisplatin induced neuropathy (CIN) presents in a stocking and glove pattern with patients reporting symptoms of allodynia. While current therapeutics show little success in treating CIN, recent studies suggest cannabis-based therapeutics as a potential treatment of CIN neuroinflammation and pain. While multiple studies have explored the effects of delta-9-tetrahydrocannabinol and synthetic cannabinoids against CIN, cannabinoids devoid of psychoactive effects would likely be a stronger candidate for drug development. Cananbichromene (CBC), a non-psychoactive cannabinoid, has shown to have antinociceptive properties across acute pain models of thermal and inflammatory pain. It is unknown if CBC can attenuate chronic allodynic pain. This study evaluated the effects of CBC against CIN induced allodynia. Our lab assessed whether CBC was able to 1) attenuate CIN (Experiment 1) and 2) prevent the development of CIN (Experiment 2). Mice in Experiment 1 received six cisplatin administrations on alternating days over 12 days with Ringers solution on non-cisplatin days. An electronic Von Frey (eVF) was used to measure withdrawal thresholds at baseline and following the 6th cisplatin administration. Two days following the last cisplatin injection, mice received either vehicle, oxycodone, or CBC (10, 25, or 50 mg/kg) then measured on the eVF 30 min later. Mice in Experiment 2 received CBC throughout the CIN induction protocol and tested on the eVF following the sixth cisplatin administration. In Experiment 1, mice receiving cisplatin showed a decrease in withdrawal thresholds, indicative of allodynia. On drug test day, vehicle treated mice continued to display low withdrawal thresholds while oxycodone increased withdrawal thresholds, indicative of analgesia. CBC produced a dose response whereas mice receiving 25 and 50 mg/kg showed increased withdrawal thresholds comparable to oxycodone. In Experiment 2, co-administration of 25 mg/kg CBC during the cisplatin regimen, significantly reduced tactile allodynia, even with a 2-day CBC wash-out period. These data suggest CBC's ability to not only attenuate the effects of cisplatin-induced neuropathic pain, but it also prevents or delay the onset of CIN. Further studies regarding the effects of CBC on the underlying pathophysiological response to CIN are ongoing. Together,

these data suggest CBC may have utility as a potential therapy for preventing or alleviating neuropathic pain.

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Poster

P398. Peripheral Mechanisms of Neuropathic Pain Including Chemotherapy Induced Peripheral Neuropathy

Location: Virtual Only

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Program #/Poster #: P398.08

Topic: D.03. Somatosensation – Pain

Support: NIH Grant R01CA231267
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Title: Oxidative DNA damage and cisplatin neurotoxicity is exacerbated by reducing the expression of 8-oxyguanosine glycosylase and AP endonuclease 1 in sensory neurons

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Abstract: Chemotherapy-induced peripheral neuropathy is a debilitating and dose-limiting adverse effect of anticancer treatment affecting cancer patients and survivors. Cisplatin, a common chemotherapeutic, is one of the most toxic chemotherapeutics and causes numbness, hypersensitivity to cold, and burning pain. Cisplatin generates DNA adducts and oxidative stress, the latter of which can result in oxidative DNA damage. The base excision repair (BER) pathway is the primary way sensory neurons repair cisplatin-induced oxidative lesions, such as 8-oxyguanosine (8-oxoG); however, the role of the different BER enzymes has not been assessed in sensory neurons. Understanding how oxidative DNA damage alters neuronal function is critical to understanding the mechanisms for cisplatin-induced neurotoxicity. Here, we utilized an in vitro model of cultured sensory neurons and siRNA technology to decrease the expression of the first two enzymes of the BER pathway, 8-oxoG glycosylase (OGG1) and AP endonuclease 1 (APE1) and measured sensory neuronal function and morphology. The content and stimulated release of the neuropeptide, calcitonin gene-related peptide (CGRP), was assessed as a surrogate for neuronal function in combination with immunohistochemical identification of cisplatin-induced DNA adducts and 8-oxoG. We found that knockdown of either BER enzyme alone did not affect CGRP release in the absence of cisplatin treatment, but increased the sensitizing effects of cisplatin on CGRP release. Surprisingly, combination knockdown had an additive effect in the presence of cisplatin, even though the enzymes are thought to participate in a linear

pathway. Decreasing OGG1 had a dramatic effect on neuronal morphology even in the absence of cisplatin, suggesting a novel role for the BER enzyme in regulating neurite homeostasis. Collectively, these data suggest that OGG1 and APE1 are neuroprotective in the presence of oxidative stress, such as that induced by cisplatin. Further research will examine how each of these BER enzymes contributes to neuronal homeostasis in conditions of high oxidative stress.

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Poster

P398. Peripheral Mechanisms of Neuropathic Pain Including Chemotherapy Induced Peripheral Neuropathy

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P398.09

Topic: D.03. Somatosensation – Pain

Title: The Role of Interleukin 23 receptor down modulator Tyrosine kinase 2, in Vincristine-induced Peripheral Neuropathy.

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Abstract: Chemotherapy-induced peripheral neuropathy (CIPN) is a common adverse effect, manifested by numbness, ticking and allodynia in 30-40% of cancer patients who have taken anticancer drugs chronically. Vincristine is a chemotherapeutic agent with activity against several of the most common cancers, including those of acute lymphocytic leukemia, acute myeloid leukemia, Hodgkin's disease, neuroblastoma and small cell lung cancer. Vincristine exerts its anti-tumor effects by binding to tubulin protein, stopping the cell from separating its chromosomes during metaphase; the cell then undergoes apoptosis. Although vincristine-induced CIPN is the most commonly reported neurotoxic and dose-limiting side effect, its mechanism remains to be fully defined. It is well understood that CIPN increases immune chemokines due to chemotherapy, and as a result, it induces inflammation and triggers pain hypersensitivity in Dorsal root ganglion (DRG). Here, we have shown that IL-23 pathway in DRG acts as an important role in vincristine-induced CIPN model. IL-23 has increased in vincristine-induced CIPN model, and when IL-23 receptor pathway was blocked, IL-23 has decreased. Especially when p19 and p40, the IL-23 subunit was neutralized using antibody, pain level has decreased only when p19 was inhibited. Moreover, when ruxolitinib, the inhibitor of JAK2, which is the IL-23 receptor down-modulation was treated, there was no significant change in the pain level. As a result, our data shows that IL-23 pathway is highly related to pain hypersensitivity in

vincristine-induced CIPN, and among the IL-23 receptors from the pathway, down-signal of IL-23R and TYk2 acts as an important role in vincristine-induced CIPN.

Disclosures: Y. Choi: None. **H. Lee:** None. **S. Jung:** None.

Poster

P398. Peripheral Mechanisms of Neuropathic Pain Including Chemotherapy Induced Peripheral Neuropathy

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P398.10

Topic: D.03. Somatosensation – Pain

Title: Chemokine receptor CXCR2 and its role in the pathway of CIPN in Dorsal Root Ganglion

Authors: *H. CHO, Y. CHOI, Y. HAN, S. JUNG;
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Abstract: Chemotherapy-induced peripheral neuropathy (CIPN) is a major drawback in the use of chemotherapeutic agents for patients with cancer. Even though diverse studies on this issue have identified a broad scale of molecules that might be related to CIPN, there has not yet been any differentiation of pathways to different chemotherapeutic agents. Since vincristine and oxaliplatin are some of the widely used chemotherapeutic agents, identifying the different pathways of these drugs and suggesting their possible treatment is crucial.
Mechanical allodynia results showed that when reparixin, the CXCR1/2 (C-X-C chemokine receptor type 1/2) blocker was treated with vincristine and oxaliplatin, only oxaliplatin-induced CIPN showed improvements. Furthermore, when imatinib, the abl-bcr inhibitor which is a critical pathway in the interleukin 23 signaling pathway was treated with, improvements were only observed in vincristine-induced CIPN. Real-time Polymerase Chain Reaction data showed that CXCR2 was increased in oxaliplatin-treated mice, but not in vincristine-treated mice, which further suggests the different pathways in vincristine and oxaliplatin.
Therefore, even though the physiological result of different chemotherapeutic agents is CIPN, since the pathway of causing CIPN between vincristine and oxaliplatin differs, it is important to pick the adequate treatment for each specific chemotherapeutic agent.

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Poster

P400. Mechanisms of Neuropathic Pain I

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Topic: D.03. Somatosensation – Pain

Support: TWU Research Enhancement Program Grant to DLH
TWU Experiential Student Scholar Grant to MPH
TWU Center for Student Research Grant to MPH

Title: Examination of neuronal arborization in the lumbar dorsal root entry zone as a potential mechanism of burn pain chronification

Authors: M. P. HUNTER, D. J. CANTU, T. P. OLAOLUWA, D. L. AVERITT, *D. L. HYNDS;
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Abstract: Burn victims experience severe pain that can become chronic and present as enhanced sensitivity to noxious (hyperalgesia) and non-noxious (allodynia) stimuli. Chronic burn pain development may involve neural plasticity, including reorganization of the central neural processes of the sensory neurons entering the spinal cord. We previously developed and characterized a rat model of full thickness thermal injury that displays persistent pain behaviors for up to 4 weeks post-injury. Thermal injury enhances the release of proinflammatory and pronociceptive (pro-pain) peptides in the spinal dorsal horn, but it is not known if sensory neuron afferent reorganization occurs and contributes to burn pain. We hypothesize that a full thickness thermal injury increases axonal sensory process arborization and synapsing in the rat spinal dorsal horn developing persistent burn pain behaviors. Adult female Sprague-Dawley rats (n=6/group; 200-300 g) received a sham injury (room temp. soldering iron tip) or a full thickness thermal injury to the plantar surface of the right hind paw (1% total body surface area) by applying a soldering iron tip at 100°C for 30 seconds. Thermal hyperalgesia and mechanical allodynia were tested at 24, 48 and 72 hours and once/week for 2 weeks. Rats were perfused with 4% paraformaldehyde and the lumbar spinal cords dorsal root ganglia (DRG) at L2-L6 were extracted. Immunohistochemistry and western blotting identified changes in markers of pain signaling, including the proinflammatory peptides calcitonin gene-related peptide (CGRP) and substance P, and the nociceptive transient receptor potential vanilloid 1 (TRPV1) ion channel. Sprouting axons were identified by nerve growth factor (NGF), beta III tubulin, and growth associated protein 43 (GAP43) labeling. Significant thermal hyperalgesia and mechanical allodynia developed at 24 hours and were maintained through 2 weeks post-injury. CGRP and substance P labeling increased in the lumbar dorsal horns and DRG and afferents displayed an increase in TRPV1 immunoreactivity in thermal-injured rats. Burn injured rats had increased beta III tubulin labeling in ipsilateral dorsal horns in processes that were also immunoreactive for CGRP. Together, these data indicate that thermal injury increases arborization of nociceptive sensory afferents in the lumbar dorsal horn. Our current studies are focused on further characterization of the sensory neuron populations and signaling pathways altered in the lumbar spinal cord that may contribute to burn pain chronification. Arborization may represent an alternative target to manage chronic pain development in burn patients.

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Poster

P400. Mechanisms of Neuropathic Pain I

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Topic: D.03. Somatosensation – Pain

Support: European Research Council (ERC) under the European Union's Horizon 2020 research and innovation program (FeelAgain grant agreement No. 759998)
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Innosuisse ICT program (n. 47462.1 IP-ICT)

Title: Ai-based decomposition of the multidimensionality of chronic pain

Authors: *N. GOZZI[1], G. PREATONI[1], F. CIOTTI[1], K. SCHALK[1], M. HUBLI[2], P. SCHWEINHARDT[2], S. RASPOPOVIC[1];
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Abstract: Chronic pain is a highly prevalent and debilitating condition which imposes an enormous burden on health care systems and society. Given its complexity, the underlying mechanisms and causes are still not fully understood. Indeed, pain is not the mere result of noxious stimuli traveling to the brain. Emotional, cognitive and behavioral components are closely interconnected with the noxious sensory experience. For this reason, it is extremely complicated to obtain an objective and reliable measure of the pain level perceived by patients. Today, the clinical standard are self-report scales. Unfortunately, they lack reliability and are biased due to their subjective nature. Advanced machine learning algorithms can be a solution, by predicting the pain experience from patterns in data obtained from patients. However, the majority of studies applying this strategy do not consider the emotional and cognitive state of the subjects, inevitably leading to the denial of this major component in the pain experience.The aim of work is to obtain a reliable and objective measurement of pain through machine learning algorithms applied on multidimensional objective and subjective data recorded in healthy and chronic pain patients. Subjects underwent experimentally induced pain through contact heat evoked potential (CHEP) and pinprick evoked potential (PEP) experiments. During the sessions, subjective (i.e., questionnaires on demographics, psychological health and coping with pain), objective (i.e., electroencephalography, skin conductance), semi-subjective (i.e., temporal summation, conditioned pain modulation, quantitative sensory testing) data has been collected. After a pre-processing step, different features have been extracted from the physiological signals. Machine Learning models were then applied to both subjective and objective features with two goals: i) classify the perceived pain level and ii) understand the contribution of each feature to

the prediction. Preliminary results confirm our hypothesis, by showing the contribution of both subjective and physiological factors to the prediction of pain level.Our framework holds promises to improve pain models. A new objective pain scale could satisfy the current search for a more precise measurement and biomarkers of chronic pain. A better understanding of the nature of the pain causes will help regulating the correct dosage of analgesics, and be a useful tool in fighting the increased opioid use. Furthermore, it would pave the way to personalized therapies accounting for inter-individual differences in pain perception, allowing for personalized control of therapies.

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Poster

P400. Mechanisms of Neuropathic Pain I

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Topic: D.03. Somatosensation – Pain

Support: NICHD R01HD083133-01A1

Title: Longitudinal MRI study of the somatosensory and motor systems in individuals with clinically confirmed neuropathic pain from ankle sprain

Authors: *A. M. S. MAALLO[1], A. KARAPANAGOU[5], J. MAR'I[1], N. LOPEZ[6], A. A. LEBEL[2,7], D. ZURAKOWSKI[3], R. BORRA[10,11,12], L. SIMONS[13], C. B. SIEBERG[4,1,8], D. BORSOOK[9], S. A. HOLMES[1,7];
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Abstract: Ankle sprain is common in the adolescent population, about 40% of whom may experience chronic pain following the injury. While the pain is primarily caused by damage to the peripheral nerves of the lower limbs, it is unclear how the same damage affects the central nervous system (CNS). Here, we systematically studied how neuropathic pain affects the CNS

and whether there are associations between CNS properties and participant's self-reported pain as measured with the painDETECT questionnaire (PDQ). To this end, we recruited patients with clinically confirmed neuropathic pain resulting from unilateral ankle sprain. We investigated the evolution of the white matter microstructure and the resting-state functional connectivity (rs-fc) of the somatosensory-motor systems. Participants were scanned twice (30 controls, 61 patients: 32 right + 29 left injury; time 1: 0-6 months, time 2: >6 months after injury) to obtain DWI and fMRI data (32-ch head coil, 3T Siemens Magnetom Trio). At time 1, compared to controls, right injury patients had lower fractional anisotropy (FA) in some segments of the left corticospinal tract (CST) only, while left injury patients had normal FA in the bilateral CST. Right injury patients had higher rs-fc of the left caudate to both the left pre- and post-central gyri, while left injury patients had lower rs-fc between the left thalamus and pre-central gyrus. At time 2, both the CST and rs-fc effects were no longer apparent in right injury patients, but there was higher axial diffusivity than controls, in the segment of the corpus callosum connecting the bilateral primary motor cortices. Also at time 2, left injury patients had lower FA than controls in some segments of the left CST only, and showed no rs-fc effects. Even though there was clinically confirmed neuropathic pain in patients, we found no associations between PDQ and the CNS properties that were different between groups. Consistent with the decussation of the limb's nerve projections to the contralateral hemisphere, we found that in right ankle injury patients only, there is an early but transient divergence from normative data in both white matter and rs-fc in the left hemisphere. However, there may be long-lasting changes to the structural connectivity of the bilateral motor cortices. On the other hand, in left ankle injury patients, there appears to be nascent structural changes in the ipsilateral (right) CST, long after sustaining the initial peripheral nerve damage. Our results demonstrate differential CNS changes concerning primarily the motor cortex after peripheral nerve injury, possibly suggesting differences in adaptive mechanisms in case of injury to either ankle.

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Poster

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Title: MIF antagonist ISO-1 prevents hypersensitivity and modulation of nociceptive synaptic transmission in a model of peripheral neuropathy

Authors: *M. PONTEARSO, P. ADAMEK, J. SLEPICKA, D. SPICAROVA, J. PALECEK; Lab. of Pain Res., Inst. of Physiology, Czech Acad. of Sci., Prague, Czech Republic

Abstract: There is an urgent need for new treatment options of chronic pain. Genetic deletion of macrophage migration inhibitory factor (MIF) prevents the development of hypersensitivity induced by peripheral nerve injury or inflammation. This proinflammatory cytokine MIF exerts its biologic functions mainly through binding to the putative membrane receptor, CD74. MIF tautomerase inhibitor (S,R)-3(4-hydroxyphenyl)- 4,5-dihydro-5-isoxazole acetic acid methyl ester (ISO-1) binds the MIF active site and blocks the MIF-CD74 binding. We have investigated the effect of ISO-1 treatment (20mg/kg i.p.) on mechanical hypersensitivity and excitatory and inhibitory synaptic transmission in the spinal cord dorsal horn in a model of neuropathic pain induced by chronic constriction injury (CCI) of sciatic nerve. Electronic Von Frey tests to assess mechanical hyperalgesia and specific optogenetic stimulation of inhibitory neurons combined with patch-clamp recording from neurons in acute mouse spinal cord slice were used. Spontaneous excitatory postsynaptic currents (sEPSCs) and light-evoked inhibitory postsynaptic currents (le-IPSCs) from superficial dorsal horn neurons were recorded. Electrophysiological experiments were performed in adult male transgenic mice VGAT-ChR2-eYFP line 8, that express an optimized channelrhodopsin specifically in GABAergic and glycinergic neurons. CCI in our experiments induced mechanical hyperalgesia, significantly increased the sEPSC frequency and decreased the amplitude of light-evoked IPSCs in excitatory dorsal horn neurons. These pathological effects of the CCI were largely diminished by the ISO-1 treatment. Our results suggest that blocking the MIF by ISO-1 inhibitor prevents the development of hypersensitivity after nerve injury. One of the underlying mechanisms of the ISO-1 treatment could be reduction of pathological macrophage modulation of excitatory and inhibitory synaptic transmission in the spinal cord dorsal horn.

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Poster

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Howard Hughes Medical Institute

Title: Spinal cord circuit dysfunction underlying over-reactivity to touch in neuropathic pain

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Abstract: In cases of nerve injury and ensuing neuropathic pain, normally innocuous tactile stimuli are often perceived as painful. This manifestation of neuropathic pain, known as mechanical allodynia, is often therapy-resistant and the responsible mechanisms for over-reactivity to tactile stimuli remain unclear. Fundamental work studying mechanical allodynia has proposed sites of dysfunction in both peripheral sensory neurons as well as within spinal cord circuits, however, how changes in either of these alters processing of somatosensory information in the intact animal is unknown. To this end, we are using large scale in vivo recordings and the SNI model of neuropathic pain to investigate how individual tuning properties and population dynamics of spinal cord interneurons and projection neurons, in both superficial and deep laminae of the dorsal horn, change in the allodynic state. Surprisingly, despite robust behavioral over-reactivity to mechanical stimuli in SNI mice, we do not observe an overall increase in sensitivity or reactivity of spinal cord interneurons. However, we do observe a decrease in sustained firing during 500ms step indentations of the skin and this change appears most prominent in spinal inhibitory interneurons, including the PV^+ inhibitory interneurons previously implicated in mechanical allodynia. Interestingly, we also observe a decrease in the synchrony of evoked firing in SNI mice across individual spinal cord interneurons and this deficit is recapitulated by specifically silencing the PV^+ interneurons. These findings suggest that decreased correlated activity across the dorsal horn may be a general feature of the neuropathic pain state and point to a circuit level dysfunction brought about by alterations in PV^+ interneurons. To ask whether changes in synchronous firing across neurons of the dorsal horn are uniquely caused by aberrant PV^+ interneurons or by disruption of feedback or feedforward inhibitory circuits in general, we are currently silencing other components of dorsal horn inhibitory pathways and monitoring evoked synchrony. These results will determine the contributions of dorsal horn inhibitory circuit motifs to alterations of spinal cord population dynamics associated with mechanical allodynia in chronic neuropathic pain states. This work is supported by NSF DGE1745303, NIH BRAIN Grant R01AT011447, and the Howard Hughes Medical Institute.

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Poster

P400. Mechanisms of Neuropathic Pain I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P400.06

Topic: D.03. Somatosensation – Pain

Support: NIDA R01 DA041809

Title: Astrocyte reaction in the lateral habenula contributes differently to spontaneous and evoked pain in paclitaxel induced peripheral neuropathy

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Abstract: The detrimental painful peripheral neuropathy induced by paclitaxel treatment (PIPN) significantly impacts the chemotherapy and the quality of life for cancer patients and may lead to the cessation of the chemotherapy and poor clinical outcomes. There is a knowledge gap in our understanding of the mechanisms underlying PIPN. The importance of glia pathophysiology, especially in terms of spinal reactive astrocytes, to the etiology of chronic pain has been investigated in multiple chronic pain models in recent several years; however, less is understood regarding their regulation at the supraspinal level. As an anti-reward center which is responsible for coding aversive emotional signals, the lateral habenula (LHb) has been extensively studied in stress related conditions such as depression and pain. However, whether reactive astrocytes in the LHb are responsible for pain signaling and how this process is regulated are not yet understood. In the current study, we investigated the astrocyte reaction in the LHb, evoked hypersensitivity (mechanical and heat hyperalgesia), ongoing spontaneous pain behaviors and comorbid anxiety-like behaviors in a mouse model of chronic PIPN. We demonstrated the presence of astrocyte reaction in the LHb, as indicated by the enhanced immunoreactivity of GFAP after 35 days in PIPN. The astrocyte reaction was diminished by the treatment with L-α-aminoadipic acid (L-α-AA), an astrocyte inhibitor. L-α-AA selectively attenuated the ongoing spontaneous pain and anxiety-like behaviors but did not alter the evoked hyperalgesia in PIPN, supporting the hypothesis that the reactive astrocytes have distinct contributions to the sensory and affective components of pain in PIPN.

Disclosures: X. Guo: None.

Poster

P400. Mechanisms of Neuropathic Pain I

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NIH Grant T32NS073548

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NIH Grant F99NS124190

Title: Spinal Neuropeptide Y1 Receptor-expressing Interneurons Represent a Pharmacotherapeutic Target for the Treatment of Neuropathic Pain

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Abstract: Neuropeptide Y (NPY) in the spinal cord dorsal horn exhibits long-lasting inhibitory control of nociceptive transmission after peripheral nerve injury (PNI). Dorsal horn neurons that express the neuropeptide Y1 receptor (Y1-INs), an inhibitory G protein-coupled receptor, are well positioned in the nervous system to mediate the anti-nociceptive actions of NPY. The aims of this study were to test if Y1-INs are both *necessary* and *sufficient* for the manifestation of PNI-induced mechanical and cold hypersensitivities via *in vivo* chemogenetic, optogenetic, and pharmacological approaches. Further we characterized dorsal horn Y1-INs via fluorescence *in situ* hybridization (FISH) and looked for conservation of gene expression in higher order species. Chemogenetic inhibition of Y1-INs with clozapine N-oxide did not alter baseline mechanical or cold sensitivity; however, following PNI, chemogenetic inhibition abolished mechanical and cold hypersensitivities ($p<0.05$, n=7 mice/group). We conclude that spinal cord dorsal horn Y1-INs are *necessary* for the full manifestation of neuropathic pain. Next, we found that chemogenetic activation of Y1-INs was *sufficient* to induce multiple signs of pain including spontaneous nocifensive behaviors (lifting of hindpaw, licking, guarding, biting) ($p<0.05$, n=3-5 mice/group) as well as heat, cold, and mechanical hypersensitivities ($p<0.05$, n=7-8 mice/group), and conditioned place aversion ($p<0.05$, n=7-8 mice/group). Similarly, *in vivo* optogenetic activation of spinal Y1-INs produced a frequency-dependent decrease in both mechanical and cold withdrawal thresholds ($p<0.05$, n=8 mice/group). Together, these results suggest that Y1-INs are a potential therapeutic target for neuropathic pain. To test this, we conducted PNI and then targeted Y1-INs with the Y1 receptor agonist, Leu^{31} Pro^{34}. We found that Leu^{31} Pro^{34} (0.1-10µg/5µL, i.t.) dose-dependently reduced mechanical and cold hypersensitivity ($p<0.05$, n=8-11); both were blocked by coadministration of the selective Y1 receptor antagonist, BIBO 3304 (10µg/5µL, i.t.). Additionally, FISH in mouse lumbar dorsal horn tissue revealed that Y1-INs segregate into three distinct populations that are demarcated by expression of *Grp* (60.61 ± 3.78%), *Cck* (16.60 ± 0.63%), or *Npff* (24.55 ± 1.75%). We found that these Y1-IN subpopulations are conserved in the porcine, macaque, and human spinal cord dorsal horn, altogether suggesting them to be promising pharmacotherapeutic targets for neuropathic pain.

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Poster

P400. Mechanisms of Neuropathic Pain I

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Topic: D.03. Somatosensation – Pain

Support: NIH Grant #1R01NS113189-01
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Title: C5a signaling in spinal mechanisms of chronic pain

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Abstract: The complement system is a principal component of innate immunity that is rapidly recruited through a cascade of enzymatic reactions to contribute to host defenses via diverse mechanisms. In spite of growing evidence implicating the complement system in various chronic pain states, the underlying mechanisms are not well understood. Recent studies suggest that neuropathic pain is associated with a robust upregulation of complement effectors in the spinal cord, which ultimately leads to production of a highly active complement product, C5a. Here we have elucidated C5a-dependent spinal mechanisms that contribute to the development of neuropathic pain induced in mice by spared nerve injury (SNI). First, we examined the effect of a potent and selective antagonist of the C5a receptor (C5aR1), PMX205 and found that its single i.t. administration produced a strong immediate analgesic effect. This analgesic effect strongly suggests that C5a/C5aR1 signaling plays an important role in the development and maintenance of neuropathic pain following SNI. Next, we generated a mouse expressing a fluorescent reporter of Ca^{2+} concentration, GCaMP5G-TdTomato, in microglia. Intact spinal cord was dissected from these mice, and used for testing the effect of complement C5a on microglia responses by multiphoton Ca^{2+} imaging. We found that C5a produced rapid transient increases of cytosolic Ca^{2+} concentration in spinal cord microglia that probably resulting in the release of mediators affecting neuronal activity. Therefore, we examined whether application of C5a mimics the effect of SNI producing an increased activity in projection or unidentified superficial dorsal horn neurons within the intact spinal cord. We have found that the number of action potentials evoked by the saturating dorsal root stimulation in about half of both neuronal populations was significantly increased after the C5a treatment. We have also observed an initiation of burst firing in some neurons and an increase in a frequency of spontaneous firing in a subpopulation of cells. Overall, these findings are consistent with the hypothesis that C5a/C5aR1 signaling and spinal cord microglia, as a primary target of C5a action, play an important role in the development and maintenance of neuropathic pain following SNI.

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Poster

P400. Mechanisms of Neuropathic Pain I

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Program #/Poster #: P400.09

Topic: D.03. Somatosensation – Pain

Support: R01DA046476

Title: Targeted manipulation of PFC 5HT2A receptors and KV7 channels attenuates chronic neuropathic pain in rats

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Abstract: Despite extensive research, chronic pain remains a disabling disease with limited therapeutic options. Whereas much is understood about pain transmission in the periphery, less is known about pain integration in the cortex. Preclinical studies associate increased pain perception with modifications in the structure and function of the prefrontal cortex (PFC). Unpublished studies from our laboratory indicate hyperexcitability in certain PFC neurons following neuropathic spared nerve injury (SNI). Other recent publications reported thatPFC pyramidal neurons innervating the pontine region of the brainstem are important for the descending regulation of pain and express both excitatory Gq-coupled 5HT2A receptors (5HT2AR) and its effector system, the inhibitory Kv7 ion channel. Although both 5HT2AR blockade or Kv7 channel activation is reported to produce antinociceptive effects in the periphery, this has not been demonstrated for cortical systems in a chronic pain model. To evaluate this possibility, we tested the *hypothesis* that local blockade of 5HT2AR or stabilization of Kv7 ion channels in the PFC would attenuate the mechanical allodynia associated with SNI. Adult male Sprague Dawley rats (P52) underwent surgery to receive either a unilateral SNI (n=18) or surgical controls (Sham; n=17). At 18-19d post-surgery, sham and SNI rats with confirmed allodynia received bilateral cannula targeting the prelimbic (A32V) region. At 21-32d, rats received counterbalanced (Latin square) bilateral infusion (0.5 uL) of vehicle (0.01 M phosphate buffer saline and 0.1 M HCL, pH adjusted to 5.8 using 0.1M NaOH), the 5HT2A antagonist M100907 (0.1 ug), the FDA approved Kv7 channel stabilizer retigabine (0.1 ug), or combined infusion of M100907 and retigabine. Relative to baseline measurements collected 15 min prior to testing, M100907, retigabine, or the combination robustly attenuated allodynia in

SNI rats at 3, 10 and 15 min after infusion. The resulting hypersensitivity thresholds in SNI rats measured using the up-down method resembled those in sham animals. By contrast, Sham-rats receiving similar drug injections showed no change in withdrawal thresholds. Baseline measurements in the SNI/Sham groups did not vary across testing days. Ongoing studies are evaluating the parallel changes in PFC mRNA and protein levels of 5HT2A and Kv7 channels using RNAscope, immunohistochemistry and proteomics. Taken together these results support a growing body of preclinical literature indicating that chronic neuropathic pain alters PFC function, warranting further investigation of the central PFC actions of 5HT2A antagonists and KV7 channel stabilizers in neuropathic pain.

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Poster

P400. Mechanisms of Neuropathic Pain I

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Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P400.10

Topic: D.03. Somatosensation – Pain

Support: NIH BRAIN Grant R01AT011447

Title: Functions of Tacr1- and Gpr83-expressing spinoparabrachial neurons and their neuropeptide signaling in acute and neuropathic pain

Authors: *S. CHOI[1], J. TURECEK[1], A. R. MAGEE[1], D. A. YARMOLINSKY[2], C. J. WOOLF[2], D. D. GINTY[1];
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Abstract: Pain is initiated by the activation of nociceptors that innervate the skin and internal organs. Nociceptive signals are propagated into the spinal cord and then transmitted to the brain by spinal cord projection neurons (PNs). These spinal PNs are attractive therapeutic targets for pain treatment because nociceptive signals emanating from the periphery are channeled through these spinal cord output neurons *en route* to the brain to produce pain sensations. Spinoparabrachial (SPB) neurons, a major population of spinal PNs that innervate the lateral parabrachial nucleus of the pons, represent an ideal neuronal population for developing new approaches to treat pain because they convey touch and pain information to higher brain centers that control the affective aspects (i.e., emotional “feelings”) of touch and pain. Previously, we showed that *Tacr1*$^+$ and *Gpr83*$^+$ SPB neurons form two largely-nonoverlapping subdivisions of the SPB pathway that cooperate to convey tactile, thermal and noxious signals from the spinal cord to the brain. To further define the contribution of each SPB subdivision to pain sensation and associated behavioral responses to noxious stimuli, we have begun to examine the effects of acute silencing of *Tacr1*$^+$ and *Gpr83*$^+$ SPB neurons, individually or simultaneously, on nocifensive behaviors using a newly generated dual recombinase-dependent SYP1-miniSOG mouse line that enables selective, light-dependent silencing of synaptic transmission. In addition, to determine if neuropeptide signaling mediated by either TACR1, GPR83 or both in the spinal cord is required for pain transmission, we also have begun to examine acute and neuropathic pain behaviors following spinal cord-specific deletions of the *Tacr1* and *Gpr83* genes using mouse lines that harbor conditional alleles of *Tacr1* and *Gpr83* in conjunction with spinal cord specific Cre lines. Collectively, these behavioral analyses will provide insights into the functions of *Tacr1*$^+$ and *Gpr83*$^+$ SPB neurons and neuropeptide signaling mediated by the TACR1 and GPR83 GPCRs in acute and neuropathic pain and may reveal novel therapeutic targets for treating pain.

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Poster

P439. Auditory Processing: Temporal, Frequency, and Spectral Processing III

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Program #/Poster #: P439.01

Topic: D.06. Auditory & Vestibular Systems

Support: F32DC016846

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National Science Foundation GFRP
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Title: Visual modulation of spectrotemporal receptive fields in mouse auditory cortex

Authors: *J. BIGELOW[1], R. J. MORRILL[1], T. R. OLSEN[1], S. N. BAZARINI[1], A. R. HASENSTAUB[2];
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Abstract: Recent studies have established significant anatomical and functional connections between visual areas and primary auditory cortex (A1), which may be important for perceptual processes such as communication and spatial perception. However, much remains unknown about the microcircuit structure of these interactions, including how visual context may affect different cell types across cortical layers, each with diverse responses to sound. The present study examined activity in putative excitatory and inhibitory neurons across cortical layers of A1 in awake male and female mice during auditory, visual, and audiovisual stimulation. We observed a subpopulation of A1 neurons responsive to visual stimuli alone, which were overwhelmingly found in the deep cortical layers and included both excitatory and inhibitory cells. Other neurons for which responses to sound were modulated by visual context were similarly excitatory or inhibitory but were less concentrated within the deepest cortical layers. Important distinctions in visual context sensitivity were observed among different spike rate and timing responses to sound. Spike rate responses were themselves heterogeneous, with stronger responses evoked by sound alone at stimulus onset, but greater sensitivity to visual context by sustained firing activity following transient onset responses. Minimal overlap was observed between units with visual-modulated firing rate responses and spectrotemporal receptive fields (STRFs) which are sensitive to both spike rate and timing changes. Together, our results suggest visual information carried by infragranular inputs influences sound encoding across cortical layers in A1, and that these influences independently impact qualitatively distinct responses to sound.

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Poster

P439. Auditory Processing: Temporal, Frequency, and Spectral Processing III

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Program #/Poster #: P439.02

Topic: D.06. Auditory & Vestibular Systems

Support: NIH Grant R01DC014101
NIH Grant R01NS116598
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Coleman Memorial Fund
Hearing Research Institute

Title: Audiovisual task switching rapidly modulates stimulus encoding and activity levels in mouse auditory cortex

Authors: *R. J. MORRILL, J. DEKLOE, J. BIGELOW, A. R. HASENSTAUB;
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Abstract: During everyday behavior, the sensory systems are in constant competition with each other for attentional resources. The brain must allocate attention to the sensory stream with the most behaviorally meaningful signals. How does modality-specific attention transform cortical sensory representations? To address this question, we created an auditory/visual rule-switching task for mice and made translaminar recordings in the auditory cortex during task performance. We show that attending to sound while ignoring visual stimuli modulates single-unit responses primarily through a reduction of stimulus-evoked and pre-stimulus activity in deep cortical cell populations. We then use a PSTH-based pattern classifier to show that the general reduction in activity does not impair decoding but instead increases mutual information encoding efficiency. Analysis of spectrotemporal receptive field nonlinearities shows that attending to sound switches auditory cortical encoding to a mode in which physical aspects of sound are more prominently represented. Finally, we show that task errors are predicted by activity levels in the same population of neurons that is modulated by modality-specific attention, suggesting that these neural effects are behaviorally meaningful correlates of task performance. Taken together, this work shows that attending to sound over vision quiets ongoing activity in the auditory cortex, shifting information transfer toward sound representation. Consistent with our previous research [Morrill & Hasenstaub J. Neurosci. 2018], this suggests that the deepest cortical layers serve as a hub for integrating extramodal contextual information.

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Poster

P439. Auditory Processing: Temporal, Frequency, and Spectral Processing III

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Program #/Poster #: P439.03

Topic: D.06. Auditory & Vestibular Systems

Support: Office of Naval Research Grant No. ONR N00014-17-1-2736

Title: Topographical distribution of spectrotemporal receptive field properties in bat primary auditory cortex

Authors: *K. BAKSHI[1], S. MACIAS[2], T. W. TROYER[3], M. SMOTHERMAN[1];
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Abstract: Spectrotemporal modulations are a prominent feature of natural sounds including animal vocalizations and human speech. The neural networks of the mammalian primary auditory cortex (A1) that encode and categorize communication sounds remain incompletely understood in part because of the heterogeneous distribution of interacting neuronal subtypes and microcircuits serving different functions throughout A1. To get around this problem, we investigated the topographical distribution of tuning properties throughout A1 in an auditory substrate homogenously specialized to process one specific acoustic feature, the downward frequency modulated (FM) sweep utilized in bat echolocation. While the majority of A1 neurons in bat auditory cortex are preferentially tuned to downward FM sweeps, the entire cortex is required to capture and categorize patterns of spectral details embedded within each biosonar echo. Here, we used linear, dual shank 32-channel translaminar microelectrode arrays to extract frequency response areas (FRAs) and spectrotemporal receptive fields (STRFs) from the A1 of adult male and female Mexican free-tailed bats (*Tadarida brasiliensis*) in response to broadband dynamic moving ripple (DMR) stimuli. We extracted a total of 170 STRFs across the A1 from spike-sorted single units (60) and multiunit activity (110) with most of our sampled STRFs at cortical depths corresponding to input layer 4. STRF best frequencies ranged between 10-55 kHz. We quantified the spectral and temporal modulation transfer functions (sMTF and tMTF, respectively) to determine neural tuning preferences in the spectral and temporal dimensions. The goal of this project is to compare STRF properties across cortical layers and throughout the tonotopic axis of A1 to reveal spatial patterns in the neuronal response properties that may in turn shed light on the functional organization of A1 neural networks processing downward FM sweeps. We found evidence of neuronal sub-categories that were classified as having simple or complex (multi-peaked) STRFs. For simple STRFs, sMTFs and tMTFs were evaluated as a function of best frequency and spatial location and revealed tonotopic trends similar to those reported for non-specialized animals.

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Poster

P439. Auditory Processing: Temporal, Frequency, and Spectral Processing III

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Topic: D.06. Auditory & Vestibular Systems

Support: Lundbeck Foundation Grant R139- 2012-12375
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Title: Sequential organization of critical periods in the mouse auditory system

Authors: *P. VALERIO, M. NAKAMURA, S. BHUMIKA, M. SOLYGA, T. R. BARKAT; Univ. of Basel, Basel, Switzerland

Abstract: Neuronal circuits are shaped by experience during time windows of increased plasticity in postnatal development. In the mouse auditory system, the critical period for simple sounds - pure frequency tones - is well defined from postnatal days 12 to 15 (Barkat et al., 2011). However, the critical period for frequency modulated sweeps (FMS), a more complex sound than pure tone, occurs weeks later, from postnatal days 31 to 38 (Bhumika et al., 2020). Whether such critical periods are timed by a temporally precise developmental program or sequentially organized, where the closing of one critical period triggers the opening of the next, was not yet known. Our work aimed to unravel the underlying neuronal mechanisms of critical periods in the auditory system, as well as their dependency on each other. We performed in vivo electrophysiological recordings and immunohistochemistry in the mouse primary auditory cortex in combination with molecular and sensory manipulators to elucidate the biological constraints on critical period timing. We demonstrated that passive exposure to FMS from postnatal days 31 to 38 leads to long-term changes in the temporal representation of sweep directions. Immunohistochemical analysis showed a decrease in parvalbumin (PV) expression in cortical layer 4 during this critical period, which was paralleled with a transient increase in responses to FMS. Despite the downregulation in layer 4 PV expression, the PV cell number was not altered. Continuous white noise exposure prevented PV expression decrease and delayed the FMS critical period onset, suggesting a reduction in inhibition as a mechanism for this plasticity. Passive sound exposure further revealed that the cortical representation of the two sound features - pure tone and FMS - are not influencing each other. Enhancing gamma-aminobutyric acid (GABA) function before the critical period for pure tones accelerated it without changing the critical period for FMS (Nakamura et al., 2020). Delaying the critical period for pure tones did also not affect FMS critical period. Together, these results demonstrate that, although sequentially organized, critical periods can be temporally shifted without affecting each other and are therefore timed by a precise developmental program. This work reveals an unexpected picture of the independence of sound features and their related developmental plasticity in the auditory cortex. Understanding the dependencies, the cortical presentations of sensory features, and their related developmental plasticity adds fundamental knowledge about how our central sensory system is built, organized, and functioning.

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Poster

P439. Auditory Processing: Temporal, Frequency, and Spectral Processing III

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Topic: D.06. Auditory & Vestibular Systems

Support: BioComp Research Initiative

Title: Neuronal population dynamics upon complex sound stimulation in the auditory cortex in an auditory processing-impaired mouse model

Authors: *S. L. WADLE[1], T. T. X. SCHMITT[1], J. ENGEL[2], S. KURT[2], J. J. HIRTZ[1]; [1]Physiol. of Neuronal Networks, Biol., TU Kaiserslautern, Kaiserslautern, Germany; [2]Biophysics & CIPMM, Saarland Univ., Homburg, Germany

Abstract: The extraction of specific features from a complex acoustic environment is essential for orientation, social interaction, and surviving in everyday life. To successfully accomplish this task, it is necessary that every station in the pathway involved functions properly. The α2δ3 auxiliary subunit of voltage-gated calcium channels (VGCCs) is expressed in spiral ganglion and principal neurons of the auditory pathway. Its loss leads to reduced VGCCs and structurally and functionally impaired endbulb of Held synapses of the auditory nerve despite normal hearing thresholds. These changes are accompanied by deficits on the behavioral level. Albeit being able to distinguish simple pure tones (PTs) from each other, α2δ3-KO mice fail to discriminate between amplitude modulated (AM) tones (Pirone et al. (2014) α2δ3 Is Essential for Normal Structure and Function of Auditory Nerve Synapses and Is a Novel Candidate for Auditory Processing Disorders. J Neurosci 34(2):434-445). We wanted to investigate how these changes in early stations of the auditory pathway affect neuronal networks in the auditory cortex (ACx). Therefore, we injected AAV-hSyn1-jGCaMP7f into the ACx of α2δ3-KO mice and littermate controls. After 13 days we performed two-photon calcium imaging of neurons in Layer 2/3 of the ACx in awake animals while presenting different sets of acoustic stimuli in a passive listening task in daily sessions over a time course of 2 weeks. Stimuli consisted of PTs, AM tones, single animal vocalizations and a complex acoustic environment, containing several animal vocalizations.

Correlation-based clustering of neuronal activity during single animal vocalizations revealed no difference between genotypes regarding the number of clusters formed, as well as the number of neurons per cluster. Interestingly, physical distances between cells in clusters in KO animals are larger compared to controls, indicating integration from a larger area, pointing to a less structured organization. In addition, activity in clusters in KO animals is less correlated than in controls, possibly indicating impairments in spectro-temporal processing. Overall, these results indicate altered processing of complex acoustic stimuli in the ACx of α2δ3-KO mice. Whether these differences are gradually increasing or decreasing with complexity of the stimulus, as well as if subfield-specific alterations are present, is currently under investigation.

Disclosures: S.L. Wadle: None. **T.T.X. Schmitt:** None. **J. Engel:** None. **S. Kurt:** None. **J.J. Hirtz:** None.

Poster

P439. Auditory Processing: Temporal, Frequency, and Spectral Processing III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P439.06

Topic: D.06. Auditory & Vestibular Systems

Support: R01 EB 028155

Title: Differential temporal modulation tuning in auditory responses between inhibitory and excitatory neurons in ferret auditory cortex.

Authors: *M. LOPEZ ESPEJO[1], S. V. DAVID[2];
[2]OHRC, [1]Oregon Hlth. and Sci. Univ., Portland, OR

Abstract: Research in the mouse auditory cortex has shown differences in sound tuning properties between excitatory and inhibitory neurons. This differential inhibitory tuning is thought to sharpen spectral and temporal tuning and to contribute to gain control in excitatory pyramidal neurons. However, the generality of differential inhibitory and excitatory tuning across species remains unexplored, particularly in the case of non-rodent species. To explore this issue we used the inhibitory interneuron-specific promoter mDlx to drive cell-type specific expression of channelrhodopsin (ChR2) in the auditory cortex of ferrets. We confirmed expression of ChR2 and colocalization with GABAergic neurons histologically. We then recorded the sound evoked activity of neuronal ensembles using a silicon multielectrode array with an integrated optic fiber. During recordings, we identified putative inhibitory interneurons by short-latency responses to a brief blue-laser stimulus. We characterized differences between ChR2+ and ChR2- neurons in spectrotemporal receptive fields (STRFs) and temporal integration properties. Preliminary results suggest that tuning differs between cell types mostly in temporal properties. Inhibitory, ChR2+ neurons respond with greater latency and duration than ChR2-. Frequency tuning, unlike in mice, showed no significant difference, but this result must be confirmed with a larger sample.

Disclosures: M. Lopez Espejo: None. **S.V. David:** None.

Poster

P439. Auditory Processing: Temporal, Frequency, and Spectral Processing III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P439.07

Topic: D.06. Auditory & Vestibular Systems

Support: NIH Grant DC018580
NIH Grant EY032230

Title: Overcoming deficits in the C57BL/6 mouse auditory spatial topography reveals that high-frequency hearing is required for frontal sound localization in the superior colliculus

Authors: *Y. SI[1], S. ITO[2], A. M. LITKE[2], D. FELDHEIM[1];
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Abstract: The ability to locate the source of a sound in a complex environment and determine its saliency is critical for survival. A topographic map of auditory space has been found in the superior colliculus (SC) of a range of species, including CBA/CaJ mice, in which high-frequency monaural spectral cues and interaural level differences are used to generate spatial receptive fields (RFs) along the azimuth. Unfortunately, C57BL/6 mice, a strain widely used for transgenic manipulation, display an age-dependent hearing loss due to an inbred mutation in the *CDH23* gene that affects cochlear hair-cell tip link stability. To overcome this problem, auditory researchers have been using young (around 3-month) C57BL/6 mice in their studies, an age that has shown to have normal hearing thresholds, despite the fact that important details of the phenotypes of the age-dependent hearing loss, such as spectral responses and spatial localization, have not been well characterized. Here, using blind analysis and large-scale in vivo physiological recordings of SC neurons in response to auditory stimuli, we show that 2-4-month C57BL/6 mice (n = 15, 3869 SC neurons recorded) of both sexes lack neurons with frontal auditory RFs and therefore do not have a topographic representation of auditory space in the SC. Analysis of the spectrotemporal receptive fields (STRFs) of the SC auditory neurons shows that C57BL/6 mouse SC neurons lack the ability to detect the high-frequency (>40kHz) spectral cues that are needed to compute frontal RFs. We also show that either crossing C57BL/6 with CBA/CaJ (n = 6, 1266 SC neurons recorded) or introducing one copy of the wildtype *CDH23* gene to the C57BL/6 mice (n = 7, 1348 SC neurons recorded) rescues both the high-frequency hearing deficit and the auditory map of space. Together, these findings 1) show that the computation of frontal RFs in the SC is correlated with the ability of SC neurons to respond to high-frequency sound, 2) should alert auditory researchers that C57BL/6 mice have significant high-frequency hearing deficits and consequently impaired auditory spatial topographic mapping in the SC even around 3 months of age, and 3) show that F1 progeny of C57BL/6 × CBA/CaJ mice or the introduction of the wild type *CDH23* gene in C57BL/6 mice restores the azimuthal auditory topographic map in the mouse SC.

Disclosures: Y. Si: None. **S. Ito:** None. **A.M. Litke:** None. **D. Feldheim:** None.

Poster

P439. Auditory Processing: Temporal, Frequency, and Spectral Processing III

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Topic: D.06. Auditory & Vestibular Systems

Support: NIDCD R01DC017516
Pew Biomedical Scholarship
Whitehall Foundation
Klingenstein-Simons Fellowship
Foundation of Hope

Title: Quantification of spatial variability in auditory cortical areas affecting stereotaxic targeting

Authors: *D. NARAYANAN, H. TSUKANO, A. M. KLINE, K. ONODERA, H. K. KATO; Univ. of North Carolina at Chapel Hill, Chapel Hill, NC

Abstract: A major goal of neuroscience is to understand the specific functions of individual brain regions. Stereotaxic coordinates based on brain atlases are widely used for targeting individual regions for experiments such as viral injections, imaging, and electrophysiology. However, in the mouse neocortex, due to the lack of cytoarchitecturally defined landmarks that delineate borders between functional areas, it remains unclear how variability across individual animals affects the precision of stereotaxic targeting. Moreover, in the auditory system, it is becoming evident that the coarse delineation of auditory cortices in the mouse atlas as “A1, AuD, and AuV” is insufficient to describe the true complexity of their functional organization. Therefore, to build a more solid understanding of the cortical area-specific functions, it is necessary to reevaluate the accuracy of area boundaries in brain atlases. Here, to evaluate the spatial variability of functionally identified auditory cortical areas across individual animals, we analyzed intrinsic imaging signal data from over two hundred mice. We quantified variabilities in both (1) inter-areal relative locations and (2) absolute stereotaxic areal locations, and examined the influences of various factors, including genotype, age, and sex. Using these two metrics, we will estimate the likelihood of failure in the stereotaxic targeting of auditory areas. Finally, we will compare the stereotaxic coordinates of “AuV” in the atlas with our targeted marking of the functionally identified secondary auditory cortex (A2) to directly quantify the deviations. Our preliminary data suggest that stereotaxic targeting is prone to inaccuracy due to the marked spatial variability across individuals, indicating the necessity of targeting based on functional mapping. Together, we hope our work sets a new standard and helps the field move forward in dissecting cortical circuits to identify the unique roles of individual auditory cortical areas.

Disclosures: D. Narayanan: None. **H. Tsukano:** None. **A.M. Kline:** None. **K. Onodera:** None. **H.K. Kato:** None.

Poster

P439. Auditory Processing: Temporal, Frequency, and Spectral Processing III

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Program #/Poster #: P439.09

Topic: D.06. Auditory & Vestibular Systems

Support: NINDS F31-NS111849
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NIDCD R01DC017516
Pew Biomedical Scholarship
Whitehall Foundation
Klingenstein-Simons Fellowship
Foundation of Hope

Title: Inhibitory gating of coincidence-dependent sensory binding in secondary auditory cortex

Authors: *A. M. KLINE, D. APONTE, H. TSUKANO, A. GIOVANNUCCI, H. K. KATO; Univ. of North Carolina At Chapel Hill, Chapel Hill, NC

Abstract: Integration of multi-frequency sounds into a unified perceptual object is critical for recognizing syllables in speech. This “feature binding” relies on the precise synchrony of each component’s onset timing, but little is known regarding its neural correlates. We find that multi-frequency sounds prevalent in vocalizations, specifically harmonics, preferentially activate the mouse secondary auditory cortex (A2), whose response deteriorates with shifts in component onset timings. The temporal window for harmonics integration in A2 was broadened by inactivation of somatostatin-expressing interneurons (SOM cells), but not parvalbumin-expressing interneurons (PV cells). Importantly, A2 has functionally connected subnetworks of neurons preferentially encoding harmonic over inharmonic sounds. These subnetworks are stable across days and exist prior to experimental harmonics exposure, suggesting their formation during development. Furthermore, A2 inactivation impairs performance in a discrimination task for coincident harmonics. Together, we propose A2 as a locus for multi-frequency integration, which may form the circuit basis for vocal processing.

Disclosures: A.M. Kline: None. **D. Aponte:** None. **H. Tsukano:** None. **A. Giovannucci:** None. **H.K. Kato:** None.

Poster

P439. Auditory Processing: Temporal, Frequency, and Spectral Processing III

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Support: Toyobo Biotechnology Foundation
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NIDCD R01DC017516
Pew Biomedical Scholarship
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Foundation of Hope

Title: Regulation of sensory processing in the primary auditory cortex by recurrent inputs from deep cortical layers

Authors: *K. ONODERA, H. K. KATO;
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Abstract: Information flow in the sensory cortex has been described as a predominantly feedforward sequence with deep layers as the output structure. Although recurrent excitatory projections from layer 5 (L5) to superficial L2/3 have been identified by anatomical and physiological studies, their functional impact on sensory processing remains unclear. Here, we use layer-selective optogenetic manipulations in the primary auditory cortex to demonstrate that feedback inputs from L5 suppress the activity of superficial layers, contrary to the prediction from their excitatory connectivity. This suppressive effect is predominantly mediated by translaminar circuitry through intratelencephalic (IT) neurons, with an additional contribution of subcortical projections by pyramidal tract (PT) neurons. Furthermore, L5 activation sharpened tone-evoked responses of superficial layers in both frequency and time domains, indicating its impact on cortical spectro-temporal integration. Together, our findings challenge the classical view of feedforward cortical circuitry and reveal a major contribution of inhibitory recurrence in shaping sensory representations.

Disclosures: K. Onodera: None. **H.K. Kato:** None.

Poster

P466. Visual System: Responses During Behavior I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P466.01

Topic: D.07. Vision

Support: Simons Collaboration on the Global Brain 542999

Title: Neuronal population representations within and across cortical areas during perceptual decision making

Authors: *A. I. JASPER[1], C. K. MACHENS[4], B. M. YU[5,6], A. KOHN[1,2,3];
[1]Dominick Purpura Dept. of Neurosci., [2]Ophthalmology and Visual Sci. Dept., [3]Systems and Computat. Biol. Dept., Albert Einstein Col. of Med., Bronx, NY; [4]Champalimaud Research, Champalimaud Ctr. for the Unknown, Lisbon, Portugal; [5]Electrical and Computer Engin. Dept., [6]Biomed. Engin. Dept., Carnegie Mellon Univ., Pittsburgh, PA

Abstract: Performing a perceptual task requires encoding sensory and decision signals in neuronal population responses and relaying these population signals between relevant brain areas. Numerous studies have investigated how sensory and decisions signals are encoded by single neurons. However, how the population representation of sensory and choice information relate to each other is poorly understood, and how these signals are relayed across brain areas on a trial-by-trial basis is nearly unexplored. To address these questions, we measured neuronal population activity simultaneously in early (primary visual cortex, V1) and midlevel (V4) visual cortex, while a macaque monkey performed a match-to-sample task. We implanted microelectrode arrays to sample ~50-100 neurons in each area, with retinotopically aligned receptive fields. We first compared the representation of stimulus and choice signals by asking how much information about each variable was available along the decoding axis of the other. We found that the representation of stimulus and decision signals were misaligned within both V1 and V4 populations: the decoding axis for one variable captured little of the available information about the other. These results appear inconsistent with proposals that decision signals reflect fluctuations in sensory neurons which drive decisions and proposals that decision signals reflect top-down beliefs about stimulus identity. Both proposals should result in aligned representations. We then used a dimensionality reduction approach—canonical correlation analysis—to understand how sensory and decision signals were relayed between areas. Consistent with previous work, we found that V1 and V4 population activity were related through a low-dimensional mapping-a communication subspace. Sensory information was robust in the communication subspace, in both areas. However, decision signals showed a marked asymmetry: nearly all decision-related activity in V1 was evident in the communication subspace, whereas in V4 much of the decision information was not. These results suggest a selective transmission of decision-related activity between visual areas. Together our findings suggest that decision signals in sensory cortex may have a different purpose than considered to date.

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Poster

P466. Visual System: Responses During Behavior I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P466.02

Topic: D.07. Vision

Support: NIH R01 Grant

Title: Interdigitating modules for visual processing during locomotion and rest in mouse V1

Authors: *A. M. S. MEIER, A. BURKHALTER;
Neurosci., Washington Univ. Sch. of Med., St. Louis, MO

Abstract: Sensory responses are not rigid representations of stimuli, but instead are modulated by behavioral context. A prominent example of this occurs in mouse primary visual cortex (V1), in which locomotion increases the gain of visual responses in excitatory neurons. This elevated responsiveness has been shown to increase the fidelity of stimulus representations across the neuronal population, enhance plasticity, improve visual object detection, and facilitate recovery of visual function after cortical damage. Multiple mechanisms driving this modulation have been described, including release of inhibition from somatostatin (SST) neurons and activation of long-range inputs from the midbrain locomotor region. The wide variability of locomotion modulation among V1 neurons raises the question of whether modulation is randomly distributed among cells, or is present in a discreet population distinguished by connectivity, location, or response tuning. Previously we showed that Layer 1 (L1) contains interdigitating modules with high or low expression of muscarinic acetylcholine receptor 2 (M2+ patches and M2- interpatches) [Ji et al. 2015]. L2/3 cells aligned with M2- interpatches are tuned for features of moving objects, including temporal frequency and motion coherence, suggesting that they may play a specialized role in processing the dynamic visual stimuli encountered during locomotion. In this study, using calcium imaging in locomoting mice, we found periodic clusters of neurons in L2/3 with high locomotion modulation which are aligned with M2- interpatches. Additionally, M2- interpatch-aligned cell pairs showed an increase in correlated trial-to-trial variability (noise correlations) at long distances across V1, suggesting a role in integrating stimuli across the visual field, possibly for detecting optic flow or large moving objects. Using viral tract tracing and immunostaining, we found that two network architectures may account for this population-specific locomotion modulation: increased expression of SST projections in M2- interpatches and preferential targeting by secondary motor cortex inputs to M2- interpatches. Finally, we elucidate a possible role of M2- interpatch cells in visual guidance of behavior by showing that somas and dendrites aligned with M2- interpatches project to primary motor cortex. These findings reveal a specialized role for M2- interpatch cells in integrating and exploiting stimuli from a wide visual field during locomotion.
Citation: Ji, W., Gămănuţ, R., Bista, P., D'Souza, R. D., Wang, Q. & Burkhalter, A. (2015). Modularity in the organization of mouse primary visual cortex. Neuron 87: 632-643

Disclosures: A.M.S. Meier: None. **A. Burkhalter:** None.

Poster

P466. Visual System: Responses During Behavior I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P466.03

Topic: D.07. Vision

Title: The lateral geniculate nucleus as a potential target for prosthetic stimulation in freely moving tree shrew

Authors: *J. WANG, H. AZIMI, M. KAESER, M. HARVEY, G. RAINER;
Dept. of Med., Univ. of Fribourg, Fribourg, Switzerland

Abstract: The visual thalamic lateral geniculate nucleus (LGN) is a potentially attractive target for visual prosthetics due to its laminar and retinotopic organization. Here we are interested in studying how visual stimulation and electrical or optogenetic activation of the LGN impact visually based behavior and neural circuit responses in the visual pathway. We chose the northern tree shrew *T. belangeri*, a diurnal mammal with a highly developed visual apparatus. In behavioral studies, we used an opaque globe containing a nose-poke at its center to deliver visual stimuli in an approximately retinotopic fashion using a projector in freely moving animals. Tree shrews readily learned to nose poke in these conditions for a juice reward in absence of food or water restriction, and performed up to 1250 trials per day. In a discrimination paradigm, animals had to discriminate 20 Hz target full field visual flicker from distractor flicker (e.g. 1Hz or 40Hz). We studied acquisition of various flicker discriminations, as well as capacity of tree shrews to generalize to previously untrained flicker frequencies. In a detection paradigm, tree shrews had to enter a nose-poke, and remain there until a visual stimulus, in this case a cloud of moving dots resembling a phosphene, was presented at a particular visual field location. Preliminary data suggests that tree shrews can quickly acquire this detection task, achieving good behavioral performance for stimuli of 2° diameter in size.We have validated expression AAV2-CaMKIIα-ChR2 constructs in tree shrew LGN, and observed robust axonal expression in layer 4 of primary visual cortex, the major thalamocortical LGN projection target. In anesthetized experiment functional validation, we have also demonstrated robust LFP and spiking activity entrainment within the LGN to blue light pulses delivered at various rates between 5 and 80 Hz. We have also performed functional validation for LGN electrical microsimulation, where we could demonstrate activity entrainment to stimulation pulses in LGN as well as V1. Our findings suggest that the tree shrew could be a useful animal model system to systematically study how electrical or optogenetic interventions in the LGN impact activations across the visual pathways.

Disclosures: J. Wang: None. **H. Azimi:** None. **M. Kaeser:** None. **M. Harvey:** None. **G. Rainer:** None.

Poster

P466. Visual System: Responses During Behavior I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P466.04

Topic: D.07. Vision

Support: ERC Grant MYSpace 948349

Title: Relativistic-like spatiotemporal distortions occur in young children at the time of saccades

Authors: *N. DOMENICI[1,2], A. TONELLI[1], M. GORI[1];
[1]Inst. Italiano di Tecnologia, Genova, Italy; [2]Dibris, Univ. degli Studi di Genova, Genova, Italy

Abstract: Adults perform more than 10,000 saccades per hour during wake, although most of them are unintentional. Interestingly, saccades produce big changes in visual perception, such as spatial mis-localization and temporal compression. Visual distortions within the peri-saccadic timeframe offered a powerful insight regarding how the human brain encodes space and time information. These effects have been explained considering receptive fields' predictive anticipatory remapping of neurons in both visual and parietal cortices. Even if incredibly fast, these remapping mechanisms are not instantaneous as they are constrained within the biophysical limit of neural communication. Nonetheless, it is reasonable to assume that the speed of remapping must be close to its limit, as predictive remapping must offset saccadic displacement. Because of this, spatial and temporal distortions observed during saccadic movement have been considered relativistic-like. In the current work, we investigated whether peri-saccadic spatiotemporal modulations also occur in children, whose maps in the brain might update at a different pace. To evaluate this possibility, we compared peri-saccadic spatial and temporal distortions between children (7-14 years old) and adults. Spatial modulations were evaluated asking participants to reproduce the position of a bar briefly presented before, during, or after the saccadic movement. Temporal distortions were instead evaluated asking participants to compare a post-saccadic fixed interval with a peri-saccadic interval, whose duration changes across trials. Regardless of their age, all the participants experienced the same level of spatiotemporal distortions, suggesting that the speed of remapping does not change between children and adults. In the end, our findings support the hypothesis that relativistic-like spatiotemporal compressions are an inherent feature of predictive remapping, and that such neural mechanisms have already reached functional maturity at 7 years of age.

Disclosures: N. Domenici: None. **A. Tonelli:** None. **M. Gori:** None.

Poster

P466. Visual System: Responses During Behavior I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P466.05

Topic: D.07. Vision

Support: Funded by CIHR grant MOP-119498 to C.B.

Title: Brain state effects on responses of early visual cortex neurons: A system identification approach.

Authors: *J. J. SOORIYAARACHCHI[1], C. ZHAN[2], C. L. BAKER[3];
[1]Physiol., McGill Univ., Montreal, QC, Canada; [2]Biomed. Engin., Southern Med. Univ., Guangzhou, China; [3]Ophthalmology, McGill Vision Res. Unit, Montreal, QC, Canada

Abstract: A visual cortex neuron's response depends not only on the characteristics of a visual stimulus in relation to its receptive field (RF), but also "brain state" fluctuations which cause substantial trial-to-trial variability. Brain state effects on sensory coding are poorly understood, potentially compromising RF estimation. Here we analyzed the interaction between a neuron's spiking response and brain state inferred from local field potentials (LFPs) to obtain better quantitative estimates of neuronal RFs. We used extracellular recordings from the primary visual cortex of anesthetized, paralyzed cats recorded with 32-channel NeuroNexus probes (polytrodes and linear arrays) which responded to rapid sequences of natural images (Talebi and Baker, 2012). Low-pass filtering (< 100Hz) was employed to extract LFPs from the raw recorded signals, and single units were isolated using SpikeSorter software (Swindale & Spacek, 2015). Phase amplitude coupling (PAC) provided an indication of the relationship between spiking responses and different frequency bands of the LFP signal. We then employed a system identification approach using machine learning, with a model architecture incorporating an LFP-driven filtering pathway in parallel with a stimulus-driven RF pathway, followed by a simple output nonlinearity. The model parameters were estimated with an iterative regression algorithm with L2 regularization, using TensorFlow and Keras on a training dataset, along with a validation dataset for regularization. The variance accounted for (VAF) was used to evaluate the model's predictive performance on a hold-back test dataset. Results from the PAC analysis showed that the spiking responses of neurons are strongly linked to various frequency band oscillations as well as the envelope of high frequency gamma oscillations (30-100 Hz) of the LFP signals. The system identification revealed the relevance of similar LFP frequency bands. However, different neurons recorded simultaneously showed significant relationships with different frequency ranges of the LFP (e.g. delta, beta), and varying degrees of contribution from the gamma envelope. Incorporating brain state effects provided significant improvements in VAF measures of predictive performance, in spite of the additional model parameters. These findings highlight the importance of incorporating the trial-to-trial variability due to brain state variations in studies of sensory coding at the single neuron level and indicate that the nature of brain state variations may vary substantially across different neurons.

Disclosures: J.J. Sooriyaarachchi: None. **C. Zhan:** None. **C.L. Baker:** None.

Poster

P466. Visual System: Responses During Behavior I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P466.06

Topic: D.07. Vision

Support: NIH Grant 1UF1NS116377-01
AFOSR Grant 19RT0316

Title: Does running modulate visual responses in primate V1?

Authors: *J. LISKA[1], J. L. YATES[2], J.-O. MUTHMANN[1], A. HUK[1];
[1]Ctr. for Perceptual Systems, Univ. of Texas at Austin, Austin, TX; [2]Univ. of Rochester, Rochester, NY

Abstract: Although activity in mouse visual cortex is strongly modulated by running, it is unknown whether similar occurs in primate V1. In fact, attentional and other-top down modulations are notoriously small and difficult to elicit in single V1 neurons in monkeys. To test whether insights from rodents translate to primates, we measured activity in V1 of marmoset monkeys when they were running and stationary. Because marmosets are a largely arboreal species, we provided them an "infinite branch" on which they can comfortably remain stationary or freely locomote. We recorded single- and multi-unit activity in foveolar V1 using chronically-implanted 64-channel 3D "N-form" electrode arrays, while presenting sinusoidal gratings of varying orientation, spatial frequency, and direction. We measured gaze position and also mapped receptive fields using reverse correlation. Both marmosets naturally alternated between running and stationary periods running ~20% of the time. Running epochs were defined as periods of forward movement at a speed greater than 3 cm/s; mean running speed was 19 and 14 cm/s for the two monkeys, respectively. We have recorded 415 unique units in 24 sessions in one marmoset and 42 units from 1 session in a second marmoset. To test for the presence of running modulations, we resampled trial-balanced firing rates both before stimulus onset and during the presentation of drifting sine wave gratings. We measured direction tuning curves for each behavioral state (running vs. stationary) and compared the difference in tuning curve peaks to a surrogate "null" distribution of differences generated by resampling after artificially shuffling the behavioral state. We then evaluated the fraction of units with significant modulation. We observed no net or unified population-level effects during running; however, 5% of units increased baseline firing rate, 9% decreased baseline firing rate, 9% increased stimulus-evoked firing rate, and 9% decreased stimulus-evoked firing rate (false positive rate = 5%). While such effects are not comparable to the large modulations observed in rodents, recent work has shown running modulations to be highly cell-type specific in rodents, and we have only begun to categorize the small subset of cells that show possible statistically-significant running

modulations in our dataset. Further analysis of the properties of the modulated cells is required for more definitive conclusions about the existence and nature of locomotion effects in primate V1.

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Poster

P475. Interactions Between the Visual Cortex and the Thalamus

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P475.01

Topic: D.07. Vision

Support: NIH F31 EY028853
NIH R01 MH063912
NIH R01 EY022577

Title: Distinct "driving" versus "modulatory" influences of different visual corticothalamic pathways

Authors: *M. A. KIRCHGESSNER[1,2,3], A. D. FRANKLIN[1,4], E. M. CALLAWAY[1,2]; [1]Salk Inst., La Jolla, CA; [2]Neurosciences Grad. Program, Univ. of California San Diego, La Jolla, CA; [3]New York Univ. Sch. of Med., New York, NY; [4]Sainsbury Wellcome Ctr., London, United Kingdom

Abstract: Higher-order (HO) thalamic nuclei, such as the pulvinar in the visual system, interact extensively with the cerebral cortex in ways that are thought to be important for sensation and perception, attention, and many other important brain functions. However, a complete mechanistic understanding of these corticothalamic interactions - especially those involving the HO thalamus - is lacking. One notable organizational feature of corticothalamic connectivity that is shared across sensory and even non-sensory systems is that HO thalamic nuclei (including the pulvinar) are innervated by two distinct excitatory corticothalamic (CT) populations in layers 5 and 6. The functional significance of having these two different CT pathways to the HO thalamus is not known but has generated significant interest and speculation. One prevailing idea in the field is that layer 5 (L5) projections may be the primary “drivers” of functional response properties in their HO thalamic targets, whereas layer 6 (L6) projections act instead as “modulators”. However, whether distinct CT projections exert different functional influences (e.g., “driving” versus “modulatory”) on the HO thalamus *in vivo* has never been directly tested. To address this question, we utilized different transgenic mouse lines and viral approaches to selectively express inhibitory opsins in L6 and/or L5 CT neurons of the primary visual cortex (V1) and recorded single unit activity from V1 and the pulvinar (also known as LP) in awake mice. We additionally recorded from the first-order (FO) visual thalamic nucleus, the

dorsolateral geniculate nucleus (dLGN), which also receives L6CT "modulatory" input but is not innervated by L5. By optogenetically inactivating one or the other CT population in V1, we demonstrate that L5, but not L6, CT projections drive visual responses in the HO visual pulvinar, even while both pathways provide retinotopic, baseline excitation to their thalamic targets. Dual optogenetics experiments confirmed that individual pulvinar neurons are "driven" by V1 L5 but not by L6. We also explored the functional influence of the superior colliculus (SC) on a region of the pulvinar that receives both V1 and SC input. Individual pulvinar neurons could be driven by the SC, by V1 L5, or even by both, suggesting that different "driving" inputs can converge onto individual cells. Altogether, these results reveal how different cortical (and subcortical) projections play distinct roles in shaping activity and sensory response properties in the HO visual thalamus.

Disclosures: M.A. Kirchgessner: None. **A.D. Franklin:** None. **E.M. Callaway:** None.

Poster

P475. Interactions Between the Visual Cortex and the Thalamus

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P475.02

Topic: D.07. Vision

Support: EY022577
EY029932-02
R01EY031517-01

Title: Functional and anatomical characterization of parallel spatiotemporal frequency channels in the mouse retinogeniculocortical pathway.

Authors: *H. WANG, O. DEY, E. M. CALLAWAY, B. STAFFORD;
Salk Inst., La Jolla, CA

Abstract: Primate retinal ganglion cells (RGCs) form anatomically and functionally segregated parallel pathways that project to the primary visual thalamus (LGN) and form distinct output channels to different layers of primary visual cortex (V1) (Nassi 2009). Whether a similar organization exists in the mouse is not well understood. Evidence suggests that a pathway originating from direction-selective RGCs transmits direction information to layer 1 of V1 via the LGN "shell" (Cruz-Martín 2014). Whether other RGCs types, like those tuned to different spatial frequencies (SFs) and temporal frequencies (TFs), that project to the LGN "core" form parallel channels to layer 4 of V1 in mice is unknown. We used two-photon calcium imaging to characterize layer 4 neurons labeled by two transgenic mouse lines (Nr5a1-Cre and Scnn1a-Tg3-Cre) and found that they are tuned to different SFs, TFs, and speeds. Mice were injected with AAV-FLEX-GCaMP6s in V1 and visual responses to drifting sine wave gratings and coherent

dot motion stimuli were imaged to reveal preferred SF, TF, and speed. We found significant differences between Nr5a1 (n=121) and Scnn1a-Tg3 (n=231) neurons in preferred SF, TF, and speed tuning. Nr5a1 neurons preferred lower SFs (median = 0.02 cpd vs. 0.04 cpd, p = 4.8e-9), and higher TFs (median = 3.1 cps vs. 0.9 cps, p = 1.4e-29) than Scnn1a-Tg3 neurons. Among speed tuned neurons, Nr5a1 neurons preferred faster speeds than Scnn1a-Tg3 neurons (median = 128 deg/s vs. 21.1 deg/s, p = 7.5e-11). Next, we labeled RGCs that provide input (via LGN) to Nr5a1 or Scnn1a-Tg3 neurons using transsynaptic rabies virus tracing. We first injected Cre-dependent AAVs expressing optimized rabies glycoprotein (AAV-FLEX-oG) and TVA (AAV-FLEX-GFP-TVA) into V1 of Nr5a1-Cre and Scnn1a-Tg3-Cre mice, and a non-Cre dependent AAV expressing oG into LGN. 21 days later, G-deleted rabies virus (EnvA-RVdG-mCherry) was injected into V1. 7 days later, mice were perfused, and brains and retinas harvested. Retinas were stained for anti-RFP and anti-ChAT. RGC type was characterized based on the stratification pattern of their dendrites relative to the ChAT bands in confocal z-stacks. The results of this study are currently being analyzed; we expect differences in RGC dendrite stratification based on RGC types that respond to different SFs, TFs, or speeds.

Disclosures: H. Wang: None. **O. Dey:** None. **E.M. Callaway:** None. **B. Stafford:** None.

Poster

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Support: NIH NEI EY025219 to F.B
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Title: Morphological evidence for multiple distinct channels of corticogeniculate feedback originating in mid-level extrastriate visual areas of macaque monkeys and ferrets

Authors: *M. ADUSEI[1], A. J. MURPHY[1], J. M. HASSE[2], E. M. CALLAWAY[3], W. M. USREY[4], F. BRIGGS[1];
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Abstract: In the visual system, retinal information reaching the lateral geniculate nucleus (LGN) of the thalamus is mainly relayed to the primary visual cortex (V1). Geniculocortical projection neurons in the LGN include magnocellular, parvocellular, and koniocellular cells that convey distinct, stream-specific visual information. While the majority of geniculocortical neurons project to V1, there are also sparse geniculocortical projections to extrastriate visual cortical

areas that are thought to originate primarily from koniocellular cells. Importantly, feedforward geniculocortical projections to V1 are complemented by reciprocal, feedback corticogeniculate projections. In macaque monkeys and ferrets, multiple morphologically distinct corticogeniculate subtypes exist in V1, and also in V2. Here we investigated whether corticogeniculate neurons are present in extrastriate visual cortex and whether these neurons are morphologically homogeneous, i.e., aligned with primarily koniocellular geniculocortical inputs, or diverse. We identified and characterized the morphology of corticogeniculate neurons in three macaque extrastriate visual cortical areas: V4, MT, and MST, and their ferret homologs: area 21a, PMLS, and PLLS respectively. We discovered morphologically diverse corticogeniculate subtypes in all three extrastriate areas across the two species. Most extrastriate corticogeniculate neurons were morphologically similar to those previously identified in V1 and V2. Whether unique corticogeniculate subtypes send stream-specific information to the LGN is not known, but prior physiological evidence based on axon conduction latencies and visual responses properties suggests that distinct corticogeniculate subtypes align with the feedforward parallel processing streams. Together, our findings suggest that V1-independent reciprocal connectivity between the LGN and extrastriate visual cortex, in ferrets and macaques, involves multiple diverse corticogeniculate cell types possibly relaying functionally distinct visual signals.

Disclosures: M. Adusei: None. **A.J. Murphy:** None. **J.M. Hasse:** None. **E.M. Callaway:** None. **W.M. Usrey:** None. **F. Briggs:** None.

Poster

P475. Interactions Between the Visual Cortex and the Thalamus

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Topic: D.07. Vision

Support: NIH R01NS104949
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Title: Motion/direction-sensitive thalamic neurons project extensively to the middle layers of primary visual cortex

Authors: *J. ZHUANG, Y. WANG, N. OUELLETTE, E. TURSCHAK, R. LARSEN, K. TAKASAKI, T. DAIGLE, B. TASIC, J. WATERS, H. ZENG, R. C. REID;
Allen Inst. For Brain Sci., Seattle, WA

Abstract: The motion/direction-sensitive and location-sensitive neurons are two major functional types in mouse visual thalamus that project to the primary visual cortex (V1). It has been proposed that the motion/direction-sensitive neurons mainly target the superficial layers in V1, in contrast to the location-sensitive neurons which mainly target the middle layers. Here, by

imaging calcium activities of motion/direction-sensitive and location-sensitive axons in V1, we find no evidence for these cell-type specific laminar biases at population level. Furthermore, using a novel approach to reconstruct single-axon structures with identified *in vivo* response types, we show that, at single-axon level, the motion/direction-sensitive axons have middle layer preferences and project more densely to the middle layers than the location-sensitive axons. Overall, our results demonstrate that Motion/direction-sensitive thalamic neurons project extensively to the middle layers of V1, challenging the current view of the thalamocortical organizations in the mouse visual system.

Disclosures: J. Zhuang: None. **Y. Wang:** None. **N. Ouellette:** None. **E. Turschak:** None. **R. Larsen:** None. **K. Takasaki:** None. **T. Daigle:** None. **B. Tasic:** None. **J. Waters:** None. **H. Zeng:** None. **R.C. Reid:** None.

Poster

P475. Interactions Between the Visual Cortex and the Thalamus

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Univ. of Chicago Pritzker Fellowship

Title: Diverging properties of trans-thalamic feed-forward and feed-back pathways in the mouse brain

Authors: *A. J. MILLER-HANSEN[1], S. SHERMAN[2];
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Abstract: Higher-order (HO) thalamic nuclei contribute to sensory processing via projections to primary and higher cortical areas, but it is unknown which of their cortical and sub-cortical inputs contribute to their distinct output pathways. We used sub-population specific viral strategies in mice to anatomically and physiologically dissect the inputs and outputs of the HO thalamic nuclei of the somatosensory and visual systems (POm and Pul respectively). Employing a complementary optogenetics and electrical stimulation strategy, we show that HO thalamic synapses in cortex have functionally divergent properties in primary (S1 and V1) vs. higher cortical areas (S2 and Higher Visual Areas or HVAs). HO thalamic projections onto excitatory targets in S1 and V1 were weakly modulatory while projections to S2 and HVAs were drivers of post-synaptic targets.
Then, using G-deleted rabies tracing verified by optogenetics to map inputs to HO thalamus, we

show that HO thalamocortical cells in POm projecting to S1 are driven by layer 5 of S1, S2, and M1, and that HO thalamocortical cells in Pul projecting to V1 are driven by layer 5 of V1 and HVAs. Therefore, in both sensory systems, layer 5 of primary and secondary sensory cortex drive trans-thalamic feedback modulation of primary sensory cortex through HO thalamus. These results highlight conserved organizational principles that may be shared by other HO thalamic circuitry, and they support the hypothesis that all direct cortico-cortical projections in the brain are paralleled by cortico-thalamo-cortical or trans-thalamic pathways, even in the feed-back direction. They also suggest that feedforward trans-thalamic pathways act as drivers well suited to information-bearing, while feed-back through HO thalamus is modulatory, as has been hypothesized for cortico-cortical projections.

Disclosures: A.J. Miller-Hansen: None. **S. Sherman:** None.

Poster

P475. Interactions Between the Visual Cortex and the Thalamus

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Topic: D.07. Vision

Support: NIH grant EY013588
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NIH Grant EY015387

Title: Center-surround spatial organization of corticogeniculate feedback

Authors: *A. N. SANCHEZ, H. J. ALITTO, W. M. USREY;
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Abstract: Neurons in the lateral geniculate nucleus (LGN) process and relay visual signals from the retina to primary visual cortex (V1). V1, in turn, gives rise to feedback pathways terminating in the LGN. Although much is known about the anatomical organization of corticogeniculate (CG) feedback, relatively less is known about its role during sensory processing. Because CG feedback can provide monosynaptic excitation and disynaptic inhibition to LGN neurons, the effects from CG feedback may be complex. We have previously shown that the filtering of retinal spike trains by the LGN depends upon both stimulus size and contrast. Specifically, the likelihood that a retinal spike is transmitted to the cortex is reduced by both large stimulus sizes (e.g., extraclassical suppression) and high stimulus contrasts (e.g., contrast gain control). Thus, current work is focused on understanding how CG feedback may contribute to these two processes in the LGN. To investigate the influence of CG feedback on visual processing in the LGN of awake primates, we infected V1 (n = 2 macaque monkeys) with either Jaws or DLX-channelrhodopsin and reversibly silenced CG neurons. To remove both the monosynaptic

excitation and disynaptic inhibition from V1 onto the LGN, we inactivated retinotopically defined regions of V1 while recording from individual parvocellular LGN neurons (median retinotopic displacement 1.5^{o}, 52 off-center cells, 25 on-center cells). Our results demonstrate that CG feedback has a functional center-surround organization that differentially modulates LGN classical and extraclassical mechanisms. Importantly, the polarity of the organization is dependent upon the presence of extraclassical suppression. When extraclassical suppression is present (a feature most common in off-center cells), CG feedback amplifies responses to stimuli that are restricted to the LGN neuron's classical receptive field and suppresses responses to stimuli that extend into the extraclassical surround. By contrast, the polarity of excitation and suppression from CG feedback is reversed when LGN neurons exhibit surround facilitation (a feature most common in on-center cells). In this case, CG feedback suppresses responses to stimuli restricted to the classical receptive field and facilitates responses to stimuli that extend into the extraclassical surround. Ongoing experiments are investigating how stimulus contrast influences this center-surround organization of cortical feedback. These results require revision to our current view of corticothalamic circuitry and have important implications for understanding the dynamics of thalamocortical interactions.

Disclosures: A.N. Sanchez: None. **H.J. Alitto:** None. **W.M. Usrey:** None.

Poster

P475. Interactions Between the Visual Cortex and the Thalamus

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Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P475.07

Topic: D.07. Vision

Support: CIHR

Title: Cortico-thalamic projections gate alpha rhythms in the pulvinar

Authors: *N. CORTES[1], L. IKAN[1], R. ABBAS FARISHTA[2], C. F. CASANOVA[1];
[2]Sch. of optometry, [1]Univ. De Montréal, Montreal, QC, Canada

Abstract: Two types of cortico-thalamic terminals reach the pulvinar nucleus of the thalamus, and their distribution varies according to the hierarchical location of the projecting cortical neurons. While type 2 terminals are more abundant at lower hierarchical levels, higher cortical areas mostly exhibit type 1 axons. Terminal types 1 and 2 evoke an excitatory postsynaptic potential (EPSP) that is facilitated (short-term facilitation, STF) or depressed (short-term depression, STD) over time, respectively. These observations yield fundamental questions about the role of these different terminal types along the cortical hierarchy. Our theoretical results support that the co-action of the two types of terminals produces different oscillatory rhythms in pulvinar neurons. More precisely, at a specific range of connectivity weights, terminals type 1

and 2 produces alpha-band oscillations. Interestingly, when the two types of terminals are empirically arranged as the projections of areas 17 and 21a into the pulvinar, the actions of these cortico-thalamic connections are antagonistic. When area 17 (21a) generates low-band oscillatory activity, cortical area 21a (17) shifts pulvinar responses to stable asynchronous spiking activity. To further investigate such oscillatory effects along the transthalamic pathway, we created a cortical feedforward network of two cortical areas with cortico-thalamic connections to a pulvinar-like network. This model simulated the interaction between cortical areas 17 and 21a with the pulvinar. Here, the transthalamic pathway allows the propagation of alpha waves from the pulvinar to area 21a. This oscillatory transfer is ceased when reciprocal connections from area 21a reach the pulvinar, so functionally closing the cortico-thalamic loop. The pulvinar shows a bi-stable spiking activity, oscillatory or irregular asynchronous spiking, whose responses are gated by the different activation of cortico-pulvinar projections from areas 17 and 21a. Preliminary experimental results are shown to reinforce such theoretical findings.

Disclosures: N. Cortes: None. **L. Ikan:** None. **R. Abbas Farishta:** None. **C.F. Casanova:** None.

Poster

P475. Interactions Between the Visual Cortex and the Thalamus

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Topic: D.07. Vision

Support: NIH NS109978
NIH NS107968

Title: Narrowband gamma oscillations propagate throughout the mouse thalamocortical visual system

Authors: *D. SHIN, J. DEL ROSARIO, K. PEELMAN, B. HAIDER;
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Abstract: Rhythmic neural activity in the “gamma” range (30 – 80 Hz) is thought to play an important role in communication between brain areas. Recent studies in awake mice show that strong narrowband gamma oscillations (NBG; 50-70Hz) in mouse primary visual cortex (V1) arise from the lateral geniculate nucleus (LGN; Niell and Stryker, 2010; Saleem et al., 2017). The strength of NBG in V1 varies with visual stimuli, but also with behavioral state and task performance (Speed et al., 2019), suggesting NBG may contribute to perception. It remains unknown if NBG activity spreads to higher visual areas (HVAs), and whether its timing is conserved across LGN, V1, and HVAs. Answering these questions requires large-scale simultaneous neural recordings from LGN, V1, and HVAs, and a framework for detecting and

quantifying population and single-neuron level NBG activity.
Here we analyzed the Allen Institute's Visual Coding dataset of simultaneous multi-site Neuropixels recording across LGN, V1, and HVAs in awake mice (33 sessions). We analyzed population signals (local field potential, LFP), single neuron spiking, and pairwise neuronal interactions to assess the strength and timing of NBG activity throughout the mouse visual system. We established a framework of identifying neurons with significant NBG spiking activity (NBG cells) using both auto-correlogram (ACG) and cross-correlogram (CCG) analysis. We performed several control measures to ensure that the identification of NBG cells was unlikely to be artefactual
We found strong evidence for NBG activity simultaneously across LGN, V1, and HVAs. First, we observed significant NBG LFP power in all recorded HVAs. Second, we found significant NBG cells in these same recordings in all HVAs (57 cells, 15 sessions). Third, we found significant pairwise correlated spiking between NBG cells in LGN ,V1 and HVAs. Many pairs showed significant CCG peaks then troughs at short latencies (< 4.5ms), suggesting direct functional interactions (Senzai et al., 2019, Siegle et al., 2021). Remarkably, the likelihood of significant pairwise interactions between pairs of NBG cells in V1 (4.72%, n = 126 pairs), was significantly higher than among all other V1 cells (1.1%, n = 2974 pairs) during the same recordings ($p<1e-8$). These effects suggest that NBG activity propagates through a preferred subnetwork of neurons across LGN, V1, and HVAs. These findings identify a substrate for rapid NBG communication across multiple visual areas in awake mice.

Disclosures: D. Shin: None. **J. Del Rosario:** None. **K. Peelman:** None. **B. Haider:** None.

Poster

P475. Interactions Between the Visual Cortex and the Thalamus

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Program #/Poster #: P475.09

Topic: D.07. Vision

Title: Pulvinar modulates cortical visual activityby increasing the gamma band contrast response.

Authors: *L. IKAN, N. CORTES, C. F. CASANOVA;
Ecole d'optométrie, Univ. de Montréal, Montreal, QC, Canada

Abstract: The pulvinar has a widespread connectivity with the visual cortex. A recent study from our group has described the impact of silencing the pulvinar on the contrast response function (CRF) of neurons in two distinct hierarchical cortical areas of the cat visual cortex, namely areas 17 and 21a (de Souza et al., 2019). To study whether oscillatory cortical signals depend upon the pulvinar activity, we studied the contrast response of the gamma- and alpha-band oscillatory responses in areas 17 and 21a during thalamic inactivation. Our data showed

that the pulvinar inactivation altered the response gain in both areas, but with more significant changes observed in area 21a. These findings were based on the linear change of the CRF position (driver effect) and the nonlinear variation in the CRF dynamic range (Rmax; modulatory effect). Extracellular responses to full-field gratings of varying contrast were recorded in both cortical areas in anesthetized cats using linear probes before, during and after the GABA inactivation of pulvinar. Local field potentials (LFPs) from low-pass filtering of raw recordings were analyzed with Wavelet to assess oscillatory gamma waves. During inactivation, gamma frequency increases as a function of increased visual contrast in area 21a. In area 17, there was no significant changes across cortical layers after pulvinar inactivation, whereas in area 21a, the inactivation yielded an increase in Rmax in most layers but mainly in layer IV as the coefficient of variation for Rmax increased by ~25% for layer IV in area 21a for gamma rhythms. These findings indicate that the pulvinar enhanced gamma-band synchronization in higher-order cortical areas, in agreement with the notion that it is involved in the attentional modulation of visual cortical neurons.

Disclosures: L. Ikan: None. **N. Cortes:** None. **C.F. Casanova:** None.

Poster

P475. Interactions Between the Visual Cortex and the Thalamus

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Program #/Poster #: P475.10

Topic: D.07. Vision

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NIH Grant RO1- EY022388
NIH Grant 1R01EB026955

Title: Learning the Feedback Connections from V1 to LGN via Information Maximization

Authors: *R. EGHBALI[1,2], F. T. SOMMER[3], S. SHERMAN[4];
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Abstract: The lateral geniculate nucleus (LGN) relay cells act as a gateway for transmitting visual information from retina to the primary visual cortex (V1). The activities of thalamic relay cells are modulated by feedback connections emanating from layer 6 of V1. This gives the cortex a degree of control over the flow of information from retina to V1. While the receptive field properties of these early parts of the visual system are relatively well understood, the function, computational role, and details of the feedback network from V1 to LGN are not. Previous

studies suggest that the feedforward Shannon information could be important for understanding the function of the feedback. Computational models of efficient coding were able to derive receptive field properties of retinal ganglion and V1 simple cells from optimizing Shannon information. Further, previous experimental results have suggested that the feedback increases the Shannon information.
Motivated by this earlier work, we try to understand the function of the feedback as optimizing the feedforward information to cortex. To model this, we set up an information maximization problem to learn a set of convolutional feedback kernels. These kernels determine the strength and sign of feedback from a V1 cell to all ON and OFF LGN relay cells in the receptive field and the surrounding ones in the parvocellular pathway. The feedback kernels resulting from our optimization have either a "Mexican hat" shape or its flipped version depending on the polarity match or mismatch. For example, the kernel suggests an excitatory feedback on an ON LGN cells from a V1 cell fed by ON LGN cells when the two receptive field locations largely overlap. Conversely, the feedback is inhibitory when the receptive fields are slightly offset. There is no feedback when the receptive fields are further offset. The feedback effect reverses when there is a mismatch in polarity. Although only driven by the maximization of Shannon information in V1, our model conforms --and thus can explain-- recent experimental results in the literature.

Disclosures: R. Eghbali: None. **F.T. Sommer:** None. **S. Sherman:** None.

Poster

P528. Reaching Control: Motor Learning: Human Psychophysics I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P528.01

Topic: E.04. Voluntary Movements

Support: IIT Gandhinagar

Title: Task-based reactivation makes visuomotor memories susceptible to interference

Authors: T. SURESH, A. KUMAR, ***P. K. MUTHA**;
Indian Inst. of Technol. Gandhinagar, Gandhinagar, India

Abstract: Learning and memory are essential functions of daily life. Memory consolidation theory states that memories are labile immediately after acquisition and become stable after consolidation, defined as resistance to interference. However, the reconsolidation theory posits that when a consolidated memory is reactivated, it becomes fragile and susceptible to interference and requires "reconsolidation" for long term storage. We aimed to study the effect of reactivation on the consolidation in motor memories. In our first experiment, we used a visuomotor adaptation task and a 3-day A-B-A study design, wherein following baseline practice, subjects learned a 30° counter-clockwise rotation A on day 1 followed by a competing

30° clockwise rotation B on day 2, and were retested on A on day 3. Subjects were divided into two groups, depending on whether the prior memory of A was briefly reactivated on day 2 just before B learning (A-AB-A) or not (A-B-A). Surprisingly, we found robust interference in both groups. Specifically, A learning on day 3 was not different from naïve A learning on day 1 in either group, suggesting that interference might be independent of task-based reactivation. We then wondered whether the effect of reactivation was being masked by anterograde effects of B learning on the learning of A on day 3. In a second experiment, we therefore introduced null rotation (N) trials prior to A learning on day 3 in two new groups with (A-AB-NA) and without (A-B-NA) reactivation. These null trials served to wash out anterograde effects of B learning. Remarkably, we now found interference only in the reactivation group. Without reactivation, the memory of A acquired on day 1 was protected and could be expressed as savings on day 3. Taken together, our findings suggest that task-based reactivation can make a memory labile and susceptible to interference from a competing memory. This view is aligned with previous studies that suggest that reactivation brings a consolidated memory from an inactive to an active state; in this active state, any new information can modify the original memory. These results hold implications for strategizing protocols aimed at improving motor memory retention in neurorehabilitation and sports.

Disclosures: T. Suresh: None. **A. Kumar:** None. **P.K. Mutha:** None.

Poster

P528. Reaching Control: Motor Learning: Human Psychophysics I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P528.02

Topic: E.04. Voluntary Movements

Support: NS116883

Title: Presenting endpoint feedback at movement onset maximizes implicit sensorimotor learning

Authors: *T. WANG[1,2], G. AVRAHAM[1,2], J. S. TSAY[1,2], R. IVRY[1,2];
[1]Psychology, [2]Helen Wills Neurosci. Inst., Univ. of California, Berkeley, Berkeley, CA

Abstract: A classic observation in sensorimotor adaptation is that learning is better with online feedback compared to endpoint feedback. There are a number of possible explanations for this difference: Online feedback is dynamic, continuous, and begins with movement onset whereas endpoint feedback involves the static presentation of feedback at the end of movement. Although these factors are typically confounded in comparisons of online and endpoint feedback, the relevance of timing is shown by the dramatic attenuation of learning that is observed when endpoint feedback is delayed, even by as little as 100 ms (Held et al. 1966; Kitazawa et al. 1995).

The cost of delaying feedback raises the complementary question, does advancing the timing of "endpoint" feedback improve learning?
To explore this problem, we used a visuomotor adaptation task in which participants reach to a visual target. To provide advanced feedback, we used the visual clamp method in which the feedback cursor is always presented at an invariant position, deviated from the target by a fixed angle (and thus independent of hand position). Despite instructions to ignore this task-irrelevant feedback, participants show implicit motor adaptation with all of the hallmarks of adaptation in response to standard feedback. An unexplored advantage of clamped feedback is that it allows us to manipulate the timing of the feedback, independent of the movement.
We first replicated the advantage of online feedback over endpoint feedback with a visual clamp, comparing when the clamp was visible throughout the reach with its radial position aligned to the hand position to a condition in which the clamp only appeared at radial position of the target. Asymptotic values of adaptation for the online and endpoint conditions were 20.5° and 14.0°, respectively. Strikingly, when the "endpoint" feedback was presented at movement onset, the asymptotic value was 20.9°, a value similar to that observed with online feedback. The duration of the advanced feedback did not impact adaptation. Learning functions were similar for advanced feedback that persisted throughout the movement or was limited to 20 ms. In subsequent experiments, we manipulated the timing of endpoint feedback, including presenting it up to a couple hundred milliseconds prior to movement initiation. We found that the range of feedback time that drives the maximal level of adaptation was narrow (±50 ms relative to movement onset). These results demonstrate that the primary advantage of online feedback is not related to the dynamic, continuous nature of the signal, but rather that the feedback signal for online feedback appears earlier than with standard endpoint feedback.

Disclosures: T. Wang: None. **G. Avraham:** None. **J.S. Tsay:** None. **R. Ivry:** None.

Poster

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Program #/Poster #: P528.03

Topic: E.04. Voluntary Movements

Support: NSERC Grant 418589

Title: Learning twice the same motor task to investigate the mechanisms of anterograde interference

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Abstract: Anterograde interference has been repeatedly attributed to a competition between differing task memories. However, recent development alternatively suggests that initial learning may trigger a refractory period that occludes neuroplasticity and impairs subsequent memory formation, consequently mediating interference independently of memory competition. Accordingly, this work tested the hypothesis that interference can emerge when participants adapt *twice to the same gradually introduced visual deviation*, that is when competition between memories is prevented. In a first experiment, the inter-session interval (ISI) between two identical adaptation sessions was manipulated to be 2 min (n = 20), 1 h (n = 20) or 24 h (n = 20). Retention, through immediate reach aftereffects, was evaluated after each session. Results revealed that retention of the second session was impaired as compared to the first one when the ISI was 2 min but not when it was 1 h or 24 h, indicating a time-dependent process. Results from a second experiment (n = 20) replicated those of the first one and revealed that adding a third adaptation session with a 2 min ISI further impaired retention, indicating a dose-dependent process. Results from a third experiment (n = 20) revealed that the retention impairments did not take place when a learning session was preceded by the simple execution of baseline reaches without concurrent adaptation, thus ruling out fatigue and confirming that retention is impaired specifically when preceded by a learning session. A fourth experiment (n = 24) behaviorally manipulated the putative overlap between adaptation-specific neural networks recruited by the two adaptation sessions. Results showed that anterograde interference emerged regardless of memory competition and along a gradient as a function of the adaptation sessions' similarity; adaptation under similar conditions generated more anterograde interference than adaptation under dissimilar conditions, indicating that putatively overlapping neural networks are required to generate interference. Altogether, the present results introduce the possibility that a time-, dose-, learning-, and network-dependent refractory period contribute to the emergence of anterograde interference.

Disclosures: R. Hamel: None. **J. Lepage:** None. **P. Bernier:** None.

Poster

P528. Reaching Control: Motor Learning: Human Psychophysics I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P528.04

Topic: E.04. Voluntary Movements

Support: IIT Gandhinagar
MHRD, Government of India

Title: Spontaneous recovery in an untrained arm: insight into interlimb generalization of motor memories

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Abstract: Multi-rate models of motor adaptation predict spontaneous recovery, or transient expression of previously learned behavior, following an adaptation-extinction paradigm. This recovery has been attributed to a slow, implicit mechanism which operates in parallel with a fast, explicit process to drive learning and formation of a motor memory. We (Kumar et al., 2020) recently showed that the memory representation acquired via the slow process is effector-independent. We found that generalization of learning to an untrained arm, the signature of an effector-independent memory, was well-predicted by the level of the slow process reached during learning with the opposite arm. We therefore asked whether spontaneous recovery would be seen in an untrained arm, following an adaptation-extinction experience with the opposite arm. Two groups of right-handed subjects first adapted to a 30-degree counterclockwise visuomotor rotation (A, 200 trials) and were then exposed to a 30-degree clockwise rotation (B, 20 trials). Subjects then performed 80 "error clamp" trials with the opposite arm; on these trials, the visual error was clamped to zero. One group (LR) adapted to rotations A and B with their left arm and then performed the error clamp trials with their right arm. This order was reversed for the second group (RL). We first observed that adaptation to A and B was not different between the groups. However, spontaneous recovery was evident only in the LR group. That is, the right arm of the LR group was directed as if it was compensating for rotation A (learned previously by the left arm), even though this rotation was not present and error had been clamped to zero. Thus, learning seemed to generalize from the left arm to the right, and was expressed as spontaneous recovery. In contrast, recovery was absent in the RL group. This result provided a confirmation on two counts: first, since spontaneous recovery is the signature of the slow process, its occurrence in the untrained arm reinforced the view that slower, implicit processes forge an effector-independent motor representation that yields interlimb generalization. Second, it bolsters the view that interlimb generalization of visuomotor learning is asymmetric. We speculate that this occurs because access to the effector-independent memory representation is asymmetric.

Disclosures: A. Kumar: None. **P.K. Mutha:** None.

Poster

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Program #/Poster #: P528.05

Topic: E.04. Voluntary Movements

Support: This research was supported (in part) by the Intramural Research Program of the NIH, NINDS.

Title: Rapid generalization during early motor skill learning

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Abstract: Skill acquired in one task can transfer or generalize to another novel task. [1–3] We conducted a series of experiments to determine transfer between tasks during early motor skill learning, [4,5] when memories may still be fragile.[1] Subjects performed an explicit motor sequence task in an online naturalistic crowdsourcing environment. We evaluated transfer of skill acquired after a short training session in task A to task B. Across a set of experiments we parametrically changed the properties of tasks A and B. In experiment 1 (4 groups; total $n = 551$), we observed substantial performance transfer after only a single practice trial of A such that performance of task B was even better than performance at the end of practice of task A. Additional practice trials of task A did not enhance transfer. In experiment 2 (5 groups; total $n = 718$), we provided longer intervals between tasks, which did not enhance transfer. Finally, in experiment 3 (5 groups; total $n = 674$) we asked what content is transferred. To address this question, we manipulated the similarity between tasks A and B. We report that rapid transfer of procedural skill relied on the ability to extrapolate a parsing rule relating salient events within the practiced and transfer sequence rather than on the specific ordinal or transitional similarities between them. While data collection is ongoing, it appears that rapid generalization of skill in early motor learning can occur following a single trial of practice, may exhibit an offline contribution, and is dependent on similar salient rules between the tasks.
References**1.**Mosha, N. & Robertson, E. M. Unstable Memories Create a High-Level Representation that Enables Learning Transfer. *Curr. Biol. CB* 26, 100–105 (2016).**2.**Krakauer, J. W., Mazzoni, P., Ghazizadeh, A., Ravindran, R. & Shadmehr, R. Generalization of Motor Learning Depends on the History of Prior Action. *PLOS Biol* 4, e316 (2006).**3.**King, B. R. *et al.* Schema and Motor-Memory Consolidation. *Psychol. Sci.* 30, 963–978 (2019).**4.**Bönstrup, M. *et al.* A Rapid Form of Offline Consolidation in Skill Learning. *Curr. Biol.* 29, 1346-1351.e4 (2019).**5.**Bönstrup, M., Iturrate, I., Hebart, M. N., Censor, N. & Cohen, L. G. Mechanisms of offline motor learning at a microscale of seconds in large-scale crowdsourced data. *Npj Sci. Learn.* 5, 1–10 (2020).**6.**Ghilardi, M. F., Moisello, C., Silvestri, G., Ghez, C. & Krakauer, J. W. Learning of a sequential motor skill comprises explicit and implicit components that consolidate differently. *J. Neurophysiol.* 101, 2218–2229 (2009).
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Poster

P528. Reaching Control: Motor Learning: Human Psychophysics I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P528.06

Topic: E.04. Voluntary Movements

Support: NASPSPA Graduate Research Grant
ASU Global Sport Institute Seed Grant

Title: Placebo Effects of tDCS on Cognitive and Motor Training

Authors: *N. K. HAIKALIS[1], P. WANG[1], A. HOOYMAN[1], L. LOSSNER[1], R. YANG[1], S. JEJUKAR[1], M. GODMAN[1], K. WOODBURN[1], A. DETTMER[2], S. Y. SCHAEFER[1];
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Abstract: Studies using transcranial direct current stimulation (tDCS) to enhance cognitive and/or motor training often produce equivocal results. Differences in stimulation parameters (e.g., stimulation site, intensity, or duration) may contribute to mixed findings; It is thus plausible that placebo effects like expectations about tDCS may interact with the true 'treatment' effect of tDCS, resulting in mixed findings. We have recently shown that the general public has higher-than-neutral expectations about tDCS to enhance motor performance. Thus, the purpose of this study was to test whether there was a placebo effect of tDCS on cognitive and motor training.

Forty-seven participants (age:23.74 ± 4.37 years; 24 female) were randomly assigned to one of three groups: active anodal tDCS (n=19), sham tDCS (n=17), or no-tDCS control (n = 11). All participants completed a pre-test of the Shepard-Metzler Mental Rotation Task (MRT), followed by 20 minutes of training on the Corsi Block Tapping Task. Participants were re-tested on the MRT, measuring changes in cognitive (visuospatial) performance. Participants then completed 30 trials of motor training on a functional reaching task (measuring improvement in motor performance), followed by an exit questionnaire that measured their expectations about tDCS. The tDCS stimulation (20-min, 2mA) was applied to the right parietal area (P4) and paired with visuospatial training only for the sham and anodal tDCS groups.

No online tDCS treatment effect on visuospatial training was observed; rather, a placebo effect was, such that the sham group demonstrated more improvement in MRT reaction times than the active tDCS ($\beta = 125.29$, $p = .046$) and the no-tDCS control ($\beta = 151.58$, $p = .037$) groups. Regarding motor training, no offline tDCS treatment effect was observed, such that the sham and active tDCS groups did not differ in performance improvement nor final performance at the end of training (all $p>.160$). However, the sham group improved more than the no-tDCS control group during training in two aspects of the functional reaching task (object manipulation, $p = .036$; object transfer, $p = .044$). Moreover, regardless of group, better final performance at the end of motor training was associated with higher expectations about tDCS ($p = .015$).

Thus, this exploratory study showed there is a measurable placebo effect of tDCS on both

cognitive and motor training. This placebo effect was stronger than the true 'treatment' effect of tDCS and was likely driven by participants' expectations about tDCS. Future studies should consider placebo effects in tDCS research and treatment, as the neural capacity for such effects is not well understood.

Disclosures: N.K. Haikalis: None. **P. Wang:** None. **A. Hooyman:** None. **L. Lossner:** None. **R. Yang:** None. **S. Jejukar:** None. **M. Godman:** None. **K. Woodburn:** None. **A. Dettmer:** None. **S.Y. Schaefer:** None.

Poster

P528. Reaching Control: Motor Learning: Human Psychophysics I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P528.07

Topic: E.04. Voluntary Movements

Support: LSU CHSE Dean's Faculty Research Grant

Title: Boost your brain? 100% oxygen supply improves motor learning processes during a visuomotor adaptation task.

Authors: Z. WANG[1], *G. SPIELMANN[1], N. JOHANNSEN[1], F. GREENWAY[2], *M. DALECKI[1];
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Abstract: Normal-baric 100% oxygen treatment (100%NbOxTr) during first aid enhances neuronal survival rates in the human brain after heart attacks or strokes. However, whether motor learning processes of the brain can be improved by this treatment is not known. Motor learning (e.g., adapting to a visuomotor distortion) requires updating existing motor programs by integrating new cognitive and motor-related sensory information. Learning or reorganization of such capabilities is crucial for skill acquisition and neurorehabilitation. The present study examined the effects of 100%NbOxTr on visuomotor adaptation. Healthy young adults (m = 21 yrs.) were split into a 100%NbOxTr group (n=13) or a control group (mixed air; n=14) and performed a visuomotor adaptation task. During this task, participant's moved a pen along a horizontal tablet to move a cursor on a vertical display from a center target to peripheral targets. Peripheral targets (1.5 cm diameter) were presented in quasi-random order at 8 positions with 7.5 cm distance to the center target. Overall, 30 sessions were performed, and each session included 24 trials with 3 trials to each peripheral target. First, 4 sessions were performed with visual feedback correctly representing the pen position (baseline phase), followed by 20 sessions with 60° clockwise rotated visual feedback (adaptation phase), 3 sessions with correct visual feedback (after-effect phase), and 3 final sessions with distorted visual feedback (refresher phase). Gas

treatment was administered via nasal cannula (5 l/min) only during the 20 adaptation phase sessions. Variables of interest were initial direction error (IDE) and pathlength (PL) of the pen stroke to the target. Participant's mean baseline was subtracted from the subsequent phases for further analysis. ANOVA's (factors: group, session) were performed for each phase. For the baseline phase, no group-related differences were found for IDE and PL (all $p>0.05$). Significant group effects existed for adaptation and refresher phase (all $p\leq0.05$), with lower values in group 100%NbOxTr (adaptation: IDE -29%; PL -56%; refresher: IDE -25%; PL -60%) than in group mixed air. PL was also lower in the after-effect phase in group 100%NbOxTr (-46%; $p<0.05$). Our study shows that movement planning and execution-related characteristics of visuomotor adaptation were improved by 100%NbOxTr. Short-term consolidation of this effect sustained in refresher and partially in after-effect phases. Our finding suggests that 100%NbOxTr may improve neuroplasticity processes during motor learning tasks. This technique may provide promising potential for neurorehabilitation or skill training approaches.

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Poster

P528. Reaching Control: Motor Learning: Human Psychophysics I

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Program #/Poster #: P528.08

Topic: E.04. Voluntary Movements

Support: NIH Grant 1R01NS096083
NSF GRFP to RM

Title: The role of effort in motor learning

Authors: R. MARBAKER, *A. AHMED;
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Abstract: In previous research, reward and punishment, monetarily valued or as points-based scores, modulate the rate of learning, how much is retained, and how much of that learning is expressed (saved) upon re-exposure (Nikooyan 2014, Galea 2015). In other studies, cognitive effort induced by random task order during learning, enhanced savings after one week (Frömer 2016). The role of physical effort, the effort associated with completing the movement in the task, is not clearly understood. In this study, we investigate the effect of effort costs on the process of motor adaptation. Specifically, we asked whether increased effort costs would accelerate learning. Participants held the handle of a robotic arm and made 10 cm out-and-back reaching movements to one of eight possible targets evenly spaced about the center circle. Participants performed a visuomotor rotation (VMR) task, which consisted of five phases:

baseline, adaptation with a 30-degree visuomotor rotation, retention without visual feedback, washout, and savings during which they were exposed to the same 30-degree visuomotor rotation. To modulate effort costs, we modified a traditional VMR task so that the robot consistently opposed the hand motion with a damping force. Participants were divided into three groups: a high effort cost group where the damping constant was 20 Ns/m (N =10), a low effort cost group where the damping constant was 5 Ns/m (N = 4), and an error-dependent effort cost where damping on the following trial increased with increasing error on the previous reach (N=11). We hypothesized that the combined mechanisms of error-reduction and effort-reduction would accelerate learning for the error-dependent group. To quantify learning rates, we fit state-space models to individual subject data during adaptation and savings to quantify learning rates and retention factors. Preliminary findings show participants in all groups adapt to the VMR and exhibit savings. Error reduction over the course of adaptation appears positively correlated with the forces experienced during adaptation. Individuals who experienced greater forces during adaptation reduced their error to a greater extent. Further data collection will help elucidate the role, if any, of effort in driving or inhibiting motor adaptation.

Disclosures: R. Marbaker: None. **A. Ahmed:** None.

Poster

P528. Reaching Control: Motor Learning: Human Psychophysics I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P528.09

Topic: E.04. Voluntary Movements

Support: 418589

Title: Implicit sensorimotor adaptation occurs during motor imagery, but execution potentiates it

Authors: *C. PAWLOWSKY, F. THÉNAULT, P.-M. BERNIER;
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Abstract: In error-driven sensorimotor adaptation, a sensory prediction error (SPE) emerges when the actual and predicted sensory feedback do not match. This discrepancy leads to adaptive motor responses. These predictive mechanisms, relying on the efference copy of the motor commands, have been shown to be involved in motor imagery (MI) when an action is internally simulated without moving. However, even if MI is commonly reported to improve motor performance, it is unknown whether it can generate SPE and lead to adaptive motor responses. Here, using a reaching task with single-trial visuomotor rotations in a within-subject design (n=30), it was hypothesized that providing a rotated visual feedback time-locked to the moment at which participants were asked to imagine reaching a target on trial n would lead to adaptive motor responses when actually reaching on trial n+1 (MI condition). In a Control condition, the

same rotated visual feedback was provided while participants were instructed to make a small non-specific inward arm movement following target onset, irrespective of its location, in order to prevent them from performing meaningful MI. In an Execution condition, participants physically performed the reaching movement on trial n while being provided with rotated visual feedback. Results revealed that the magnitude of the post-rotation bias in hand direction was largest in the Execution condition. Interestingly, smaller but significant adaptive biases opposite to the direction of the perturbation were also observed in the MI and Control conditions. This implies that SPEs can be generated in absence of movement execution. It also suggests that sensorimotor adaptation is further improved by movement execution and/or dynamic proprioceptive signals, probably reflecting refined state estimation.

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Poster

P550. Neurophysiology: Implanted Electrodes and Other Direct Interactions with Neurons VI

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P550.01

Topic: E.05. Brain-Machine Interface

Support: NIH Grant 5R01NS116464
CNT (Center for Neurotechnology) EEC-1028725

Title: Smart dura: a large scale flexible semi-transparent interface for non-human primates

Authors: *T. BELLOIR[1], S. MONTALVO VARGO[2], D. J. GRIGGS[1], I. KIMUKIN[2], Z. AHMED[2], J. REDDY[2], S. FISHER[1], Z. ANDERSON[1], M. CHAMANZAR[2], A. YAZDAN-SHAHMORAD[1];
[1]Univ. of Washington, Seattle, WA; [2]Carnegie Mellon Univ., Pittsburgh, PA

Abstract: Studying large scale cortical circuits in non-human primates (NHPs) is important to gain insight on a variety of neural phenomena from fundamental mechanisms to neural disease and mechanisms of recovery. To do so, there is a need for chronically stable neural interfaces that are capable of recording neural activity with high spatial and temporal precision from large areas, while permitting the integration of additional modalities such as imaging or optogenetics manipulation. In that context, we have developed a novel neural interface, called *smart dura* that monolithically incorporates a micro-electrocorticographic (µECoG) array in an artificial dura. The µECoG arrays include recording electrodes that have been patterned on a Parylene C substrate embedded in polydimethylsiloxane (PDMS). PDMS is also the material used for the structure of the artificial dura since it is biocompatible, transparent and castable for realizing large and flexible structures. This original approach not only allows for large scale high density

electrophysiological recording, it also equips our interface with optical access and chronic stability. Here we discuss *in vivo* experiments to assess the utility and electrophysiological recording capacity and performance of our *smart dura*. Electrical recordings were acquired acutely in macaque sensorimotor cortex using arrays with electrode diameters ranging from 20 to 40 µm. We will discuss acute electrophysiological recordings from our interface in response to tactile stimulation on each finger of the monkey's right hand, enabling mapping its digit representation. The transparency of our interface allowed us to photochemically induce a focal ischemic lesion targeting a specific digit location determined with the somatotopy map. Electrical activity recorded before, during and after the lesion produces somatotopy maps that reveal the predicted functional changes of the lesioned circuits. These experiments show that our *smart dura* is capable of recording electrophysiology signals while maintaining optical access over large regions of NHP cortex (up to 300 mm^2). In addition, demonstrating the somatotopy maps acquired with our interface is a proof of concept that we can link a sensory function to the corresponding circuit activity, as well as induce a targeted lesioning of the underlying circuit. Our *smart dura* is well suited both for acute and chronic experiments and has the potential to accommodate higher recording sites density. Paired with complimentary stimulation modalities, it can be used to study more complex large-scale neural phenomena associated with the behaviors and cognitive abilities of non-human primates.

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Poster

P550. Neurophysiology: Implanted Electrodes and Other Direct Interactions with Neurons VI

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P550.02

Topic: E.05. Brain-Machine Interface

Support: NWO INTENSE (Grant No: 17619)

Title: The influence of pre-processing methods on the reconstruction of continuous speech from stereo-electroencephalography

Authors: *M. VERWOERT[1], M. C. OTTENHOFF[1], S. GOULIS[1], J. P. VAN DIJK[2], A. COLON[2], S. TOUSSEYN[2], L. WAGNER[2], P. KUBBEN[1,3], C. HERFF[1];
[1]Dept. of Neurosurgery, Sch. for Mental Hlth. and Neuroscience, Maastricht Univ., Maastricht, Netherlands; [2]Academic Ctr. for Epileptology, Kempenhaeghe, Heeze, Netherlands; [3]Maastricht Univ. Med. Ctr., Maastricht, Netherlands

Abstract: Reconstructing speech directly from neural signals has the potential to be an intuitive tool for restoring communication in individuals who have lost the ability to speak. Recent advances have been made to reconstruct speech with techniques that are fast enough to be applied in real-time for a closed-loop brain-computer interface (BCI) system. However, the reconstructed speech has yet to reach high intelligibility and the feasibility of reconstructing speech with stereo-electroencephalography (sEEG) has not been extensively studied. Here, we evaluate if sEEG can be used for speech synthesis and how different pre-processing techniques impact the quality of the reconstructed speech. We aim to gain an understanding of the underlying neural processes and contributing regions within the sparse sampling provided by sEEG. We simultaneously recorded sEEG and acoustic data in epileptic patients undergoing assessment for resective surgery, while articulating 100 uniquely prompted Dutch words continuously for five minutes. The acoustic data was transformed into the frequency domain using Fast Fourier Transform to create a speech spectrogram. The sEEG data samples were divided into overlapping windows which were time-aligned to the corresponding speech spectrograms. A timeframe of 400ms of neural data centered around the spectrogram was considered for the reconstruction. Regression approaches were applied to reconstruct the complete speech spectrogram from the neural data alone. We further investigated the influence of several pre-processing techniques on the reconstruction and on signal quality metrics. Results show that the speech spectrogram can be reconstructed using sEEG. We furthermore found that local referencing methods are beneficial compared to broader referencing in terms of apparent signal quality, however, this does not necessarily translate to improvements in the reconstruction of speech. This work encourages the potential use of sEEG electrodes for a speech neuroprosthesis and contributes to the knowledge on sEEG pre-processing methods.

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Poster

P550. Neurophysiology: Implanted Electrodes and Other Direct Interactions with Neurons VI

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P550.03

Topic: E.05. Brain-Machine Interface

Title: The brain interchange evaluation kit allows bench-top preparation for clinical studies with a wireless implantable brain-computer interface

Authors: *M. SCHUETTLER, C. STOLLE, M. OBERT, F. KOHLER, T. LEDWIG, J. F. QUICENO QUINTERO, J. RICKERT;
Cortec GmbH, Freiburg Im Breisgau, Germany

Abstract: We developed a brain-computer interface which is an implanted device that wirelessly communicates with a personal computer. The device is capable of brain recording and stimulation on 32 channels and allows the investigation of new closed-loop neuromodulation therapies. To de-risk clinical studies, clinical researchers can evaluate the system using a bench-top version of the implant. Together with an electronic representation of brain implanted electrodes, this bench-top implant comes as a kit suitable for investigation of system behavior, such as programming features (C, C++ or Python using an application programming interface API), stimulation output, recording properties, noise and timing aspects, wireless connectivity, etc. The bench-top implant can also be connected to electrodes in saline, e.g. for investigation of artifact pickup when stimulation and recording is carried out at the same time. Besides these in-vitro evaluations, the bench-top implant can be connected to patients having implanted electrodes with externalized leads as a first feasibility phase towards a complete implantation study. By this way, clinical brain-computer-interface studies can have two de-risking phases: 1. In-vitro system setup, programming and evaluation, 2. Connection of the bench-top version of the implant to externalized leads of electrodes implanted in patients, to eventually begin the 3. phase: chronic implantation of the electronic implant.

Disclosures: M. Schuettler: A. Employment/Salary (full or part-time):; CorTec GmbH, Freiburg, Germany. **C. Stolle:** A. Employment/Salary (full or part-time):; CorTec GmbH, Freiburg, Germany. **M. Obert:** A. Employment/Salary (full or part-time):; CorTec GmbH, Freiburg, Germany. **F. Kohler:** A. Employment/Salary (full or part-time):; CorTec GmbH, Freiburg, Germany. **T. Ledwig:** A. Employment/Salary (full or part-time):; CorTec GmbH, Freiburg, Germany. **J.F. Quiceno Quintero:** A. Employment/Salary (full or part-time):; CorTec GmbH, Freiburg, Germany. **J. Rickert:** A. Employment/Salary (full or part-time):; CorTec GmbH, Freiburg, Germany.

Poster

P550. Neurophysiology: Implanted Electrodes and Other Direct Interactions with Neurons VI

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Program #/Poster #: P550.04

Topic: E.05. Brain-Machine Interface

Support: NIH RN01NS092726
NIH R01NS110893
NSF GRFP DG31746891

Title: Galvanic stimulation evokes more natural neural responses than pulsatile stimuli.

Authors: C. STEINHARDT, ***G. FRIDMAN**;
Johns Hopkins Univ., BALTIMORE, MD

Abstract: Implantable pulse generators used ubiquitously in neural implants are constrained to delivering charge balanced biphasic pulses to modulate neural activity. Until recently low frequency galvanic (non-pulsatile) current modulation has not been possible without violating charge injection safety constraints. Safe direct current stimulator (SDCS) implant technology (Aplin and Fridman, 2019) supplants this restriction allowing for ionic galvanic current (iGC) modulation of the neuronal activity delivered via electrolyte-filled flexible conduits. The effects of iGC and pulsatile neuromodulation of the peripheral vestibular system have been examined experimentally in both behavioral assays and single afferent recordings. We developed a vestibular afferent computational model and replicated the experimental results of both stimulation paradigms in-silico. In verification with experimental data, pulsatile stimulation trains evoked a wide range of neural firing rates. Also, in agreement with previously obtained experimental data iGC could smoothly excite or inhibit neural activity while maintaining natural stochasticity of inter-spike intervals. The computational model allowed us to identify the complete range of neural responses to both pulse trains and iGC modulation that would not be possible otherwise. Pulse-train effects on the neural firing rate strongly depend on the pulse rate, pulse amplitude, and spontaneous activity of the neuron. For sub-threshold amplitudes (<~60uA), pulse-effects add to produce a single spike for multiple pulses, with threshold defined as the minimum amplitude at which a single pulse will evoke a single action potential. For supra-threshold mid-range amplitudes (~60-250uA) pulses evoked a single spike for each pulse at low pulse rates (<~150pps). At higher pulse rates however pulse-evoked AP refractory period interferes with the subsequent pulse to evoke one AP for every two pulses. High amplitudes (<250uA) will block the ability of subsequent pulses to produce spikes at higher rates (<~250pps). Spontaneous activity strongly interferes with the ability of pulses to evoke spikes. In contrast, iGC delivery changes the afferent's extracellular membrane potential. Depending on the polarity of the iGC, it smoothly depolarizes or hyperpolarizes the resting membrane potential, effectively modulating the threshold of AP generation in response to its synaptic inputs. This ability to modulate sensitivity of the afferent to its inputs could account for the improvement in behavioral assays that use iGC over its pulsatile correlate.

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Poster

P550. Neurophysiology: Implanted Electrodes and Other Direct Interactions with Neurons VI

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Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P550.05

Topic: E.05. Brain-Machine Interface

Support: NSF GRFP grantDGE-1842473

Title: Radiculopathy treated with Kilohertz Frequency Alternating Current Stimulation

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[1]Univ. of Florida, Gainesville, FL; [2]university of florida, gainesville, FL

Abstract: We evaluated the ability of kilohertz frequency alternating current (KHFAC) stimulation of the sciatic nerve to block behavioral evidence of pain and disability in a rat model of sciatic nerve lumbar disc herniation. Approximately 90% of clinical sciatica is caused when an intervertebral disk is herniated or slips. When this happens, nucleus pulposus (NP) slips out from the protective ring of the annulus fibrosus and causes inflammation of the nerve root through both compression and inflammatory signaling factors present in the NP. KHFAC applied to a nerve causes a nerve conduction block which is rapidly reversible when applied for less than 5-10 minutes. If applied to an injured sciatic nerve, KHFAC may limit hyperalgesic signals reaching the central nervous system, thereby limiting painful sensation from the innervated area distal to the stimulation. To mimic a physiologic radiculopathy, autologous tail NP was harvested and placed upon the right L5 nerve root. Male Lewis rats (3 mo.) (n=24) were separated into 3 groups: NP injury + KHFAC (n=7), NP injury + sham cuff (n=6), and sham injury + sham cuff (n=5). 6 rats were used for baseline measurements only. The surgeon was blinded to group assignment until the NP placement step. Electrode cuffs were placed around the ipsilateral sciatic nerve trunk. During blocked trials, a 50 kHz, 1 mA peak-to-peak sinusoidal waveform was delivered via the sciatic cuff. An optically isolated stimulus isolation unit (model 2200; A-M Systems) was used in conjunction with a high-pass filter circuit to limit direct current contamination of the waveform. Prior to surgery and for 2 weeks following surgery, animal tactile sensitivity, gait, place preference, and standing weight distribution were evaluated. Tactile sensitivity was evaluated through Chaplan's method for von Frey filaments. Gait was measured by recording freely moving animals. Gait kinematics were assessed using DeepLabCut and dynamics through force plate readings. The researcher testing behavior was blinded. 50 percent Paw withdrawal thresholds show that KHFAC applied to a distal cuff electrode can treat allodynia caused by this NP model. Without KHFAC, injured animals had increased mechanical sensitivity compared to baseline ($p<0.05$). Thresholds obtained with KHFAC were significantly higher than unstimulated thresholds ($p<0.01$), indicating KHFAC decreased hypersensitivity. Following our hypothesis, KHFAC decreases hypersensitivity but does not cause additional gait disability. This supports the idea that KHFAC applied to a peripheral nerve may be able to treat chronic pain resulting from sciatica.

Disclosures: L. Dewberry: None. **K. Allen:** None. **K.J. Otto:** None.

Poster

P550. Neurophysiology: Implanted Electrodes and Other Direct Interactions with Neurons VI

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Program #/Poster #: P550.06

Topic: E.05. Brain-Machine Interface

Support: NIH/NINDS RO1 NS109552

Title: Mechanisms of electrical stimulation of spinal cord micro circuits: Rebound suppression after acute transection

Authors: M. K. CHARDON, *M. JOHNSON, A. A. MAHROUS, J. MILLER, C. HECKMAN; Physiol., Northwestern Univ., Chicago, IL

Abstract: Spinal cord stimulation is widely used in patients with chronic pain, and electrical activation of spinal circuits has potential as an assistive therapy for individuals with spinal injury (Formento et al., 2018; Harkema et al., 2011; Minassian et al., 2004; Roy, Harkema, & Edgerton, 2012). However, the neurophysiology of spinal stimulation is still relatively unknown. Here we explore the mechanisms of direct electrical stimulation (DES) of the dorsal surface of the spinal cord. We use DES to stimulate the dorsal surface of the lumbar enlargement and record the behavior of the canonical motor micro circuit (CMM) of the ankle flexor and extensor pair (Sol/TA) in the decerebrate cat. CMMs are comprised of Ia sensory axons, interneurons (IN) and motoneurons (MN), are repeated segmentally throughout the spinal cord and form the basis of reciprocal inhibition between agonist/antagonist muscle pairs and their proper function is critical for inter-joint coordination. Stimulation of the Sol/Ta CMM (10Hz-40Hz, 500µA, 5s) generated distinct force patterns comprised of 3 phases: an initial transient spike at the start of the stimulus, a profound inhibition during the duration of the stimulus and a long (< 15 s) sustained phase (rebound) after removal of the stimulus in the Sol muscle, as well as a single phase, strong activation during the stimulation period, in the TA muscle. The force responses of TA and Sol are essentially mirror opposites during stimulation due to their reciprocal inhibitory relationship. The rebound force in Sol could illustrate the activation of persistent inward currents (PICs) in spinal motoneurons (MNs) or the interaction among inter neurons, allowing strong muscle activation when the stimulation ceased (rebound). We also previously showed that these patterns could be modulated by electrode location, favoring activation of TA or Sol. In the acute thoracic transection (T12) the responses change : transection abolished rebound activity in the Sol muscle but did not alter the interplay between Sol and TA. Rebound could not be induced even at stimulation intensities 4 times the stimulation used on the intact spinal cord suggesting dependence on a supra spinal mechanism. These results suggest that the excitatory/inhibitory balance between motor pools can be effectively modulated by electrical stimulation of the CMM via dorsal spinal stimulation and that there exists a fundamental difference between intact and transected spinal cord.

Disclosures: M.K. Chardon: None. **M. Johnson:** None. **A.A. Mahrous:** None. **J. Miller:** None. **C. Heckman:** None.

Poster

P550. Neurophysiology: Implanted Electrodes and Other Direct Interactions with Neurons VI

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P550.07

Topic: E.05. Brain-Machine Interface

Support: NIH Grant 25B0874

Title: Vagus nerve stimulation (VNS) enhances skilled motor learning through temporally-precise cholinergic neuromodulation

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Abstract: INTRODUCTION: Vagus nerve stimulation (VNS) paired with motor rehabilitation improves functional recovery from stroke and other neurologic injuries (Khodaparast et al., 2016; Dawson et al., 2021). Paired-VNS drives plasticity in both the injured and healthy nervous systems, but it is unknown if paired-VNS can improve motor performance in healthy animals. Moreover, the mechanisms by which paired-VNS drives changes in behavior remain unclear. Cholinergic signaling plays a critical role in learning, plasticity and reinforcement signaling, making it an appealing candidate to mediate the effects of VNS on motor learning and plasticity. In this study, we investigate the ability of paired-VNS to enhance motor learning in healthy animals and explore the role of cholinergic signaling in VNS-driven functional improvement. We hypothesize that paired VNS will improve motor learning in healthy animals through activation of cholinergic signaling. METHODS: To test the effects of VNS on motor learning, we implanted chronic stimulating electrodes on the left cervical vagus nerve of mice (Cortec; Microleads). Mice were trained to perform a dexterous forelimb reach for a food pellet using a novel automated behavioral system, CLARA. CLARA utilizes DeepLabCut to track real-time kinematics, which allows for high throughput, closed-loop stimulation based on behavioral metrics. Animals received VNS only after successful reach attempts (30hz, 0.6mA, 0.5s). To understand the role of cholinergic signaling during VNS, we performed *in vivo* extracellular recordings in the basal forebrain (BF) while delivering VNS in awake and anesthetized mice. In separate studies, mice learned to perform the skilled reach in the CLARA system, and VNS applied simultaneously with optogenetic inhibition of cholinergic neural activity within the basal forebrain (continuous, 0.5s, 0.5mW). RESULTS: In healthy animals, VNS paired with successful reaches enhanced acquisition of the task. Detailed kinematic analysis revealed that mice with paired-VNS made fewer targeting errors and displayed increased reach consistency compared to

controls. Additionally, VNS drove neural activity in the BF in awake and anesthetized mice. Moreover, optical inhibition of cholinergic BF neurons during VNS was sufficient to prevent VNS-driven enhancement of motor learning and eliminated the effects on reach kinematics. CONCLUSION: Our results demonstrate that VNS paired with reach success enhances dexterous motor learning in healthy animals, mediated in part, through the BF cholinergic neuromodulatory system.

Disclosures: S.G. Bowles: None. **J.L. Hickman:** None. **X. Peng:** None. **W. Williamson:** None. **C.G. Welle:** None.

Poster

P550. Neurophysiology: Implanted Electrodes and Other Direct Interactions with Neurons VI

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Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P550.08

Topic: E.05. Brain-Machine Interface

Support: DARPA HAPTIX N66001-15C-4017
NIH Grant UG3NS107688

Title: Long-term stability of Utah Slanted Electrodes Arrays as bidirectional peripheral nerves interfaces in human subjects

Authors: L. RIETH[1], B. BAKER[2], S. KELLIS[7], R. SHARMA[3], R. CALDWELL[8], D. KLUGER[7], J. A. GEORGE[4], A. NEIBLING[4], D. J. WARREN[5], G. A. CLARK[6];
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Abstract: The limited lifetime of penetrating microelectrode arrays for effective recording and stimulation is a critical impediment for their clinical use. Both biotic and abiotic performance degradation mechanisms have been identified. This study focuses primarily on the abiotic mechanism of tip metal degradation and lead wire breakage for Utah Slanted Electrode Arrays (USEAs) that have been implanted in peripheral arm nerves of 3 human subjects for up to 17 months, compared with an unimplanted array as a control. The effects of in-dwelling time, stimulation dose, and spatial position of the electrodes are evaluated. This report compares 7 USEAs implanted in subjects S5, S6, and S7 for 3, 14, and 17 months, respectively, with one unimplanted control array. The tip metal condition was analyzed by SEM and scored based on observed damage ranging from 1 (no damage) to 4 (severe). Differences in metallization damage by array and subject were analyzed using ANOVA and post-hoc tests. The effects of stimulation

dose delivered by each electrode on tip metal degradation were also statistically evaluated. Additionally, correlations between saline and in-vivo impedances and tip damage were analyzed. Spatial statistics were used to evaluate patterns in the tip metal degradation . All implanted arrays had statistically more tip metal degradation compared with the control array. A gap between the tip metal and silicon shank was often observed in SEM. This gap likely resulted from Si etching in the physiological environment. Additionally, arrays implanted in S5 (3 months) had statistically less damage than those implanted in S6 and S7. Considerably improved electrode impedance stability was achieved for S7 compared with S6[1], despite no significant differences in their tip metal damage, highlighting the strong impact of leads and device architecture. Surprisingly, stimulation dose was not found to correlate with tip metal degradation. Spatial analysis of tip degradation revealed a statistically significant clustering of tip metal damage, suggesting spatial factors do impact tip metal stability. Significant IrOx tip metal degradation occurred in USEAs implanted in human subjects, and damage was worse in arrays implanted for longer periods (14 and 17 months). Damage was spatially clustered, not randomly distributed. The damage did not correlate with stimulation doses up to 19 mC, and did not have a simple relationship with impedance. Dissolution of Si that undercuts the tip metallization is a potentially important degradation mechanism for the tip metallization of USEAs. [1] George et al. JNE pg. 056042, 2020, https://doi.org/10.1088/1741-2552/abc025

Disclosures: L. Rieth: None. **B. Baker:** None. **S. Kellis:** A. Employment/Salary (full or part-time):; Blackrock Microsystems. **R. Sharma:** A. Employment/Salary (full or part-time):; Applied Biosensors. F. Consulting Fees (e.g., advisory boards); Blackrock Microsystems. **R. Caldwell:** A. Employment/Salary (full or part-time):; Avanos Medical. **D. Kluger:** A. Employment/Salary (full or part-time):; Blackrock Microsystems. **J.A. George:** None. **A. Neibling:** None. **D.J. Warren:** None. **G.A. Clark:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Utah Array IP.

Poster

P550. Neurophysiology: Implanted Electrodes and Other Direct Interactions with Neurons VI

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P550.09

Topic: E.05. Brain-Machine Interface

Support: NIH Grant R01NS111518

Title: Magnetically aligned regenerative tissue-engineered electrical nerve interfaces: functional and histological evaluation and future directions

Authors: *A. S. LIM[1], E. W. ATKINSON[2], L. JIRACEK-SAPIEHA[3], M. KASPER[1], V. RIVERA-LLABRES[4], C. KULIASHA[3], C. M. RINALDI-RAMOS[4], C. E. SCHMIDT[1], J. W. JUDY[3], K. J. OTTO[1];
[1]Biomed. Engin., [2]Neurosci., [3]Electrical and Computer Engin., [4]Chem. Engin., Univ. of Florida, Gainesville, FL

Abstract: Achieving a high-bandwidth recording and stimulation interface with the peripheral nervous system (PNS) is crucial for the success of potential therapeutics for the amputee population. Higher information resolution at the peripheral nerve level, i.e., recording and stimulating isolated nerve fibers and fascicles, may allow finer control of the bidirectional communication taking place. In regards to prosthetic limbs for amputees, high-bandwidth PNS implants may ideally support sophisticated, modern prosthetics with multiple degrees of freedom for both sensory information and motor control.
Our approach to a peripheral nerve interface takes advantage of the regenerative ability of the PNS while treating amputee phantom limb pain, i.e., excision of peripheral neuromas. Doctors often excise the neuroma from the nerve and redirect the distal end to nearby musculature to minimize recurrence of neuroma formation. With regards to the Magnetically Aligned Regenerative Tissue Engineered Electrical Nerve Interface (MARTEENI), the neuroma will be excised and the distal end inserted into our device with hopes to inhibit neuroma reformation. Regenerating nerve fibers will encounter a growth promoting environment via a tissue-engineered hydrogel composed of glycidyl methacrylate-hyaluronic acid and guided through magnetically-aligned microchannels. After natural hydrogel degradation, the regrown nerve fibers will be in close proximity to uniformly spaced microelectrodes. Our implant, referred to as the MARTEENI, may allow the field of neuroprostheses a reliable, chronic PNS interface with a sufficient input-output information bandwidth to support next-generation prosthetic devices.
The findings here are two-fold. 1) we demonstrate the functional and histological evaluation of MARTEENIs in regenerated Lewis rat sciatic nerves using a multimodal analysis approach including: electrochemical impedance spectroscopy (EIS), electrophysiological recordings, and immunohistological techniques. Our data suggests stable impedance magnitudes at 1 kHZ up to 2 months after nerve implantation. This is validated with 2D histology and 3D imaging. 2) we report our electrode characterization findings as we transition from platinum metal to devices incorporating sputtered-iridium oxide (SIROF). The data suggests that SIROF microelectrodes have lower impedance across the frequency domain and greater charge storage capacity relative to platinum microelectrodes of identical surface area which in turn will result in decreased noise floor, i.e., improving electrophysiology recording quality, for better microstimulation at the PNS.

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Poster

P564. Cellular Properties of CPGS: Interneurons and Motor Neurons

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P564.01

Topic: E.07. Rhythmic Motor Pattern Generation

Support: DFG DU 331/6-2 to C. Duch
Carl Zeiss Foundation

Title: Decoding the network architecture and function of the central pattern generator for asynchronous flight

Authors: *S. HÜRKEY, S. RYGLEWSKI, C. DUCH;
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Abstract: Decades of research on the neural control of locomotion provide insight into the architecture of central pattern generating (CPG) networks. In brief, patterned motor output is typically generated by premotor interneuron synaptic connections and membrane properties. However, the most prevalent form of animal locomotion, asynchronous insect flight, comes with specific constraints to CPG architecture. By combining electro- and optophysiology with Drosophila genetics we demonstrate that coordinated motoneuron (MN) firing does not rely on patterned activity of premotor interneurons, but instead is shaped by electrical synapses between MNs. This de-synchronizes MN firing to generate specific firing sequences, which in turn, are required for stable wing beat frequencies. Asynchronous flight as employed by >600,000 species requires neuronal firing de-synchronization by electrical synapses. Asynchronous flight muscles are specialized to generate high mechanical power at fast contraction frequencies (~100-1000Hz) to compensate for low Reynold numbers. They do not contract in synchrony with neurogenically controlled cycling of myoplasmic Ca^{2+}, but rather are stretch activated. However, muscle power is tightly controlled by the CNS, because MN firing at every ~20-40th wingbeat tunes muscular stretch activatability. Although the MN firing patterns are known in multiple species, the mechanisms for pattern generation and the resulting function remain unknown. Each of the 6 Drosophila wing depressor muscle (DLM) fibers is innervated by one MN. During flight all MNs fire at the same frequencies, though not simultaneously but in preferred sequences. Common MN firing frequencies result from shared but unpatterned, excitatory cholinergic input. The specific MN firing sequences are generated by ShakB mediated weak, non-rectifying, reciprocal electrical synapses between MNs, thus de-synchronization firing of electrically coupled MNs and bypassing pattern generation by premotor interneurons. Both *in vivo* and in models knock-down and overexpression of ShakB causes MN firing synchronization. Forced MN firing synchronization by targeted overexpression of ShakB results in fluctuating wingbeat frequencies, demonstrating that splay state firing serves even power production.

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Poster

P564. Cellular Properties of CPGS: Interneurons and Motor Neurons

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Program #/Poster #: P564.02

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NSF IOS 1755098
ISU Dissertation Completion Grant

Title: Modulation of gap Junction voltage-dependence by the hyperpolarization-activated inward current

Authors: *M. DEMAEGD[1,2], C. STAEDELE[2,4], A. L. HARRIS[3], W. STEIN[2];
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Abstract: Voltage-dependent electrical synapses have an important role in shaping the direction and strength of electrical current that flows between neurons. The voltage-dependence often supports rectification between neurons of different types and modifies the synaptic drive as neuronal activity changes. In the crustacean stomatogastric ganglion, voltage-dependence has long been well-described for the electrical synapse between the descending projection neuron MCN1 (Modulatory Commissural Neuron 1) and the LG (Lateral Gastric) neuron in the gastric mill central pattern generator. Specifically, the electrical postsynaptic potentials (ePSPs) that LG receives from MCN1 increase in amplitude when LG's membrane voltage depolarizes, prolonging the duration of LG's bursts.Here, we show that this voltage-dependence is altered by the hyperpolarization-activated inward current (I_h). Specifically, when I_h was blocked with CsCl, ePSPS not only increased in amplitude, but also showed very little voltage-dependence between LG's resting membrane potential (-60mV) and spike threshold (-40mV). Instead, the LG membrane potential had to be hyperpolarized below resting potential to observe any voltage-dependence. Therefore, the dynamic range of the voltage-dependence shifted to more hyperpolarized values (Saline: EC50 = -64.98±5.06mV, N = 8; CsCl: EC50 = -82.87±10.37mV, N = 7). Modelling results indicate that such a shift may be enabled through presynaptic expression of I_h and the subsequent depolarization of the presynaptic membrane potential, which reduces the difference in membrane potential between the presynaptic and the postsynaptic neuron. Indeed, we found that I_h was present in the MCN1 axon presynaptic terminal and depolarized its membrane potential, while the postsynaptic LG membrane potential remained unaffected (N=4). Consequently, the dynamic range of the MCN1-LG electrical synapse voltage-dependence shifted toward the voltage range at which LG operates. Our data thus demonstrate that a presynaptic ionic current can modulate the dynamic voltage range over which an electrical synapse functions.

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Poster

P564. Cellular Properties of CPGS: Interneurons and Motor Neurons

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Program #/Poster #: P564.03

Topic: E.07. Rhythmic Motor Pattern Generation

Support: DeLisle Departmental Grant
Goethe University Grant

Title: Variability in encoding in identified leech neurons

Authors: *T. WRIGHT, J. AMMARI, V. PASMURTSEV;
Biol. Sci., California State University, Sacramento, Sacramento, CA

Abstract: It is now well established that there is considerable animal to animal variability in the production of rhythmic motor patterns. Recent work in the medicinal leech, *Hirudo medicinalis* has quantified the variability in the production of the motor pattern that drives the rhythmic constrictions of a bilateral pair of heart tubes. This variability begs the question: what parameters must be fixed in order to produce stereotyped behaviors despite the variability inherent within the nervous system? To address this question, we are exploring variability in the firing properties of the mechanosensory P (pressure,) and N (nociceptive) cells, which have been shown to have an influence on several behaviors in the leech (heartbeat, swimming,) and thus variability in their firing properties, while not directly involved in the cycle-to-cycle variability of the heartbeat pattern may never the less contribute to that variability. Because the output of many neural circuits is the firing of action potentials, the variability in the way that these neurons encode their inputs will contribute to the variability in the behaviors that we observe. There are several ways to assess encoding, including ramp current injections and FI Curves. Each of these protocols, while effective at measuring how neurons integrate their inputs, do not always capture the temporal dynamics likely to be observed in the nervous systems. To assess how leech neurons integrate inputs with different frequency characteristics, we recorded from several neuron types in adult leeches. For each neuron, we injected a white-noise signal (Gaussian White Noise (GWN) with a mean of 0 and an SD of 1 nA; cut-off frequency was 25 Hz) with a gain sufficient to produce at least 50 action potentials per injection. From this data we constructed a spike-triggered average of the input (STI; also known as a “Reverse Correlation”) and measured several parameters, including the delay to the peak of the STI, width, etc. We also assessed the normalized frequency response of the neurons to the stimulus. Finally, we computed return maps of the inter-spike intervals to assess how variable the ISIs were for each cell type. We show that the return maps for N cells are more variable than return maps for P Cells, and that the frequency response for both cell types are highly variable, with cells of each type acting as low-, band-, or high-pass filters.

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Poster

P564. Cellular Properties of CPGS: Interneurons and Motor Neurons

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Topic: E.07. Rhythmic Motor Pattern Generation

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NSERC-PGS-D (SAS)

Title: Dynamic mechanisms underlying continuous and episodic bursting patterns produced by a half-center oscillator

Authors: *G. CYMBALYUK[1], J. PARKER[1], R. J. ERAZO TOSCANO[1], S. A. SHARPLES[2], P. ELLINGSON[1], A. VARGAS[1], M. A. MASINO[3], P. J. WHELAN[4], R. L. CALABRESE[5]; [1]Neurosci. Inst., Georgia State Univ., Atlanta, GA; [2]Univ. of St Andrews, St Andrews, United Kingdom; [3]Neurosci., Univ. of Minnesota, Minneapolis, MN; [4]Univ. Calgary, Calgary, AB, Canada; [5]Emory Univ., Atlanta, GA

Abstract: Specialized oscillatory neuronal circuits controlling rhythmic movements, central pattern generators (CPGs), are capable of producing a wide variety of rhythmic patterns [1-6]. For example, leech heart beat CPG is based on pairs of mutually inhibitory interneurons forming half-center oscillators (HCOs) and producing continuous bursting (period ~ 1 s) while isolated neonatal rodent spinal cord preparations exhibit continuous locomotor bursting rhythms (period ~ 1 s) and a complex pattern evoked by dopamine: a very slow episodic bursting rhythm (period ~ 50 s) in which episodes of the fast-bursting rhythm are interrupted by long pauses [1]. Recent studies provide evidence that the Na^+/K^+ pump current (I_{pump}) participates in the dynamics of neurons and is a target of neuromodulation [2,6,7]. Intracellular Na^+ concentration ([Na]i) controlling I_{pump} plays a pivotal role in the generation of both continuous and episodic bursting rhythms. We build a general model describing dynamics of [Na]i, which led us to unveiling general mechanicms implicated in episodic and continuous bursting patterns of the CPGs on different timescales across species: leech, zebrafish, and mouse. Drugs affecting ($[Na^+]i$ like monensin and pharmacological blockade of currents carrying Na^+ fluxes such as I_{pump} and h-current were compared with corresponding manipulations of parameters in a family of biophysical models. For example, we describe general mechanisms producing episodic bursting patterns by a mammalian CPG model composed of a basic HCO, and a zebrafish CPG, composed of mutually excitatory neurons. These mechanisms may be applicable to other

biological systems that engage in continuous and episodic activity.

References

1. Sharples SA, Whelan PJ. Eneuro. 2017, 4(1).
2. Picton LD, et al. Journal of Neuroscience. 2017 Jan 25;37(4):906-21.
3. Issberner JP, Sillar KT. European Journal of Neuroscience. 2007 Nov;26(9):2556-64.
4. Parker J, et al. Journal of neurophysiology. 2018 Sep 1;120(3):1074-89.
5. Wiggin TD, et al. Journal of neurophysiology. 2012 Aug 1;108(3):925-34.
6. Sharples SA, et al. bioRxiv. 2021 Jan 1:2020-09.
7. Kueh D, et al. Elife. 2016 Sep 2;5:e19322.
8. Picton LD, et al. Current Biology. 2018 Dec 17;28(24):3911-23.

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Poster

P564. Cellular Properties of CPGS: Interneurons and Motor Neurons

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Program #/Poster #: P564.05

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NIH Grant U01 3823261

Title: New tools and refinements to interpret the role of spinal interneurons in motor modularity

Authors: *T. S. SMITH[1], M. ABOLFATH-BEYGI[2], T. D. SANGER[2], S. F. GISZTER[1]; [1]Neurobio. & Anat., Drexel Univ. Col. of Med., Philadelphia, PA; [2]Biomed. Engin., USC, Los Angeles, CA

Abstract: Novel motor plans are translated through the spinal cord, to control the musculature quickly and efficiently. In the motor modularity model of movement, dynamic ‘building blocks’ are combined in sequence or parallel to compactly construct most motor commands. We define a motor ‘module’ as a neural element evoking stereotyped motor activity, extracted from kinetic or biological features (e.g. synergist muscle EMG). In the spinal frog, modularity can be studied without influence of higher centers using the hindlimb-to-hindlimb wiping reflex as an inducible behavior composed of adjusted phases and amplitudes of three motor modules.During wiping, the spinal cord must control 1) when modules are recruited and 2) how modules coordinate the motor pools to produce coordinated muscle synergies. Previously we have discovered interneurons whose spiking activity coincided with these observed muscle synergies, but how these interneurons precisely contribute to modularity is unclear. One method of elucidating this

relationship is to investigate patterns of motoneuron recruitment within motor pools and across synergist muscles. Our new braided intramuscular EMG electrodes can record the activity of multiple single motor units within a muscle, and thus the spiking of individual motoneurons. By concurrently recording single motor units within and across synergist muscles, with associated interneurons, we will test for subfractionated recruitment of motor units within motor pools and concerted recruitment across motor pools. Because the role of single neurons in the ongoing motor pattern may be minor and spinal-state dependent, we utilize the stochastic dynamic operator (SDO) framework to evaluate relationships between point-processes and continuous state (e.g. neural spikes to EMG) and point-processes to point-processes (e.g. neural spikes to single motor units). We have stably recorded interneurons and motor units in spinal frogs. Currently we are evaluating best practices for maximizing the predictive utility of the SDO to map the activity of neurons and recruited single motor units to ongoing motor state. We demonstrate here that incorporating background dynamics is necessary for best interpreting and predicting the state-dependent behavior and specificity of interneurons. We have also found that single motor units may be strongly tuned to activity of a synergist muscle, suggesting fixed patterns of muscle coactivations. As we continue to collect and interpret this data, we expect to further elucidate the neural underpinnings of spinal modularity. Furthermore, we anticipate our innovative tools to be broadly applicable to other realms of neuroscience.

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Poster

P564. Cellular Properties of CPGS: Interneurons and Motor Neurons

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Topic: E.07. Rhythmic Motor Pattern Generation

Support: Wellcome Trust

Title: Involvement of the α3-subunit of Na^+/K^+-ATPase in the regulation of spinal motor network output

Authors: *F. SORRELL[1], F. W. HOOPER[1], E. AKKURATOV[2], A. APERIA[3], K. T. SILLAR[4], G. B. MILES[4];
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Abstract: The α3 subunit of Na^+/K^+-ATPase generates a large hyperpolarising current (the ultra-slow afterhyperpolarisation or ‘usAHP’) in lumbar motor neurons of neonatal mice following periods of prolonged, intense neuronal firing due to the rapid expulsion of intracellular sodium

(Picton et al., 2017). The neuron-specific α3 subunit is generally co-expressed with the ubiquitous α1 subunit, but its lower Na^+ affinity allows it to remain inactive or submaximally active until a considerable increase in Na^+ occurs (Azarias et al., 2013). The usAHP is thought to have a protective role as a fatigue-prevention mechanism in the locomotor network by facilitating a short-term 'motor memory' (STMM). Selectively blocking the α3 subunit using an acute application of the cardiac glycoside ouabain (1-3μM) reduces the amplitude of the usAHP and modulates the frequency of fictive locomotor-like rhythms in mouse spinal cords *in vitro* (Picton et al., 2017). To elucidate further the role of α3 Na^+/K^+-ATPase we here utilise an animal model harbouring the Rapid-Onset Dystonia-Parkinsonism gene mutation T613M in the α3-isoform encoding gene *ATP1A3*. This mutant α3 has a Na^+ affinity similar to wild-type α1 and is less efficient than wild-type α3 at rapidly restoring Na^+ load. We show, using whole-cell patch-clamp recordings, that lumbar motor neurons harbouring T613M fail to produce usAHPs (T613M n=38, WT n=35) and show a small but significant hyperpolarisation of the resting membrane potential (3-4 mV). At the network level, T613M does not affect spontaneous spinal motor activity (T613M n=17, WT n=14) or canonical drug-induced locomotor-like output (T613M n=14, WT n= 10). Similarly, when using a dorsal root stimulation to elicit locomotor-like output, T613M does not affect general properties of the locomotor rhythm but does produce a modest reduction in short-term 'motor memory' (T613M n=8, WT n=11). These data support a role for the Na^+/K^+-ATPase α3 subunit in the production of usAHPs in spinal neurons. At the network level, these data also support the involvement of α3 in STMM. However, the function of α3 in locomotor rhythm/pattern generation is less clear. We suggest that a compensatory mechanism, such as upregulation of α1-containing Na^+/K^+-ATPase may be able to rescue essential locomotor network behaviour in mutant models. Many movement disorders are linked to mutations in *ATP1A3*. These data advance our understanding of the fundamental role of Na^+/K^+-ATPase in motor networks and disease progression.

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Poster

P564. Cellular Properties of CPGS: Interneurons and Motor Neurons

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P564.07

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Support: NINDS Grant R01NS123116
ALSAC

Title: Molecular architecture of spinal V1 interneurons revealed by single-nucleus transcriptomic analysis

Authors: *A. J. TREVISAN[1], K. HAN[1], P. D. CHAPMAN[1], M. OKROS[1], Y. DHUNGANA[2], J. YU[2], V. MENON[3], M. GABITTO[4], J. B. BIKOFF[1];
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Abstract: Movement is essential for survival, enabling animals to explore their environment, obtain food, engage in social interactions, and escape predation. The neural circuits directly responsible for movement reside within the spinal cord, where networks of interneurons shape motor neuron activity to control the spatial and temporal patterns of peripheral muscle contraction. Despite recent insights into the developmental origin and function of spinal interneurons, their molecular identities and gene expression profiles remain enigmatic. A more thorough understanding of their identity will help provide insight into fundamental aspects of interneuron circuit organization and therapeutic potential in developmental motor disorders, neurodegenerative diseases, and traumatic spinal cord injury. The most abundant population of inhibitory interneurons in the spinal motor system is the evolutionarily conserved, ipsilaterally-projecting class of V1 interneurons, defined by expression of the homeobox transcription factor Engrailed1. Genetic ablation or silencing of V1 interneurons reduces the speed of locomotion and results in limb hyperflexion, indicating they influence diverse aspects of vertebrate motor output. In prior work, we found that V1 interneurons in neonatal mice constitute a molecularly heterogenous population that segregates into at least four mutually exclusive clades identified by the expression of the transcription factors Foxp2, MafA, Pou6f2, and Sp8. How this developmental molecular and cellular diversity relates to the complexity of mature neural circuits in the spinal cord remains unknown. Using single-nucleus RNAseq of V1 neurons isolated by flow cytometry at cervical, thoracic, and lumbar spinal segments across postnatal development (p0, p14, p28, and p56), we have identified approximately two dozen putative V1 cell types, revealing subtype-specific regulation of transcription factors, cell-adhesion molecules, and ion channels. Moreover, integration of transcriptomic data across development revealed that virtually all proposed cell types present at birth are preserved in adult neural circuits, suggesting their unique transcriptional profiles may confer distinct functional roles required for fine and adaptive control of complex movements. To our knowledge, this is the most detailed single-cell transcriptomic dataset focused on a single class of spinal interneuron to date, and will serve as a valuable resource for understanding the molecular architecture of neural circuits in the spinal motor system.

Disclosures: A.J. Trevisan: None. **K. Han:** None. **P.D. Chapman:** None. **M. Okros:** None. **Y. Dhungana:** None. **J. Yu:** None. **V. Menon:** None. **M. Gabitto:** None. **J.B. Bikoff:** None.

Poster

P564. Cellular Properties of CPGS: Interneurons and Motor Neurons

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Topic: E.07. Rhythmic Motor Pattern Generation

Support: NIH grant R01 NS047357

Title: V1 interneuron expressing the mafb transcription factor include renshaw cells and display bursting firing phenotypes and unique properties

Authors: E. K. BICHLER, A. F. RIVARD, *F. J. ALVAREZ;
Emory Univ., Atlanta, GA

Abstract: V1 cells are ventral inhibitory interneurons defined by the transcription factor (TF) En1 and are divided into four groups defined by MafA, Pou6f2, Foxp2 and Sp8 TFs. MafB is a TF expressed by MafA-Renshaw cells (RC) and some dorsal Pou6f2 V1s. MafB V1s uniquely show capacity for burst spiking. To analyze commonalties and differences in firing among different MafB V1s and along postnatal development we recorded whole-cell MafB V1s in spinal cord slices of P1 to P17 mice. MafB V1s were identified by GFP and tdT expression in *en1cre::mafbGFP::R26lsl-tdT* mice. RC identity was confirmed by post-hoc calbindin-IR. Recorded cells were subject to various stimulation paradigms to reveal different firing properties. These included short (400 ms) and long (2 s) depolarizing square pulses, hyperpolarizing pulses (1 s), slowly depolarizing triangular pulses to target slowly activating and inactivating mechanisms and very short pulses (3-5 ms) to evoke single action potentials. Bursting was found at all ages. It depends on TTA-P2 sensitive T-currents generating transient low threshold spikes (LTS during subthreshold depolarization) and afterdepolarizations (ADP after single Na+ spikes). They are best revealed with membrane hyperpolarization prior to depolarizing pulses. They are present in 100% RCs at any age with any depolarization protocol. In dorsal MafB-V1s, ADPs are evident after single spikes following short pulses (3-5 ms), but in longer pulses they are balanced by outward currents that block LTS and bursting in 50 to 70% of cells. In both cells, triangular pulses revealed firing hysteresis with higher excitability during ascending depolarizing ramps compared to descending hyperpolarizing ramps. We did not find evidence for persistent inward currents at any age. RCs showed higher excitability, more hyperpolarized action potential thresholds, higher gains in f-I curves, and higher steady-state firing. They also showed larger post-train afterhyperpolarizations (AHP) and at intermediate depolarizations large variability in interspike intervals (irregular firing) suggesting decorrelated inhibitory output with maintained excitatory drive. Dorsal MafB-V1s have lower excitability, more regular firing and spike-dependent accommodation. Properties that changed with age included amplitude and time course of post-spike AHPs and, in particular for RCs, the speed of action potentials rise and decay, significantly increasing firing rates. Thus, MafB V1s can burst spike but also display cell-specific plasticity that can alter their excitability and switch these cells between bursting and non-bursting and in the case of RCs between regular and irregular firing.

Disclosures: E.K. Bichler: None. **A.F. Rivard:** None. **F.J. Alvarez:** None.

Poster

P564. Cellular Properties of CPGS: Interneurons and Motor Neurons

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Program #/Poster #: P564.09

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Support: Swedish Research Council
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Ragnar Söderberg Foundation

Title: Single cell transcriptomic analysis of spinal dI6 neurons in zebrafish and mouse reveals functional distinct subtypes and novel subpopulations

Authors: *A. B. IGLESIAS GONZALEZ, J. JAKOBSSON, J. VIEILLARD, M. LAGERSTROM, K. KULLANDER, H. BOIJE;
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Abstract: The spinal locomotor network is frequently used for studies into how neuronal circuits are formed and how cellular activity shape behavioral patterns. A population of dI6 interneurons, marked by the Doublesex and mab-3 related transcription factor 3 (*Dmrt3*), has been shown to participate in the coordination of locomotion and gaits in horses, mice and zebrafish. Analyses of *Dmrt3* neurons based on morphology, functionality and the expression of transcription factors suggest the existence of different subtypes. Here, we have analyzed the transcriptomes of individual cells of the *Dmrt3* lineage from zebrafish and mice to unravel the molecular code that underlies their subfunctionalization by single cell RNA-sequencing. Indeed, clustering of *Dmrt3* neurons based on their gene expression verified known subtypes and revealed novel dI6 populations expressing unique markers. Differences in birth order, expression of axon guidance genes, neurotransmitters and their receptors, as well as factors affecting electrophysiological properties, provide the basis for their functional diversity and help explain functional and morphological differences. Our analysis provides the basis for experimentally determined parameters; early born *Dmrt3* neurons are ventrally located, active during fast swim, contact the soma of contralateral motor neurons, have low input resistance and high amplitude, to provide a strong mid-cycle inhibition to the opposite side. Meanwhile, late born *Dmrt3* neurons have more dorsal somas, are active during slow swims, contact contralateral dendrites of motor neurons, have higher input resistance and lower amplitude, to provide fine control of movements at slower speeds. In addition, the comparison between fish and mice provides insight into the evolutionary driven subspecialization concomitant with the emergence of limbed locomotion.

Disclosures: A.B. Iglesias Gonzalez: None. **J. Jakobsson:** None. **J. Vieillard:** None. **M. Lagerstrom:** None. **K. Kullander:** None. **H. Boije:** None.

Poster

P564. Cellular Properties of CPGS: Interneurons and Motor Neurons

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Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P564.10

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NIH R01 DC016413

Title: Clonally related V2a/b neuron pairs in the spinal exhibit similar morphologies

Authors: *S. BELLO-ROJAS[1], M. W. BAGNALL[2];
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Abstract: The ventral spinal cord houses a variety of excitatory and inhibitory neurons. Precisely timed excitation and inhibition allow organisms to perform locomotor behaviors. However, it is unclear how spinal circuits achieve proper balance between excitation and inhibition. Spinal premotor neurons are divided into cardinal classes based on progenitor group of origin and differences in transcription factor expression. The p2 progenitor group gives rise to the broad class of V2 precursors, which are labeled by the early transcription factor *vsx1*. Interestingly, individual Vsx1+ precursors undergo terminal differentiation into an excitatory, Chx10+ V2a neuron and an inhibitory, Gata3+ V2b neuron. It is unknown whether these clonally related sister V2a/b neurons have shared fates, including location in the spinal and position within the spinal circuit. To label clonally related V2a/b neurons, we injected titrated amounts of a *vsx1*:GFP plasmid in zebrafish embryos at the one cell stage. *In vivo* time-lapse imaging of stochastically labeled *vsx1*+ V2a/b sister neuron pairs demonstrated that sister neurons stay relatively close to each other, with their somata remaining within 10-20 μm of each other within 3D space. Axon analysis revealed similar trajectories of ipsilateral, descending projections, showing areas of axonal overlap between sister neurons. Furthermore, total axon length of a V2a neuron was consistently longer than its sister V2b, suggesting differences in the structure of excitation and inhibition along the rostrocaudal axis of the spinal cord. Collectively, these data indicate that there are anatomical relationships unique to these clonally related pairs, suggesting an innate developmental relationship between sister neurons. Currently, we are performing electrophysiological experiments between sister and non-sister V2a/b neurons to evaluate the impact of a shared origin on circuit connectivity.

Disclosures: S. Bello-Rojas: None. **M.W. Bagnall:** None.

Poster

P567. Respiratory Rhythm and Pattern Generation

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Topic: E.07. Rhythmic Motor Pattern Generation

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CNPQ grant 313719/2020-9

Title: Chemogenetic manipulation of astrocytes in the lateral parafacial region induces active expiration and sympathoexcitation in mice.

Authors: *D. MORAES[1], B. H. MACHADO[2], M. P. SILVA[3];
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Abstract: Previous studies demonstrated that the lateral aspect of the parafacial (pFL) region, which contains expiratory neurons, drives abdominal muscles (active expiration) and sympathetic outflow during high chemical drive (hypercapnia/acidosis). Astrocytes modulate neuronal activity/excitability and synaptic transmission as well as control the activity of motor circuits. Herein, we investigated the contribution of pFL local astrocytes to the central generation of the expiratory neuronal and neural activities and their impact on the sympathetic outflow and cardiovascular function of mice (7 weeks old - C57Bl/6). The pFL astrocytes were unilaterally activated by virally driven expression of designer receptors exclusively activated by designer drugs (hM3Dq) using the glial fibrillary acidic protein (GFAP) promoter. The *in situ* preparations of pFL-GFAP-hM3Dq mice received systemically clozapine N-oxide (90 µM) while the activities of pFL expiratory neurons, inspiratory (phrenic), expiratory (abdominal), and sympathetic (thoracic) nerves were extracellularly recorded. We observed that GFAP-hM3Dq-signaling activation induces an active expiratory pattern, with increased abdominal activity at the end of expiration, and evokes the pFL expiratory neurons firing. GFAP-hM3Dq-signaling activation also enhanced the sympathetic activity and perfusion pressure levels, without affecting the heart rate. On the other hand, GFAP-hM3Dq-signaling activation did not affect the hypercapnia-evoked active expiration and the concomitant response of sympathetic outflow. Besides, clozapine N-oxide did not affect the respiratory or sympathetic outflows in the absence of hM3Dq expression. These findings demonstrate that astrocytes modulate the activity of pFL neuronal circuit generating the expiratory and sympathetic activities in mice.

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Poster

P567. Respiratory Rhythm and Pattern Generation

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Topic: E.07. Rhythmic Motor Pattern Generation

Support: NIH HL111215
NIH NS110169
Kentucky Spinal Cord and Head Injury Research Trust
Commonwealth of Kentucky Challenge for Excellence
Craig F. Neilsen Foundation CNF546714

Title: Swallowing disorder following a clinical dose of buprenorphine in adult cats

Authors: *M. FRAZURE[1,2], K. YOUNG[2], N. MUSSELWHITE[2], C. GREENE[2], K. ICEMAN[2], T. PITTS[2];
[1]Dept. of Physiol., [2]Dept. of Neurosurg. and Kentucky Spinal Cord Injury Res. Ctr., Univ. of Louisville, Louisville, KY

Abstract: Opioid use has been associated with increased risk of infectious disease, including pneumonia. Individuals on opioids are more susceptible to chest infection due to opioid suppression of immune function, ventilation, cough, and secretion mobilization. While buprenorphine is a commonly used post-operative analgesic in veterinary medicine, its effects on the pharyngeal phase of swallow are unknown. Intramuscular buprenorphine (0.015 mg/kg) was administered to female cats (*n*=4; 12 months old) and videofluoroscopy was performed in the lateral plane 1 hour later. Animals were presented with 40% by volume barium sulfate in tuna water mixed to a thin consistency, and all ate voluntarily. Images were recorded at 30 frames/second and were compared to control feeding assessments using the Penetration Aspiration Scale (PAS). The PAS is an 8-point scale: 1-2 is considered normal, 3-6 is penetration of material into the larynx, and 6-8 is aspiration of material into the trachea/lungs. During control assessments, 75% (3 of 4 animals) had a PAS of 1, and the other animal had a PAS of 5 (material entered the airway, contacted the vocal folds, and was not ejected from the airway). After opioid administration, 50% (2 of 4 animals) exhibited significant aspiration (PAS=8); the most severe was the animal with penetration during the control period. One animal had a PAS of 4, and the final animal showed no significant deterioration of function. These results are consistent with the work of the Jackson Lab (doi: 10.1038/s41598-020-71804-2), reporting that specific genetic variance can lead to significant variability of opioid-induced effects on breathing. In 2 animals, swallow function failed with no appreciable response from the animal (i.e. no cough, throat clear, cessation of eating, etc.), one had worsened function which would not lead a clinician to withhold food/drink, and one animal showed no change. Our success in producing dysphagia (significant but varying by animal) with relatively small doses of opioid has potential implications for predicting aspiration pneumonia in post-surgical clinical cases.

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Poster

P567. Respiratory Rhythm and Pattern Generation

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Program #/Poster #: P567.03

Topic: E.07. Rhythmic Motor Pattern Generation

Support: NIH NS101737

Title: Development of an automated method for detection of onset and offset of multiunit inspiratory burst activity

Authors: *R. M. WADOLOWSKI, I. C. SOLOMON;
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Abstract: In animal studies, inspiratory electromyogram (EMG) multiunit burst activity is commonly used for assessing various aspects of central respiratory control. In rats, however, breathing frequencies can range from 50-100 bursts/min, and it is often difficult to accurately capture the individual bursts using automated approaches due to the stochastic nature of motor unit recruitment and gradual onset and offset behaviors of motor unit activity. Accurate automated burst onset and offset timing detection would facilitate burst analysis, and approaches commonly used to estimate these timings include threshold and/or window-based smoothing or binning methods that attempt to minimize signal variance around the overall trend of the recorded signal. These approaches are limited by reliance on conservation of burst-to-burst characteristics so that a generalized parameter (e.g., threshold, rate of rise) can be applied to multiple bursts and prior knowledge about the record (e.g., burst duration, non-stationarity, signal/noise ratio (SNR)), and therefore can result in inadequate capture of the onset and/or offset. To overcome these limitations, we present a method for automated estimation of burst onset/offset that minimizes the need for prior knowledge of the record, and assess its capabilities using diaphragm EMG activity recorded from urethane-anesthetized rats with artificially varied SNRs. Our method operates in two phases: (1) detection of the occurrence of a burst in order to create an arbitrary window around the burst and (2) scanning within the window for the timepoint at which the precise burst onset and offset occur. Each phase is achieved by clustering of a time-delay embedded matrix of the original record so that each point, regardless of continuity, is discretized into burst or noise categories. Features of these discretized points are then used to inform probability density functions, which are used to find optimum timepoints for burst onset/offset. This approach allows the original record to inform the algorithm with individualized parameters per burst, which is necessary for accurate detection within a non-stationary signal. Comparison of the algorithm onset/offset timings against 3 user annotated versions of the same sequences of diaphragm EMG bursts with various SNRs revealed that in most cases, the algorithm produced timings that were within or close to user timing ranges at all SNRs, but at a more rapid processing speed (30 burst/second). We believe that this algorithm is a

viable alternative to manually annotated burst onset/offset timings, particularly for large data sets.

Disclosures: R.M. Wadolowski: None. **I.C. Solomon:** None.

Poster

P567. Respiratory Rhythm and Pattern Generation

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Program #/Poster #: P567.04

Topic: E.07. Rhythmic Motor Pattern Generation

Support: FAPESP
CNPq

Title: Intrinsic and synaptic mechanisms controlling the expiratory activity of excitatory lateral parafacial neurones of rats

Authors: *K. S. MAGALHÃES[1], M. P. SILVA[1], A. S. MECAWI, SR[2], J. PATON[3], B. H. MACHADO[1], D. J. MORAES[1];
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Abstract: Active expiration is essential for increasing pulmonary ventilation during high chemical drive (hypercapnia). Several studies demonstrated that the lateral aspect of the parafacial (pFL) region, which contains expiratory neurones, drives abdominal muscles during active expiration in response to hypercapnia. However, the electrophysiological properties and synaptic mechanisms determining the activity of pFL expiratory neurones, as well as the specific conditions for their emergence, are not fully understood. Using whole cell electrophysiology and single cell qRT-PCR techniques, we evaluated the intrinsic electrophysiological properties, the phenotype and the respiratory-related synaptic inputs to the pFL expiratory neurones, as well as the mechanisms for the expression of their expiratory activity under conditions of hypercapnia induced active expiration in *in situ* preparations of juvenile rats. We also evaluated whether these neurones possess intrinsic $CO_2/[H^+]$ sensitivity and burst generating properties. GABAergic and glycinergic inhibition during inspiration and expiration suppressed the activity of glutamatergic pFL expiratory neurones in normocapnia. In hypercapnia, these neurones escape glycinergic inhibition and generate burst discharges at the end of expiration. Evidence for the contribution of post-inhibitory rebound, Cav3.2 isoform of T-type Ca^{2+} channels and intracellular $[Ca^{2+}]$ are presented. Neither intrinsic bursting properties mediated by persistent Na^+ current, nor $CO_2/[H^+]$ sensitivity or expression $CO_2/[H^+]$ sensitive ion channels/receptors (TASK or GPR4) were observed. On the other hand, hyperpolarisation-activated cyclic nucleotide-gated and twik-related K^+ leak channels were recorded. Post-synaptic disinhibition and the intrinsic electrophysiological

properties of glutamatermic neurones both play important roles in the generation of the expiratory oscillations in the pFL region during hypercapnia in rats.

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Poster

P567. Respiratory Rhythm and Pattern Generation

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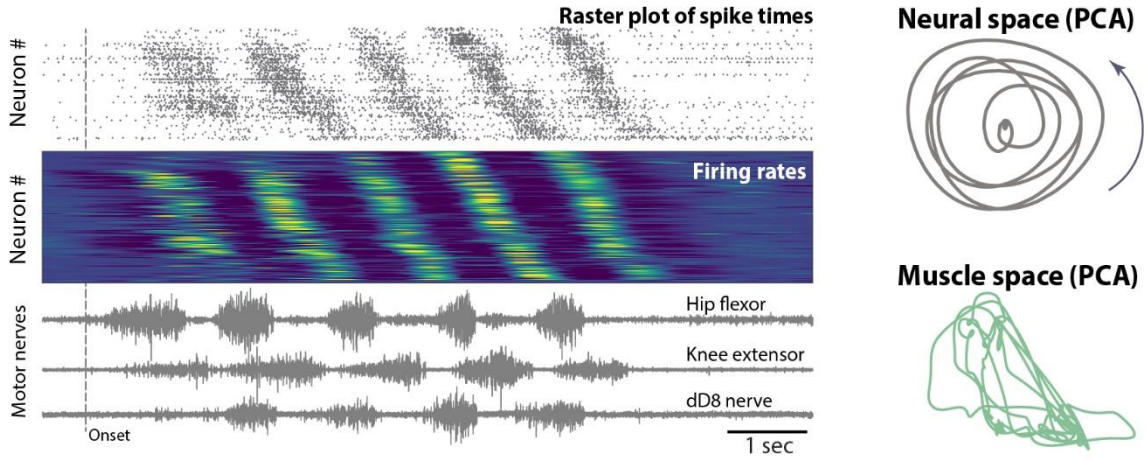
Topic: E.07. Rhythmic Motor Pattern Generation

Support: Independent research fund Denmark
Carlsberg foundation

Title: Rhythmic movement by spinal motor networks is governed by rotational population dynamics

Authors: H. LINDÉN[1], P. C. PETERSEN[2], M. VESTERGAARD[3], ***R. W. BERG**[1];
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Abstract: Most of the investigations on spinal rhythm generation are based on motor nerve recordings and single neuron recordings. Since flexor/extensor muscles alternate during movements, it is often assumed that the generation is accomplished by neuronal modules that alternate in opposition, which single neuron recordings seem to support. However, here we argue that when many neurons are monitored simultaneously a different picture emerges. We recorded hundreds of neurons from the lumbar spinal cord of turtles during rhythmic scratching and found that, rather than alternating, the neuronal population is performing a "rotation", i.e. cycling continuously through all phases (Figure). Rotational dynamics are observed across trials as well as behaviors. Since such rotation is difficult to explain with existing models of alternating neuronal groups, we propose a new theory that accounts for the rotational dynamics. Using a simplified network model, we show that in spinal networks with recurrent excitatory and inhibitory connectivity, there is no need for pacemaker activity or modular structures. Tonic input to the network controls the rhythm and pattern depending on the task. The model reproduces other experimental observations and provides a mechanism for multifunctionality. It also predicts that subsets of neurons, with both inhibitory and excitatory members, have the capacity to either speed up or slow down the movement. Figure caption: *Top left: Raster of neuronal population activity sorted according to phase. Middle left: Sorted population firing rates from top. Bottom left: Nerve activity of 3 motor nerves. Right top: neural space exhibit rotational activity (principal components). Bottom: muscle activity has less rotational activity.*



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Poster

P567. Respiratory Rhythm and Pattern Generation

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Topic: E.07. Rhythmic Motor Pattern Generation

Support: HL-126523
HL-144801
HL-090554
HL-145004
HL154558-01

Title: Dynamic network states of the brainstem respiratory rhythmic network

Authors: *N. J. BURGRAFF[1], N. E. BUSH[1], J. RAMIREZ[1,2], N. A. BAERTSCH[1,3];
[1]Ctr. for Integrative Brain Res., Seattle Children's Res. Inst., Seattle, WA; [2]Neurolog. Surgery, [3]Pediatrics, Univ. of Washington, Seattle, WA

Abstract: Within the brainstem, rhythmic respiratory activity is produced and shaped through interactions between a distributed network located bilaterally along the ventrolateral medulla and certain areas within the pons. Of particular importance is the preBötzinger Complex, which produces periodic synchronized bursting activity that coincides with inspiratory activity fundamental to mammalian breathing. Perturbing this region *in-vivo* can result in respiratory failure and fatal apnea, and when isolated *in-vitro*, the preBötC remains sufficient for

rhythmogenesis. The interval between synchronized bursts is regulated by a gradual increase in excitation among preBötC neurons that eventually synchronize to produce a population burst. Implicated in this process are network-based mechanisms involving synaptic interactions between interconnected excitatory and inhibitory neurons, as well as intrinsic-based mechanisms within the preBötC involving burst-promoting inward cation currents such as the persistent sodium current (I_{NaP}). The contributions of these intertwined mechanisms are not static, but can be dynamically tuned by e.g. changes in excitability and neuromodulatory inputs. Here, we test the hypothesis that the dynamic nature of these rhythmogenic properties plays a key role in determining the stability of rhythmic bursting produced within the network. We find that manipulating network excitability by changing extracellular [K^+] altered rhythm stability. Increasing [K^+] increased rhythm stability to a maximum point (stability^{max}). Further increases in [K^+] above stability^{max} destabilized the rhythm, resulting in a bell-shaped stability curve. Understanding this fundamental property that the respiratory network can assume different rhythmogenic states may allow for better predicting responses to drugs that alter network excitability such as opioids. Accordingly, we exposed isolated preBötC networks within horizontal brainstem slices to opioids (DAMGO) at each point along the bell-curve of rhythm stability. Brainstem slices displaying rhythmic activity that was at, or below the point of stability^{max} became highly unstable and were silenced with low levels of DAMGO. In contrast, brainstem slices displaying rhythmic activity beyond the point of stability^{max} became stabilized by DAMGO and were resistant against silencing at high levels of DAMGO. The concept that the respiratory network can assume different rhythmogenic states with different opioid tolerances may provide a novel framework for improving the predictability of drug response and inspire new strategies for prevention and reversal.

Disclosures: N.J. Burgraff: None. **N.E. Bush:** None. **J. Ramirez:** None. **N.A. Baertsch:** None.

Poster

P567. Respiratory Rhythm and Pattern Generation

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Commonwealth of Kentucky Challenge for Excellence
Rebecca F. Hammond Endowment
Craig F. Neilsen Foundation CNF546714

Title: Swallow motor synchrony is restored with a 5HT1a agonist after cervical spinal cord injury

Authors: *M. N. MUSSELWHITE[1], M. FRAZURE[1], K. YOUNG[1], C. GREENE[1], N. MELLEN[2], K. E. ICEMAN[1], D. R. HOWLAND[3], T. PITTS[1];
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Abstract: Oral ingestion requires synchronous motor activation to produce a rostral-caudal pressure cascade while ensuring protection of the airway. We have hypothesized that feedforward afferent information is necessary from bulbar and spinal pathways to promote precise motor recruitment and coordination with other airway behaviors (breathing). To test this hypothesis acute and sub-acute cervical C2-3 lateral hemisection was performed in adult cats while bilaterally recording from airway and chest wall musculature. Swallow was stimulated with water infusion into the oropharynx in sodium pentobarbital anesthetized adult cats. After hemisection, swallow motor pattern has distinct left-right asymmetries, with an increase in motor recruitment contralesionally and a delay in motor initiation ipsilesionally. The altered motor pattern produced pressure waveforms indicative of significant swallow dysfunction. This form was completely ameliorated with intravenous administration of a 5HT1a agonist, 8-OH-DPAT. To further confirm the specificity of action, the selective and competitive antagonist WAY-100635 returned the swallow pattern to the disordered state. While prior work has supported the importance of 5HT in swallow excitability, these new findings support involvement of serotonin in proper execution of motor actions.

Disclosures: M.N. Musselwhite: None. **M. Frazure:** None. **K. Young:** None. **C. Greene:** None. **N. Mellen:** None. **K.E. Iceman:** None. **D.R. Howland:** None. **T. Pitts:** None.

Poster

P567. Respiratory Rhythm and Pattern Generation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P567.08

Topic: E.07. Rhythmic Motor Pattern Generation

Support: R01 HL126523
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R01 HL151389

Title: The postinspiratory complex (PiCo), a hub for postinspiratory laryngeal closure and swallow related behaviors.

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Abstract: Breathing consists of three distinct phases: inspiration, postinspiration (postI) and active expiration. PostI is important for breathing, vocalization, cough, swallow and laryngeal control (closure). These behaviors involve laryngeal activity, which serves dual purposes for airway protection during swallow and mechanical slowing of expiration. Dysregulation of laryngeal activity during postI can result in foreign material to enter the larynx and trachea (aspiration) and cause pneumonia, a leading cause of death among many CNS disorders. PiCo, a medullary region composed of neurons that uniquely co-express both cholinergic and glutamatergic markers, has been shown to be active during postI, reset the respiratory rhythm when stimulated, and is proposed as the pattern generator for postI. The role of PiCo in other postI behaviors such as swallow is poorly understood. Our preliminary data revealed activation of PiCo can trigger not only postI, but also swallow, and both behaviors reset the respiratory rhythm. To evaluate the role PiCo plays in postI behaviors we targeted cholinergic and glutamatergic neurons by optogenetically stimulating PiCo in anesthetized spontaneously breathing adult ChATcre/Ai32(ChR2) and Vglut2cre/Ai32 mice. The ventral surface of the brainstem in the region of PiCo was exposed to optical pulses from a 447nM laser for durations ranging from 40-200ms. Swallow was evoked by peripheral stimulation of water injected into the oral cavity and by central stimulation, laser activation of PiCo. Swallow and postI laryngeal activity were measured via monopolar suction electrodes of the hypoglossal (XII) and vagus (X) nerves as well as bipolar electromyogram (EMG) of the submental, laryngeal complex and costal diaphragm muscles. We found a significant increase in swallow duration of water versus PiCo stimulated swallows (*p=0.02)*. Optogenetic stimulation of ChAT or Vglut2 neurons applied at random across different phases of breathing resulted in a significantly higher probability of triggering a swallow during inspiration than triggering laryngeal closure (*p=0.02, p=0.05 respectively).* Water swallows delayed the respiratory rhythm up to two times the normal respiratory rhythm, whereas PiCo stimulated swallows delayed up to half of the respiratory rhythm. Our data indicates PiCo is a hub for the generation of both postI activity and swallowing. Peripheral stimulation of swallow sends afferent information to the nTS shaping the swallow motor pattern. It is plausible that the absence of this afferent feedback in swallow triggered by PiCo stimulation bypasses the dorsal swallow group, directly activating the ventral swallow group.

Disclosures: A. Huff: None. **M. Karlen-Amarante:** None. **J. Ramirez:** None.

Poster

P567. Respiratory Rhythm and Pattern Generation

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Topic: E.07. Rhythmic Motor Pattern Generation

Support: NSF DMS 1951095

Title: Biophysical mechanisms of bursts and burstlets in the respiratory preBötzinger complex

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Abstract: Inspiratory breathing rhythms arise from synchronized neuronal activity in a bilaterally distributed brainstem structure known as the preBötzinger complex (preBötC). In *in vitro* slice preparations containing the preBötC, extracellular potassium must be elevated above physiological levels (to 7-9mM) to observe regular rhythmic respiratory motor output in the hypoglossal nerve to which the preBötC projects. Reexamination of how extracellular K^+ affects preBötC neuronal activity has revealed that low amplitude oscillations persist at physiological levels (1,2). These oscillatory events are sub-threshold from the standpoint of transmission to motor output and are dubbed burstlets. Burstlets arise from synchronized neural activity in a rhythmogenic neuronal subpopulation within the preBötC that fails to recruit the larger network events, or bursts, required to generate motor output. The fraction of preBötC oscillatory events that are subthreshold (burstlet fraction) decreases sigmoidally with increasing extracellular potassium (2). These observations underlie what is known as the burstlet theory of respiratory rhythm generation. In this computational study, we show how intracellular calcium dynamics in conjunction with a calcium-activated non-selective cationic current, modeled based on previous observations (3,4,5), can explain the key observations underlying the burstlet theory of respiratory rhythm generation. Thus, we provide a mechanistic basis to unify the experimental findings on rhythm generation and motor output recruitment in the preBötC.
1) Kam, Kaiwen, et al. "Distinct inspiratory rhythm and pattern generating mechanisms in the preBötzinger complex." *Journal of Neuroscience* 33.22 (2013): 9235-9245.
2) Kallurkar, Prajkta S., et al. "Evaluating the burstlet theory of inspiratory rhythm and pattern generation." *Eneuro* 7.1 (2020).
3) Koizumi, Hideo, et al., “Transient receptor potential channels TRPM4 and TRPC3 critically contribute to respiratory motor pattern formation but not rhythmogenesis in rodent brainstem circuits.” *Eneuro* 5.1 (2018).
4) Phillips, Ryan S., et al. "Biophysical mechanisms in the mammalian respiratory oscillator re-examined with a new data-driven computational model." *Elife* 8 (2019): e41555.
5) Picardo, Maria Cristina D., et al. "Trpm4 ion channels in pre-Bötzinger complex interneurons are essential for breathing motor pattern but not rhythm." *PLoS Biology* 17.2 (2019): e2006094.

Disclosures: R.S. Phillips: None. **J.E. Rubin:** None.

Poster

P634. Sleep: Pharmacological and Cellular Manipulations

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Program #/Poster #: P634.01

Topic: F.08. Biological Rhythms and Sleep

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Title: Peripheral sensory stimulation elicits global slow waves by recruiting somatosensory cortex bilaterally

Authors: *Z. P. ROSENTHAL[1], R. V. RAUT[2], R. BOWEN[3], A. Z. SNYDER[5], J. P. CULVER[4], M. E. RAICHLE[6], J.-M. LEE[7];
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Abstract: Slow waves (SWs) are globally propagating, low-frequency (0.5 - 4 Hz) oscillations that are prominent during sleep and anesthesia. Three decades of local electrophysiology and EEG recordings have demonstrated that SWs are essential to neural plasticity, memory, and grouping of other oscillatory events. However, much remains unknown about the mechanisms coordinating SW propagation at the macroscale - in particular why SW patterns change globally after focal injuries like stroke. Here, we investigate how specific cortical modules and their interhemispheric connectivity contribute to the initiation and global propagation of somatosensory-evoked SWs. To this end, we recorded cortical activity in awake and ketamine/xylazine anesthetized mice, using widefield optical imaging with fluorescent calcium indicator GCaMP6f (n=33, 10-12 week old males, JAX 024276*)*. We then characterize the spatiotemporal properties of cortical waves with space-frequency singular value decomposition and power spectral density analysis. We demonstrate that unilateral somatosensory stimulation evokes bilateral waves that travel across the cortex with state-dependent trajectories. Under anesthesia, we observe that rhythmic stimuli elicit globally resonant, front-to-back propagating SWs. By delivering focal photothrombotic lesions to right forepaw somatosensory cortex (S1), we show that somatosensory-evoked global SWs depend on bilateral recruitment of homotopic S1 cortices. Specifically, unilateral lesions of forepaw S1 disrupt the global coherence of forepaw-evoked SWs evoked from both contralesional (left paw $*P = 0.02$) and ipsilesional (right paw $*P = 0.03$) hemispheres. In contrast, ablation of forepaw S1 spares spontaneous global SWs outside the perilesional area ($^{NS}P = 0.85$), as well as SWs evoked by neighboring

somatosensory modalities (^{NS}P = 0.34). These results show that evoked slow waves may be triggered by bilateral activation of modality-specific, homotopically-connected cortical hubs. Our findings further highlight the existence of mechanisms by which focal injury may disrupt global brain network activity.

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Poster

P634. Sleep: Pharmacological and Cellular Manipulations

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Program #/Poster #: P634.02

Topic: F.08. Biological Rhythms and Sleep

Support: EU H2020 Research and Innovation Programme, Grant 785907 (HBP SGA2)
EU H2020 Research and Innovation Programme, Grant 945539 (HBP SGA3)

Title: Emerging slow waves in premotor cortex of behaving monkeys

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Abstract: The richness of brain dynamics has been often associated with criticality typically observed in quiescent-to-active state transitions, as an optimal condition for cognitive functions like sensory-motor control and information processing. However, boundaries between different dynamical states were also associated with a different critical transition, as in the sleep-wake cycle, when brain activity goes from asynchronous, highly irregular, to rhythmically regular with low complexity. In the latter condition neuronal assemblies oscillate synchronously, giving rise to the so-called slow waves activity (SWA), characteristic of the deepest sleep stages and under general anesthesia. Disentangling which of these two kinds of critical dynamics occur and what role plays criticality in brain activity still needs to be conclusively elucidated. In particular, criticism has been raised against the assessment of criticality based on coarse experimental measures resulting from the coalescence of local neuronal signals.
Here we address this issue using *in vivo* experiments, data analysis and theoretical tools that have the power to determine whether, and in which condition the brain activity reaches criticality. By measuring intracortical multi-unit activity (MUA) and local field potential (LFP) with multi-electrode arrays chronically implanted in the premotor cortex of two macaque monkeys, we found a rich repertoire of spatiotemporal patterns spontaneously emerging in resting epochs following overt movements in delayed-reaching tasks. Surprisingly, these patterns consisted of MUA Down-to-Up transitions coordinated in space as traveling waves, closely resembling SWA activity during NREM sleep-like anesthesia. We found that during the short resting-like

instances of the activity, but not during the execution of the task or the SWA, neural avalanches of the MUA displayed a scale invariant distribution of their size following power-laws with exponents predicted by the theory of synchronization phase transition.
We reproduced this dynamics in a network of spiking neuron models set to work close to the critical point separating the synchronous and asynchronous global states. Importantly, our neuronal data confirmed the model prediction that the size of the avalanches depends on the preceding level of activity, which determines the amount of "fatigue" accumulated by the network before its sudden activation.
Overall, these findings provide compelling evidence that the premotor cortex, while performing reaching tasks, only sporadically approaches the critical point separating synchronized (sleep) and desynchronized (wake) brain states.

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Poster

P634. Sleep: Pharmacological and Cellular Manipulations

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Program #/Poster #: P634.03

Topic: F.08. Biological Rhythms and Sleep

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Title: Proposing a duty cycle model for alpha rhythms in human EEG

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Abstract: We propose a duty cycle model for human alpha rhythm in scalp-recorded EEG. In the pulse width modulation (PWM) technology, a rectangular pulse wave with a fixed peak amplitude is used. The output is the time-averaged amplitude by duty cycle, which is the ratio between two discrete states, on and off. Today, quantifying EEG power in the frequency domain is a common practice. For example, the recently published Fitting Oscillations & One Over F (FOOOF) is an algorithm to model the power spectral density (PSD). However, as the single-trial EEG properties were 'discovered' after prevalence of trial-averaged event-related potentials (ERPs), recent studies on single-trial time-frequency domain EEG power studies discovered sporadic and discrete 'events' that disappears after averaging. In the same vein, we propose a new model of the classical human occipital alpha rhythm to free it from the implicit assumption of the stationary Gaussian process. We analyzed 34ch scalp EEG data recorded from 150 healthy

adults during eyes-open resting. Total of 270 brain independent components showed dominant alpha peak in the PSD plot that allowed model fitting. We developed the novel peak-power frequency (PPF) ribbon analysis, which sorts the time frames of the spectrogram so that PPF is sorted from low to high. We found the emerging PPF ribbon was fitted with a sigmoid with an extended lower and upper plateaus, which corresponds to off and on states of the alpha rhythm, respectively. The duty cycle ratio across the group was 10%-70% with flat distribution with the peak at 20%. The length of the upper plateau and the height of the PSD alpha peak were positively correlated ($r = 0.53$, $p < 10^{-16}$). Importantly, the peak power distribution across the upper plateau (average range 6.3-13 Hz) was flat with no Gaussian bump, indicating that Gaussian model is not supported. Thus, our duty cycle model is supported by the data, and the Gaussian model was rejected. The results shed light on the distribution of the temporal dynamics of thalamo-cortical network observed at the macroscale electrophysiology. The novel ribbon analysis provides more insights into the generative mechanism of EEG than PSD, which brings the richness of the unaveraged data back to the conventional mean-based statistics.

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Poster

P634. Sleep: Pharmacological and Cellular Manipulations

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Program #/Poster #: P634.04

Topic: F.08. Biological Rhythms and Sleep

Support: R01MH111601

Title: Altered 12 hour rhythms in the dorsolateral prefrontal cortex of subjects with schizophrenia

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Abstract: Schizophrenia (SZ) is a chronic neuropsychiatric illness associated with cognitive dysfunction and disrupted circadian behaviors. Our lab recently observed a distinct set of diurnally rhythmic transcripts in SZ subjects relative to a cohort with no history of psychiatric illness (non-psychiatric; NP) using rhythmic analysis of RNA-sequencing (RNA-seq) data from the dorsolateral prefrontal cortex (DLPFC), a region associated with executive dysfunction and heavily implicated in SZ. This study found that genes which gained 24 h rhythmicity in SZ were associated with mitochondria dysfunction and GABA-ergic signaling. In the current study, we

examine ultradian rhythms in the same DLPFC data using two types of analysis, a harmonic analysis that accounts for multiple rhythms at once (eigenvaluepencil analysis) and a sinusoidal nonlinear regression analysis that specifically tests for 12 h rhythms in transcript expression. In the eigenvaluepencil analysis, transcripts in the DLPFC of both NP and SZ subjects are enriched for rhythmic components with 12 and 24 h periods. Ingenuity pathway analysis indicates that transcripts with a 24 h rhythmic component are associated with circadian rhythm signaling and various intercellular communication pathways in NP subjects, but mitochondria and GABA receptor signaling in SZ subjects, consistent with our previous findings. In both cohorts, transcripts with 12 h rhythmic components are associated with mitochondria and EIF2 signaling pathways. In the NP cohort, these transcripts peak in expression at ZT 2-3 (~9 AM/PM), but in SZ we observe an anti-phasic shift, with these transcripts peaking in expression at ZT 8-9 (~3 PM/AM). Using the nonlinear regression analysis we observe significant 12 h rhythms in transcript expression and confirm that these are associated with mitochondria and EIF2 signaling pathways in both cohorts, peak at two distinct timepoints (ZT 2-3 and ZT 9), and have an anti-phasic shift in timing in SZ. However, this analysis also uncovered a significant loss of 12 h rhythms associated with the unfolded protein response pathway in SZ. These data suggest alterations at multiple levels in the rhythmic regulation of mitochondria, as well as an overall disruption in the 12 h rhythm system in SZ.

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Poster

P634. Sleep: Pharmacological and Cellular Manipulations

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Topic: F.08. Biological Rhythms and Sleep

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Title: Increase in sleep slow waves after cortical injections of sodium oxybate and the role of SOM+ interneurons.

Authors: *G. M. SPANO[1], W. MARSHALL[1,2], M. L. CAVELLI[1], G. TONONI[1], C. CIRELLI[1]; [1]Dept. of Psychiatry, Univ. of Wisconsin Madison, Madison, WI; [2]Dept. of Mathematics and Statistics, Brock Univ., St. Catharines, ON, Canada

Abstract: The systemic administration of sodium oxybate (SXB), the sodium salt of gamma-hydroxybutyric acid, promotes slow wave activity (SWA, 0.5-4 Hz EEG power) and increases non-rapid eye movement (NREM) sleep. These effects are mediated by GABAb receptors, which are expressed pre- and postsynaptically in cerebral cortex and many other brain regions. Because

slow waves are mainly a cortical phenomenon, we hypothesized that the effects of SXB are partly mediated at the cortical level. Because we found that SOM+ interneurons play a key role in SWA generation (Funk et al. JN 2017), we also hypothesized that the inhibition of SOM+ cells would prevent SXB from enhancing SWA. Adult mice (SOM-Cre [B6N.CgSsttm2.1(cre)Zjh/J; either sex) were implanted with a microdialysis cannula targeting left frontal cortex, and with EEG electrodes and laminar silicon probes (NeuroNexus, A1x16; 50um site spacing) in frontal and parietal cortex ipsi or contralateral to the injection site. In mice used for chemogenetic inhibition of SOM+ cells, the adeno-associated virus AAV8-hSyn-DIO-hM4D(Gi)-mCherry was injected in right frontal cortex. After the microdialysis probe was implanted at Day0, artificial extracellular fluid (aECF) was perfused (0.5ul/min) for 2hrs at Day1, followed by 90min of either aECF or SXB (500µg/ml). For chemogenetic experiments, clozapine-N-oxide (CNO; 0.1mg/kg) or saline was injected i.p. 20min before the infusion of SXB. EEG and local field potential signals were analyzed offline by visual inspection of 4-sec epochs to score sleep/wake states and perform power spectra analysis. During NREM sleep, SWA increased during the first 30min post-SXB injection relative to the 30min pre-injection (11 mice), while it did not change after the infusion of aECF or SXB preceded by CNO (7 mice). The SWA increase was present ipsi and contralateral to the SXB injection in frontal and parietal cortex (paired sample t-tests; SXB, all $p < 0.007$, Cohen's $d > 1.1$; CNO-SXB all $p > 0.43$, Cohen's $d < 0.32$). Using linear mixed effects models, we found no interaction between injection and electrode position and a significant effect of injection ($p = 2.1e\text{-}11$; $f^2 = 0.6625$). Post-hoc tests revealed SXB > CNO-SXB ($p = 1.2e\text{-}10$) and SXB > aECF ($p = 3.9e\text{-}10$). In 5 other mice whose microdialysis cannula was misplaced, the injection of SXB failed to increase SWA. These results show that SBX promotes SWA, at least in part, by acting directly on the cortex, and this effect involves GABAergic SOM+ interneurons. Our working hypothesis is that SXB potentiates the ability of these cells to inhibit all other cortical cell types via a GABAb mechanism, thus promoting the transition from ON to OFF periods during NREM sleep.

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Poster

P634. Sleep: Pharmacological and Cellular Manipulations

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Program #/Poster #: P634.06

Topic: F.08. Biological Rhythms and Sleep

Support: Department Of Anesthesiology, University of Virginia Health Systems

Title: Intensive Care Unit delirium is associated with impaired expression of key genes that regulate Rapid Eye Movement sleep and circadian rhythm

Authors: H. OSURU, E. DULKO, J. KLOS, N. ATLURI, M. ILLENDULA, T. LE, Z. ZUO, N. LUNARDI;
Anesthesiol., Univ. of Virginia Hlth. Systems, Charlottesville, VA

Abstract: Background: Delirious patients admitted to the Intensive Care Unit (ICU) after anesthesia and surgery commonly experience loss of Rapid Eye Movement (REM) sleep and circadian rhythm disruption. Experimental studies indicate that *BDNF* plays a crucial role in the gating of light inputs to the brain circadian pacemaker and that muscarinic acetylcholine receptors *Chrm1* and *Chrm3* are essential for REM sleep.**Objective**: To validate our recently established mouse model of ICU delirium by testing for loss of REM sleep and circadian integrity, and for impairment of *BDNF, Chrm1* and *Chrm3* expression, in aged mice subjected to anesthesia, surgery and Intensive Care environment (A/S/I). **Methods:** Aged C57BL/6J mice were randomized to insult (A/S/I) or control group and implanted with electroencephalographic (EEG) and electromyographic (EMG) electrodes for sleep recording. After 2 weeks, A/S/I animals were subjected to 3 h of sevoflurane anesthesia and laparotomy, followed by 2 h of propofol sedation and 12 h of ICU environment. Control mice did not receive A/S/I. 24 hour-long EEG/EMG recordings were obtained at the end of ICU conditions. Hippocampal tissue was collected at completion of EEG recordings for gene and protein expression analysis. **Results:** A/S/I mice exhibited a significant decrease in REM sleep during 12 hours of lights on (rest phase in mice) following ICU conditions. A/S/I mice also spent more time awake with lights on and showed a tendency to spend more time asleep with lights off (active phase), suggesting dysfunction of the internal circadian pacemaker. The expression level of several *BDNF* gene exons was decreased significantly in the hippocampus of A/S/I mice relative to controls (*exon I*:*, p=0.014; *exon IIa*:**, p=0.001; *exon IIb*:*, p=0.01; *exon IV*:**, p=0.006; *exon VI*:**, p=0.005; *exon VII*:**, p=0.004). *BDNF exons IIc, III, V, VIII* and *IXA* levels were unchanged. The expression of *Chrm1* and *Chrm3* genes was also reduced in A/S/I mice, while that of *chAT, Chrm2, Chrm4,* and *Chrm5* remained unchanged. Total *BDNF* protein levels were decreased by 22% and protein levels of *Chrm1 and Chrm3* by 16.4% and 15.5%, respectively, in A/S/I mice compared to controls.**Conclusions:** A/S/I caused disruption of circadian rhythm and loss of REM sleep in aged mice. These sleep-wake changes were associated with impaired expression of *BDNF, Chrm1* and *Chrm3*, key genes for the regulation of circadian rhythm and REM sleep. The changes in circadian rhythm and REM sleep induced by A/S/I are consistent with those experienced by patients with ICU delirium, and support the use of our animal model for future mechanistic studies on the role of sleep disturbances in the pathogenesis of delirium.

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Poster

P634. Sleep: Pharmacological and Cellular Manipulations

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Program #/Poster #: P634.07

Topic: F.08. Biological Rhythms and Sleep

Title: Detection and pharmacological modulation of sleep spindles in rat: a tool for safety/efficacy pharmacology using EEG telemetry recording in the conscious rat

Authors: A. SALIN[1], **F. ADRAOUI**[1], S. LOIODICE[1], N. KAUSHAL[2], P. ODONNELL[3], J. BLIESATH[2], M. PAPAKOSTA[2], D. BARTKOWSKI[2], B. JEFFY[2], S. MURPHY[2], S. NOURRY[1], J. MODOLO[4], F. WENDLING[4], E. CAYRE[1], R. HODGSON[2], *C. DRIEU LA ROCHELLE[1];
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Abstract: Sleep spindles (SPI) are physiological events that occur during Non-Rapid Eye Movement (NREM) sleep in both human and rodents. They are thought to be critically involved in cognitive functions and can be measured using electroencephalography (EEG), in rodents and humans using similar modalities. From a drug development perspective, the evaluation of a drug effect on SPI can therefore provide valuable information on target engagement, efficacy and/or safety. Cav3.3 (T-type calcium channel) is known as an important risk gene for schizophrenia and Cav3.3 knock out mice show sleep spindle deficits similar to schizophrenia patients. The aim of the present study was to evaluate the effect of acute administration of a Cav3.3 blocker Compound-5j (Benzecon et al. 2017), in comparison to gamma-butyrolactone (GBL) and Zolpidem on SPI density in rats (i.e. number of SPI over NREM sleep duration).
Eight Sprague-Dawley male rats were instrumented with a telemetry device (TL11M2-F40-EET, Data Sciences International, St Paul MN, USA) under isoflurane anaesthesia. Global cortical EEG and electromyogram (EMG) signals were recorded from leads of the telemetry implant fixed on the skull and the dorsal muscles of the neck, respectively. After a post-surgery recovery period, animals were dosed on separate occasions during the light phase with vehicle (0.5% methylcellulose, *po*), Zolpidem (30 mg/kg, *ip*), GBL (250 mg/kg, *ip*) and Compound-5j (0.3, 3, 30 or 100 mg/kg, *po*) according to a cross-over study design with a 48h wash-out period between two recording sessions. EEG and EMG recording were performed during a 1-hour baseline period, and over two hours post-dosing. Visual EEG/EMG analysis of the whole recording for SPI number and density was performed by a trained-experimenter blinded to the treatment.
No difference in baseline values for sleep duration and SPI density was observed between groups. Although at the dose used, Zolpidem had no effect on NREM sleep duration and SPI density, an overall decrease in NREM sleep duration and SPI density was measured in animals dosed with GBL when compared to controls. Interestingly, Compound-5j was associated with a dose-dependent and significant decrease of SPI density, but without any modification of NREM sleep duration.
In conclusion, the present study is the first to identify a significant modification of SPI activity following pharmacological inhibition of Cav3.3 channels using acute oral dosing with the Cav3.3 blocker Compound-5j. This result is in line with previous work evidencing the critical role of Cav3.3 channels in SPI activity using genetic models.

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Poster

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Title: Unique EEG signature of a $GABA_A$-slow agonist; Propofol vs BB

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Abstract: IV anesthetic agents like propofol produce increased power in slow delta frequencies (0.1 to 3 Hz), with a general decrease in EEG frequencies above 30 Hz. Propofol is non-selective for $GABA_A$ response subtypes, it enhances all three $GABA_A$-subtypes (slow, fast and tonic). A new anesthetic, BB, selectively targets $GABA_A$-slow synapses to depress brain responsiveness. We hypothesized that a selective GABAA-slow agonist, BB, would produce a different EEG signature compared to the broad spectrum $GABA_A$ agonist (propofol), and tested this using rat EEG recordings. Male rats were used following IACUC approval from the US Army Medical Research Institute of Chemical Defense. Rats were anesthetized using isoflurane (3-5% induction, 1-3% maintenance; with oxygen @ 0.5-1.0 L/min. Stainless steel screw electrodes placed in drilled holes served as cortical EEG leads. Propofol produced an increased power in slow delta frequencies (0.1 to 4 Hz) with a general decrease in EEG frequencies above 30 Hz at loss of consciousness (LOC). BB increased theta activity markedly (3-5 Hz and 8-10 Hz), and slightly increased delta power, but did not depress high frequency responses above 30 Hz. Neither agent produced burst suppression activity at LOC. Both anesthetics produced a characteristic flattening of chaotic attractors, similar to volatile and dissociative anesthetics at LOC. Propofol's EEG effects were in agreement with those seen in previous studies across individuals and species. At LOC a generalized slowing in EEG was seen with increased power in frequencies below 4 Hz. BB produced a markedly different EEG pattern, with a selective increase observed in the theta frequency range. This is interesting because $GABA_A$ slow

synapses have previously been suggested to underlie theta frequency oscillations, while fast synapses control high, gamma frequency oscillations (30-60 Hz). Tonic $GABA_A$ responses produce a generalized depression of neuronal activity across all frequencies. BB and propofol shared the ability to flatten EEG chaotic attractors at LOC, similar to volatile and dissociative anesthetics. Flattened attractors are also seen in humans and thought to reflect a decrease in EEG information content at LOC. It appears that propofol's effects on fast and/or tonic responses contribute to it's respiratory and cardiovascular unwanted side effects, since these were not produced by BB.

Disclosures: B. MacIver: None. **S.L. Eagleman:** None. **M.F. Davies:** None. **H.S. McCarren:** None. **E.J. Bertaccini:** None.

Poster

P634. Sleep: Pharmacological and Cellular Manipulations

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P634.09

Topic: F.08. Biological Rhythms and Sleep

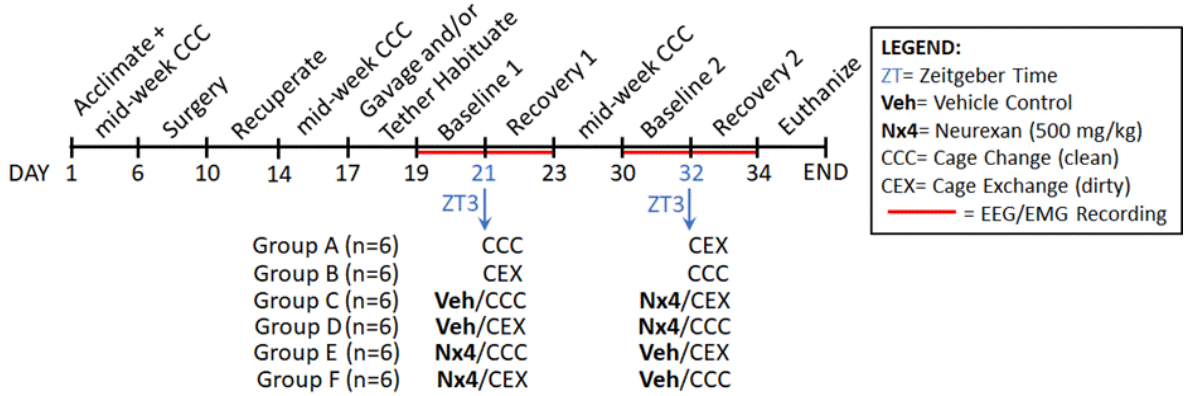
Support: Fully funded by Heel GmbH

Title: Multicomponent drug Neurexan mitigates stress-induced acute insomnia

Authors: *C. J. DAVIS[1], M. A. SCHMIDT[1], N. KRÖEMMELBEIN[2], B. SEILHEIMER[2], K. HEMMER[2];
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Abstract: Fast-paced lifestyles can lead to decreased health and wellbeing, stress-related symptoms, and in many cases sleep disturbances including insomnia. The objective of the current study was to determine whether the natural, multicomponent drug Neurexan (Nx4) could mitigate acute insomnia resulting from exposure to an acute stressor. We administered Nx4 orally to rats and examined the sleep-wake EEG phenotypes with the male rat dirty cage exchange method (CEX) reported by Cano et al., (2008). We selected the CEX model as the molecular and electrophysiological correlates are characterized, and best represents human insomnia conditions. Adult male Sprague-Dawley rats (n=36; CEX;) were randomly assigned as part of a crossover design to one of 6 treatment groups: 1. change over to a clean cage (CCC), or 2. exchange to a dirty cage soiled for a week by another rat (CEX), or 3. vehicle gavage followed by transfer to a clean cage (Veh CCC), or 4. a dirty cage (Veh CEX), or 5. Nx4 gavage followed by transfer to a clean cage (Nx4 CCC) or 6. a dirty cage (Nx4 CEX). Rats were surgically instrumented with cortical EEG electrodes, habituated to a cable tether, and biopotentials recorded (Fig 1). Non-rapid eye movement sleep (NREMS), rapid eye movement sleep (REMS),

and waking states were determined manually off-line in 10 sec epochs. Investigators were blinded to treatments during sleep scoring. One-way ANOVAs compared treatment means for sleep latency and for 2 hrs after cage change for time in each state, state-specific episode duration and frequency. Mixed ANOVAs of time and treatment compared NREMS delta (0.5-4 Hz), NREMS gamma (30-60 Hz) and REMS theta (5-9 Hz) spectral windows. Nx4 CEX rats fell asleep 30.4 mins faster than Veh CEX rats and spent less time awake, with shorter and more waking episodes than Veh CEX rats. Moreover, Nx4 CEX rats were not different from Nx4 CCC rats on any outcomes. No changes between any treatment groups were detected in the EEG spectral windows examined. This research confirms that Nx4 treatment has somnogenic effects and ameliorate psychological stressor-induced acute insomnia.



Disclosures: **C.J. Davis:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Independent PI for subcontract by drug maker. **M.A. Schmidt:** None. **N. Kröemmelbein:** A. Employment/Salary (full or part-time):; employed by Heel GmbH at an early stage of study implementation. **B. Seilheimer:** A. Employment/Salary (full or part-time):; employed by Heel GmbH. **K. Hemmer:** A. Employment/Salary (full or part-time):; employed by Heel GmbH.

Poster

P634. Sleep: Pharmacological and Cellular Manipulations

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Program #/Poster #: P634.10

Topic: F.08. Biological Rhythms and Sleep

Support: Wellcome Trust PhD studentship 203971/Z/16/Z

Title: Effects of clozapine-N-oxide and Compound 21 on sleep in laboratory mice

Authors: *J. TRAUT[1,2,4], J. MENGUAL[1,2], E. MEIJER[1,2], L. E. MCKILLOP[1,2], H. ALFONSA[3], A. HOERDER-SUABEDISSEN[1], S. M. SONG[5], Z. MOLNÁR[1], C. J. AKERMAN[3], V. V. VYAZOVSKIY[1,2], L. B. KRONE[1,2];
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Abstract: Designer Receptors Exclusively Activated by Designer Drugs (DREADDs) are chemogenetic tools to activate or inhibit a targeted cell population. However, recent reports suggest that the prototypical DREADD agonist, Clozapine-N-Oxide (CNO), can affect brain activity and animal behaviour, putatively through conversion to clozapine. Despite the common use of DREADDs in neuroscience and sleep research, the potential effects of CNO upon sleep have never been systematically tested.We assessed sleep in 16 male C57BL/6 mice after intraperitoneal injections of commonly used doses of CNO and Compound 21 (Cmpd-21), a DREADD agonist which cannot convert to clozapine. Mice were housed under a 12/12 h light/dark cycle, chronically recorded with electroencephalography (EEG) and electromyography (EMG), and injected at light onset. All mice received saline and 5 mg/kg CNO in a counterbalanced design. Some animals also received 10 mg/kg CNO (n=14), 1 mg/kg CNO (n=12), and 3 mg/kg Cmpd-21 (n=7). Conditions were separated by a rest interval of at least 72 hours. Compared to saline, CNO suppressed rapid eye-movement (REM) sleep over the entire six hour observation period ($F_{1.788,22.05} = 7.382$, $p = 0.005$). Changes in sleep architecture consisted of reduced sleep onset latency and increased REM latency, particularly in the 1 mg/kg condition, as well as an increased duration and reduced number of non-REM (NREM) episodes for the 5 mg/kg and 10mg/kg CNO conditions in line with a report by Varin et al. 2018. We also found an increased duration and reduced number of REM episodes for 10 mg/kg CNO. EEG NREM power spectra showed a significant power increase between 0.5-1.25 Hz and decrease above 6 Hz in the 5 mg/kg CNO condition. In addition, CNO reduced the frequency of brief awakenings (4-16s wakefulness, $F_{1.968,23.62} = 10.38$, $p < 0.001$) during the first two hours after injection. Interestingly, Cmpd-21 also suppressed REM sleep, increased the duration and reduced the number of NREM episodes, and reduced the frequency of brief awakenings. Our preliminary results indicate that CNO and Cmpd-21 affect sleep. The pattern is similar to sleep after clozapine injections in rats (Sorge et al., 2004). However, the effects of Cmpd-21 suggest that even without clozapine back-metabolism, DREADD agonists can exert behavioural effects. This might be due to off-target binding at endogenous receptors (Jendryka et al., 2019). Our results require replication with alternative CNO and Cmpd-21 products and in an optimised experimental design. However, they indicate that chemogenetic experiments should include a DREADD-free control group and highlight the need for novel DREADD agonists and chemogenetic tools.

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Poster

P667. Neural Systems IV

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Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P667.01

Topic: G.02. Reward and Appetitive Learning and Memory

Support: R01MH110594
R01MH116937
McKnight Foundation to IEM

Title: Integrating information and reward into subjective value: lateral habenula activity predicts and causally influences online decision-making

Authors: *Y.-Y. FENG[1,2], E. S. BROMBERG-MARTIN[1], T. OGASAWARA[1], J. WHITE[1], K. ZHANG[1,2], I. E. MONOSOV[1,2,3,4];
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Abstract: Every day, humans and other animals make decisions based on our preferences. Prevalent economic and computational theories posit that to make these decisions, individuals must integrate each option's attributes to compute its total subjective value. This is thought to be supported by neural systems for reinforcement learning, which use reward prediction errors (RPE) to update the values of states and actions to guide future decisions. A promising neural substrate for such value learning is the lateral habenula (LHb), known to broadcast RPEs and regulate the activity of midbrain dopamine neurons and other neuromodulators. However, the precise function of LHb RPE signals in decision-making is unclear. In particular, it is unknown whether LHb RPE signals truly reflect overall subjective value, accounting for all attributes defining an option, and it remains unclear whether the LHb RPEs thought to control learning also influence online decision-making. To answer these questions, we designed a multi-attribute decision-making task for monkeys and humans which requires integrating preferences for both primary reward and advance information about uncertain future outcomes. This task allowed us to precisely model the computation of subjective value, by observing subjects' choices between options with independently varied attributes including reward amount, uncertainty, delay, and timing of information to resolve uncertainty. To test LHb's role in this computation, we recorded neurons in LHb and in one of its key inputs, the anterior/ventral pallidum (Pal). We find that many LHb neurons integrate attributes in a manner consistent with subjective value, while many Pal neurons signal attributes such as information and primary reward in distinct manners (see companion poster). We next asked what role the value-related signals in LHb play in determining the ultimate decision. Consistent with previous work, LHb activity was negatively related to value. Therefore, we hypothesized that stronger LHb responses to an option would decrease the option's value and discourage its selection. Indeed, analysis of trial-to-trial response variance indicated that stronger LHb responses to an option predicted a lower probability of

choosing that option. To assess whether LHb value-related activity causally influences decision-making, we used weak electrical stimulation. We find that stimulating LHb during an option's presentation biases choice away from that option in a manner consistent with decreasing its value. These results suggest that RPEs signaled by LHb reflect subjective value integrated across multiple attributes and may influence online decision-making.

Disclosures: Y. Feng: None. **E.S. Bromberg-Martin:** None. **T. Ogasawara:** None. **J. White:** None. **K. Zhang:** None. **I.E. Monosov:** None.

Poster

P667. Neural Systems IV

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Program #/Poster #: P667.02

Topic: G.02. Reward and Appetitive Learning and Memory

Support: R01MH110594
R01MH116937
McKnight Foundation Award to I.E.M.

Title: Integrating information and reward into subjective value: common computations in humans, monkeys, and basal ganglia-lateral habenula circuitry

Authors: *E. S. BROMBERG-MARTIN[1], Y.-Y. FENG[1], T. OGASAWARA[1], J. WHITE[1], K. ZHANG[4], I. E. MONOSOV[1,2,3,4,5];
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Abstract: When faced with an uncertain future, humans and animals can be highly motivated to seek information to resolve their uncertainty. Some individuals pay a great deal for this information even when it apparently has no 'objective' value, suggesting that information has a subjective value of its own. However, we know little about the algorithms the brain uses to (i) compute an internal estimate of uncertainty, (ii) compute the subjective value of information to resolve that uncertainty, and (iii) integrate both information and primary reward to guide decision making. To begin to address these questions, we trained humans and monkeys to perform multi-attribute decision making tasks. Individuals chose between pairs of options that provided different probability distributions of primary rewards (money for humans, juice for monkeys). In addition, once chosen, different options provided either informative cues that indicated the upcoming reward outcome, or non-informative cues that did not. Importantly, this design allowed individuals to trade off primary reward to gain information about their future outcomes, while ensuring that the information had no 'objective' value (i.e. information could be used to predict reward outcomes but not to control them). We then fit models to each

individual's choice data to estimate the subjective value of information. We found that in both humans and monkeys (1) most individuals treated information as valuable, (2) the value of information was greatest when reward was uncertain and the information would resolve the uncertainty sooner rather than later, (3) the value of information was tied to a specific form of uncertainty that was remarkably similar across species. To uncover how this subjective value of information is computed by neurons in the brain, we recorded neuronal activity in monkeys. We targeted the lateral habenula (LHb), a nucleus implicated in reward prediction error coding and value learning, as well as its major input sources in the basal ganglia, including the anterior/ventral pallidum (Pal). Many Pal and LHb neurons responded to opportunities to gain information, juice reward, or both. However, there was a key difference: Pal neurons commonly responded to information and juice reward in distinct manners, while LHb neurons commonly integrated them together in a manner closely resembling their total subjective value. Thus, our data sheds new light on the computations humans and animals use to value information, and suggest that pallidum-habenula circuitry may perform a key step in this computation: integrating information and primary reward into subjective value.

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Poster

P667. Neural Systems IV

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Program #/Poster #: P667.03

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIH Grant R01 DA033123

Title: The role of the prelimbic cortex in renewal of extinguished behavior based on a return to a satiated interoceptive context

Authors: *C. M. P. THOMAS, E. A. THRAILKILL, M. E. BOUTON, J. T. GREEN; Psychological Sci., Univ. of Vermont, Burlington, VT

Abstract: We have previously shown that the rat prelimbic cortex (PL) is necessary for acquisition contexts to promote the performance of their associated instrumental behaviors, whether that context is physical (operant chamber) or behavioral (recent performance of a chained behavior). In the present experiment, we investigated the role of the PL in satiety level as an interoceptive acquisition context. Rats were trained to lever-press for sweet/fat pellets while sated (22 hrs continuous food access) followed by the extinction of the response while hungry (22 hrs food deprived). Pharmacological inactivation of the PL (with baclofen/muscimol infusion) appeared to attenuate renewal of the response upon a return to the sated context. In

contrast, animals that received a vehicle (saline) infusion showed renewal of the previously extinguished response. In a follow up test, we examined the sensitivity of the response to a change in physical context while animals were sated (interoceptive acquisition context). Interestingly, no effect of a physical context change was detected in either group (PL-inactivated or control), although overall responding in the PL-inactivated group was lower than controls. This pattern of results is consistent with the interpretation that during the initial training and subsequent extinction of the response both groups learned that the relevant contextual element was satiety state and not the physical context. Collectively, these preliminary results support the hypothesis that the PL monitors the relevant contextual elements (physical, behavioral, or satiety state) associated with reinforcement of a response and promotes the subsequent performance of that response in their presence.

Disclosures: C.M.P. Thomas: None. **E.A. Thrailkill:** None. **M.E. Bouton:** None. **J.T. Green:** None.

Poster

P667. Neural Systems IV

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Program #/Poster #: P667.04

Topic: G.02. Reward and Appetitive Learning and Memory

Support: MRC Studentship to KF MR/N013867/1
Sir Henry Dale Fellowship to AL 213465
Human Science Frontiers Program grant to ABS RGY0076/2018
Sir Henry Dale Fellowship to ABS 200501

Title: Midbrain dopamine neurons provide teaching signals for goal-directed navigation

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Abstract: Successful goal-directed navigation requires learning to accurately estimate location and select optimal actions in each location. Midbrain dopamine neurons are known to be involved in reward value learning, action selection, and have also been linked to reward location learning. Dopamine neurons are therefore ideally placed to provide teaching signals for goal-directed navigation. To test this, we imaged dopamine neural activity using a genetically-encoded calcium indicator and Miniscope as mice learned to navigate in a closed-loop virtual reality corridor and lick to report the reward location. Across learning, phasic dopamine responses developed that resembled reward prediction errors (RPEs) and indicated the animal's estimate of the reward location. We also observed the development of pre-reward ramping activity, the slope of which was modulated by both learning stage and task engagement. The

ramp slope was inversely correlated with locomotor speed, indicating that the ramp did not reflect motor vigour. The slope of the ramp was correlated with the accuracy of reward zone licks in the next trial, suggesting that the ramp sculpted accurate location-specific action during navigation. Both phasic and ramping activity were consistent with temporal difference error in a Q-learning model that incorporated belief about the current state of the agent. Preliminary analysis also shows that the ramping activity was also present in individual dopamine neurons, with variance in ramp slope across neurons. Our results indicate that midbrain dopamine neurons, through both their phasic and ramping activity, provide teaching signals for improving goal-directed navigation.

Disclosures: K. Farrell: None. **A. Lak:** None. **A.B. Saleem:** None.

Poster

P667. Neural Systems IV

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Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIH F99NS115270
NSF IOS 1557987
NIH R01DA044199

Title: Activity dynamics in the central nucleus of the amygdala during habit formation

Authors: *K. A. AMAYA, J. E. CARMICHAEL, J. A. PALMER, K. S. SMITH; Psychological and Brain Sci., Dartmouth Col., Hanover, NH

Abstract: Habits are behaviors that are notoriously automatic, consistent, and inflexible and emerge with repeated performance of an action. Neural recordings of key habit-promoting brain areas like the infralimbic cortex, dorsolateral striatum (DLS), and substantia nigra have characterized activity dynamics while a habit is formed and executed (Graybiel and Smith, 2013; Jin and Costa, 2010). Meanwhile, as the central nucleus of the amygdala (CeA) has been extensively studied with respect to threat learning, a smaller body of literature has implicated this amygdalar nucleus as being involved in reward seeking and habits as well. In particular, the CeA has been shown to be necessary for the general motivational influence of Pavlovian cues over instrumental behaviors and, separately, lesions to the CeA abolish habit formation (Corbit and Balleine, 2005; Lingawi and Balleine, 2012). Additionally, although there are no known direct projections between the two areas, contralaterally lesioning DLS and CeA similarly abolishes habit formation, further solidifying the idea that the CeA is involved in the habit formation process. To formally address this, we trained animals on an elevated plus-maze task that consistently required a specific response to earn rewards and subsequently quantified c-Fos

expression in the CeA. In the next experiment, we implanted animals with silicon probes targeting the CeA and recorded neural activity throughout training and during post-outcome devaluation extinction and reacquisition sessions. Our findings show that CeA neurons are recruited during habit formation (as evidenced by c-Fos expression) and that CeA neural activity may fluctuate as habits are formed to accentuate pre- and post-reward delivery.

Disclosures: K.A. Amaya: None. **J.E. Carmichael:** None. **J.A. Palmer:** None. **K.S. Smith:** None.

Poster

P667. Neural Systems IV

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Program #/Poster #: P667.06

Topic: G.02. Reward and Appetitive Learning and Memory

Support: WT 095495
WT 204811
ERC 293549

Title: Dopamine neurons signal 'true' subjective value (willingness-to-pay) prior to bidding in a Becker-DeGroot-Marschak auction-like mechanism in rhesus macaques.

Authors: *D. F. HILL, R. W. HICKMAN, W. SCHULTZ;
Univ. of Cambridge, Cambridge, United Kingdom

Abstract: Dopamine neurons play a prominent a role in choice behavior by signaling the utility of choice options when animals are presented with their associated cues. Dopamine neurons have also been shown to encode the relative value of chosen goods prior to choices being made, altering activity based on which good will be chosen. Whether these responses reflect the learned probability of reward, or an intrinsic, subjective value estimation remains unclear. To test whether dopamine neurons encode an estimate of one's 'true' subjective value, we trained rhesus monkeys to bid in a Becker-DeGroot-Marschak (BDM) auction to elicit their willingness-to-pay for juice rewards while recording from single midbrain dopamine neurons. The BDM is unique in that it is incentive compatible; that is, the best strategy is to bid one's 'true' value. Critically, the BDM offers a trial-by-trial measure of utility that more accurately resembles real-world moment-to-moment value fluctuations. Monkeys were trained to associate images of fractals with varying amounts of juice reward. They were then allowed to bid for the juice indicated by a specific fractal while paying the associated price from a fixed 'budget' of water. After their bid, they were shown the opponent's bid (i.e., random computer bid), revealing whether they had won or lost the auction. In two monkeys, dopamine responses to fractal cues reflected the bid that the animals would subsequently make, even when reward magnitude was held constant. That

is, when presented with fractals representing three separate reward levels, dopamine neuron responses varied with the monkeys' bids prior to the bids being made. Moreover, responses correlated with bids when analyzing reward levels individually. These data show that dopamine responses reflect an intrinsic value estimate of goods prior to actions taken to acquire them. These responses allow dopamine neurons to be involved in guiding actions to obtain the highest utility.

Disclosures: D.F. Hill: None. **R.W. Hickman:** None. **W. Schultz:** None.

Poster

P667. Neural Systems IV

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Program #/Poster #: P667.07

Topic: G.02. Reward and Appetitive Learning and Memory

Support: P50 MH096889
T32 DA050558
T32 GM008620
F30 MH126615-01A1

Title: The paraventricular nucleus of the thalamus contributes to early-life adversity-induced deficits in reward-related behaviors

Authors: *C. L. KOOIKER[1], M. T. BIRNIE[2], Y. CHEN[3], T. Z. BARAM[4];
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Abstract: Rationale: Early-life adversity (ELA) is associated with poor cognitive and emotional health and increased risk for a variety of affective disorders, such as major depressive disorder and bipolar disorder. Many of these disorders are characterized by impairments in reward-related behaviors, and we find that these same deficits are provoked by rodent models of ELA. However, the brain regions and processes underlying these long-term consequences of ELA remain largely unknown. The paraventricular nucleus of the thalamus (PVT) is an important component of the reward circuit that encodes remote emotionally salient experiences to influence future motivated behaviors. We hypothesize that the PVT encodes adverse experiences as remote as the early postnatal period in mice, and that ELA-engaged PVT neurons subsequently contribute to deficits in reward-related behaviors in adults. **Methods:** We employ TRAP2 mice, which we exposed to a week of simulated ELA in a limited-resource cage between postnatal days 2-9. We induced the TRAP2 system using tamoxifen on P6, triggering Cre-dependent recombination in neurons activated during P6-P8. This leads to permanent marking of neurons activated during this epoch. We validated our findings using routine cFos immunohistochemistry

in WT mice. **Results:** ELA robustly and selectively activates PVT neurons to a degree much higher than typical rearing conditions. We are now characterizing the molecular phenotype of these ELA-engaged neurons, focusing on the stress-sensitive neuropeptide CRH and its receptors. Finally, we aim to silence these ELA-engaged PVT neurons during reward-related tasks in adult mice, with the possibility of ameliorating the observed ELA-induced deficits in reward-seeking behaviors. **Conclusion:** The PVT robustly and almost uniquely expresses cFos in response to emotionally salient events in neonatal mice. The PVT is thus poised as a potential contributor to deficits in reward-related behaviors following ELA.

Disclosures: C.L. Kooiker: None. **M.T. Birnie:** None. **Y. Chen:** None. **T.Z. Baram:** None.

Poster

P667. Neural Systems IV

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Program #/Poster #: P667.08

Topic: G.02. Reward and Appetitive Learning and Memory

Support: K08 MH109735
NARSAD grant from the Brain Behavior Research Foundation

Title: Acute stress impacts reward consumption via different VTA neural activity than reward anticipation

Authors: D. C. LOWES[1], A. A. AMILCAR[2], E. S. HOLT[3], L. YUSUFOVA[2], ***A. HARRIS**[4]; [1]Pharmacol., [2]Columbia Univ., New York, NY; [3]Res. Fndn. for Mental Hyg., New York, NY; [4]Psychiatry, Columbia University/Research Fndn. for Mental Hyg., New York, NY

Abstract: Acute stress impairs reward seeking, but the underlying neural circuitry deficits are incompletely understood. We recently demonstrated that acute stress impairs reward anticipation ventral tegmental area (VTA) GABA neurons projecting to the nucleus accumbens (NAc), but until now there has been little evidence of the effect of stress on reward consumption. We recorded single unit activity in the VTA as mice engaged in a cued-reward task after exploring a familiar environment and after exposure to restraint stress. We found that stress decreases reward consumption independent of its effects on reward anticipation (N=13 mice, $p<0.01$). Surprisingly, while we recently demonstrated that restraint-induced 4 Hz NAc LFP oscillation strength correlates with reward anticpation deficits, this oscillation did not correlate with the effect of restraint on reward consumption. In addition, while stimulating NAc-projecting VTA GABA neurons impairs reward anticipation, it spares reward consumption (N=5 mice, $p=0.47$). Finally, we found that restraint impairs the firing of putative VTA DA neurons during reward consumption ($n_{Familiar} = 80$ neurons, $n_{Restraint} = 80$ neurons, $p<0.05$) but had no effect on putative VTA GABA neurons during reward consumption ($n_{Familiar} = 29$ neurons, $n_{Restraint} = 22$

neurons, p>0.5). Together, these data suggest that stress impairs reward consumption via altered VTA DA activity, while stress impairs reward anticipation via altered VTA GABA activity.

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Poster

P667. Neural Systems IV

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Program #/Poster #: P667.09

Topic: G.02. Reward and Appetitive Learning and Memory

Title: Role of Ventral Midbrain Dopamine Neurons in Familiarity

Authors: *R. KOLARIC[1], S. FLEURY[1], A. TOFT SØRENSEN[2], U. GETHER[3], S. MINGOTE[1];
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Abstract: It is well established that dopamine (DA) neurons in the ventral midbrain signal novelty by increasing their bursting activity, while decreasing it after repeated inconsequential stimulus presentations. Thus, the lack of DA neuron burst activity is a physiological signature that a particular stimulus has become familiar. Here, we tested the hypothesis that decreasing DA neuron activity while animals explore objects for the first time enhances familiarization and improves subsequent novel object recognition (NOR).
NOR is dependent on the amount of pre-exposure to the familiar object, and we showed that one familiarization session results in poor novelty discrimination. The lack of increased exploration of a novel object is thought to reflect a stage of equal attention allocated to the familiar and novel stimulus. To investigate whether decreasing DA neuron activity prompts gain-of-function in NOR, we chemogenetically inhibited DA neurons in the ventral tegmental area (VTA) during the familiarization session of this NOR paradigm.
We injected an AAV *flp*-dependent hM4Di into the VTA of TH-*flp* transgenic mice to selectively inhibit DA neurons. The selectivity of the viral strategy was verified via immunohistochemistry staining. The virus was then activated via injection of Clozapine N-oxide dihydrochloride (CNO; i.p.) 30 minutes before trial. We found that inhibiting DA neurons during the familiarization phase resulted in enhanced novel object recognition in hM4Di+CNO male mice compared to controls, using the discrimination index (DI) for the time exploring the objects. Additionally, we observed similar effects in hM4Di+CNO female mice compared to controls, although sex-mediated differences in exploratory behavior emerged. Female mice tended to explore through shorter but more frequent burst-like visits to the object compared to

male mice whose visits to the objects tended to be longer but less frequent. Although hM4Di+CNO female mice showed no increase in DI for time exploring the objects, the DI for the average time per visit revealed a significant difference between groups, indicating that females also show increased discrimination. Finally, to confirm these results were due to an effect on familiarization processes, we conducted further trials in which DA neurons were only inhibited after the familiarization phase and during subsequent NOR session. Under this manipulation, we did not see an improvement in discrimination. These results suggest that suppressing DA activity ameliorates novelty discrimination by enhancing familiarity in both females and males despite differences in patterns of exploratory behavior.

Disclosures: R. Kolaric: None. **S. Fleury:** None. **A. Toft sørensen:** None. **U. Gether:** None. **S. Mingote:** None.

Poster

P667. Neural Systems IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P667.10

Topic: G.02. Reward and Appetitive Learning and Memory

Title: A data driven approach to analyze rat ultrasonic vocalizations

Authors: K. LAWSON[1], A. FLORES[1], R. HOEKENSON[1], C. M. RUIZ[1], S. V. MAHLER[2]; [1]Univ. of California, Irvine, Irvine, CA; [2]Neurobio. and Behavior, Univ. of California Irvine Dept. of Neurobio. and Behavior, Irvine, CA

Abstract: Rats emit ultrasonic vocalizations (USVs) to convey emotional arousal, and/or as a form of social communication about salient rewards and threats in the local environment. In adult rats, these USVs fit into two broad categories; “22kHz” calls that are produced when the rat is in a painful or dangerous situation, and “50kHz” calls, which are emitted during experience or anticipation of rewards like play, mating, or certain drugs of abuse. Relatively recent advances in USV detection hardware and analysis software have made it possible to examine more nuanced aspects of rat USVs, for example showing that “50kHz” calls occur at a range of frequencies and patterns—with up to 14 subtypes (Wright et al., 2010). It is not presently clear whether these complex signals contain information that could be of use to behavioral neuroscientists for interrogating the subjective states of rats.
Here, we examined USVs emitted by male and female Long Evans rats under a variety of circumstances, in hopes of narrowing down the information that is potentially contained within these mysterious vocalizations. We recorded USVs after administration of drugs like amphetamine, heroin, and ketamine, and examined how chemogenetic inhibition of nucleus accumbens or ventral tegmental area dopamine neurons impacted the rate of USV production. Further, we validated and employed Deepsqueak (Coffey et al., 2019) software to more precisely

identify the acoustical features of USVs, generating large datasets which allowed us to employ high dimensionality reducing analyses to determine how the characteristics of USVs are altered by these pharmacological and chemogenetic manipulations. We expect that these studies will facilitate further data-driven investigations into the insights USVs may afford into rats' subjective states during behavioral neuroscience experiments.

Disclosures: K. Lawson: None. **A. Flores:** None. **R. Hoekenson:** None. **C.M. Ruiz:** None. **S.V. Mahler:** None.

Poster

P719. Stress, Fear, and Anxiety: Animal Models

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Program #/Poster #: P719.01

Topic: F.04. Stress and the Brain

Support: NIH (R01MH104373)
VA (I01 BX005118)

Title: Increased Startle Response and Anxiety-Like Behavior in a Mouse Model of Traumatic Stress

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Abstract: Post-traumatic stress disorder (PTSD) is a stress-and trauma-related disorder affecting approximately 8% of the population, with higher incidence rates among women and the military. PTSD is associated with hyperactivity of the amygdala, a key region for adaptive stress and fear processing. Amygdala hyperactivity is thought to be the cause of hyperarousal symptoms of PTSD, which include hypervigilance, increased startle response, heightened anxiety, and perturbed sleep. Specifically, traumatic stress disrupts the excitatory and inhibitory balance within the basolateral nucleus of the amygdala (BLA), resulting in long-term maladaptive stress processing. The purpose of this study is to determine the impact of intense, uncontrollable stress on hyperarousal symptoms and fear learning in a mouse model. Five-week-old male and female C57Bl6/J mice were tested on day 1 for their baseline acoustic startle response (ASR) to 15 110 dB white noise bursts delivered at a 30-s inter-tone interval. On days 2-4, the mice were exposed to 3 consecutive days of traumatic stress, which consisted of 40 2-mA, 2-s shocks while restrained over a 2-h period. The mice were retested on days 9-11 for ASR and were assessed for anxiety-like behavior using the elevated plus maze (EPM) and open field test. Cued fear conditioning was performed on day 15 and fear memory extinction was assessed on days 16-19. Finally, on day 22, mice were tested for ASR a final time. Mice exposed to the traumatic stress paradigm had significantly higher ASR on days 11 ($p<0.05$) and 22 ($p<0.001$) compared to

baseline, while control mice showed a similar ASR across the three time points. Mice exposed to traumatic stress showed an increase in anxiety-like behavior, spending less time in the open arms of the EPM ($p<0.05$) and more time in the perimeter of the open field compared to control mice ($p<0.05$). Finally, traumatic stress-exposed mice showed greater fear memory acquisition ($p<0.05$) but no difference in fear extinction compared to control mice. Our results indicate that this traumatic stress paradigm induces a hyperarousal phenotype and increases anxiety-like behavior in the mouse. We are using this model to further assess the cellular and molecular mechanisms underlying these behavioral endpoints induced by traumatic stress.

Disclosures: B.L.W. Sweeten: None. **M.T. Watson:** None. **J.G. Tasker:** None.

Poster

P719. Stress, Fear, and Anxiety: Animal Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P719.02

Topic: F.04. Stress and the Brain

Support: NIDDK 1-R01 DK105826

Title: Estrogen receptor beta in oxytocin neurons mediates social behavior and hypothalamic-pituitary-adrenal axis stress response in novel transgenic mouse line

Authors: *J. A. SHENG, R. M. DANIELS, A. M. MILLER, S. A. MYERS, N. J. BALES, R. J. HANDA;
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Abstract: Estrogens (E) influence a myriad of neuroendocrine and behavioral functions. The dichotomous effects of E are regulated by two specific receptors in the brain, E receptor alpha (ERα) and E receptor beta (ERβ). These receptors have been shown to induce opposing actions on the hypothalamic-pituitary-adrenal (HPA) axis stress response and related behavior. While ERα is generally shown to produce an anxiogenic effects, ERβ produces anxiolytic effects. Reports further demonstrate ERs in modulating neuropeptide systems that drive sex differences in stress regulatory systems. Estrogen receptor β is predominantly co-expressed with oxytocin (OT) in the paraventricular nucleus of the hypothalamus (PVN), the core regulator of the HPA axis. Therefore, Es have been highly implicated in the OTergic system in mediating the stress response and associated behaviors. In this study, we used Cre/loxP recombination to delete exon 3 of the ERβ gene in OT neurons, generating a novel $OT^{cre}/ER\beta^{loxP/loxP}$ mouse model on C57BL/6 background. Controls used were $ER\beta^{loxP/loxP}$ or OT^{cre} mice. To validate this model, droplet digital PCR was used to quantify ERβ mRNA in the PVN, which showed ~50% reduction ($p<0.0001$) in adult M and F mutants (n=8/sex). Using a 3-chamber apparatus (n=12-15/sex), we next examined social behavior. In the social discrimination test, M and F mutants

showed decreased social interaction with the stimulus mouse (S1; p=0.0481). In the social recognition test, a second stimulus mouse (S2) was placed at the opposite end of S1 in the apparatus. While control F preferred interacting with S2 (p=0.0461), mutant F preferred the familiar S1 (p=0.0218). Interestingly, control M preferred S2, but mutant M interacted with S1 and S2 equally. In the social memory test, interaction with a 3rd stimulus mouse (S3) versus S1 was examined. Preference for familiar mouse in this test was observed in only mutant F (p=0.0313). Social behavior results implicate ERβ in the OTergic system in promoting social behavior in F but not M. Finally, stress-induced corticosterone secretion was decreased in mutant versus control F following acute restraint (p=0.0121), but not in mutant M (n=12-15/sex). This suggests Es actions on ERβ in OT neurons stimulate the HPA axis stress response in F, but not M. Taken together, these data indicate an important role of ERβ in the OTergic system in regulating sex differences in behavioral and neuroendocrine physiology. Understanding actions of E on these physiologic functions could be important for determining the underlying role of sex in stress-related diseases in humans, such as social disorders, anxiety, and depression. Supported by NIDDK 1-R01 DK105826

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Poster

P719. Stress, Fear, and Anxiety: Animal Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P719.03

Topic: F.04. Stress and the Brain

Support: NIH Grant R21MH122862 to JMS

Title: Impact of Peripartum Stress on Postpartum Maternal Behavior and Associated Endocrine and Neurobiological Substrates

Authors: *J. PLUCHINO, J. GIFFORD, J. SCHWARZ;
Univ. of Delaware, Newark, DE

Abstract: The purpose of this study is to measure the impact of prenatal stress on postpartum maternal behavior in addition to the levels of estrogen receptor alpha (ERα), specifically in the medial preoptic area (mPOA), as well as measuring the effects of stress on anhedonia by sucrose preference test. A limited bedding and nesting (LBN) condition was applied to half of the cohort. They received 250 mL of bedding compared to the control group which received 4000 mL of bedding. The purpose of this condition is to simulate a stressful environment which simulates stressors a new mother might experience when lacking adequate resources to care for her infant. Maternal behaviors of LBN and control animals were recorded twice daily for 30 minute time

periods, once in the morning and once in the evening. The footage was then analyzed and hand-scored on a minute-by-minute basis, choosing from a list of 17 behaviors. A mixed model analysis was used to analyze the data. A sucrose preference test was performed to measure for anhedonia using a 1% sucrose solution for 48 hours at 4 timepoints throughout the study (prior to breeding, E20-22, P0-2, P7-9). Additionally, tissue samples were collected from a total of 60 animals (same animals that were observed with maternal behavior) and were sliced at 30 μm. Slices were then stained, mounted and analyzed via microscope to examine cell count of ERα in the mPOA. The analysis of maternal behavior, although ongoing, is anticipated to exhibit a decrease in positive maternal behaviors within the LBN group, characterized by postures such as arch back nursing, pup licking, and retrieving pups. In regards to the sucrose preference test, we found the most significant decrease in preference among both LBN and control groups during the P0-P2 and P7-P9 time points when compared to baseline measurements. Specifically, in the P7-P9 time point, there are distinct groups of dams with high and low preference, suggesting that stressors do not have a uniform effect on the entire cohort, regardless of group condition. In regards to the stained tissue, we anticipate a larger proportion of ERα found in the mPOA to be correlated with more positive maternal behavior. This analysis is ongoing. With a lack of research which specifically focuses on postpartum depression, the use of an animal model proposes a new approach in observation. The findings of this study should prove to be beneficial in elucidating biomarkers of PPD to aid in the proper and accurate diagnosing of cases.

Disclosures: J. Pluchino: None. **J. Gifford:** None. **J. Schwarz:** None.

Poster

P719. Stress, Fear, and Anxiety: Animal Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P719.04

Topic: G.04. Emotion

Title: Characterizing the Role of Dopamine D3 Receptors on Dopaminergic Neurons on Anxiety like Behavior

Authors: *M. SHAW[1], M. BOCARSLY[2], V. A. ALVAREZ[1];
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Abstract: Dopamine transmission in the striatum plays an important role in alcohol reward and reinforcement and is mediated by a family of G protein-coupled dopamine receptors, dopamine D1 - D5. The D1 and D2 receptors are often the focus of addiction research, and while the dopamine D3 receptor (D3R) has been indicated in alcohol seeking and relapse, very little is known about the specific role of this receptor. The purpose of this project is to behaviorally characterize the role of the dopamine D3R on midbrain dopaminergic neurons in alcohol intake.

We developed a novel transgenic mouse with a selective knockdown of D3Rs on the midbrain dopamine neurons (DATD3R KO). Because these studies employ a new transgenic model, potential behavioral phenotypes including, locomotion and anxiety-like behavior were explored. No differences were seen between DATD3R KO mice and littermate controls in basal locomotion or anxiety-like behavior using the elevated zeromaze, light/dark box, open-field and novelty suppressed feeding. We then assessed alcohol intake in the DATD3R KO mouse. No differences were seen in alcohol intake using two alcohol intake paradigms, an ethanol preference test and intermittent ethanol drinking task. These data indicate that D3Rs on dopaminergic neurons are not necessary in promoting increased preference for or consumption of ethanol.

Disclosures: M. Shaw: None. **M. Bocarsly:** None. **V.A. Alvarez:** None.

Poster

P719. Stress, Fear, and Anxiety: Animal Models

Location: Virtual Only

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Program #/Poster #: P719.05

Topic: G.04. Emotion

Support: Jim and Betty Ann Rodgers Fund

Title: Aspartame exposure of male mice produces heritable behavioral phenotypes

Authors: *S. JONES[1], D. M. MC CARTHY[1], P. G. BHIDE[2];
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Abstract: The use of low-calorie sweeteners as weight loss tools is highly prevalent. In the US, approximately 25% of children and 40% of adults report using low-calorie sweeteners. Aspartame is among the most widely used low-calorie sweeteners worldwide. We examined whether aspartame exposure produces behavioral changes in male mice, and if offspring derived from the aspartame-exposed males display behavioral alterations. We exposed adult (6-8 week) male C57BL/6 mice to drinking water containing aspartame (0.03% or 0.015%; to deliver aspartame at doses equivalent to 100% and 50%, respectively, of FDA's Acceptable Daily Intake for humans). Following 12 weeks of aspartame exposure, we analyzed anxiety-like behavior (open field test), spatial working memory (Y-maze), and spatial learning and recall memory (Barnes maze). The data were analyzed for statistical significance by ANOVA followed by *post hoc* pairwise comparisons among the different experimental groups, whenever a significant main effect or interaction effect was present. The mice exposed to either dose of aspartame displayed significant anxiety-like behavior, spatial working memory deficit, and a significant deficit in acquisition of spatial learning. A single administration of diazepam (3 mg/kg; i.p.) produced significant downregulation of the anxiety-like behavior, suggesting the involvement of GABA

neurotransmission in the aspartame-induced phenotype. The aspartame-exposed and control mice were bred with drug-naïve females. Male and female offspring derived from the aspartame-exposed males displayed anxiety-like behavior and spatial working memory deficit demonstrating heritability of these behaviors. Ongoing studies are examining whether the anxiety-like phenotype in the offspring responds to diazepam and if aspartame-induced changes in GABA neurotransmission are heritable as well. In summary, our data show that aspartame exposure even at levels equivalent to 50% of the FDA recommended human daily intake, carries the risk of adverse behavioral outcomes not only in individuals exposed to the aspartame but also their descendants.

Disclosures: S. Jones: None. **D.M. Mc Carthy:** None. **P.G. Bhide:** None.

Poster

P719. Stress, Fear, and Anxiety: Animal Models

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Program #/Poster #: P719.06

Topic: G.04. Emotion

Support: H2020-EU Grant 847826, Brain Involvement iN Dystrophinopathies (BIND) project
Ministère de l'Enseignement Supérieur et de la Recherche (France)
Association Monégasque contre les Myopathies (AMM, Monaco)
Centre National de la Recherche Scientifique (CNRS, France)
Université Paris-Saclay (France)

Title: Enhanced fear- and anxiety-related behaviours and impaired fear learning in mdx52 and mdx^{5cv} mouse models of Duchenne muscular dystrophy

Authors: *M. D. MITSOGIANNIS[1], A. SAOUDI[3,4], F. ZARROUKI[3], C. FERGUS[2], E. STOJEK[1], S. TALAVERA[1], V. P. KELLY[2], A. GOYENVALLE[4], F. MONTANARO[5], F. MUNTONI[5], E. SOKOLOWSKA[1,6], C. VAILLEND[3];
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Abstract: Duchenne muscular dystrophy (DMD) is a neuromuscular disorder in which dystrophin loss, caused by mutations in the *DMD* gene, affects muscle and cognitive-affective functions. In the past 30 years new DMD mouse models have emerged to investigate how

specific *DMD* mutations, impacting different dystrophin isoforms, contribute to neurobehavioural comorbidities. We examined anxiety-like behaviours, emotional reactivity and fear conditioning (FC) in two such models, bred on a C57BL/6 background: the *mdx52* mouse, in which exon 52 deletion eliminates production of both Dp427 and Dp140 brain dystrophins, and the *mdx*^{5cv} mouse, carrying a point mutation in exon 10 that exclusively impairs Dp427 expression. Hemizygous *mdx52* or *mdx*^{5cv} male mice and wild-type littermate controls (WT) aged 6-7 weeks and 3-4 months (n=8-17/genotype) were tested in the elevated zero maze (EZM) for 5 min to assess state anxiety; unconditioned fear responses (UFR) were then assayed by scruff-restraining mice for 15 s followed by 5 min of locomotor activity monitoring. In the EZM, *mdx52* and *mdx*^{5cv} mice displayed heightened anxiety, e.g., a significantly lower time spent in open maze sections compared to WT, at all ages. Enhanced anxiety was confirmed in 2-month-old *mdx52* mice compared to WT, as significant decreases in time spent in the central area of an open field (n=14 WT, 15 *mdx52*), or in the lit chamber in a light-dark choice test and in open arms of an elevated plus maze (n=9 WT, 12 *mdx52*). In the UFR assay, restraint-induced freezing time was found significantly higher compared to WT in young and adult *mdx*^{5cv} mice (>78% of test time in *mdx*^{5cv} vs <20% in WT) and *mdx52* mice (>80% of test time), indicating that cumulative Dp427 and Dp140 loss does not worsen fear responses in this test compared to selective Dp427 loss. Acquisition of cued FC was significantly impaired in *mdx52* and *mdx*^{5cv} mice, as reflected by a strongly reduced amount of freezing compared to WT in response to the conditioned stimulus (tone predicting negative reinforcement delivery). These findings resemble those obtained in analogous paradigms in the well-characterised *mdx* mutant mouse lacking Dp427 (*mdx23*), yet behaviour abnormalities were more clearly expressed in the present study. This indicates that loss of dystrophins at brain level is associated with enhanced emotional and stress reactivity and impaired amygdala-dependent associative learning for both proximal (*mdx*^{5cv}) and more distal (*mdx52*) *Dmd* mutations. Phenotypic commonalities observed across DMD mouse models in the assays described make emotional reactivity and fear learning optimal neurobehavioural readouts to investigate brain function-targeted therapies in DMD.

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Poster

P719. Stress, Fear, and Anxiety: Animal Models

Location: Virtual Only

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Program #/Poster #: P719.07

Topic: F.04. Stress and the Brain

Support: GUTMOM grant 01EA1805

Title: Deletion of FKBP51 in POMC cells shapes the stress response and whole-body metbolism

Authors: *L. M. BRIX[1], A. S. HÄUSL[1], I. TOKSZOEZ[1], J. BORDES[1], L. VAN DOESELAAR[1], C. ENGELHARDT[1], H. YANG[1], M. SPRINGER[1], S. NARAYAN[1], M. L. PÖHLMANN[1], S. MITRA[1], J. DEUSSING[2], N. C. GASSEN[3], A. CHEN[4], M. V. SCHMIDT[1]; [1]Neurobio. of Stress Resilience, [2]Res. Group Mol. Neurogenetics, Max Planck Inst. of Psychiatry, Munich, Germany; [3]Dept. Psychiatry and Psychotherapy, Universitätsklinikum Bonn, Bonn, Germany; [4]Dept. of Neurobio., Weizmann Inst. of Sci., Rehovot, Israel

Abstract: There is increasing evidence of a high co-morbidity of metabolic and mental health disorders, but the underlying mechanism linking these disorders is largely unresolved. The co-chaperone FKBP51 is a very promising candidate in this context, as it has previously been genetically and functionally linked to metabolic and psychiatric disorders and plays a central role in the regulation of the stress system, which is a common risk factor for both disorders. Furthermore, FKBP51 was shown to be expressed in POMC-positive cells, which play a key role in metabolic control in the arcuate nucleus (ARC) and are also central in mediating the body's stress response in the pituitary gland. We therefore here tested the hypothesis that FKBP51 expression in POMC-positive cells may be causally involved in the regulation of stress and metabolism.
Using a Pomc$^{Fkpb5\text{-}/\text{-}}$ knockout line or virally-mediated region-specific FKBP51 overexpression, we assessed the standard diet (SD) and high fat diet (HFD) induced metabolic phenotypes as well as basal and stress-induced hypothalamic-pituitary-adrenal (HPA) activity.
Our results confirmed that lack of FKBP51 in POMC cells of the pituitary leads to an enhanced Glucocorticoid receptor (GR)-mediated negative feedback and thereby a suppression of HPA axis function. At the same time, deletion of FKBP51 in POMC neurons of the ARC is driving resistance to diet-induced weight gain, which is independent of food intake.
Taken together, our data underline the cell type specific effects of FKBP51 in regulating the stress response and metabolism and position this protein as a promising molecular player and pharmacological target that acts at the interface of metabolism and stress related physiological changes.

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Poster

P719. Stress, Fear, and Anxiety: Animal Models

Location: Virtual Only

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Program #/Poster #: P719.08

Topic: F.04. Stress and the Brain

Support: MH049698
MH119814
F31MH125541-01

Title: Molecular neurobiology of enrichment loss: role of microglia and the extracellular matrix

Authors: *M. A. SMAIL[1], R. SHUKLA[2], J. L. BOLLINGER[1], R. PARIKH[1], R. E. MCCULLUMSMITH[2], J. P. HERMAN[1];
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Abstract: Psychological loss can erode well-being and precipitate depression. Here, we emulate loss by exposing rats to protracted environmental enrichment and subsequently removal to an impoverished environment. Previously, multi-omics analyses revealed that enrichment removal (ER) dysregulates immune signaling and cell-matrix communication in the basolateral amygdala (BLA) of male rats. Here, we further investigate these microglia and extracellular matrix-related phenotypes to more clearly define their role in the mechanisms underlying loss. All studies feature 3 groups of adult male rats: environmentally enriched (EE), enrichment removed (ER), and control. EE and ER animals were housed in groups of 10 in large, multi-level cages with toys. After 4 weeks, ER animals were moved to single-housing, simulating the experience of loss. Two weeks after removal, brains were collected for analysis of microglia and matrix-related endpoints in the BLA. Immunohistochemistry revealed that microglia were smaller in EE and ER, and expressed significantly less CD68, a marker of phagocytosis, in ER alone. This supports the role of microglia previously identified in the multi-omics analysis and suggests that ER impairs the ability of microglia to interact with their surroundings. One such interaction suggested by the omics is digestion of the extracellular matrix. ER increased the total amount of WFA, a marker of extracellular matrix, in the BLA, as well as the number of perineuronal nets around inhibitory parvalbumin interneurons. This increase in perineuronal nets was accompanied by decreased synaptic input, both excitatory (vGLUT) and inhibitory (vGAT), onto the parvalbumin cells, providing a potential link between impaired microglia-matrix interactions and altered BLA output in ER. Altogether, these results suggest that ER impairs microglia activity, allowing for accumulation of extracellular matrix, blocking synaptic input onto parvalbumin interneurons, impairing their ability to inhibit the excitatory output of the BLA. In a region like the BLA that is critical for driving stress responses, this decreased plasticity could precipitate loss phenotypes and play a key role in driving subsequent behavioral impairments. This mechanism not only deepens our understanding of the molecular effects of psychological loss, but also reveals several intriguing targets, including microglia and perineuronal nets, that could be used to develop novel therapeutics aimed at ameliorating loss.

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Poster

P719. Stress, Fear, and Anxiety: Animal Models

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P719.09

Topic: F.04. Stress and the Brain

Title: Assessing mouse behavior and microglia morphology in a prolonged stress model of post-traumatic stress disorder

Authors: *J. HOLSTEN, T. COTHREN, K. PRESCOTT, D. MACQUEEN, T. PETERSON; Univ. of North Carolina Wilmington, Wilmington, NC

Abstract: Post-Traumatic Stress Disorder (PTSD) is characterized as exposure to a traumatic event, which manifests into dysfunctional behaviors such as hyperarousal and intrusive thoughts that can disrupt a person's well-being. Currently, PTSD is classified by the DSM-V criteria of outwardly expressed behaviors (i.e., anxiety, negative alterations in cognition and mood, and altered arousal and reactivity). However, there are various anxiety- and fear- related psychiatric disorders that share common symptomology with PTSD. Recent data suggests microglia alter their morphology and function in primary brain regions associated with the disorder (i.e., prefrontal (PFC), hippocampus, and amygdala) that accompany the dysfunctional behaviors. The current study used C57Bl/6j mice (male and female) to measure dysfunctional behavior and assess microglia morphology in a PTSD mouse model of prolonged stress. This stress model consisted of 2 hr restraint stress, 20 min forced swim, exposure to diethyl ether until unconscious, and single housing for the remainder of the study. This PTSD model has been shown to induce behavioral and physiological deficits that mimic the human condition. Behavior, corticosterone, and neuropathology measures were assessed across four groups (naïve; n = 11, single housed; n = 9, single prolonged stress (SPS); n = 9, and repeated prolonged stress (rSPS); n = 9). Stress groups displayed significant differences in anxiety-like ($p < 0.05$), social ($p < 0.05$), and nest building ($p < 0.05$) behavior tasks. Corticosterone levels had roughly a three-fold increase in stress groups compared to naïve animals ($p < 0.01$) suggesting the stress hormone was upregulated in stress groups compared to naïve animals. The data collected thus far have replicated behavioral and corticosterone levels from past animal and human studies. Immunofluorescence was conducted with ionized calcium binding adaptor molecule 1 (Iba1) to label microglia and DAPI, and imaged with a confocal microscope to create a 3-D image using z-stacks. Currently, we are imaging and analyzing branch (i.e., process length, number of processes, end points, etc.) and fractal data (i.e., fractal dimension, span ratio, complexity, etc.) of microglia in the PFC, hippocampus, and basolateral amygdala. Establishing the morphological and functional changes of microglia in these commonly used mouse models of PTSD is crucial for developing possible future treatments, as well as other disorders where microglia are maladaptive.

Disclosures: J. Holsten: None. **T. Cothren:** None. **K. Prescott:** None. **D. MacQueen:** None. **T. Peterson:** None.

Poster

P721. Addictive Drugs: Developmental Effects I

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Program #/Poster #: P721.01

Topic: G.09. Drugs of Abuse and Addiction

Support: TRDRP Grant 22RT-0103
Brain & Behavior Grant 21517

Title: Role of a Human Polymorphism (rs2304297) in the 3'-UTR of the CHRNA6 Gene in Nicotine-Modulated Behaviors in Adolescent Sprague-Dawley Rats

Authors: *A. CARDENAS[1], S. LOTFIPOUR[2];
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Abstract: The initiation of nicotine-containing product use typically occurs prior to the age of 18. Adolescence is a period in development where the maturation of brain neurocircuitry is vulnerable to nicotine exposure. With recent escalation of e-cigarette use among adolescents, it is critical to identify at-risk populations and potential therapeutic targets to prevent and treat nicotine dependence. In human adolescents, a single nucleotide polymorphism (SNP), rs2304297, at base pair 123 in the 3'-untranslated region (UTR) of the CHRNA6 gene (encodes the alpha(*α)*6 nicotinic receptor subunit), has been associated with increased smoking and substance use. Moreover, *α*6 nicotinic receptor subunits reach peak expression during adolescence in the mesolimbic dopaminergic neurons. To study the CHRNA6 3'-UTR SNP *in vivo*, our lab generated a human CHRNA6 3'-UTR mutant rodent line via CRISPR-Cas9 genome engineering where we replaced the 3'-UTR of the CHRNA6 gene in the rat with the human CHRNA6 3'-UTR containing either the non-risk allele, C ($\alpha6^{CC}$), or the risk SNP allele, G ($\alpha6^{GG}$). To evaluate the role of the CHRNA6 3'-UTR SNP in adolescent substance use, we assessed functionality of the SNP in nicotine-induced locomotor activity and anxiety-like behavioral measures that depend on the mesolimbic dopamine pathway. We hypothesize the *α*6 3'-UTR knock-in interacts with nicotine to influence locomotion and anxiety-like behavior with $\alpha6^{GG}$ rodents being more affected. Furthermore, in behavioral matched tissue, we hypothesize nicotine will enhance *α*6 mRNA expression in $\alpha6^{GG}$ versus $\alpha6^{CC}$ rats. To test our hypotheses, we used a low dose nicotine pretreatment paradigm in male and female rats where animals received either nicotine (2x, 30 μg/kg/0.1 mL, i.v., equivalent to a few cigarettes) or saline for one (acute) or four consecutive days (sub-chronic) during early adolescence (PN 31, 28-31). Immediately following nicotine exposure on P31, animals were placed into an open-field chamber. Our studies illustrate acute and sub-chronic nicotine exposure enhances locomotion and anxiolytic

behavior in a genotype-dependent manner. Additionally, sub-chronic nicotine exposure effects were sex-dependent. Preliminary results for α6 mRNA expression show no genotype, nicotine pretreatment, or sex effects, indicating that our observed behavior is not due to mRNA levels in the mesolimbic tissue. Taken together, our findings illustrate that our human CHRNA6 3'-UTR SNP knock-in is functional *in vivo.* Future studies are underway to evaluate mechanisms underlying our observed effects in our genetic variant rodents including neuronal activity and protein expression pattern in brain tissue.

Disclosures: A. Cardenas: None. **S. Lotfipour:** None.

Poster

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Topic: G.09. Drugs of Abuse and Addiction

Support: MoE 1-441-122

Title: Effects of prenatal gabapentinoids exposure on human cortical neurons

Authors: M. ALHOMRANI[1], *A. ALAMRI[1], W. ALJUAID[1], O. BAHRI[1], E. ALOSIMI[1], H. HABEEBALLAH[1], E. ALMALKI[1], N. ALSOFUANI[1], Y. ALTHOBAITI[2], A. ALSHEHRI[1], N. ALTHIBITY[1], A. GABER[1], J. HAYNES[3], C. POUTON[3], ***W. ALSANIE**[1];
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Abstract: Prenatal substance exposure is a major public health concern associated with many detrimental fetal consequences. Unfortunately, polysubstance use in pregnancy is common. Gabapentinoids are widely used as treatments in psychiatry and neurology; however, they have been increasingly reported as having potential for misuse. Moreover, gabapentinoids can cross the placental barrier. Due to difficulties in accessing fetal brains exposed to gabapentinoids, we used the human embryonic stem cell (hESC) line H9 to generate early, intermediate cortical progenitors and cortical neurons to modulate prenatal gabapentinoid exposure *in vitro*. Since the cortex is responsible for cognition and behavior, we focused on cortical development. We analyzed treated (10uM) and untreated (control) cultures for gene expressions, neurogenesis, and morphogenesis. At the early patterning stage, there was a significant increase in Tbr2+ intermediate progenitors in pregabalin- and gabapentin-treated cultures. In addition, there was a significant increase in the expression of cortical related genes *Pax6*, *Foxg1*, and *Tbr2* in pregabalin-treated cultures, whereas gabapentin significantly increased *Tbr2* expression solely. At the maturation stage, the number of mature cortical neurons was unchanged in pregabalin-treated cultures. At early maturation, gabapentin significantly increased Tbr1+ neurons, but not

Ctip2+ neurons. At the genetic level, we screened the effects of pregabalin on different cortical layer related genes. Pregabalin significantly increased expression of *Brn2* without significant effects on other screened genes. Meanwhile, gabapentin did not alter any cortical layer related genes. Regarding morphogenical analysis, both pregabalin and gabapentin significantly decreased neurite length, branches, and neurites of human cortical neurons. Our data also shows that the effects of pregabalin and gabapentin on the morphogenesis of cortical neurons differ based on the presence of maturation factors, such as GDNF and BDNF, suggesting a possible interaction mechanism. Our study demonstrates that exposure to gabapentinoids during early brain development may interfere with the neurogenesis and morphogenesis of various neuronal subpopulations. Currently, we are investigating gabapentinoids' effects on cortical neuron functionality.

Disclosures: M. Alhomrani: None. **A. Alamri:** None. **W. Aljuaid:** None. **O. Bahri:** None. **E. Alosimi:** None. **H. Habeeballah:** None. **E. Almalki:** None. **N. Alsofuani:** None. **Y. Althobaiti:** None. **A. Alshehri:** None. **N. Althibity:** None. **A. Gaber:** None. **J. Haynes:** None. **C. Pouton:** None. **W. Alsanie:** None.

Poster

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Program #/Poster #: P721.03

Topic: G.09. Drugs of Abuse and Addiction

Support: WSU ADARP Grant

Title: Impact of maternal cannabis vapor self-administration on offspring neurodevelopment and anxiety-like behavior

Authors: *H. V. WEIMAR, A. N. MALENA, D. E. GINDER, A. M. BROWN, R. J. MCLAUGHLIN;
Washington State Univ., Pullman, WA

Abstract: Cannabis is the most used illicit drug during pregnancy, yet little is known about the lasting impacts of prenatal cannabis exposure on neurodevelopmental endpoints. To address this issue, our laboratory recently used a novel cannabis vapor exposure model to show that vaporized cannabis exposure in pregnant rat dams alters the cognitive, emotional, and social phenotype of offspring across the lifespan. However, we also found significant differences between air-exposed and vehicle-exposed offspring on several of these endpoints, which suggests that forced vapor delivery in pregnant dams may produce an aversive state that likely confounds the effects of cannabis per se. To address this, we developed a novel model of prenatal cannabis exposure involving volitional intake of cannabis (or vehicle) vapor in pregnant

rat dams. We used this model in the current study to interrogate effects of maternal cannabis vapor self-administration on offspring development and anxiety-like behavior.
Female Sprague Dawley rats were trained to self-administer either Δ9 tetrahydrocannabinol (THC)-rich cannabis vapor (69.9% THC; 150 mg/mL) vapor or vehicle (VEH; 4:1 propylene glycol:vegetable glycerol) vapor on a fixed-ratio-1 schedule during one-hour sessions twice daily throughout mating and pregnancy. Another cohort of dams were included that did not self-administer either vapor (AIR) to assess potential effects of VEH vapor self-administration. Beginning after postnatal day (P) 80, offspring were tested in the elevated plus maze (EPM) test for 5 minutes to evaluate anxiety-like behavior in adulthood.
Results indicate that dams readily self-administered cannabis vapor throughout pregnancy. Cannabis self-administering dams showed greater discrimination for the vapor-associated operandum and received significantly more vapor deliveries than VEH self-administering dams. Cannabis-exposed offspring weighed significantly less than both VEH and AIR pups on P1, which aligns with findings in human studies. Surprisingly, female cannabis-exposed offspring spent less time in the open arms of the EPM compared to VEH and AIR offspring, whereas male cannabis-exposed offspring spent more time exploring the open arms than AIR offspring.
These results support vapor self-administration in pregnant rat dams as a feasible approach for modeling prenatal cannabis exposure. Moreover, our data indicate that maternal cannabis use may have sexually dimorphic effects on anxiety-like behavior in adulthood. Altogether, validation of this approach will provide a more translationally relevant way to assess effects of maternal cannabis use on offspring neurodevelopment.

Disclosures: H.V. Weimar: None. **A.N. Malena:** None. **D.E. Ginder:** None. **A.M. Brown:** None. **R.J. McLaughlin:** None.

Poster

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Program #/Poster #: P721.04

Topic: G.09. Drugs of Abuse and Addiction

Support: MoE 1-441-122

Title: Effects of prenatal gabapentinoids exposure on mouse cortical neurons

Authors: *M. ALHOMRANI[1], A. ALAMRI[1], S. ABDELRAHMAN[3], Y. ALTHOBAITI[2], A. ALSHEHRI[1], N. ALSOFUANI[1], E. ALOSIMI[1], N. ALTHIBITY[1], W. ALJUAID[1], E. ALMALKI[1], A. GABER[1], J. HAYNES[4], H. HABEEBALLAH[1], O. BAHRI[1], R. FELIMBAN[5], A. CHAUDHARY[5], C. POUTON[4], C. HAUSER[3], W. ALSANIE[1];
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Abstract: Though widely used as treatments in psychiatry and neurology, gabapentinoids have been increasingly reported as having potential for misuse. Due to a lack of safety studies, gabapentinoids at therapeutic doses are the last treatment option for various neurological diseases, such as neuropathic pain, during pregnancy. Taking this into consideration, gabapentinoid abuse in pregnant women, or even use at therapeutic levels, may impair fetal development. Here, we used primary mouse embryonic neurons to investigate whether prenatal exposure can impair fetal brain development. We focused on cortical and ventral midbrain development, as they are responsible for cognition and behavior. Early neurogenesis and morphogenesis were analyzed *in vitro* using cortical and ventral midbrain neurons isolated from E12.5 embryonic mouse brains (n=6) in 3D cultures (using ultra-short peptides). We investigated the expression of different cortical layers and ventral midbrain specific proteins in gabapentinoid-treated 3D cultures (using different doses), comparing these with untreated 3D cultures. In addition, we studied the morphogenesis *in vitro* in the 3D cultures. In embryonic mouse cortical neurons, pregabalin and gabapentin (in all tested doses) did not alter early development of preplate and deep layer cortical neurons in treated cultures, in comparison with untreated cultures. However, pregabalin and gabapentin significantly decreased neurite length of both preplate neurons and other deep cortical layer neurons. Meanwhile, in pregabalin-treated cultures, the number of branches significantly increased only in the other deep layer neurons not belonging to the preplate population. Nevertheless, in both cortical populations, the number of neurites and branches significantly decreased in gabapentin-treated cultures. In ventral midbrain neurons, pregabalin and gabapentin significantly increased neurite length of dopaminergic neurons, while the number of branches significantly increased in non-dopaminergic neurons. Pregabalin, but not gabapentin, significantly increased the number of neurites in non-dopaminergic ventral midbrain neurons. Our study demonstrates that exposure to gabapentinoids during early brain development may interfere with the neurogenesis and morphogenesis of various neuronal subpopulations. We are currently investigating gabapentinoids' effects on the functionality of the aforementioned neuronal populations.

Disclosures: M. Alhomrani: None. **A. Alamri:** None. **S. Abdelrahman:** None. **Y. Althobaiti:** None. **A. Alshehri:** None. **N. Alsofuani:** None. **E. Alosimi:** None. **N. Althibity:** None. **W. Aljuaid:** None. **E. Almalki:** None. **A. Gaber:** None. **J. Haynes:** None. **H. Habeeballah:** None. **O. Bahri:** None. **R. Felimban:** None. **A. Chaudhary:** None. **C. Pouton:** None. **C. Hauser:** None. **W. Alsanie:** None.

Poster

P721. Addictive Drugs: Developmental Effects I

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Program #/Poster #: P721.05

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant K99DA049908

Title: Amygdala-related behavioral phenotype of prenatal opioid exposure depends on history of early life stress

Authors: *B. L. SMITH, T. A. GUZMAN, T. M. REYES;
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Abstract: In the past decade, U.S. maternal opioid use has increased by 130%. Children with prenatal opioid exposure (POE) have an increased risk for impaired emotional control and increased functional connectivity in the amygdala. Consequently, we used mice to study amygdala-related behavioral consequences of POE. C57/Bl6 dams were given access to 0.3 mg/ml morphine in drinking water sweetened with 0.2% saccharin (MO, n = 24) from 10 days prior to conception until offspring were weaned on postnatal day (P)21, and control dams were given 0.2% saccharin (SCH, n = 16). A subset of MO dams were given limited bedding material from P2-9 to model early life stress (MO LB, n = 7). Adolescent MO LB offspring of both sexes had reduced time spent in the light compartment of the light dark test. In contrast, male MO offspring had increased social time in the social interaction test. In the elevated zero maze test, group differences were not observed but maternal MO consumption in the 3rd gestational week related to the amount of time male offspring spent in the open sections. In MO LB males, maternal MO consumption negatively correlated with time spent in the open sections but in the male MO offspring not exposed to LB, there was positive correlation between dose and open section time. Importantly, this replicates our prior studies demonstrating that prenatal MO exposure increases behavior related to exploration or novelty seeking, while prenatal MO combined with early life stress reduces exploration. This indicates that the behavioral effects of POE manifest differently if combined with early life stress.

Disclosures: B.L. Smith: None. **T.A. Guzman:** None. **T.M. Reyes:** None.

Poster

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Topic: G.09. Drugs of Abuse and Addiction

Support: NIH DP1 DA039658
NIH R01 DA051831
TRDRP T30FT0967

Title: Effect of prenatal nicotine and THC on cognitive behaviours in adolescent male and female rats.

Authors: *V. LALLAI, L. MANCA, Y. SHERAFAT, C. D. FOWLER;
Univ. of California, Irvine, Irvine, CA

Abstract: Although there has been a decrease in the prevalence of tobacco smoking in many developed countries, exposure to nicotine/tobacco during pregnancy remains a substantial problem worldwide. Further, given the recent escalation in e-cigarette use among various age groups and legalization of cannabis, it has become essential to better understand the effects of nicotine and cannabinoid co-exposure on neural development and function, especially in consideration of vulnerable populations. These studies sought to systematically examine the effects of nicotine and THC prenatal exposure on cognitive behaviors in both male and female offspring rats. Dams were exposed to nicotine vape (5 mg/kg) or vape vehicle control, and oral THC (5 mg/kg) or vehicle control (sesame oil). Offspring were then tested in the pre-pulse inhibition test at PND21, novel object recognition task at PND35, and novelty suppressed feeding task at PND37. During adolescence, prenatal vape exposure in males and females resulted in a changed baseline startle reactivity, and significant effects were found in pre-pulse inhibition. Deficits in novel object recognition were also found in males prenatally exposed. Finally, modestly increased anxiety-associated behaviors were found with THC or nicotine exposure in the latency to approach novel food in an open field. Together, these studies demonstrate differential effects of prenatal exposure to e-cigarette nicotine vape and/or edible THC on cognitive processing tasks, with differing effects for males and females.
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Poster

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Program #/Poster #: P721.07

Topic: G.09. Drugs of Abuse and Addiction

Title: Adolescent THC Persistently Alters Medial Prefrontal Cortex-Dependent Cognition in Rats

Authors: *M. X. MARTINEZ[1], V. ALIZO-VERA[1], C. M. RUIZ[1], S. B. FLORESCO[2], S. V. MAHLER[1];
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Abstract: Epidemiological studies suggest a link between early adolescent cannabis use and risk of developing schizophrenia, but whether Δ^9-tetrahydrocannabinol (THC) actually precipitates schizophrenia symptoms remains unclear. Moreover, the neurobiological mechanisms underlying this relationship remains unknown. Evidence from rodents suggests that adolescent THC alters the function of medial prefrontal cortex (mPFC) circuits, and human studies also suggest a link between PFC abnormalities and schizophrenia. Here, we tested the hypothesis that adolescent THC causes mPFC changes that may have relevance to schizophrenia. We exposed male and female Long-Evans rats to THC (5mg/kg i.p.) or vehicle daily from postnatal day 30 to 43, and after they grew to adulthood (PD 67+), we examined performance on two mPFC-dependent cognitive tasks, operant set-shifting and probability discounting. After lever training, rats were trained on a set shifting task. They were required to press whichever lever was under a light cue, in order to receive a banana pellet reward. They then experienced a rule reversal, such that correct responses consisted of pressing either the right or left lever, irrespective of the light cue—a set shifting response indicative of cognitive flexibility. After completion of the set shifting task, rats then began the probabilistic discounting task. They were trained to choose between a small/safe lever which always delivered one pellet, or a large/risky lever that sometimes delivered four banana pellets, with a probability that decreased parametrically across the six blocks in a 50min session (100%, 50%, 25%, 12.5%,6.25%). Prior work showed that the dopamine-enhancing drug amphetamine facilitates prepotent "risky" responding on this task (St Onge and Floresco, 2009), and given the link between dopamine and schizophrenia, and our prior findings of mPFC dopamine changes after adolescent cannabinoid exposure, we examined how amphetamine (0, 0.25, and 0.5mg/kg) impacts behavior in adolescent THC- or vehicle-exposed groups. We found that adolescent THC had persistent effects on performance in both operant set-shifting and probabilistic discounting tasks, suggesting THC can persistently alter adolescent brain development of mPFC circuits. These findings may provide insight into the observed link between adolescent cannabis use and schizophrenia.

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Poster

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Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant AA019967
NIH Grant AA007474

Title: Projection specific changes in dopamine 1 receptor function following adolescent intermittent ethanol

Authors: *J. D. OBRAY[1], L. CHANDLER[2];
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Abstract: We have previously demonstrated that adolescent intermittent ethanol (AIE) reduces medial prefrontal cortex (mPFC) dopamine 1 (D1) receptor function in adult male rats. These effects are believed to be related to deficits in executive function observed in adulthood following AIE. To further investigate these effects, male and female rats underwent an AIE paradigm starting at PD28 and ending at PD56. After completing the AIE paradigm the rats were injected with red retrobeads in the basolateral amygdala (BLA) and green retrobeads in the nucleus accumbens (NAc). Beginning 3 weeks after the injections the rats were euthanized and brain slices containing the mPFC were prepared for whole cell patch clamp electrophysiology. Electrophysiological measurements were made in visually identified neurons projecting to the BLA or NAc. Current clamp recordings (K-gluconate internal) were performed, and evoked firing was measured to assess intrinsic excitability. D1- and dopamine 2 (D2) receptor function was then assessed by measuring evoked firing in response to an injected current while bath applying SKF38393 or Quinpirole. We found that in AIE animals D1-receptor, but not D2-receptor function was reduced in neurons projecting to the BLA. This effect was consistent across both male and female rats. Female rats, however, displayed greater activation of BLA projecting pyramidal neurons during bath application of SKF38393 in both the AIR and AIE conditions. There were no clear changes in D2-receptor function based on treatment group or sex. Analysis is underway to determine the effects of AIE on D1- and D2-receptor function in mPFC pyramidal neurons projecting to the NAc.

Disclosures: J.D. Obray: None. **L. Chandler:** None.

Poster

P721. Addictive Drugs: Developmental Effects I

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Program #/Poster #: P721.09

Topic: G.09. Drugs of Abuse and Addiction

Title: Effects of adolescent methamphetamine exposure on perineuronal net and parvalbumin+ cell density in the rodent mPFC

Authors: *A. S. BRINKS[1], L. K. CARRICA[2], D. J. TAGLER[3], J. M. GULLEY[2], J. M. JURASKA[2];
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Illinois At Urbana-Champaign, Champaign, IL; [3]Psychology Dept., Univ. of Illinois at Urbana-Champaign, Champaign, IL

Abstract: Adolescence is a time of enhanced vulnerability to several psychiatric disorders, including substance use disorder. While there are likely social factors that contribute to this vulnerability, ongoing maturation of brain regions critical for cognitive function may also contribute. Across adolescence, the medial prefrontal cortex (mPFC) undergoes significant reorganization, with parvalbumin+ (PV) inhibitory interneurons gaining their fast-spiking phenotype and perineuronal nets (PNN's) aggregating primarily around PV cells. Previous studies have shown PNN's are influenced by female, but not male, puberty with post pubertal females having fewer PNN's than prepubertal littermates. PNN's are also influenced by drug administration, though this is highly specific with heroin resulting in fewer PNN's and cocaine resulting in more PNN's. To assess the effects of methamphetamine (METH) on PV cells and PNNs rats were dosed with METH around female puberty (EA), male puberty (LA), and early adulthood. Male and female Sprague Dawley rats were exposed to 3mg/kg methamphetamine via intraperitoneal injection from either postnatal day (P) 30-38 (EA), P40-48 (LA), or P60-68 (Adult). Brains were collected one day after dosing and fluorescently labeled for PNN's and PV for density quantification. Preliminary results show that METH moderately increased PNN density after adult exposure in both sexes and during the pubertal times for each sex: EA for females and LA for males. PV density was decreased following exposure in both males and females in the LA timepoint but only males in early adolescence. While these results indicate an effect of METH exposure on both PV cell and PNN density, further results will include measures of mPFC volume to control for changing volume over adolescence and increased N. This will allow for further comparisons across age and sex.

Disclosures: A.S. Brinks: None. **L.K. Carrica:** None. **D.J. Tagler:** None. **J.M. Gulley:** None. **J.M. Juraska:** None.

Poster

P732. Neural Mechanisms of Addictive Drugs III

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Support: Instituto de Salud Carlos III, Fondos FEDER, Red de Trastornos Adictivos (RTA). Grant RD16/0017/0014.

Title: Cannabidiol modulates behavioral and gene expression alterations induced by spontaneous heroin withdrawal

Authors: *F. NAVARRETE[1], A. GASPARYAN[2], J. MANZANARES[3];
[1]Inst. de Neurociencias, Univ. Miguel Hernández - CIF: Q5350015C, San Juan de Alicante, Spain; [2]Inst. de Neurociencias, Univ. Miguel Hernández, San Juan de Alicante, Spain; [3]Inst. De Neurociencias,, Univ. Miguel Hernandez, San Juan de Alicante, Spain

Abstract: Background: Previous clinical data suggest that cannabidiol (CBD), a non-psychoactive constituent of the Cannabis sativa plant with anxiolytic, antidepressant, antipsychotic properties, and anti-addictive properties, may hold promise as a therapeutic tool for managing opioid use disorder (OUD). **Aims:** The objective of this study was to evaluate the effects of CBD on behavioral disturbances induced by spontaneous heroin withdrawal in CD1 male mice. Furthermore, quantitative real-time PCR (qPCR) experiments were carried out to evaluate the effects of CBD on gene expression changes induced by spontaneous heroin withdrawal in specific key targets. **Methods:** Spontaneous heroin withdrawal was evaluated 30 h after cessation of 8-days heroin treatment in CD1 male mice (5, 10, 20, and 40 $mg \cdot kg^{-1}$; every 12 h; 2 days each dose; s.c.). CBD effects (5, 10, and 20 $mg \cdot kg^{-1}$; i.p.) on withdrawal-related behavioral signs were evaluated by measuring anxiety-like behavior, motor activity, and somatic signs (rearing, jumping, rubbing, grooming, digging). Furthermore, qPCR experiments were carried out to evaluate gene expression changes of mu-opioid receptor (Oprm1), proopiomelanocortin (Pomc), cannabinoid CB1 receptor (Cnr1) and cannabinoid CB2 receptor (Cnr2) in the nucleus accumbens (NAcc), and tyrosine hydroxylase (TH) and Pomc in the ventral tegmental area (VTA). Statistical analyses were performed using one- and two-way analysis of variance (ANOVA) with repeated measures (RM). **Results:** Anxiety-like behavior, motor activity, and withdrawal-related somatic signs were significantly increased in heroin-treated mice. In addition, Oprm1 and Cnr2 in the NAcc and TH and Pomc in the VTA were upregulated, whereas Cnr1 and Pomc in the NAcc were significantly reduced in abstinent animals. Interestingly, the treatment with CBD significantly reduced anxiety-like behavior (10 and 20 mg/kg; $p<0.001$), hyperactivity (10 and 20 mg/kg, $p<0.001$), and somatic signs (5, 10 and 20 mg/kg; $p<0.001$) induced by spontaneous heroin withdrawal. Furthermore, CBD normalized the gene expression of Cnr1 and Pomc in the NAcc and of TH in the VTA, induced an additional up-regulation of Cnr2 gene expression in the NAcc, and did not change the increased gene expression of Oprm1 in the NAcc. **Conclusion:** The present study results provide evidence about the efficacy of CBD to modulate the behavioral and gene expression disturbances associated with spontaneous heroin withdrawal. Further studies are warranted to evaluate the potential therapeutic actions of CBD for the management of heroin withdrawal and to elucidate the precise underlying neurobiological mechanisms involved.

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Poster

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Program #/Poster #: P732.02

Topic: G.09. Drugs of Abuse and Addiction

Support: NIDA
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BBRF

Title: Epigenetic priming drives aberrant gene expression in cocaine relapse

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Abstract: Substance use disorders represent an enormous public health crisis and are among the most intractable illnesses in our society. An ongoing focus of research into the molecular pathology of addiction are mechanisms by which neuronal gene regulation is altered in a central brain region of reward, the nucleus accumbens (NAc). Stable changes in chromatin are proposed to underlie the maladaptive transcriptional states in this brain region, which persist despite long-term drug withdrawal. However, there is no direct link between drug-induced epigenetic marks and aberrant gene expression programs that drive relapse. A fundamental challenge is determining which neuronal subtypes are responsible: the NAc is composed primarily of two opposing types of medium spiny neurons (MSNs), the D1 and D2 dopamine receptor-expressing subtypes, which exhibit dramatic differences in activity and effects on drug reward. In these distinct subtypes, we examined how chronic cocaine modifies chromatin structure and characterized immediate versus persistent changes in gene regulation. We surveyed circuit-specific chromatin accessibility genome-wide in combination with unbiased histone modification profiling by mass spectrometry and ChIP-sequencing. We discovered that chronic cocaine persistently ‘scars’ chromatin structure in D1 MSNs, involving dramatic depletion of the histone variant H2A.Z at key neuronal genes related to synaptic plasticity. Genome accessibility is prominently increased at these genes even after prolonged withdrawal, linked to aberrant gene expression upon drug relapse. The histone chaperone ANP32E promotes the removal of H2A.Z, and we demonstrate that D1 circuit-selective ANP32E knockdown prevents cocaine-induced H2A.Z depletion and effectively blocks cocaine conditioned place preference. In contrast, the D2-specific knockdown of ANP32E enhances cocaine-related reward learning in this animal model. These findings provide new insight into circuit-specific epigenetic priming as a critical mechanism and promising clinical target employed by drugs of abuse to modify brain function and behavior in lasting ways.

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Poster

P732. Neural Mechanisms of Addictive Drugs III

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Program #/Poster #: P732.03

Topic: G.09. Drugs of Abuse and Addiction

Support: CVF2019-PS-B02

Title: Electrophysiological correlates of affective processing in people with chronic consumption of psychoactive substances

Authors: *J. AVILA MORENO[1], J. D. RODRÍGUEZ[1], Y. V. MOLINA[1], D. LEÓN[2], S. ZARATE[1], F. CARDENAS[2], J. M. DURAN[1];
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Abstract: Polydrug use of psychoactive substances is a worldwide problem, it has been shown that drug use is associated with deficits in attention, processing and identification of emotional stimuli that can be identified by changes on Event-related potentials (ERPs). Most studies of chronic drug use effects on ERPs have focused on a given class of addictive substances. However, this single-use drug profile represents only one part of consumptions spectrum since clinical population report the use of more than one drug. Although the high prevalence of polyconsumption, there are few studies evaluating emotional processing in that condition. The aim of the study was to evaluate patterns of cortical activity of affective processing in polyconsumers of psychoactive substances. Fifty Colombian men and women between 18 and 55 years old participated in this research, (25) consumers and (25) non-consumers of psychoactive substances. ERPs were obtained from EEG data recorded while individuals scored the valence of a set of stimuli taken from the International Affective Pictures System (IAPS). We found that, polyconsumers expressed lower average amplitude for the later P1- in posterior electrodes during 90-130 latency range for both valences, also non-consumers N1-Cz were more negative than for consumers during 170-210 latencies, particularly to pleasant images. In the posterior region polyconsumers had attenuated the P2 potential, while in the anterior zone they had higher negativity in N2 ERP. Non-consumer had more negative N3 potential than polyconsumers, but this negativity was bigger to unpleasant images. Finally, in the latency range of 700-1000 milliseconds non-consumer showed a more positive potential to aversive images than polyconsumers in parietal areas. Previous reports have been shown the critical role of P1, N1 and N2 on the attentional process underpinning allocation, orientation, and awareness of emotional stimuli as well as cognitive control and behavioral inhibition. These results could indicate that polyconsumption is associated with an insufficient ability for early visual processing of affective images, a reduction in the attentional orientation to the affective stimuli, a poor valence

processing specially of pleasant images and a worse ability to focus and process the task and select the appropriate response by inhibiting involuntary, impulsive, and inappropriate actions.

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Poster

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Program #/Poster #: P732.04

Topic: G.09. Drugs of Abuse and Addiction

Support: PCSI2020-B007

Title: Electrophysiological biomarkers in polyconsumers of psychoactive substances before and after a psychotherapeutic process

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Abstract: Alterations in affective processing, especially depressive symptoms, increased negative affect, difficulties in emotion regulation, alterations in reward processing, and high rates of impulsivity are common among drug addicts. These changes in emotional appraisal and response can be identified by changes in event-related potentials (ERPs). Although advances in neuroscience research have helped to describe the neurobiological changes that occur during a substance use disorder or addiction, only a few studies have attempted to examine changes in functional brain activity that may accompany changes in behavior before and after treatment. Therefore, ERP measurement might fill a gap in treatment-response biomarkers to psychotherapeutic interventions, consider that the therapeutic goal in the treatment of addictions is not only abstinence as behavior, but an affective reprocessing, acquisition impulse control and decision-making skills, among others. This study aimed to evaluate the patterns of cortical activity before and after psychological treatment of polyconsumers of psychoactive substances during the assessment of affective images. Twenty-three Colombian polyconsumers between 18 and 55 years old who voluntarily initiated inpatient psychological treatment participated in this study. The ERPs were obtained from electroencephalographic data recorded before and after psychotherapeutic process while the participants scored the valence of a set of visual stimuli from the International Affective Pictures System (IAPS). We found that polyconsumers expressed a higher P1 posterior during 90-130 latency range, and the P2 were more positive during latencies 270-320, both for unpleasant images before treatment. Also, in the anterior

region polyconsumers had higher P3b positivity before treatment compared to post-treatment, particularly to pleasant images. In the latency range of 1000-1250 analysis showed that polyconsumers had a more positive LPP potential to pleasant images, however in the latency range of 1250-2490 subjects expressed LPP potential with higher negativity to aversive images in the parietal areas, both before treatment. These preliminary results can be associated with a process of self-regulation, cognitive control, and response inhibition besides emotion regulation and decrease in craving related to psychological treatment in polyconsumers. This interpretation helps to demonstrating the need for studies using ERPs that show the changes in neural activity associated with affective processing before and after psychotherapy and thus contribute to better practices and effective treatments.

Disclosures: J. Rodríguez Animero: None. **J.C. Avila:** None. **V. Molina:** None. **D. León:** None. **S. Zarate:** None. **F. Cárdenas:** None. **J.M. Duran:** None.

Poster

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Program #/Poster #: P732.05

Topic: G.09. Drugs of Abuse and Addiction

Support: NIDA

Title: Capturing and profiling cocaine-recruited neuronal ensembles in the nucleus accumbens.

Authors: *M. SALERY, A. GODINO, Y. Q. XU, J. F. FULLARD, P. ROUSSOS, E. J. NESTLER;
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Abstract: The ability to first learn associations between the rewarding effects of a drug and the context in which it is experienced is decisive for precipitated drug-seeking and relapse in abstinent individuals. As learned associative memories have been proposed to be stored in sparse and highly discriminative populations of concomitantly activated neurons, emerging evidence supports the idea that drug-recruited neuronal ensembles could similarly encode addiction-related pathological memories. However, little is known regarding the dynamics and molecular mechanisms of both their recruitment upon initial drug exposure and their later contribution to relapse. Here, we postulate that subsequent reactivation of an initially-activated ensemble could support the encoding, the strengthening and ultimately the expression of drug-associated memories. A related goal is to explore the intrinsic (phenotypical) or acquired (plastic) cellular properties that would favor the allocation of specific cells to these functional ensembles and predict their further reactivation. Capitalizing on the activity-dependent labeling of neuronal ensembles in Arc-CreER^{T2} mice (Denny et al., 2014), we were able to capture cocaine-activated

cells in the nucleus accumbens and permanently tag them with fluorophores or channel-rhodopsin for further characterization, optogenetics, and single-nuclei sorting. We identified a subset of neurons activated at both early and late stages of drug exposure and show that the level of reactivation of the initial ensemble correlates with the amplitude of behavioral sensitization. Similarly, re-exposure to a cocaine-paired context in a conditioned place preference (CPP) paradigm was associated with an increased reactivation of cocaine-recruited ensembles. The behavioral consequences of such reactivation was further assessed using optogenetics-mediated artificial reactivation. We found that the reactivation of ensembles recruited at early- versus late-stages of drug exposure had opposite effects on CPP expression. We then isolated tagged nuclei with FACS and performed single nucleus RNA sequencing to analyze their transcriptional signature. Using activity-dependent transcriptional programs as a marker of recent activation, we successfully isolated a cluster of reactivated cells within the initially activated ensemble. Together, such multiscale and ensemble-specific approaches represent a pivotal step towards a better understanding of the cellular and molecular processes involved in the encoding of pathological memories associated with drug addiction. Supported by NIDA

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Poster

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Topic: G.09. Drugs of Abuse and Addiction

Support: NIDA Grant P01DA047233

Title: Transcriptomic analysis of the nucleus accumbens after volitional heroin intake implicates Mbd3 as a key regulator in relapse susceptibility

Authors: *R. FUTAMURA[1], C. J. BROWNE[1], X. ZHOU[2], A. RAMAKRISHNAN[1], A. M. MINIER-TORIBIO[1], A. GODINO[1], M. SALERY[1], O. SIAL[4], B. ZHANG[2], Y. L. HURD[3], L. SHEN[1], E. J. NESTLER[1];
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Abstract: Repeated opioid use causes molecular changes in the nucleus accumbens (NAc), a brain region critical for coordinating motivated behavior and reward processing. Dysfunction of the NAc can perpetuate compulsive drug-seeking and drug-taking behaviors to the extent of which voluntary control of these behaviors is lost. Further, molecular changes within the NAc

may support long-term susceptibility to relapse despite cessation of opioid use, induced by drug-related cues or re-exposure to the drug itself triggering intense drug craving. However, transcriptional regulation within the NAc induced by drug-seeking behavior has not been previously examined. Ongoing studies in our laboratory have identified broad patterns of transcriptional regulation throughout the brain reward system induced by volitional opioid exposure and drug-seeking behavior using heroin intravenous self-administration in mice. In these studies, mice self-administered saline or heroin for 15 days and underwent a 30-day homecage forced abstinence period, after which mice were challenged with either saline or heroin and underwent a drug-seeking test. Here, we focus on how transcriptional regulation within the NAc contributes to drug-seeking behavior. Using multiscale embedded gene co-expression network analysis (MEGENA), we identified a particular gene network in this brain region that is enriched with upregulated genes induced by heroin-primed drug-seeking. Within this network, methyl-CpG binding domain protein 3 (Mbd3), one of the most significantly upregulated genes within the NAc in this condition, was found to be a key hub gene. In addition, exploratory factor analysis linking behavioral outcomes from self-administration with gene expression uncovered a positive association between Mbd3 and addiction-like behavior. Taken together, these data suggest that Mbd3 is an important driver in controlling this gene network that associates with relapse-like behavior. Ongoing efforts include determining the cell-type-specificity of Mbd3 regulation in NAc D1 or D2 medium spiny neurons, manipulating Mbd3 using a knockdown approach, and examining changes to behavior that promote drug craving, and ultimately, relapse.

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Poster

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Topic: G.09. Drugs of Abuse and Addiction

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NIH Grant R01DA07359

Title: Phosphodiesterase 1b is a key driver of a gene network in nucleus accumbens associated with addiction-like behaviors

Authors: *C. D. TEAGUE[1], X. ZHOU[1], Y. VAN DER ZEE[1], A. GODINO[1], D. M. WALKER[2], B. ZHANG[1], E. J. NESTLER[1];
[1]Icahn Sch. of Med. At Mount Sinai, New York, NY; [2]Oregon Hlth. & Sci. Univ., Portland, OR

Abstract: Cocaine use disorder (CUD) is a serious public health issue with no effective pharmacotherapies. Identifying treatments for CUD is hindered in part by an incomplete understanding of the coordinated changes in gene expression that drive long-lasting maladaptive plasticity in the brain and promote addiction-like behaviors. In this study, we conducted unbiased gene co-expression network analysis on a published RNA sequencing (RNA-seq) dataset from 6 different brain regions of animals that underwent cocaine self-administration followed by prolonged withdrawal plus context- or cocaine-induced reinstatement (Walker et al., *Biol Psychiatry*, 2018). We ranked gene networks by their fold enrichment in genes whose expression is significantly correlated with an "addiction index" (AI) - a composite score developed by machine learning to capture maladaptive, addiction-like behaviors during cocaine self-administration. We identify the gene encoding phosphodiesterase 1b (*Pde1b*), a Ca^{2+}/calmodulin-dependent enzyme that catalyzes the hydrolysis of cAMP and cGMP, as the strongest regulator of a gene network in the nucleus accumbens (NAc) that shows the highest magnitude association with the AI of all gene modules in this brain region. Our data suggest that *Pde1b* is primed in *Drd1*-expressing medium spiny neurons (D1 MSNs) following withdrawal from chronic cocaine. To investigate the role of *Pde1b* in regulating addiction-like behaviors, we are performing virus-mediated *Pde1b* overexpression or knockdown in the NAc followed by conditioned place preference (CPP), locomotor sensitization, and self-administration assays for cocaine in male and female rodents. Additionally, we will use RNA-seq to measure the effect of *Pde1b* manipulations on gene network activity and cocaine-induced transcriptomic changes in the NAc. Given successful drug discovery efforts focused on other PDE isoforms, this work may present a novel therapeutic approach for CUD.

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Poster

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Topic: G.09. Drugs of Abuse and Addiction

Support: NIDA P01DA047233
NIDA R01DA007359

Title: Molecular basis of ΔFosb action in drug addiction: mapping ΔFosb binding sites in nucleus accumbens medium spiny neurons

Authors: *S.-Y. YEH, M. S. ESTILL, L. SHEN, E. J. NESTLER;
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Abstract: Drug addiction exacts a devastating impact on drug users, their family members, and the nation's public health. Although different in chemical structures and initial mechanisms affecting the brain, all classes of drugs of abuse induce the expression of the transcription factor ΔFosB in nucleus accumbens (NAc), a central node of the reward circuitry. Studies with ΔFosB genetic and viral manipulations in mice and rats suggest that ΔFosB in NAc neurons is involved in the development of addiction. However, the molecular mechanisms by which ΔFosB controls addiction remain incompletely understood. Here, we extend earlier work on revealing ΔFosB's transcriptional targets, which previously interrogated promoter regions only, by leveraging the CUT&RUN (cleavage under targets and release using nuclease) method to pinpoint ΔFosB binding genome-wide upon chronic cocaine exposure. Thousands of ΔFosB peaks were revealed, virtually all of which corresponded to consensus AP1 sites. Interestingly, one third of the loci are positioned at distal intergenic enhancers, while only ~15 percent of the peaks occur within known promoter areas, suggesting that a primary function of ΔFosB is coordinating distal regulatory elements with the transcription machinery. In addition, we identified ΔFosB peaks, as well as histone modifications linked to enhancer activity, specific to medium spiny neurons (MSNs) expressing either dopamine receptor D1 or D2 - the two major neuronal types in NAc, which adds to the early discovery that ΔFosB directs differential molecular and synaptic plasticity in D1- versus D2-type MSNs. Hundreds of only partly overlapping ΔFosB binding sites were identified in D1- and in D2-type MSNs that were also surrounded by active enhancer histone marks. Interestingly, ΔFosB enrichment was found at multiple genes associated with genetic risk for substance use disorders in human, supporting ΔFosB's key role in the genetic architecture of drug addiction. Further extension of this CUT&RUN approach to other chromatin regulatory mechanisms in D1- and D2-type MSNs within NAc will set the groundwork for understanding distinct roles of these MSN subtypes, and their adaptations to chronic drug exposure, in drug addiction.

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Poster

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Program #/Poster #: P732.09

Topic: G.09. Drugs of Abuse and Addiction

Support: NIDA

Title: Elucidation of the astrocyte-specific transcriptome following exposure to cocaine

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Abstract: Drug addiction represents an enormous healthcare burden. To better understand its biological underpinnings, investigations of the transcriptional response to drugs of abuse have demonstrated lasting changes in gene expression throughout the brain's reward circuitry. Historically focused on neurons, emerging evidence increasingly indicates that astrocytes are also involved in disorders of the nervous system, including addiction. Indeed, candidate genes in astrocytes have been identified and, furthermore, manipulation of astrocyte function has been demonstrated to influence rodent behavioral responses to cocaine administration. However, the astrocyte-specific transcriptome following exposure to drugs of abuse has not yet been investigated. As an initial approach, we examined previously published work (Walker et al., *Biol. Psychiatry*, 2018) and identified numerous astrocyte-enriched genes within RNA-sequencing datasets from bulk homogenates of several brain reward regions that are associated with behavioral responses to cocaine self-administration. In parallel, to obtain more comprehensive and selective data, we utilized whole cell sorting of astrocytes and RNA-sequencing to investigate the astrocyte-specific transcriptome in several key brain regions involved in reward-processing, including the nucleus accumbens and prefrontal cortex, following chronic exposure to cocaine. Subsequent gene ontology analysis revealed a variety of pathways, including synaptic regulation, calcium signaling, and GPCR signaling in both brain regions as being prominently regulated by cocaine exposure. Additional analysis revealed several deduced upstream regulators of this abnormal transcription, such as CREB and several members of the STAT family. Furthermore, our analysis indicates a robust regional-specific response of astrocytes following exposure to cocaine. Current studies are directed at extending our findings utilizing cocaine self-administration in mice to establish the astrocyte transcriptome in response to drugs of abuse and to subsequently study the role of specific transcripts in contributing to the pathophysiology of addiction.

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Poster

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Topic: G.09. Drugs of Abuse and Addiction

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Title: Proteomic Profiling of Nucleus Accumbens Synaptosomes Following Withdrawal From Cocaine or Heroin Self-Administration.

Authors: *Y. YIM[1], C. J. BROWNE[1], A. GODINO[1], A. MINIER-TORBIO[1], A. LY[2], J. CALLENS[1], F. MARTINEZ-RIVERA[1], R. FUTAMURA[1], J. LANDRY[1], R. S. WILSON[3], A. C. NARIN[4], Y. L. HURD[1], E. J. NESTLER[1];
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Abstract: Addiction is a devastating disorder that is exceptionally difficult to treat due to the high propensity for relapse even long after prolonged abstinence. The persistence of addiction is mediated at least partly by drug-induced changes in the physiology of reward-processing regions of the brain. Dysregulated signaling within the nucleus accumbens (NAc) is thought to play a critical role in promoting drug-seeking and relapse. Determining these changes may reveal more effective targets to treat substance use disorders. However, the molecular details underlying these adaptations remain incompletely understood. Here, we extend previous work focused mainly on candidate proteins of interest by contrasting whole-proteome changes induced by cocaine or heroin self-administration that persist through extended abstinence. Rats underwent 10 days of extended-access (6 hr/day) intravenous cocaine, heroin, or saline self-administration followed by 30 days of forced abstinence, after which they were euthanized from their homecages, and the NAc was extracted. Synaptosomes from these punch dissections were purified and analyzed by lipid chromatography-tandem (LC-MS/MS) mass spectrometry followed by label-free quantification. Using this approach, we identified unique patterns of induction or repression of numerous synapse-enriched proteins in NAc following prolonged withdrawal from cocaine or heroin self-administration. While most of these drug-regulated proteins are expressed by neurons, a significant subset is enriched in astrocytes or microglia, consistent with the intimate association of processes from these cell types with synapses. Interestingly, long-term withdrawal is associated with a greater degree of repression rather than induction of protein expression overall. We are now validating these findings and characterizing particular synaptic proteins that directly contribute to drug-seeking behavior. These studies will dramatically enhance our understanding of the molecular restructuring of synapses in the reward circuitry that underlie susceptibility to relapse.

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Poster

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Program #/Poster #: P732.11

Topic: G.09. Drugs of Abuse and Addiction

Support: NIDA P01DA047233

Title: Transcriptome profiling of the brain's reward circuitry in heroin self-administration identifies a ventral hippocampus gene network related to relapse susceptibility

Authors: *C. J. BROWNE[1], R. FUTAMURA[1], A. RAMAKRISHNAN[1], X. ZHOU[2], A. MINIER-TORIBIO[1], F. MARTINEZ-RIVERA[1], M. ESTILL[1], A. GODINO[1], A. TORRES-BERRÍO[1], E. M. PARISE[1], A. M. CUNNINGHAM[1], P. J. HAMILTON[4], D. M. WALKER[5], B. ZHANG[2], Y. L. HURD[3], L. SHEN[1], E. J. NESTLER[1];
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Abstract: Opioid addiction exacts a devastating toll on individuals, their families, and the healthcare system. Treatment is made exceptionally difficult by prolonged susceptibility for relapse into compulsive drug-seeking and taking, often triggered by re-exposure to drug-associated cues or the drug itself. Lasting relapse susceptibility is thought to be mediated in part by persistent changes in gene expression within interconnected reward-processing regions of the brain. However, few studies have performed transcriptome-wide analyses throughout brain reward regions following volitional opioid intake. Here, we combine heroin self-administration in mice, RNA sequencing (RNA-seq), and advanced bioinformatics approaches to identify novel genes and gene networks throughout the brain's reward system that are regulated by opioid intake and drug-seeking. First, mice underwent 15 daily 4 hr self-administration sessions wherein lever pressing lead to intravenous delivery of saline or heroin (FR1, 0.05 mg/kg/inf). Mice were then euthanized either from their homecage 24 hr after the last session or after a 30-day homecage forced abstinence period. In the 30-day group, mice also received a challenge injection of saline or heroin (1 mg/kg) and were placed back into self-administration chambers for a 2 hr drug-seeking test, after which mice were immediately euthanized. This design enabled comparisons of multiple addiction-relevant outcomes, including first-ever heroin exposure, early withdrawal from chronic use, context-induced drug-seeking, and combined drug/context-induced drug-seeking. RNA-seq was conducted on six brain regions involved in reward-processing: medial prefrontal cortex, nucleus accumbens, dorsal striatum, basolateral amygdala, ventral hippocampus, and ventral tegmental area. Bioinformatic analysis of this rich dataset has uncovered numerous patterns of differential gene expression in a region- and condition-dependent manner. Further, exploratory factor analysis was used to link gene expression patterns across brain regions with behavioral profiles relevant to an addiction-like state. Employing multiscale embedded gene co-expression network analysis (MEGENA), we also identified gene networks in the ventral hippocampus that associate with susceptibility to relapse. Current work

focuses on modulating expression of genes within this network and studying the impact on heroin self-administration and drug-seeking behavior.

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Poster

P751. Cocaine Circuits

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Title: Infralimbic-cerebellum anatomical map

Authors: *J. GUARQUE-CHABRERA[1,2], M. MIQUEL[1,2], K. KHODAKHAH[1];
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Abstract: Reciprocal pathways connecting the cerebellum and the medial prefrontal cortex (mPFC) provide a biological and functional substrate of their codependence to modulate cognitive and motivational functions. Dysfunction of both areas underlie the phenotypes of several neuropsychiatric disorders, and may operate as a risk factor for drug addiction. Extensive literature shows that the prelimbic (PL) and the infralimbic (IL) cortices, two areas included in the mPFC, play opposite roles not only over the initiation and inhibition of drug seeking but also regarding the control of goal directed and habitual behaviors respectively. Miquel's lab has reported that either a lesion in the vermal lobule 8 (L8) of the cerebellum or an IL deactivation facilitated the establishment of conditioned preference for cocaine-related cues. However, this effect was not replicated by PL deactivations. Cerebellum-IL functional relationships appear to be compensatory because simultaneous cerebellum-IL deactivations abolished the facilitative effect of separate deactivations. Moreover, unpublished results showed that IL but not PL deactivations during a repeated cocaine experience upregulates neuronal activity in the posterior vermis in rats. One of the relays for the cerebellum to modulate mPFC functions is the VTA. Indeed, we have shown that optogenetic manipulation of the cerebellar outputs targeting VTA

were able to modulate reward-related behaviors. However, anatomical projections from the IL to the cerebellum have been poorly described and it is far to be clear through which brain regions the IL might regulate cerebellar activity. Neither, if IL projections are monosynaptic or involve disynaptic pathways. Therefore, our goal is to create an anatomical map of the IL-cerebellum pathway in mice. We used a combination of monosynaptic and transneuronal viral (Herpes and Rabbies virus and AAVs) and classic (Fluorogold and BDA) tracing agents. Our results show that IL might send projections to both interposed DCN and inferior olive and these areas then will reach cerebellar cortex of L8 in the vermis.

Disclosures: J. Guarque-Chabrera: None. **M. Miquel:** None. **K. Khodakhah:** None.

Poster

P751. Cocaine Circuits

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Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P751.02

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant DA000467-17
NIH Center for Compulsive Behaviors
Janelia Visiting Scientist Program
NIDA IRP Scientific Director's Fellowship for Diversity in Research

Title: In vivo labeling and molecular characterization of cocaine relapse ensemble neurons in the infralimbic cortex using a photo-activatable calcium integrator

Authors: *O. R. DRAKE[1], R. MADANGOPAL[1], M. B. BRENNER[1], S. J. WEBER[1], V. A. LENNON[1], L. E. KOMER[1], K. E. SAVELL[1], F. J. RUBIO[1], L. WANG[2], K. SCHAEFER[2], A. LEMIRE[2], E. R. SCHREITER[2], V. MENON[3], B. T. HOPE[1];
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Abstract: In abstinent drug users, cues previously associated with drug-taking maintain the ability to provoke drug craving and precipitate relapse long after the last instance of drug use. These persistent, maladaptive cue-drug associations are thought to be encoded in sparse patterns of strongly activated neurons termed '*neuronal ensembles'*. Our lab and others have previously used the activation of the immediate early gene *Fos* as a marker to identify and characterize ensemble neurons specifically activated during events of interest (e.g. the presentation of a drug-associated cue or the execution of a learned drug-seeking behavior). However, these approaches have poor temporal specificity, and the relationship between *Fos* and neuronal activity is not well understood. To address this issue, we will use a genetically encoded photo-activatable

calcium integrator, CaMPARI2, to permanently label active neurons in the infralimbic cortex (IL) of rats during cocaine relapse. UV light exposure permanently converts CaMPARI2 from green to red fluorescent state only in active neurons and allows permanent *in vivo* labeling of these neurons with sub-second temporal resolution. Following cocaine self-administration, we will label active neurons during cocaine-seeking (cocaine-memory ensemble) or novel context exposure (control ensemble) and use FACS to isolate CaMPARI2 labeled (red, active) neuronal nuclei. We will then use single-nuclei RNA sequencing to identify unique molecular alterations within cocaine-memory ensemble neurons and investigate if their enrichment is restricted to specific cell type clusters within the IL. Understanding how drug memories are maintained could enable us to prevent relapse by selectively weakening these persistent drug-related memories without influencing other memories.

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Poster

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Program #/Poster #: P751.03

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH/NIDA Intramural funds

Title: Contributions of zincergic neurons to drug-seeking behaviors

Authors: *O. SOLIS CASTREJON, E. VENTRIGLIA, B. LAING, J. BONAVENTURA, Y. APONTE, M. MICHAELIDES;
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Abstract: Glutamatergic neurons to the striatum are thought to be implicated in drug seeking behavior. The striatum receives glutamatergic inputs from the prefrontal cortex, basolateral amygdala, hippocampus and paraventricular thalamus (PVT). Within a subset of glutamatergic neurons (zincergic), zinc (Zn^{2+}) is co-released with glutamate. These zincergic neurons that release Zn^{2+} exclusively express the Zn transporter 3 (ZnT3), a protein essential for Zn^{2+} transport into synaptic vesicles and necessary for synaptic Zn^{2+} release. A growing body of evidence shows that synaptic Zn^{2+} acts as a neuromodulator of both excitatory and inhibitory synaptic transmission to influence synaptic plasticity and regulate cognition, memory and affective processing. On a systems level, there is a lack of knowledge concerning the functional role of zincergic neurons on drug seeking behavior. The distribution of zincergic neurons in the

brain has been previously described using histochemical procedures based on Zn-selenide stains. However, to date there is no information regarding the molecular phenotype of zincergic neurons that project to the striatum. While it has been demonstrated that zincergic neurons are predominantly glutamatergic, recent studies have also reported a population of zincergic inhibitory neurons (interneurons) in the auditory cortex. By using fluorescent In situ hybridization, we found that in the neocortex, ZnT3 is mainly expressed in glutamatergic neurons. Interestingly, ZnT3 is expressed in parvalbumin and somatostatin interneurons but not in VIP interneurons. To further characterize zincergic neurons, we recently developed the Cre-ZnT3/DreO transgenic mouse. We injected into the prefrontal cortex a Cre-dependent virus driving synaptophysin-mRuby to label putative synaptic output sites. Preliminary results show that prefrontal cortical zincergic project to direct and indirect pathway striatal neurons. We are currently working on the functional role of zincergic neurons in the striatum.

Disclosures: O. Solis Castrejon: None. **E. Ventriglia:** None. **B. Laing:** None. **J. Bonaventura:** None. **Y. Aponte:** None. **M. Michaelides:** None.

Poster

P751. Cocaine Circuits

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Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant R01AA023183
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Title: Brain-wide 3D profiling of neuronal ensembles reactive to relapse-promoting vs. relapse-suppressing cues in rats

Authors: *H. NEDELESCU[1], N. J. O'CONNOR[2], B. E. EASTWOOD[2], A. H. THAN[1], G. E. WAGNER[1], H. C. CHANG[1], G. L. DE NESS[1], A. CARROLL[1], Y. LIU[1], E. WHITNEY[1], N. E. EMERSON[1], Z. MIKULSKI[1], F. WEISS[3], J. R. GLASER[2], N. SUTO[4];
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Abstract: Drug addiction is a chronically relapsing brain disease. While environmental stimuli signaling drug availability (S+) are well-known to promote relapse, we have found that environmental stimuli signaling drug omission (S-) can suppress relapse in rats. This bidirectional modulation of drug relapse is regulated by two functionally distinct coactive groups of infralimbic cortex (IL) neurons – neuronal ensembles or engram cells – with each group

selectively reactive to S+ or S- (as visualized by Fos protein). However, the neuroanatomical source of afferents that activate these neurons remains unknown. We have thus conducted brain-wide analysis to identify neuronal ensembles that send axonal projections (visualized by AAV2retro-GFP) to the IL and that are evoked by S+ or S- in different groups of rats trained to self-administer cocaine or alcohol. While automated brain-wide profiling is available for mice, similar procedures for rats are not yet established, even though rats are a preferred animal model for studying more complex behaviors to model drug addiction. Hence, we developed automated 3D brain-wide profiling for rats where image data and cell counts were registered using NeuroInfo to the Waxholm rat atlas (ref: https://www.nitrc.org/projects/whs-sd-atlas). Briefly, [1] whole slide images of serial brain sections were captured by a ZEISS Axioscan slide scanner microscope, [2] these 2D images were aligned and registered using NeuroInfo to the Waxholm rat atlas coordinate system, [3] Fos- and GFP-positive IL-projecting neurons were detected with single cell resolution in NeuroInfo using deep learning methods with anatomic specificity conferred by matching image data to the atlas coordinate space. This method provides superior cellular tagging (staining) and image resolution – especially for the larger rat brains – than similar methods using brain clearing and light-sheet microscopes. In my poster presentation, I will further describe our new NeuroInfo tool for rat brains by applying it to identify brain-wide neuronal networks reactive to relapse-promoting vs. relapse-suppressing stimuli. In short, this study provides a new tool for automated 3D brain-wide profiling of rats and expands our knowledge of brain circuitry mediating environmental modulation of drug relapse.

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Poster

P751. Cocaine Circuits

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Support: NIMD Grant R01-MH111604

Title: Calreticulin as a target for cocaine in a ventral hippocampus to accumbens circuit

Authors: *A. L. EAGLE[1], M. A. DOYLE[1], H. M. KUHN[1], B. R. MURRAY[1], M. DYKSTRA[1], R. M. BASTLE[2], I. S. MAZE[2], A. ROBISON[1];
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Abstract: Ventral hippocampus neurons that project to the nucleus accumbens (vHPC-NAc) are critical to cocaine seeking and the expression of cocaine reward. However, how drugs like cocaine modulate the physiology and gene expression within this circuit is unknown. We have identified that chronic cocaine, both experimenter- and self-administered, decreases the intrinsic excitability of vHPC-NAc neurons in *ex viv*o brain slices from male transgenic *Rosa26*$^{eGFP/L10a}$ mice. We next sought to identify gene targets of cocaine that may underlie this phenomena. Using innovative retrogradely expressing HSVs to drive the Cre-dependent expression of GFP-tagged ribosomes in vHPC-NAc neurons of *Rosa26*$^{eGFP/L10a}$ mice along with circuit-specific TRAPSeq (Translating Ribosomal Affinity Purification followed by 3rd-generation sequencing) we identified actively translating mRNAs differentially regulated by chronic cocaine. Following the trail of our TRAPSeq findings, we validated the ER resident chaperone protein, calreticulin, as a target for cocaine in vHPC-NAc neurons using traditional molecular assays: RT-PCR and Western blotting in whole vHPC punches from male C57Bl6/J mice and immunofluorescence in circuit-tagged transgenic male *Rosa26*$^{eGFP/L10a}$ mice. Ongoing studies using calreticulin overexpression AAVs and floxed calreticulin mice will identify whether calreticulin regulates cocaine-induced changes in excitability in vHPC-NAc neurons. These studies are poised to identify a novel ER protein that may regulate cocaine-induced changes in this circuit that would likely also underlie the relapse to cocaine seeking and reward.

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Poster

P751. Cocaine Circuits

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Topic: G.09. Drugs of Abuse and Addiction

Support: P30DA013429
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T32DA007237

Title: Localizing circuits and molecular mediators of cocaine contextual memories

Authors: *C. CABAN RIVERA[1], X. SHI[1], J. BARR[1], C. K. DO[1], C. LI[1], C. CHEN[1], L.-Y. LIU-CHEN[1], L. G. KIRBY[1], G. M. SMITH[2], E. M. UNTERWALD[1];
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Abstract: Addiction is characterized by compulsive drug seeking and taking behaviors. Cravings triggered by the presence of environmental cues that were once associated to drug use are a main

contributor of relapse and once these drug-cue memories are formed they are difficult to erase. Addictive drugs like cocaine engage molecular pathways of associate learning, in which recall of an established memory is followed by its reconsolidation, strengthening that memory. The reactivation period is a critical time wherein memories are labile and vulnerable to behavioral and pharmacological interference. Given the widespread negative impact of drug use in our society, it is of great importance to understand the mechanisms underlying drug seeking behaviors to generate effective preventions and treatments. The goal of this study is to elucidate the anatomical and molecular substrates crucial for the maintenance of cocaine contextual memories, with a focus on the nucleus accumbens and GSK3b signaling. Designer receptors exclusively activated by designer drugs (DREADDs) were used to inhibit neurons at the nucleus accumbens. To accomplish this, adult male C57Bl6 mice underwent bilateral intracranial injections of an inhibitory DREADD (AAV2-hsyn-hm4D(Gi)-mcherry) followed by a cocaine conditioned place preference paradigm. Nucleus accumbens neurons were inhibited via systemic injection of clozapine N-oxide immediately following reactivation of cocaine contextual memory, and mice were re-tested for place preference 24 hours and 7 days later. A separate cohort of mice underwent cocaine conditioned place preference but instead received intracranial infusions of a selective GSK3 inhibitor, SB216763, into the nucleus accumbens following reactivation of cocaine contextual memory and then re-tested for place preference 24 hours later. Results show that chemogenetic inhibition of the nucleus accumbens immediately following reactivation of cocaine contextual memory significantly reduced a once established preference for the cocaine context. Furthermore, pharmacological inhibition of GSK3 in the nucleus accumbens following reactivation of said memory abolished preference for the cocaine paired context. These findings suggest the nucleus accumbens is important for the reconsolidation of cocaine contextual memories and that GSK3 is a key molecular mediator of this process.

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Poster

P751. Cocaine Circuits

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Topic: G.09. Drugs of Abuse and Addiction

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Title: Presence of a peer, like subthalamic nucleus optogenetic manipulation, reduces escalated cocaine intake

Authors: C. VIELLE[1], L. VIGNAL[1], M. DEGOULET[1], Y. PELLOUX[2], ***C. BAUNEZ**[1]; [1]INT UMR7289 CNRS & Aix-Marseille Univ., Marseille, France; [2]Dept. of Physiol. and Neurosci., ITT, Genova, Italy

Abstract: Drug use disorders are a major worldwide public health problem for which no medication has been developed yet. The subthalamic nucleus (STN) high-frequency deep brain stimulation has been shown to prevent cocaine escalation and re-escalation, but its effects are still poorly understood. Besides, social context is known to be a key influential factor in drug use. Indeed, in both rats and humans, the immediate presence of a peer decreases recreational cocaine use. Yet, its effect on later stages of addiction's cycle has not been elucidated. Here, we thus assessed whether the presence of a peer can reduce the cocaine intake of rats having previously lost the control over their drug intake. Rats were first subjected to a cocaine escalation procedure, consisting in extended access (6h) to the drug over 20 days and were then tested for 2h self-administration sessions alone or in presence of a stranger observing peer. The effects of STN optogenetic inhibition and high-frequency stimulation were tested in these two various conditions. In the alone condition, both optogenetic STN inhibition and stimulation drastically reduced cocaine intake. The presence of a peer reduced the level of cocaine intake of the control rats, but did not reduce it further in the STN inhibited and stimulated groups. Those results show the importance of social factors in the study and cure of drug use disorders and highlight a possible role for the STN as a promising target for the treatment of addiction.

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Poster

P751. Cocaine Circuits

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Program #/Poster #: P751.08

Topic: G.09. Drugs of Abuse and Addiction

Support: NIMH (R01-MH111604, AJR)

Title: Lateral entorhinal cortex neurons that project to nucleus accumbens mediate the expression of cocaine reward

Authors: *H. KUHN[1], B. R. MURRAY[1], D. BERMUDEZ[2], A. ROBISON[1], A. L. EAGLE[1]; [1]Physiol., Michigan State Univ., East Lansing, MI; [2]Neurosci. and Cognitive Sci., Univ. of Arizona, Tucson, AZ

Abstract: Neuronal circuits that synapse onto nucleus accumbens (NAc) medium spiny neurons are critical for regulating motivated behaviors, including those underlying drug reward and drug seeking. Lateral entorhinal cortex (LEC) is activated by craving in cocaine-dependent individuals and by cocaine-associated cues in rats. The LEC also sends axonal projections to the NAc (LEC-NAc), but how LEC-NAc neurons regulate drug reward and seeking behavior is currently unknown. We sought to determine how LEC-NAc neurons mediate drug seeking behavior and reward in mice. We used blinded behavioral experiments in 8-12 week old male and female C57Bl6J mice and replicated our findings across multiple cohorts to ensure scientific rigor. As has been previously shown for LEC neurons in general, we found that LEC-NAc neurons are activated by acute cocaine in mice as measured by cFos expression. We also used viral-mediated, circuit-specific DREADDs (Designer Receptors Activated by Designer Drugs) to show that LEC-NAc activity necessary for the expression of cocaine reward in conditioned place preference. Ongoing studies will determine whether this circuit also regulates relapse to cocaine seeking in a cue-dependent intravenous self-administration mouse model. These findings suggest that the previously unexplored LEC-NAc circuit is necessary for reward and implicate an entirely novel key pathway in the regulation of motivated behaviors relevant to cocaine addiction.

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Poster

P751. Cocaine Circuits

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Harvey Family Endowment

Title: A single acute fear experience redirects projections from the anterior cingulate: Evidence for a biological mechanism of anxiety and substance abuse disorders

Authors: E. L. BEARER[1,2], T. W. USELMAN[1], C. S. MEDINA[1], *R. E. JACOBS[3];
[1]Dept. of Pathology, UNM Sch. of Med., Albuquerque, NM; [2]Div. of Biol. and Biol. Engin., Caltech, Pasadena, CA; [3]Zilkha Neurogenetics Inst., USC Keck Sch. of Med., Los Angeles, CA

Abstract: Pyramidal neurons from anterior cingulate area (ACA) in the prefrontal cortex (PFC) project into striatum, thalamus, hypothalamus and reach as far as dorsal raphe (midbrain) and locus coeruleus (pons). Neural activity in ACA is reciprocally modulated by monoamine systems

arising in these same caudal brain regions: noradrenergic, dopaminergic and serotonergic. We previously reported that projections from PFC are redirected when these monoaminergic systems a genetically disrupted. Here we report that a single experience of acute threat also redirects ACA projections in wild type mice, and this change correlates with multiple subtle behavioral effects. Mice with or without acute threat (30m exposure to predator odor) (2,3,5-Trimethyl-3-thiazoline, TMT) were group-housed in an environmentally controlled facility for 3 weeks and then underwent manganese-enhanced MRI tract-tracing. Behavior was video recorded before odor, during odor and before tract-tracing. $MnCl_2$ (3-5nL of 0.6M in saline) with 0.5 mg/ml 3k rhodamine dextran was injected into right ACA and the location confirmed by post-injection MRI: $x = 0.59 \pm 0.13$ (R of midline), $y = +0.72 \pm 0.49$mm (bregma), $z = -1.04 \pm 0.36$ mm (deep) ($n = 24$). Longitudinal images were collected at 0.5, 6 and 24h after injection. Between imaging sessions animals were returned to their home cages. At conclusion of imaging, mice were sacrificed and perfusion-fixed brains processed for microscopy. Voxel-wise SPM comparisons between time points revealed progression of Mn(II) accumulation distally ($p<0.001$ FDR). Application of our new in vivo atlas to automated measurements of Mn(II) signal revealed dramatic anatomical differences in projections between animals experiencing acute threat and those without, including stronger connections to the PAG and hypothalamus after threat. Comparisons of projections between threat-exposed and mice with genetic disruptions of monoaminergic transporters suggested that fear-induced ACA projections were most similar to those in the norepinephrine transporter (NET) knock-out, where the interstitial amount of noradrenaline is increased. Acute threat further impacted ACA projections in mice lacking the serotonergic transporter (SERT). Future work will explore whether a single dose of cocaine, which inhibits all three monoamine reuptake transporters, has similar effects on ACA projection anatomy. Plasticity of ACA projections evidenced by this remodeling would produce enduring changes in reward circuitry, explaining how life-threatening experiences or cocaine exposure pose risks for anxiety and substance use disorders. Supported by NIH RO1 MH096093 (ELB) and DA018184 (REJ).

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Poster

P786. Neural Mechanisms of Decision-Making in Rodents

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Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

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Topic: H.03. Decision Making

Support: NIH Grant EY022979

Title: Animal movements can be decoded to reveal choice, reward outcome, and the presence of optogenetic stimulation

Authors: *C. YIN[1], A. KOSTIUK[1], S. MUSALL[2], X. R. SUN[3], S. GLUF[4], A. K. CHURCHLAND[1];
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Abstract: Developments in automated movement tracking open the door to understanding how movements on multiple timescales relate to diverse aspects of behavior and internal state. These tools are especially relevant to decision making: recent studies have revealed the considerable influence of uninstructed movements on neural activity (Musall, Kaufman et al., 2019). However, these findings have left unanswered how these movements relate to the other aspects of decision-making behavior at both the single-trial and session level. Further, the impact of optogenetic stimulation on such movements is not fully understood. To address this, we analyzed video datasets from 7 head-fixed mice trained on a two-alternative perceptual decision-making task. We deployed DeepLabCut to track the movements of 26 points on the mice (Mathis et al., 2018). We report three observations: 1. The movement pattern of mice tracked by DeepLabCut could predict upcoming choice, reward outcome, and trial history. This indicates that videos contain rich task-related information, much of which is hardly perceptible to human observers. 2. The variance remaining in the video tracking after task-related information was regressed out (termed “task-independent variance”) was associated with worse overall performance in 6/7 mice. This indicates that some spontaneous, uninstructed movements may reflect disengagement or may interfere with the animal’s ability to do the task effectively. 3. We found that movements could distinguish the presence vs. absence of transient optogenetic stimulation. This was true even when traditional movement metrics (e.g. reaction time) were unaffected. Interestingly, optogenetic stimulation of different pyramidal neuron cell types (FezF2, PlexinD1 and EMX) and brain areas (frontal vs. parietal cortex) triggered distinct movement changes, implying these changes are not merely the byproduct of stimulation, but contain rich information about the neurons being manipulated.These observations suggest that fluctuations of spontaneous movements on multiple timescales may uncover rich information about the internal state and neural activity of animals. Further, our analyses argue that fully understanding the impact of optogenetic perturbations can require a careful analysis of diverse movements.

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Poster

P786. Neural Mechanisms of Decision-Making in Rodents

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Title: Cortex-wide decision circuits are shaped by distinct classes of excitatory pyramidal neurons

Authors: *S. MUSALL[1], X. R. SUN[2], H. MOHAN[3], S. GLUF[4], Z. J. HUANG[3], A. K. CHURCHLAND[4];
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Abstract: To understand the neural circuits that maintain complex behavior, it is crucial to investigate the role of the distinct cellular subtypes that comprise them. Much effort has focused on inhibitory neuron subtypes but the functional roles of excitatory pyramidal neuron (PyNs) subtypes are less well understood. We used widefield calcium imaging to measure the cortex-wide activity of distinct PyN types and investigated their functional role in mice making auditory decisions. We studied neural activity in two knock-in driver mouse lines by expressing the calcium indicator GCaMP6 in two non-overlapping PyN populations: FezF2 (pyramidal tract, or PT) and PlexinD1 (intratelencephalic, or IT) neurons. Cortical activity was compared to a third line, EMX, which expressed GCaMP6 in all excitatory neurons. Using dimensionality-reduction methods, we isolated cortex-wide activity patterns from PT, IT and EMX mice. These analyses revealed major differences in the complexity and spatial layout of cortical activity patterns in each PyN subtype, both at a cortex-wide and a within-area scale. Next, we used linear encoding models to test for differences across PyN subtypes in relation to decision-making. In all PyN subtypes, the linear model predicted neural variance with high accuracy: up to 90% of variance in some areas. Interestingly, the relative importance of task variables differed across PyN subtypes. PT activity was better explained by the mouse's choice, especially in secondary motor cortex (M2), while IT activity was better explained by sensory and task history variables, especially in parietal cortex. Temporally restricted and cell-type specific optogenetic suppression confirmed a causal role for parietal cortex in sensory processing and M2 for formation of choice. Finally, we further probed the representation of choice by training decoders to predict the animal's decision based on the neural activity of each PyN cell type. Robust choice-predictive signals were found in EMX and PT populations in area M2. These signals ramped up throughout the stimulus and delay period. By contrast, M2 choice signals continuously decreased after the stimulus for IT neurons. For activity from an intersectional viral strategy that selectively labeled corticostriatal projection PyNs, the choice signal in M2 was again negatively weighted by the model. Our results argue that during decision-making, PyNs do not act as a monolithic group, but instead have distinct roles that are linked to their developmental lineage and projection target.

These distinct roles are evident across cortex and throughout a decision, and are especially clear in frontal structures late in the decision.

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Poster

P786. Neural Mechanisms of Decision-Making in Rodents

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Leonard and Isabelle Goldenson Postdoctoral Fellowship

Title: Mixed representations of memory and visual information for flexible navigation decisions

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Abstract: The survival of many animals requires navigation by making decisions based on sensory cues and past experience. In particular, identical sensory cues can carry distinct meanings depending on the context, such as memories of recent experiences, and thus can require different behavioral actions to achieve a rewarding outcome. A key computation for flexible navigation decisions is therefore the integration of internal contextual signals with sensory signals to inform a goal location and guide an appropriate behavioral action. Here we aimed to reveal the cortical areas and neural mechanisms that are central to combining contextual information stored in memory and visual signals to mediate flexible decision-making during navigation.

We trained mice to perform a delayed match-to-sample task in a virtual reality T-maze. As the mouse ran through the maze, the mouse sequentially observed two cues separated by a short delay (~1 s). The mouse was required to combine the contextual memory established by the first cue and visual information of the second cue to choose an appropriate turn direction at the T-intersection. For a given visual stimulus in the second cue, the mouse responded flexibly with a different turn direction based on the contextual memory of the first cue.

We performed an optogenetic screen to identify cortical areas that are involved in flexible navigation decisions. We bilaterally inhibited different sites across the dorsal cortical surface by optogenetically activating inhibitory neurons in VGAT-ChR2 mice. Inhibition of V1, posterior parietal cortex (PPC), or retrosplenial cortex (RSC) induced a large decrease in the task performance, suggesting that these areas were critical for mice to perform the task.
Two-photon calcium imaging revealed that navigation goals were signaled by neurons that nonlinearly mixed specific combinations of contextual information from short-term memory and visual information. These mixed selectivity neurons formed efficient, easy-to-decode population codes that appeared to govern accurate decision-making because they were informative before correct choices but degenerated during errors. Such mixed selectivity neurons were present in most parts of posterior cortex, even in V1. Surprisingly, these cells were distributed across posterior cortex, including V1, and were densest in RSC and sparsest in PPC. We propose the flexibility of navigation decisions arises from distributed neurons that mix context and visual information within a visual-parietal-retrosplenial network, centered in RSC.

Disclosures: S. Kira: None. **H. Safaai:** None. **A.S. Morcos:** None. **S. Panzeri:** None. **C.D. Harvey:** None.

Poster

P786. Neural Mechanisms of Decision-Making in Rodents

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P786.04

Topic: H.03. Decision Making

Title: Lesions of the anterior cingulate cortex impairs task engagement and ensemble encoding in dorsomedial striatum during reward-guided decision-making, with minimal impact on single-unit firing

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Abstract: The anterior cingulate cortex (ACC) sends projections to the dorsomedial striatum (DMS), constituting part of the corticostriatal circuit—a circuit which has been extensively implicated in the development of goal-directed behaviors. However, there is little understanding of how the ACC might modulate downstream DMS signaling during reward-guided decision-making. In this study, we aimed to elucidate the role of the ACC in the modulation of downstream response-outcome encoding and action planning in DMS. To address this issue, rats were trained on a reward-guided decision-making task; reward value was manipulated by independently varying the size of or the delay to reward over several trial blocks. Subsequently, we unilaterally injected either ibotenic acid (experimental group) or saline (control group) into the rat ACC, and rats performed the previously learned reward-guided decision-making task

while we recorded from single neurons in DMS. We found that ACC lesions resulted in impairments across all trial-types and behavioral measures; lesioned rats performed significantly worse across all trial types, chose high-valued rewards less frequently than control rats, and had significantly slowed reaction and movement times. Ensemble analysis also seems to suggest that ACC lesions resulted in impaired population encoding in the DMS, but did not significantly disrupt encoding at the level of single units. These results suggest that the deleterious behavioral effects observed following ACC dysfunction do not arise as a result of single-unit disruptions of downstream action-outcome encoding when attention is necessary for learning (i.e., during block transitions), but rather are related to a breakdown in neural ensemble encoding during complex reward-guided decision-making.

Disclosures: D. Vazquez: None. **N. Peña-Flores:** None. **M. Roesch:** None.

Poster

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Topic: H.03. Decision Making

Support: Simons Collaboration on the Global Brain 543005
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Title: Mesoscale Activity Map (MAP) underlying a memory-guided decision acquired with standardized, multiplexed, brain-wide Neuropixels recordings

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Abstract: Our brains can maintain information over timescales of a few seconds to guide future actions. Such 'short-term memory' (STM) serves as the brain's post-it note and is essential for cognition, such as motor planning, decision-making, and foraging. The neural correlates of STM have been studied mostly one brain region at a time. For instance, neurons in the frontal cortex and related brain regions show persistent and slowly varying changes in spike rate that correlate with the maintenance of STM. Yet, how coordinated neuronal activity across inter-connected

brain regions collectively maintains STM remains poorly understood.We used up to five Neuropixels probes to simultaneously record the activity of hundreds of neurons across more than a dozen brain regions during a memory-guided delayed response task in the mouse. Anterior lateral motor cortex (ALM; part of M2) is an essential hub for the planning and execution of memory-guided directional licking[1]. Taking an anatomy-guided approach, we targeted regions that form multi-regional networks with ALM, including nuclei in thalamus, basal ganglia, midbrain, pons, medulla, cerebellum and others. Each recording was aligned into the Allen Mouse Common Coordinate Framework to assemble an activity map of STM-related activity[2]. These measurements were combined with detailed movement tracking and precise optogenetic perturbations. We developed pipelines for processing[3], analyzing, and sharing the large-scale neurophysiological and behavioral data (datajoint.io). 50,000 high-quality single units were obtained in 692 penetrations from 29 animals. Activity encoding sensory stimuli, choice, movement, and outcome, were distributed across the brain. Choice-selective neurons were more prevalent in the ALM network. At the level of population activity, choice encoding was correlated at the level of single trials across the brain regions of the ALM network. Movements were encoded more strongly in brainstem and midbrain compared to frontal cortex and thalamus. These data provide a foundation for understanding how STM and other neural computations are produced by multi-regional interactions across the brain (Yi Liu et al SFN 2021).
Reference1. Guo et al, Neuron 2014.2. Liu et al, BioRxiv. 2020.02.25.965210.3. https://github.com/jenniferColonell/ecephys_spike_sorting

Disclosures: S. Chen: None. **Y. Liu:** None. **Z. Wang:** None. **L.D. Liu:** None. **H. Hou:** None. **N. Tien:** None. **T.T. Nguyen:** None. **N. Li:** None. **S. Druckmann:** None. **K. Svoboda:** None.

Poster

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Program #/Poster #: P786.06

Topic: H.03. Decision Making

Support: CRCNS Grant R01DA053014

Title: Striatal control of action switching and decision bias

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Abstract: When selecting actions in noisy and volatile scenarios, animals exhibit a high degree of flexibility with response to environmental contexts, quickly adapting to feedback to better

exploit future opportunities. These decisions are also made in the context of internal biases the animal possesses, which may be beneficial or maladaptive depending upon the environment. The neural underpinnings of these biases and how they can be modified is not well understood, but considerable work suggests that the representation of the decision environ relies critically on cortical-striatal pathways, whose circuit-level architecture is ideally suited for adapting decision policies via positive and negative reinforcement mechanisms. This reinforcement mechanism has been shown to be under the control of the direct and indirect pathways, respectively. To understand how these neural populations contribute to decision making across varying levels of reward conflict, we trained mice to push or pull a joystick within the context of a two-armed bandit task. Importantly, any changes in decision-making strategy across the different conflict levels was influenced purely by reward feedback information - the mice were not explicitly cued regarding optimal decisions nor environmental contexts. Decision strategies were altered by conflict level, but reach kinematics were not. Further, direct pathway (dSPN) stimulation exacerbated choice bias. Together, the data suggests that decision-making behavior is subject to the environment in which the decision is being made (even when that environment is only revealed through feedback information), and dSPN activity may serve to reinforce internal biases regarding decision-making.

Disclosures: J.K. Badyna: None. **E.A. Yttri:** None.

Poster

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Program #/Poster #: P786.07

Topic: H.03. Decision Making

Support: ERC Grant PRIORS-683209

Title: Neural signatures of vigor and proactive responses during perceptual decisions in dorsomedial striatum

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Abstract: Standard perceptual decision-making models postulate that responses are triggered in reaction to the stimulus, when the accumulated evidence reaches a decision threshold. This excludes, however, responses that are generated proactively at a time independent of stimulus. We have recently shown that, in a free reaction time (RT) auditory discrimination task in rats, reactive and proactive responses coexist and are triggered when either of two competing processes, respectively Action Initiation (AI) or Evidence Accumulation (EA), reaches a bound.

Importantly, the timing of proactive responses is strongly modulated by both physical fatigue and water satiety. However, the neural substrate of the AI process remains unclear although recent studies have shown ramping dopaminergic activity preceding proactive actions. The dorsomedial striatum (DMS) receives extensive dopaminergic innervation and it is involved in determining the response vigor. Hence, we performed population recordings in the DMS of 4 rats (n= 652 single units) to seek for neural substrates of the proactive AI process. A GLM analysis isolated the different contributions to trial-to-trial variations of individual spike rates and revealed that (1) a large fraction of DMS neurons encodes the current choice from response initiation to the response in the next trial, but we found no encoding of the stimulus information in any trial epoch; (2) a significant subpopulation already encodes RT during fixation, so that the timing of the response can be predicted from before stimulus onset; and (3) a large fraction of neurons show slow modulation of their baseline firing across trials, which parallels slow changes in RT along the session and strongly correlates with motor time and self-paced inter-trial interval, two common behavioral proxies of vigor. Overall, these results suggest that DMS activity, just as it seems to control vigor, may also play an important role in modulating the timing of proactive responses in perceptual decisions.

Disclosures: L. Hernández-Navarro: None. **A. Hermoso-Mendizabal:** None. **J. de la Rocha:** None. **A. Hyafil:** None.

Poster

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Support: Supported by Marie Skłodowska-Curie Fellowship
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Title: Bayesian approach to pathological and physiological decision making: involvement of dorsal raphe nucleus serotoninergic neurons

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Abstract: Current theories do not satisfactorily explain the function of the neuromodulator serotonin (5-HT) on processing rewarding, aversive and ambivalent stimuli. Therefore, new theoretical perspectives are required to define 5-HT contribution to behavior. Our study aimed to assess through a Bayesian approach the contribution of serotonergic neurons of the dorsal raphe

nucleus ($DRN_{5\text{-}HT}$) in decision-making processes that require weighing recent observations and prior beliefs built upon previous experiences. Specifically, we applied Bayesian analysis on a probabilistic discrimination task in which mice are presented two wheels they can turn clockwise or anticlockwise for reward; wheel turning preference is then shaped by contrasting the probability of reward upon the different options. We validated the Bayesian analysis of probabilistic discrimination to be sensitive to the physiological conditions of uncertainty (evoked by tempering reward signaling) as well as pathological conditions (evoked by acute systemic injection of ethanol). By using fiber photometry, we found activation of $DRN_{5\text{-}HT}$ neurons after turning the wheels and at reward collection,, that that is when animals are provided feedback from their motor response. , This feedbackthat allows them to switch between consecutive actions or switch wheel turning preference their choice (behavioral switching). Antagonism of the 5-HT4 receptor subtype in the dorsolateral striatum reduced behavioral switching, as exemplified by an increase in perseverative responses and reduction in the unfavorable use of recent observations. Optogenetic silencing of the DRN prevented further reduction of the use of unfavorable recent observation upon following sessions, suggesting integrated function of the different DRN neuronal populations, and their target regions, in the acquisition and expression of probabilistic discrimination. Together, our results demonstrates the use of Bayesian approach of decision making in pathological and physiological conditions and suggest that $DRN_{5\text{-}HT}$ neurons respond to expected events to switch motor action, and possibly decisions.

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Poster

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Title: Dopamine dynamics during evaluation and reevaluation of neuroeconomic decisions

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Abstract: Dopamine signaling in the nucleus accumbens core (NAc core) is an important neural substrate for value-based decision making. Positive changes in dopamine reflect reward

expectation and motivation-driven processes, but dopamine dynamics under economic conditions of varying flavor preference, value, and time budgets have not been extensively studied. Furthermore, decreases in dopamine have been seen in conditions of noxious stimuli or exogenous disappointment, but the dopamine dynamics in settings of internally-driven decision reevaluation are unknown.

Using fiber photometry and viral expression of dLight1.3b to monitor dopamine dynamics in mice during a neuroeconomic foraging task (Restaurant Row), we investigated how dopamine encodes evaluation and reevaluation of decisions. Mice were given 1 hr to forage for food rewards with varying time costs as they ran around a square maze with each corner ("restaurant") dispensing a differently flavored pellet. Upon entering a corner, mice first encountered an Offer Zone (OZ) in which a tone sounded, signaling the delay before reward would be delivered. After the tone presentation in the OZ, the animal could either accept or reject the offer. Mice could reject the offer by continuing to the next corner to receive an offer for a different flavored pellet. Mice could accept the offer by advancing into a Wait Zone (WZ), where the countdown to reward began. During the countdown, the animal had the opportunity to quit the trial at any point and only earned reward if it waited out the entire delay. Early in training, mice indiscriminately accepted most offers. As mice learned to budget their time, they increased their earnings by becoming more selective, rejecting offers for longer delays. The proportion of rewarded trials among accepted offers increased, as mice gradually reduced their quitting behaviors mid-countdown. The time invested before quitting also declined with training.

Dopamine dynamics in the OZ scaled with the value of the offer, consistent with descriptions of dopamine's role in valuation. Early in training, when mice initially accept low-value offers and quit the trial mid-countdown in the WZ, we observed a decrease in dopamine, reflecting re-evaluation processes. As training progressed, and mice transitioned from quitting to rejecting suboptimal offers, decreases in dopamine shifted from the time of quit to the time of offer rejection. This suggests that decreases in dopamine may transition to the earliest decision to reject, reminiscent of the established phenomenon that, with training, dopamine transients shift from the time of reward receipt to the earliest predictive cue.

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Poster

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Title: Cerebellar activity modulates spatial working memory decisions and neuronal interaction between the prefrontal cortex and hippocampus in mice

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Abstract: Spatial working memory (SWM) is a cognitive skill supporting survival relevant behaviors, such as foraging for food. In rodents, the medial prefrontal cortex (mPFC) and dorsal hippocampal CA1 region (dCA1) are jointly necessary for SWM. SWM-based decisions about maze-arm entry are associated with increased coherence of local field potential (LFP) oscillations between the mPFC and dCA1. Coherence facilitates neuronal communication, suggesting that SWM decision making involves increased mPFC-dCA1 communication. The mechanisms controlling task-specific modulation coherence are unknown. Several studies have implicated the cerebellum in SWM and we recently showed that Purkinje cells in cerebellar lobulus simplex (LS) and Crus I represent phase differences between oscillations in the mPFC and dCA1. Here, we asked whether the role of the cerebellum in SWM involves the modulation of mPFC-dCA1 coherence. We tested SWM performance in 9 adult mice (5 females, 4 males) by observing spontaneous exploration behavior in a plus maze. All mice expressed Channelrhodopsin2 in cerebellar Purkinje cells, had extracellular recording electrodes implanted into the mPFC and dCA1 and an optic fiber (465 nm) implanted over LS for optogenetic stimulation. Photoactivating LS Purkinje cells in the early stages of the SWM decision process impaired SWM performance and multiple aspects of neuronal activity in the mPFC and dCA1, suggesting a causal involvement of the cerebellum in SWM decision making and in the modulation of multiple aspects of decision related activity in the mPFC and dCA1. LFP peak activity in mPFC and dCA1 reflected decision outcome, with correct decisions associated with higher peak values than incorrect decisions. Stimulation of LS disrupted the LFP activity and decision related differences. The power of LFP oscillations in both the mPFC and dCA1 reached peak values during decision making in control trials, which were disrupted in trials with cerebellar stimulation. The coherence of beta and gamma range oscillations in mPFC and dCA1 increased during SWM decision making with higher coherence values observed during correct decisions in control trials. Cerebellar stimulation disrupted decision related coherence increases, which also no longer differentiated between decision outcomes. Granger causality showed no SWM related changes in control trials. LS activation caused increased dCA1->mPFC causality in the gamma range during correct vs. incorrect decisions. Our results suggest a causal involvement of the cerebellum in SWM, possibly through the modulation of multiple aspects of neuronal activity and functional connectivity between the mPFC and dCA1.

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Poster

P800. Inhibitory Control in Human and Non-Human Primates

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Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

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Topic: H.04. Executive Functions

Support: NIH R01 NS102201

Title: Processing of attentional cues is selectively increased after action errors in the service of behavioral adaptation

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Abstract: Many theories of action error processing (e.g., Wessel, 2018; Purcell & Kiani, 2016) propose that errors trigger a processing cascade that involves attentional processes - such as orienting to the source of the error or the selective retuning of attentional representations of task relevant stimuli. Here, we aim to test whether attentional cues in the context of the Posner's attention network test (ANT; Posner & Rothbart, 2007) selectively benefit error processing. In the ANT, spatial cues provide valid information about where subsequent task-relevant stimuli will occur, thus directing attention to the cued location. The orienting effect is defined as the accelerated RT and increased accuracy with spatial (informative) vs. center cues (noninformative). However, little is known about the orienting effect and its underlying neural activities in post-error processing. We hypothesized that errors lead to an amplification of the beneficial effect of the attentional cues. Preliminary analyses of behavioral and EEG data (Fig.1) showed increased orienting effect on RT in post-error short RSI condition, and increased orienting effect on accuracy in post-error long RSI group. Time-frequency decompositions showed increased beta power (13-20 Hz) within 200ms following spatial vs. center cues in post error vs. post-correct condition. The increased beta power difference between spatial and center cues condition in post-error condition could reflect the enhanced attention triggered by spatial cues after error commission. This is consistent with Cómez et al. (2009), where increased beta power was found at the attended vs. unattended condition. Nonhuman studies with cats also showed sustained beta synchronization during attentional expectancy of stimuli (Roelfsema et al., 1997). In summary, we found that the orienting effect was modulated by error commission, in that errors were followed by greater orienting effect. Together with the beta effect, the results suggest processing of attentional cues is selectively enhanced after action errors in the service of behavioral adaptation.

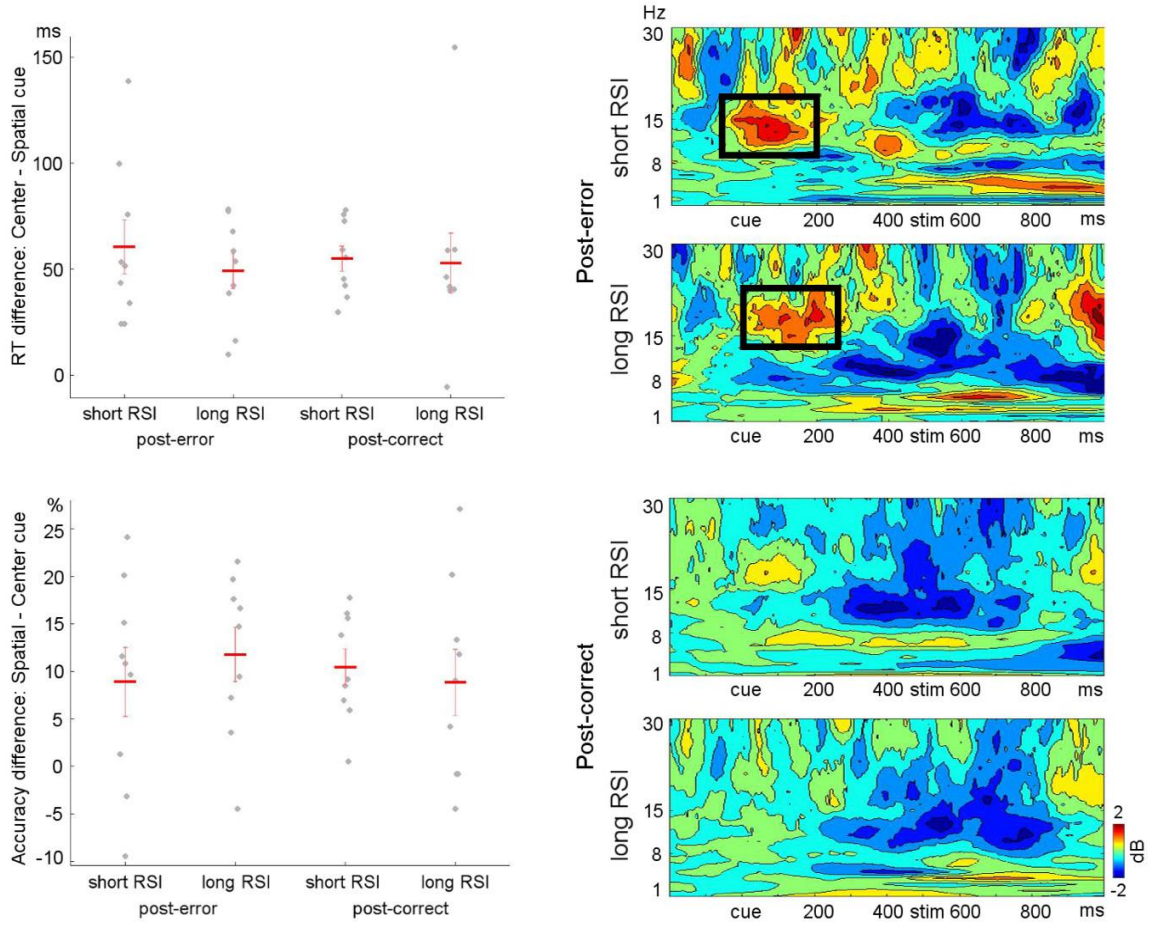


Figure 1. Behavioral results (left side) for reaction time (top left) and accuracy (bottom left) difference showing the orienting effect as a function of trial type (post-error vs. post-correct) and response-stimulus intervals (RSI; 1500 or 500ms) conditions. Grand mean time-frequency planes (right side) showing dynamic power changes at frontal midline electrodes (FCz Cz) relative to a 200ms pre-cue baseline as a function of trial type (post-error vs. post-correct) and RSI (long vs. short) conditions. Power values were obtained via subtracting power in center cue trials from spatial cue trials. Note the increased power at the beta band (13-20 Hz) after cue onset in the post-error conditions, which suggests orienting triggered by spatial cue facilitates the post-error processing.

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Poster

P800. Inhibitory Control in Human and Non-Human Primates

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Program #/Poster #: P800.02

Topic: H.04. Executive Functions

Support: NIH Grant R37 MH066078

Title: Assessing test-retest reliability of fronto-parietal fMRI activity patterns associated with cognitive control

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Abstract: A major goal of cognitive neuroscience is to understand the neural bases of individual differences in cognitive control abilities. Neural measures useful for individual differences analyses need to exhibit both high psychometric reliability and convergent validity. Task-activation fMRI has recently been questioned in terms of its suitability for studying individual differences, due to poor test-retest reliability (Elliot et al., 2020, *Psych. Sci.*). But, prior work has primarily assessed reliability using (a) classical test theory statistics (e.g., intra-class correlation), (b) traditional 'univariate' measures of neural activity, and (c) data from single cognitive control tasks (e.g., N-back). Here, we tested whether added sensitivity and reliability could be gained through a multivariate, multi-task approach. We adopted the "neural fingerprinting" approach (Finn et al., 2015, *Nat. Neurosci.*), leveraging the Dual Mechanisms of Cognitive Control dataset (Braver et al., 2021, *J. Cog. Neurosci.*), consisting of fMRI data acquired from human participants each performing four classic cognitive control tasks (AX-CPT, task switching, Sternberg, and Stroop). The sample of participants was split into two subsets: a subset (N = 15) that completed two testing sessions ("test" and "retest", >2 months apart), and a subset (N = 92) that completed only the first. A set of cortical regions commonly recruited across the four tasks by trials with high control demand was identified, primarily within fronto-parietal and cingulo-opercular areas. The neural activation pattern extracted from the high vs low-demand contrast, distributed across these regions, was treated as a multivariate analytic unit (one per task*participant). Pattern similarity was compared between and within-individuals, testing whether the pattern from an individual at one timepoint ("retest") was more similar to their pattern at another timepoint ("test"), relative to patterns from all other individuals at that timepoint — thus accurately identifying them via their neural fingerprint. In all comparisons, participants could be identified with high accuracy (80–100%), but critically, only when aggregating data across tasks into a single, task-general measure. Activation patterns extracted from matched-control brain regions did not demonstrate this sensitivity. Additional analyses explicitly compared multivariate and univariate measures in their reliability and sensitivity to individual differences Together, the results suggest that multivariate, multi-task fMRI may be a more sensitive and reliable tool for studying individual differences of cognitive control.

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Poster

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Topic: H.04. Executive Functions

Title: Forcing the winning horse: how the amount of force on a push button affects the inhibition of finger release in a stop signal task.

Authors: *S. COLANGELI[1], S. RAMAWAT[2,1], I. B. MARC[2,1], P. PANI[1], S. FERRAINA[1], E. BRUNAMONTI[1];
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Abstract: Suppressing a pending action is an essential ability when someone must cope with an unpredictable situation. This component of executive control has been extensively studied by the stop-signal task (SST). SST is a task requiring to respond with a movement to a go signal (go trials) but to refrain from the movement when occasionally a stop signal occurs after the go signal (stop trials). A theoretical model describes the outcome in stop trials as a race towards a threshold between a go and a stop process. Every time the go process runs faster than the stop process the movement cannot be interrupted (stop error trials). In addition, it has been hypothesized the go process as a two-stage process including: 1) a controlled stage (CS), that can be inhibited at any time; 2) a ballistic stage (BS), that cannot be inhibited. A point-of-no-return defines the crossing line between CS and BS. It has been reported that perceptual and cognitive variables can affect the duration of CS and influence the probability of the stop process to win the race. Besides these factors, for some effectors, the state of biomechanics variables could influence the start of the go process. For example, we can hypothesize that if we ask someone to push a button and then release it in response to a go signal, the strength applied to push the button at the beginning of a given trial could play a role in influencing the CS of button release and, consequently, the run of the go process against the stop process. Here we tested this hypothesis by studying 20 healthy volunteers performing a button release SST while monitoring the time evolution of CS by a force sensor resistor (FSR) fixed on a mouse button. The analysis of the perturbation of FSR revealed in a proportion of correctly stopped trials, defined as “partial errors”, an initial decline of the force applied on the button (as in go trials), followed by a rising back of it. This FSR pattern was never observed in stop error trials. Looking at the amount of force exerted on the button at the beginning of trials, it was significantly higher in partial error than in stop error trials, suggesting that trials started with an initial high force on the button have higher probability to turn in correct for higher differences between the force at the beginning of CS and that at the time of movement onset. Moreover, in partial errors the velocity of decay of the FSR signal was lower than in stop error trials, allowing subjects to stay within the CS for longer. These results point out that models of movement inhibition need to account for the biomechanical state of the effector before the go-signal.

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Poster

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Robin and Richard Patton through the E. Bronson Ingram Chair in Neuroscience

Title: Anterior cingulate cortex proactively controls and monitors action during saccade countermanding

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Abstract: Anterior cingulate cortex (ACC) is known as a hub for cognitive control. Convergent evidence across species and methodologies has supported its role in adapting behaviors and evaluating the outcome and value of actions. However, neurophysiological evidence for how ACC contributes to reactive and proactive control is sparse, and its role in conflict monitoring is debated. To address this we analysed a preliminary sample of 538 units recorded from 992 linear electrode contacts (50, 100, 150 µm spacing) across 18 independent sites. Sites spanned an 8 mm rostro-caudal zone, and contacts captured the dorsal (area 6/32) and ventral (area 24c) banks of the anterior cingulate sulcus of two macaque monkeys. Whilst recording neural signals, monkeys performed a saccade countermanding (stop signal) task, in which they were required to generate a saccade to a target on its appearance, but to inhibit this planned saccade when an infrequent stop-signal appeared. The time at which the stop-signal appeared varied, resulting in an equal probability of successful inhibition or committing an error. As such, this task offers a clear method in which neural signals can be attributed to the generation of actions and their monitoring. This analysis quantified the incidence of neurons (1) exerting reactive control, (2) detecting response conflict, or (3) nudging proactive control. We found no neurons meeting criteria to accomplish gaze-shifting or gaze-holding, nor modulating systematically enough within the relevant timeframe to exert reactive control of response inhibition. Likewise, we found no neurons meeting criteria to signal response conflict. Instead, we found numerous neurons that could enact proactive control by differentiating between successful and failed inhibition before presentation of the stop-signal. These neurons operated over different timescales: some recruited ~100 ms before the stop-signal and others predicting performance

during the preceding inter-trial interval. These results contradict a dominant theory of ACC function and offer new insights into the contributions of ACC to proactive control of behavior.

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Poster

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Title: Magnetoencephalography shows increase of right-inferior frontal gyrus beta power for response conflict - A generalized response inhibition function?

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Abstract: It is well established that the right-inferior frontal gyrus (R-IFG) is recruited during outright action stopping, and that this relates to increases in beta (12-30 Hz) power. Some work has suggested that inhibition of incorrect response tendencies contributes to overcoming response conflict. However, very little research has implicated R-IFG in conflict, and to our knowledge, no one has shown this R-IFG beta signature of response inhibition during conflict. In the current study, we analyzed existing high-resolution magnetoencephalography (MEG) data recorded from a small cohort (N = 8) that completed many trials (n > 10,000 total) of a response conflict paradigm (Little et al., 2019). Subjects saw a random dot motion that varied in coherence (high, medium, low) and whether it accurately (70%; no conflict) or inaccurately (30%; conflict) predicted a subsequent movement cue. We tested the novel hypothesis that R-IFG MEG sensors would show increased beta on conflict trials, particularly so when the level of conflict was strongest (i.e., on higher coherence trials). We first computed average event-related spectral perturbations (ESRPs) at the group level for conflict and no conflict trials. We saw increased R-IFG low beta power during conflict. This was specific to R-IFG, as we did not see a similar increase in left-IFG (L-IFG) beta power. We then ran linear mixed effects models for subject-

specific low beta activity at the single trial level. Supporting the qualitative picture from the ERSP results, there was a significant main effect of response conflict on subject-specific low beta in R-IFG ($X^2(1) = 12.11$, $p < .001$), and not in L-IFG. Additionally, there was a significant interaction between conflict and coherence on subject-specific low beta in R-IFG ($X^2(2) = 6.76$, $p < .05$). To visualize this interaction, we computed average ERSPs for conflict and no conflict trials, for high, medium, and low coherence trials separately. We saw increased R-IFG low beta power during conflict for high and medium coherence trials, but not for low coherence trials. Pairwise comparisons revealed the difference in R-IFG subject-specific low beta was significant for high coherence trials only ($Z = -4.02$, $p = .0001$). Our results show that R-IFG beta is recruited on response conflict trials, especially those that would appear to require punctate response inhibition. This suggests a more generalized role for response inhibition, beyond stopping tasks to conflict scenarios.

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Poster

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Topic: H.04. Executive Functions

Title: Beta-band in the premotor cortex of non-human primates predicts the strategy adopted in a selective stopping task

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Abstract: Among motor cortical oscillations, those in the Beta (β) frequency range [~12–30 Hz] have received particular attention. Different theories arose about the role of sensorimotor β activity in cognitive control, including those suggesting that β-activity subserves the maintenance of a sensorimotor set, like, for example, a strategy adopted by an agent moving in a changing environment. Here, we focused on the role of β- activity in encoding the strategy adopted by two rhesus monkeys (Macaca mulatta) trained in the Stop Selective Task (SST). In this task, participants are required to cancel the planned movement after a particular stimulus (Stop signal) but not after a different one (Ignore signal) occurring randomly after a Go signal during the reaction time (RT). Behavioural studies suggest that participants can adopt different strategies to adapt to the task demands, inhibiting the response only upon discriminating the Stop signal, or whenever detecting an imperative signal. We hypothesize that these different strategies rely on the relative prevalence of one of two inhibitory controls (i.e. proactive and reactive inhibition)

and that the level of β- activity can be used to predict which strategy will be adopted at the single trial level. To test this hypothesis, we recorded Local Field Potentials, by means of Utah arrays (96 Channels) implanted on the dorsal premotor cortex of the monkeys performing the SST, and used β-power as predictor of the strategy adopted. First, we confirmed that β-power decay is positively correlated with RTs, independently of the strategy emerging behaviorally, suggesting that β-activity is regulating the level of motor responsiveness. Secondly, we found that β-power exhibited different patterns of response to the presentation of the Ignore signal, increasing only when a certain strategy was behaviorally emerging. Then we analysed the after-Ignore single trial β-activity via a multiclass-classification algorithm and found that strategies can be described based on β-activity similarity between different trial types. Finally, we used a binary linear classifier to test if the level of β-activity before the Go signal could account for the strategy adopted. We found that, independently of the level of movement preparation (modelled by RT duration), β-band is a reliable predictor of the strategy. We therefore suggest that the β-activity level before the Go signal indicates the amount of proactive control and drives the subsequent response pattern to an imperative stimulus. Our results are thus in agreement with the hypothesis that β-band subserves the maintenance of a sensorimotor set.

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Poster

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Title: Cortical β-bursts following unexpected events in auditory, visual, and tactile modalities

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Abstract: The human brain constantly monitors the environment for unexpected events in the service of potentially adapting behavior. For example, in a cross-modal oddball (CMO) task, in which motor responses are made after (sometimes unexpected) sensory stimuli, the brain tracks their occurrence and adapts behavior by slowing reaction times. Using the CMO, Wessel and Huber (2019, PLoS Comput Biol) found that while the brain tracks Bayesian surprise terms separately for auditory and visual stimuli, a crossmodal fronto-central P3 event-related is

triggered by both events. Independent component analysis revealed that the neural generator underlying this P3 was also active after stop-signals in a separate task that required active motor inhibition, suggesting that surprising events trigger a common inhibitory network through which they slow reaction times. The present study fulfilled two purposes: First, we replicated and extended Wessel and Huber's findings to include a tactile modality. Second, we tested whether recently discovered indices of inhibition - namely, burst-like beta activity at fronto-central scalp sites (Wessel, 2020, J Neurosci) - also occurred after unexpected events. We recorded extracranial EEG from participants who completed a CMO task with unexpected auditory, visual, and tactile events. Like unexpected auditory and visual events, tactile events also induced motor slowing, particularly early in the experiment, when Bayesian surprise was high. Replicating Wessel & Huber, our analysis suggested that the fronto-central P3 reflects separate Bayesian surprise terms across the three sensory domains. In terms of beta bursts, both the original Wessel and Huber dataset (n = 55) and our current three-domain dataset (n=21) showed increased β-burst rates at the FCz electrode following unexpected events. These differences were significant within the first 200ms following unexpected auditory events in both experiments, and as early as 75ms following tactile unexpected events in the new data. These findings suggest β-bursts as a potential early marker of inhibition after unexpected events across sensory domains—i.e., even in contexts that do not explicitly require inhibition.

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Poster

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Support: Iowa State University

Title: Greater Adiposity Influences Working Memory and Cognitive Inhibition

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Abstract: INTRODUCTION: Obesity rates of adults in the U.S. have increased from 30.5% in 1999-2000 to 42.4% in 2017-2018, with a further projected prevalence to nearly 50% by 2030. Greater adiposity has been consistently associated with exacerbated brain atrophy and central glucose hypometabolism in prefrontal cortices that govern executive functions, including inhibitory control, working memory, emotion regulation, and cognitive flexibility. However, there is a paucity of research elucidating how neural physiology that governs these executive

functions is impacted by obesity. Thus, our primary aim of this study was to investigate whether such electrophysiological changes due to obesity could be detected in healthy young adults. **METHODS**: In this cross-sectional study, we recruited 60 participants (18-40 years, 55% female) and examined real-time neural electrophysiology using electroencephalography (EEG) on 34 lean participants (mean body mass index (BMI)=22.1±2) and 26 overweight/obese participants (mean BMI=34.9±6.1) while they completed four cognitive tasks which separately focused on inhibitory control, working memory, emotion regulation, and cognitive flexibility. Serum proteomics were collected, and glycemic control was quantified from glycated hemoglobin (HbA1c) serum measures. We directly assessed body composition via dual energy X-ray absorptiometry (DXA). Structural equation modeling was utilized in to probe direct/indirect associations between body composition, cardiometabolic biomarkers, and EEG waveforms. **RESULTS**: After controlling for age, sex, HbA1c, and DXA android/gynoid ratio, we found that higher body fat percentage was associated with a significantly slower response in order to reach trough amplitudes on the FC1 electrode (196.3±134.4 milliseconds (ms) versus 204.4±311.1ms; β=0.94, p<0.001), as well as a significantly slower response in order to reach peak amplitudes on the FT7 electrode (215.9±129.9ms versus 334.1±327.2ms; β=-0.537, p=0.019) during a Stroop task. Furthermore, we found that higher body fat associated with significantly slower responses in order to reach trough and peak amplitudes on the FT7 electrode (202.9±95.4ms versus 567.2±434.5ms, β=-0.87, p=0.032; and 543.4±205.2ms versus 613±333.9ms, β=-0.79, p=0.049, respectively) during an Operation Span task. No differences were observed between obese and lean subjects for emotion regulation or cognitive flexibility tasks. **CONCLUSION**: Young adults with greater adiposity may have impaired cognitive inhibition and decreased working memory capacity compared to lean counterparts.

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Poster

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Topic: H.04. Executive Functions

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Title: Task-instruction modulates intra-individual variability and motor response inhibition to negative stimuli in older and younger adults

Authors: *S. MIRAJKAR, S. E. WILLIAMS, J. D. WARING;
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Abstract: Intra-individual variability (IIV) refers to inconsistencies in an individual's behavioral task responses. Increased IIV may indicate inefficient cognitive processing and decreased central nervous system integrity. Previous literature has identified a relationship among higher IIV, age-related neurodegeneration and decline across several cognitive domains. In the domain of executive functioning for example, higher IIV is linked with poorer response inhibition. However, the impact of aging on the relationship between IIV and emotional response inhibition is an open question given reported age-related changes in emotion regulation. The present study examined if age and/or task instruction modulated the relationship between IIV and motor response inhibition to emotional stimuli. We used an emotional stop signal task that displayed human faces with happy, fear, or neutral expressions in 'Stop' trials. Older (age = 60-86 yrs) and younger (age = 17-27 yrs) men and women (N = 237) completed one of three versions of the task, which differed in instructions about the stop-signal: emotion was task-irrelevant for the stop-signal for two versions, and task-relevant in the third version. Stop signal reaction times (SSRT) were calculated for each facial expression separately, and fear and happy SSRTs were also compared to neutral. IIV was measured using the standard deviation of the response time for accurate 'Go' trials. We observed a significant relationship between response consistency and inhibition only when emotional attributes of the stop-signal were relevant to task goals; when task instructions required overt focus on emotion, the people with the more consistent responses (i.e., lower IIV) had more efficient response inhibition (i.e., lower SSRT) for fear expressions compared to neutral. Negative stimuli can promptly elicit attention and are more effective measures of involuntary interference to tasks than positive stimuli. Task-goals may amplify these characteristics of negative stimuli. Although IIV differed between age groups, age did not moderate the relationship between IIV and response inhibition, suggesting neural resources similarly support response consistency and inhibition in both older and younger adults.

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Poster

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Topic: H.04. Executive Functions

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Title: Functional connectivity dynamics of the frontoparietal and default mode networks during cognitive control in children with attention-deficit hyperactivity disorder

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Abstract: Cognitive control deficits in children with attention-deficit hyperactivity disorder (ADHD) have been associated with altered functional connectivity relative to typically developing (TD) children. Much functional connectivity work in ADHD has focused on stable connectivity assessed across several minutes. However, children with ADHD experience fluctuations in behavior on a much shorter timescale. Therefore, dynamic functional connectivity approaches that assess patterns of functional connectivity on the order of seconds are well-suited to understand how disrupted brain function underlies cognitive control deficits in ADHD. These approaches have found that more variable functional connectivity is associated with better cognitive control performance in TD children and reduced inattention in children with ADHD, however it is presently unclear whether variability in functional connectivity is associated with cognitive control performance in children with ADHD. To examine this, children with ADHD and TD children completed a Go/NoGo task in an MRI scanner. We used an edge-time series approach, which temporally unwraps traditional measures of functional connectivity and yields pairwise cofluctuation time series for each pair of brain regions in a network, to quantify task-based functional connectivity dynamics within the frontoparietal and default mode networks. We predicted that less variable functional connectivity in these two networks would be associated with fewer commission errors on the Go/NoGo task (i.e., better cognitive control performance) in both groups. In the TD group, Pearson correlations did not reveal significant relationships between functional connectivity dynamics and cognitive control performance. In the ADHD group, contrary to our hypothesis we found that more variable frontoparietal network, but not default mode network, connectivity was related to fewer commission errors. This analysis represents one of the first examinations of dynamic functional connectivity during active engagement of cognitive control in children with ADHD, and suggests that reduced variability (i.e., greater stability) of functional connectivity in the frontoparietal network may represent a mechanism underlying cognitive control impairments in children with ADHD. Future analyses will examine functional connectivity dynamics between the frontoparietal and default mode networks and during successful versus failed attempts at cognitive control in children with and without ADHD to further uncover patterns of functional connectivity dynamics that underlie cognitive control deficits in ADHD.

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Poster

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Title: Transient beta modulates decision thresholds during human action-stopping

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Abstract: Prefrontal beta oscillations are a mystery when it comes to their role in executive control. Action-stopping, an extensively studied executive function in humans, involves bursts of beta oscillations in prefrontal-basal ganglia regions. To determine the functional role of these beta bursts we utilized the well-developed computational framework, i.e. the Race Model describing action-stopping.
We incorporated beta bursts in three race model variants, hypothesizing that a transient increase in beta could 1) modulate decision thresholds, 2) change stop accumulation rates, or 3) promote the interaction between the Stop and the Go process. We then tested the model predictions using EEG recordings in humans (two datasets; N=12 and N = 23) performing a Stop-signal task, and found that the model variant in which beta increased decision thresholds for a brief period of time best explained the empirical data. This was confirmed by several observations. First, the threshold variant of the model predicted that the probability of responding during the stop trials would decrease if a beta burst occurred close and prior to latency of stopping, i.e. the stop-signal reaction time, SSRT. The experimental data revealed a similar trend. Next, this model variant also predicted an effect on go trials, unlike the other two variants, such that the time when a beta burst ended or the threshold reset is when the response was more likely to occur. In the experimental data, we observed this as an increase in the probability of burst end times close and prior to the go reaction times in comparison to a null distribution where the burst end times were distributed uniformly (Kolmogorov-Smirnov test, $p<0.001$). Finally, the model predicted during successful stop trials, the threshold modulation would likely occur just prior to the stopping time. The experimental data corroborated that finding with an increased probability of bursts occurring 50ms before the stopping time. The fitted model parameters indicated that beta bursts modulate Go process thresholds stronger than that of the Stop process (0.7 ± 0.1 vs 0.3 ± 0.1, $p=0.008$). These results suggest that prefrontal beta influences stopping by temporarily holding the response from execution. Our study further suggests that beta could act as a pause, where the Stop process can intervene and modulate behavior during the time beta increases the response thresholds, thus implying that human action-stopping could be a multi-staged process.
Overall, by introducing transient oscillations into the race model and testing against human electrophysiological data, we provide a novel functional account of the role of prefrontal beta in executive control.

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Poster

P826. Thalamic and Brainstem Circuits II

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Stanley Center at the Broad Institute
Hock E. Tan and K. Lisa Yang Center for Autism Research at MIT
James and Patricia Poitras Center for Psychiatric Disorders Research at MIT

Title: Anterior thalamic dysfunction underlies cognitive deficits in neuropsychiatric disease models

Authors: *Y. ZHANG[1], D. ROY[2], T. AIDA[2], S. CHOI[2], Q. CHEN[2], Y. HOU[2], N. LEA[2], K. SKAGGS[2], J. QUAY[2], M. LIEW[2], H. MAISANO[2], V. LE[2], C. JONES[2], J. XU[2], D. KONG[2], H. SULLIVAN[2], A. SAUNDERS[2], S. MCCARROLL[2], I. WICKERSHAM[2], G. FENG[2];
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Abstract: Neuropsychiatric disorders are often accompanied by cognitive impairments/intellectual disability (ID). It is not clear whether there are converging mechanisms underlying these debilitating impairments. We found that many autism and schizophrenia risk genes are expressed in the anterodorsal (AD) subdivision of anterior thalamic nuclei, which has reciprocal connectivity with learning and memory structures. CRISPR-Cas9 knockdown of multiple risk genes selectively in AD thalamus led to memory deficits. While AD is necessary for contextual memory encoding, the neighboring anteroventral (AV) subdivision regulates memory specificity. These distinct functions of AD and AV are mediated through their projections to retrosplenial cortex, using differential mechanisms. Furthermore, knockdown of autism and schizophrenia risk genes *PTCHD1*, *YWHAG*, or *HERC1* from AD led to neuronal hyperexcitability, and normalization of hyperexcitability rescued memory deficits in these models. This study identifies converging cellular to circuit mechanisms underlying cognitive deficits in a subset of neuropsychiatric disease models.

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Poster

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Title: Responses to local and global deviance in subcortical structures

Authors: *A. MAZANCIEUX[1], F. MAUCONDUIT[2], A. AMADON[2], F. MEYNIEL[1];
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Abstract: The brain faces a stochastic environment, the ability to detect regularities and their violations is thus crucial to adapt our behaviour. Previous proposals [1] posit that the noradrenergic (NA) system plays a key role in the detection of surprising events and the adjustment of behavior. Supportive evidence comes from non-human animals and pharmacological manipulation or recording of the Locus Coeruleus (LC), the major NA nucleus; in humans it comes from indirect measures like the P300 component and pupil dilation, which are NA-dependent [2]. Few studies more directly measured the LC activity with fMRI [3], some of them did not use procedures to locate this nucleus accurately. Here, we tested in 15 human subjects whether NA increases for unexpected stimuli using pupilometry and dedicated fMRI methods (including neuromelanin-sensitive images to locate the LC in each subject). We extended previous findings by investigating the anatomical specificity in the pons but also neuromodulator specificity by comparing with the substantia nigra (SN) related to dopamine (DA). We also extended the classical auditory oddball task [2, 3] and used the "local-global" paradigm [4] that distinguishes two temporal scales for deviance. Stimuli consisted of patterns of four identical sounds plus a fifth one that could be either identical or different, corresponding to a local (within-pattern) standard and a local-deviant respectively. Each pattern type was either frequent (80%) or rare (20%) in a sequence, corresponding to a global (across patterns) standard and a global deviant, respectively. . Pupil size transiently increased for local standards and local deviants when they were global deviants. Using event-related analyses, we found the same result in the LC activity and SN. We demonstrate anatomical specificity by showing that the effect of deviants decreased in voxels further from the LC. Overall, our results confirm the role of catecholamines in the detection of unexpected stimuli and validate a method to investigate neuromodulator-specific subcortical structures to test other hypotheses in future studies.

[1]Aston-Jones & Cohen (2005) *J Comp Neurol* [2] Nieuwenhuis et al (2011) *Psychophysiology* [3] Murphy et al (2014). *Human brain mapping* [4] Bekinschtein et al. (2009) *PNAS*

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Poster

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Title: Neural ensemble dynamics of auditory thalamus upon sensory learning

Authors: *M. HASEGAWA[1,2], Z. HUANG[2], J. GRÜNDEMANN[1,2];
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Abstract: Sensory cortices receive and process uni- and multisensory information to guide behaviours. However, so far, the encoding of uni- and multisensory inputs during complex behaviours in sensory thalamic nuclei preceding cortex is poorly understood. Here, we established a gradient refractive index lens-based two-photon calcium imaging approach to monitor the activity of large populations of individual auditory thalamus (medial geniculate body, MGB) neurons in mice during a multisensory go/nogo task. We tracked the same populations of MGB neurons across learning and found distinct ensembles of MGB neurons that encode auditory, visual and multisensory stimuli as well as task features. The activity of individual MGB neurons was plastic and their sensory responses changed dynamically across reward-association and task learning. Furthermore, using dimensionality reduction, go/no-go stimuli could be well distinguished on the population level across the learning phases independent of the stimulus modality. Our data suggests that MGB neurons flexibly encode multisensory information across learning supporting the view that sensory thalamic nuclei are actively involved in cognitive processes beyond a classic relay function.

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Poster

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Title: Sex-specific effects of paraventricular thalamic nucleus functional connectivity on depression and anxiety symptoms in humans

Authors: *S. M. KARK[1], S. J. GRANGER[1], J. G. ADAMS[1], L. MCMILLAN[1], I. CHEN[2], R. M. BENCA[2], M. A. YASSA[1];
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Abstract: The paraventricular thalamic nucleus (PVT) is a subnucleus of the dorsal midline thalamus. The PVT is a sensor and integrator of salient experiences and memories—both rewarding and aversive—functions made possible partly by its reciprocal connections with the nucleus accumbens (NAc) and amygdala (AMY). We recently established that resting-state functional connectivity (RSFC) of the human PVT is highly consistent with this region's known anatomical and functional connections observed in rodents. Disruption of this circuitry is associated with depressive- and anxiety-like behavior in rodents, often as a function of sex, suggesting aberrant RSFC of the PVT might contribute to the severity of psychopathological symptoms in humans. Here, we collected self-reported depression and anxiety scores (Beck Depression Inventory-II and Beck Anxiety Inventory), and resting-state functional magnetic resonance imaging (fMRI) scans in a human sample (n=75, 55 females) that endorsed a range of depression and anxiety symptom severity. Factor analysis applied to the depression and anxiety symptom scores empirically differentiated symptom classes in this sample. RSFC was calculated as the time-series correlations between the activity of the PVT with the AMY and NAc. In females, but not in males, greater PVT-AMY RSFC was associated with higher anxiety symptoms. In males, but not in females, greater PVT-NAc RSFC was associated with more severe symptoms overall, but with anhedonia-like symptoms in particular. These findings suggest that changes in the PVT circuitry impact depression and anxiety symptoms in sex-specific ways.

Disclosures: S.M. Kark: None. **S.J. Granger:** None. **J.G. Adams:** None. **L. McMillan:** None. **I. Chen:** None. **R.M. Benca:** None. **M.A. Yassa:** None.

Poster

P826. Thalamic and Brainstem Circuits II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P826.05

Topic: H.08. Learning and Memory

Support: NIH Grant R01EY028219

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R01MH126351, and the Picower Institute Innovation Fund

Title: Temporal dynamics and function of locus coeruleus norepinephrine during learned behaviors

Authors: *V. BRETON-PROVENCHER, G. T. DRUMMOND, M. SUR;
Dept. of Brain and Cognitive Sci., MIT, Cambridge, MA

Abstract: The release of norepinephrine (NE) from locus coeruleus (LC) neurons has been linked to multiple functions including arousal, action and sensory gain, and learning. How and whether specific components of behavior are encoded by LC-NE activation remains poorly understood. Here we show that distinct temporal dynamics of LC-NE activity enable two functions during learned behavior - facilitating task execution and encoding reinforcement to improve performance accuracy. To examine these functions, we trained mice on a go/no-go auditory task with graded stimulus detection and task performance. Optogenetic inactivation demonstrated that LC-NE affects this behavior in two ways. First, silencing LC-NE activity affects the animal's ability to respond to low stimulus intensities. Second, silencing LC-NE activity changes response bias, or the animal's ability to adjust its behavior on subsequent trials based on the outcome of their decision. Targeted recordings of LC-NE neurons using photo-tagging showed that transient LC-NE activation preceded behavioral execution and followed reinforcement. Signal levels of LC-NE execution or reinforcement activation depended on stimulus evidence. The LC-NE execution signal scaled up with the go stimulus intensity, hence encoding reward expectation. The LC-NE reinforcement signal was highest after a punishment and scaled up with surprise, including reward following low stimulus evidence. Together, our results suggest that differential timing of LC-NE activity is critical in supporting complementary roles in learned behavior. In a separate study (*see companion poster Drummond et al.*), we show that these distinct temporal signals are also spatially segregated among LC-NE cortical outputs, and that these spatiotemporal dynamics further refine LC-NE roles in learned behavior.

Disclosures: V. Breton-Provencher: None. **G.T. Drummond:** None. **M. Sur:** None.

Poster

P826. Thalamic and Brainstem Circuits II

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Topic: H.08. Learning and Memory

Support: NIH Grant R01EY028219
NIH Grant R01MH126351
Picower Institute Innovation Fund

Title: Targeted functions of locus coeruleus norepinephrine in a learned behavior

Authors: *G. DRUMMOND, V. BRETON-PROVENCHER, M. SUR;
Dept. of Brain and Cognitive Sci., MIT, Cambridge, MA

Abstract: The locus coeruleus (LC) serves as the primary source of the neuromodulator, norepinephrine (NE), in the brain. Through a widely divergent set of projections, LC neurons have been suggested to release NE throughout the brain to regulate arousal and attention. However, LC-NE is also implicated in more precise roles such as mediating learning, promoting task execution, and signaling unexpected uncertainty. How LC-NE activity facilitates distinct aspects of behavior is unknown. We have found that in mice performing an instrumentally conditioned go/no-go task with graded auditory stimulus detection, LC-NE activity dually reflects both behavioral execution and reinforcement encoding (*see companion poster by Breton-Provencher et al.)*. Silencing this LC-NE activity decreases performance by reducing responses on low stimulus evidence trials, and by diminishing the effect of trial history on task performance. Here, we explored whether these temporally distinct task execution and performance optimization signals are also spatially segregated, such that spatiotemporal targeting of LC-NE release could enable modulation of multiple aspects of a learned behavior. Using retrograde tracing, we found that LC-NE neurons exhibit a modular organization with a subset of LC-NE neurons projecting to either motor cortex or prefrontal cortex, but not the other cortical region. Using two-photon calcium imaging of LC-NE axons, we found that the two phasic components of LC-NE activity were heterogeneously distributed among cortical regions such that the task execution signal was higher in motor cortex while the negative reinforcement signal was widely distributed among cortical targets. Optogenetic silencing of LC-NE axons specifically in the motor cortex recapitulated the effects of whole LC inactivation on task execution, but did not alter the effect of trial history, whereas optogenetic silencing of LC-NE axons specifically in the prefrontal cortex alone did not affect task performance. Together, these results suggest that modular targeting of LC outputs enables diverse functions whereby some NE signals are segregated amongst targets to facilitate task execution, while others are broadly distributed to promote reinforcement encoding and performance optimization.

Disclosures: G. Drummond: None. **V. Breton-Provencher:** None. **M. Sur:** None.

Poster

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The Picower Institute Innovation Fund

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Title: Rodent Lateral Posterior (LP) thalamus input to Anterior Cingulate Cortex (ACC) tracks ongoing and post-factum perceptual uncertainty

Authors: *Y. LEOW[1], T. LUO[1], M. SUR[2];
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Abstract: Perceptual uncertainty guides sensory-driven actions, and can be conveyed through multiple sensory and associative brain areas. The LP thalamic nucleus, is a higher order visual thalamus and the rodent homolog of the primate pulvinar. LP is extensively connected to multiple visual and associative cortical areas. The medial subdivision of the pulvinar/LP also has reciprocal connections with the prefrontal ACC, an area important for action selection and top-down modulation of visual processing. Interaction between the pulvinar and ACC has been associated with impaired sensory filtering and over-responsiveness to stimuli. We hypothesized that by integrating input from multiple visual areas and the parietal cortices, higher order visual information conveyed by LP to ACC contribute to decision-making computations performed within the ACC. Here, we imaged calcium activity of LP→ACC axons with two-photon microscopy in mice (n = 6 mice, 10 sessions, ~400-500 boutons) trained on a global motion direction discrimination task under a two-alternative forced choice paradigm. To serve as a proxy for sensory-driven perceptual uncertainty, we varied the proportion of coherent dots moving in target directions. Task-responsive LP→ACC activity varied with multiple task variables on different timescales. To account for time-varying task events and motor activity, we employed a linear encoding model (Generalized Linear Model, GLM) to predict LP→ACC axonal activity with behavioral variables, as well as to examine their relative contributions to LP→ACC encoding. We found that a significant proportion (~30%) of LP→ACC axons show coherence-dependent responses throughout different epochs of the task. Furthermore, perceptual uncertainty-associated activity often persisted beyond the visual stimulus presentation period, into time of choice and post-reinforcement. Some LP→ACC axons also exhibited reward-associated responses that scaled with the coherence of the preceding stimulus. Our results suggest LP involvement in decision-making beyond ongoing visual processing and perceptual evidence accumulation to include a potential involvement in conveying information on post-factum perceptual uncertainty.

Disclosures: Y. Leow: None. **T. Luo:** None. **M. Sur:** None.

Poster

P904. Schizophrenia: Molecular Mechanisms III

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Program #/Poster #: P904.01

Topic: H.13. Schizophrenia

Support: NIMH MH107487
NIMH MH121102

Title: Altered purinergic receptor transcript expression in the frontal cortex in Schizophrenia

Authors: *A. LUNDH[1], J. A. HOEFLINGER[3], S. ASAH[1], K. ALGANEM[1], R. ALNAFISAH[1], R. E. MCCULLUMSMITH[1], S. M. O'DONOVAN[2];
[2]Neurosci., [1]Univ. of Toledo, Toledo, OH; [3]Ohio State Univ., Columbus, OH

Abstract: Schizophrenia (SCZ) is a neuropsychiatric disorder that is estimated to affect around 1% of the United States population and is characterized by psychotic symptoms such as hallucinations and delusions, as well as reduced emotional expression, motivation, cognitive and motor impairment. SCZ is difficult to diagnose and the mechanisms behind it are poorly understood. Dysregulation of the purinergic system is implicated in SCZ, including the receptors which modulate the activity of purine neurotransmitters like adenosine triphosphate (ATP) and ADP. ATP acts on the purinergic P2X receptors, which consists of 7 different receptors (P2X1-7). ATP and other purines also act on the P2Y receptors (P2Y1, 2, 4, 6, 11, 12, 13, & 14). However, expression of P2Rs in neuropsychiatric disorders has not been explored. To address this gap in our knowledge, we assayed P2X (P2X4, 5, 7) and P2Y (P2Y12, 13) receptor transcript expression in the frontal cortex of SCZ and non-psychiatrically ill controls (n=20/group) using qPCR. We also mined postmortem brain transcriptomic datasets using the R shiny application Kaleidoscope (kalganem.shinyapps.io/Kaleidoscope/), to assess P2 receptor expression in different brain regions (anterior cingulate cortex (ACC) and dorsolateral prefrontal cortex (DLPFC)) in different diseases (SCZ, Major depressive disorder (MDD) and bipolar disorder) and a model (chronic unpredictable stress (CUS)) of neuropsychiatric disorders. We found that P2X7 and P2X4 receptor transcript expression was significantly altered (Student's t-test, $p<0.05$) in the frontal cortex in SCZ. There was no significant change in expression of P2Y receptors. In silico analysis also confirmed alterations in the expression (Log2FC) of different P2X receptor transcripts in SCZ. The use of antipsychotic medications and purinergic receptor expression was also analyzed in silico and alterations in the expression (Log2FC) of different P2X receptor transcripts were found with antipsychotic treatment. Protein expression and localization of P2Rs will also be qualitatively assessed in the frontal cortex of SCZ subjects (n= 3-5 slides/subject, n=3 subjects/grp) using immunohistochemistry. In summary, these data suggest that purinergic receptor expression is altered in a region-specific manner in SCZ and may contribute to the pathophysiology of disease. Future studies will expand upon these findings, including an analysis of cell-specific alterations of the purinergic receptor expression in SCZ.

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Poster

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Program #/Poster #: P904.02

Topic: H.13. Schizophrenia

Support: NIMH MH107487
NIMH MH121102

Title: Perturbation of Cell-subtype specific Active Kinome Networks in Schizophrenia

Authors: *A. S. IMAMI[1], E. S. SHEDROFF[1], A. W. JOYCE[1], S. SAHAY[1], K. ALGANEM[1], W. RYAN, III[1], N. D. HENKEL[1], R. E. MCCULLUMSMITH[1,2];
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Abstract: Abstract
Perturbation of Cell-subtype specific Active Kinome Networks in Schizophrenia
Imami AS, Shedroff ES, Joyce AW, Sahay S, Alganem K, Ryan W, Henkel ND, McCullumsmith RE
Schizophrenia is an illness with significant consequences for patients and their families. Since the disease affects executive function, the Dorsolateral Pre-frontal Cortex (DLPFC) has been an area of interest. We previously reported perturbed protein kinase activity of kinases, including AKT, in postmortem brain samples of patients with schizophrenia. We are now extending this hypothesis to the cell level, with a focus on frontal cortical pyramidal neurons. We used laser capture micro dissection to isolate DLPFC pyramidal neurons from matched pairs of schizophrenia and control PM brain samples (n = 20). We then utilized the PamChip STK kinome array assay for high throughput characterization of kinase activity in the samples. We then deployed well-characterized bioinformatics approaches to identify upstream kinases implicated in schizophrenia. We identified several kinases of interest, including c-Jun N-terminal kinases (JNK), extracellular signal-regulated kinases (ERK) and p38 mitogen-activated protein kinases (P38). These kinases belong to the mitogen-activated protein (MAP) kinase family and are associated with inflammatory responses. We then applied a novel technique of identifying the kinase interaction networks in high throughput kinase activity data. This modelling allowed us to identify the network-level changes in pyramidal neurons in schizophrenia. This is the first study to examine the subkinome at the cellular level in schizophrenia and to identify active kinome network changes in this often devastating illness. Our data provides an important new starting point for understanding how signaling networks are perturbed in severe mental illness.
Keywords: Active Kinome, Computational Modelling, Schizophrenia, Bioinformatics, MAP Kinases

Disclosures: A.S. Imami: None. **E.S. Shedroff:** None. **A.W. Joyce:** None. **S. Sahay:** None. **K. Alganem:** None. **W. Ryan:** None. **N.D. Henkel:** None. **R.E. McCullumsmith:** None.

Poster

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Program #/Poster #: P904.03

Topic: H.13. Schizophrenia

Support: MH107487

Title: Limited common gene enrichment in astrocyte subpopulations and psychiatric disorder associated biological pathways

Authors: X. ZHANG, A. WOLFINGER, R. SHUKLA, A. LUNDH, R. MCCULLUMSMITH, **S. M. O'DONOVAN**;
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Abstract: Astrocytes play an essential role in normal and pathological brain function. Clinical and experimental evidence implicate critical roles for astrocytes in the pathogenesis of serious mental illnesses but our understanding of the role of astrocytes in neuropsychiatric disorders is still limited. There is also growing evidence that astrocytes are targets of psychotropic medications. Using a bioinformatic approach, we conducted enrichment analysis of publically available gene signatures from murine astrocyte cell subtypes (https://holt-sc.glialab.org/sc/), human psychiatric (Schizophrenia, major depression) and neurological disease signatures (Alzheimer's disease, epilepsy) obtained from DisGeNet (www.disgenet.org) and drug signatures from DSigDB (http://dsigdb.tanlab.org/DSigDBv1.0/). In enrichment analysis of disease and astrocyte subpopulations, the density index score, indicating similarity of gene enrichment across datasets, was highest for major depression related signatures and astrocyte subpopulations. However, enrichment of astrocyte-subpopulation gene signatures and different psychotropic drug (e.g antipsychotics) signatures was low. Interestingly, gene ontology analysis identified enrichment of immune, metabolic and signaling processes in different astrocyte subpopulations and across psychiatric disorders and Alzheimer's disease, in line with previous reports of dysregulated biological processes in these disorders. In particular, common enrichment of an astrocyte subpopulation which may be associated with neurogenesis, and depression signatures were observed at the biological pathway level. Overall, similarities in gene enrichment were found in astrocyte subpopulations and neurological disorders, but there was limited common enrichment with psychotropic drug signatures.

Disclosures: X. Zhang: None. **A. Wolfinger:** None. **R. Shukla:** None. **A. Lundh:** None. **R. McCullumsmith:** None. **S.M. O'Donovan:** None.

Poster

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Program #/Poster #: P904.04

Topic: H.13. Schizophrenia

Support: NIH R01 MH107487

Title: The role of the ATPome in cognitive disorders

Authors: *X. WU, X. ZHANG, R. SHUKLA, A. JOYCE, H. E. EBY, R. E. MCCULLUMSMITH, S. M. O'DONOVAN;
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Abstract: Major depressive disorder (MDD) and schizophrenia (SCZ) are devastating cognitive disorders that greatly reduce patients' life quality and are costly to our society. Despite decades of investigation, available treatments for these disorders are only partially efficacious. To develop more effective pharmacotherapies, a more sophisticated understanding of disease pathophysiology is required. Adenosine triphosphate (ATP) is the brain's main energy carrier. A large family of enzymes called ATPases utilize ATP to carry out many essential cellular functions. ATPase dysfunction is widely reported in a variety of cognitive disorders, including SCZ and MDD. To assess how ATPase dysfunction may contribute to the pathophysiology of SCZ and MDD, we conducted bioinformatic analysis. We analyzed 24 publically available MDD and SCZ transcriptomic datasets (14 SCZ and 10 MDD, male and female postmortem brain tissue) using two ATPase targeting gene lists, one bioinformatically predicted and one experimentally demonstrated. We extracted significant gene expression data for the ATPase targeting genes from the transcriptomic datasets. Next, we performed pathway enrichment and pathway clustering (parent-child analysis) to determine which biological processes these significant ATPase genes are involved in. We identified approximately 300 significantly altered genes from each dataset based on the median number of significant ATPase related genes, confirming dysregulation of ATPases in MDD and SCZ at the transcript level. We found similar significant pathway clusters across all disease and sex groups. However, pathway analysis identified significant enrichment of metabolic and stress response pathway clusters in male MDD datasets, suggesting that ATP utilization by ATPases is dysregulated across cognitive disorders but with unique disease- and sex-specific differences. These findings provide insight into dysregulation of ATPase and ATP utilization in the pathophysiology of cognitive disorders.

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Poster

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Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P904.05

Topic: H.13. Schizophrenia

Title: Assessing the validity of methods to report a schizophrenia diagnosis.

Authors: *G. WOOLWAY, S. LEGGE, I. JONES, J. WALTERS;
Cardiff Univ., Cardiff, United Kingdom

Abstract: Background: Participant diagnoses in psychiatric research are derived from a variety of sources, ranging from self-report to those determined following lengthy diagnostic research interviews. Schizophrenia case definitions usually rely on detailed research interviews, which impacts sample sizes and generalisability of clinical cohorts. In this study we aimed to (i) assess the reliability and validity of self-report, clinician report and diagnostic interview methods of diagnosing schizophrenia within a research context and, (ii) investigate whether schizophrenia cases ascertained via these different methods display differences in genetic liability to schizophrenia. **Method:** Participants were taken from the National Centre for Mental Health (NCMH; n=292) and CardiffCOGS (n=600) cross-sectional studies. These cohorts collected multiple sources of diagnoses including self-reported, clinician diagnosis and research diagnosis obtained from a clinical assessment based on the SCAN interview. We investigated the positive predictive values (PPV) of self-reported and clinician diagnosis compared to the gold standard research diagnosis. Polygenic risk scores (PRS) were calculated using the latest schizophrenia GWAS to compare the genetic liability of participants that only provided a self-reported diagnosis with participants that continued participation to obtain the research diagnosis. **Results:** The PPV of self-reported schizophrenia to a formal research diagnosis of schizophrenia (n=78) was 76%, and increased to 85% when including both schizophrenia and schizoaffective depressed research diagnoses. The PPV of a clinician diagnosis to a research diagnosis of schizophrenia (n=22) was 82%. In comparison, the PPV of self-reported (n=59) broader psychosis to a research diagnosis of schizophrenia was only 41%. We found no evidence for a significant difference in the schizophrenia PRS between participants who had a self-reported diagnosis only and participants who had a research diagnosis in NCMH (OR=0.93; 95% CI=0.72,1.19; P=0.55) or CardiffCOGS (OR=1.02; 95% CI=0.86,1.22; P=0.78). **Discussion:** These findings show that a self-reported diagnosis of schizophrenia has a high PPV of receiving a schizophrenia diagnosis from research interview and that there are not significant differences in genetic liability to schizophrenia. Less stringent methods of assessing a schizophrenia diagnosis may be useful to gain larger sample sizes.

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Poster

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Title: Substance use in individuals at high genetic and clinical high risk of psychosis

Authors: *C. AMIR[1], C. BEARDEN[1], S. WOODS[2], D. PERKINS[3], E. WALKER[4], T. CANNON[2], B. CORNBLATT[5], K. CADENHEAD[6], J. ADDINGTON[7], D. MATHALON[8], B. STONE[9], M. KESHEVAN[9];
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Abstract: Substance abuse occurs at elevated rates in patients with chronic schizophrenia and a role for cannabis has been suggested in the onset of psychosis, but substance use in populations at high risk for developing psychosis is limited. Here, we aim to elucidate the relationship between substance use and psychosis in two distinct high risk populations. Specifically, we examined substance use in both youth at clinical high risk (CHR) of developing psychosis (n = 735; mean age = 18.50; 42.45% female) and age-and sex-matched typically developing (TD) controls (n= 278; mean age = 19.60, 49.64% female), and participants with chromosome 22q11.2 deletion syndrome (22q11DS, n= 82; mean age= 16.67, 55% female), one of the strongest genetic risk factors for psychosis, compared to age-and sex-matched TD controls (n= 72; mean age = 15.93, 58% female). We found significantly greater alcohol and THC use severity in CHR participants compared with controls at baseline. While TD males had higher tobacco, alcohol, and THC use severity than females, there were no sex differences in CHR drug use severity. Results of mixed modeling of substance use over time indicated that CHR youth had higher alcohol and tobacco use severity compared with controls, and higher THC use severity, with significant increases in use over time. Clinical status was assessed at the two-year follow-up period:109 CHR participants were in remission, 92 were symptomatic, 71 met criteria for prodromal progression, and 90 had developed psychosis. All CHR subgroups, except those whose symptoms remitted, had greater baseline tobacco use than controls. CHR participants who were symptomatic, those who were in remission, and those who developed psychosis had greater baseline THC use than controls. These results together support higher rates of substance use in CHR youth relative to controls. However, in youth with 22q11DS, we did not find elevated rates of substance use relative to controls. This points to a potential difference in substance use

patterns between these two high risk populations. These results suggest that there may be secondary risk factors at play in CHR substance use, and to the need to examine symptom progression as it relates to substance use.

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Poster

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Title: No difference in cavum septum pellucidum prevalence, length, or volume in a large psychosis spectrum sample compared to healthy controls

Authors: *M. Y. GOTRA[1], J. WREN-JARVIS[2], H. CHURCH[2], C. BIRCH[2], E. N. HERMS[2], C. A. TAMMINGA[3], E. I. IVLEVA[3], G. D. PEARLSON[4,5], M. S. KESHAVAN[6], J. A. SWEENEY[7], E. S. GERSHON[2], B. A. CLEMENTZ[8], J. E. MCDOWELL[8], J. R. BISHOP[9], S. K. HILL[1], S. K. KEEDY[2];
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and Neurosci., Univ. of Georgia, Athens, GA; [9]Departments of Pharm. and Psychiatry, Univ. of Minnesota, Minneapolis, MN

Abstract: Background: Cavum septum pellucidum (CSP) is an atypical brain structure which, by some accounts, has higher prevalence in psychiatric disorders. CSP is hypothesized to be a sign of early abnormal neurodevelopment which may also be associated with psychotic disorders. Some reports suggest that an abnormally large CSP (≥6 mm) is associated with cognitive deficits in psychosis samples. However, this has not been systematically investigated in a large sample. The present study utilized standard as well as novel measurements of CSP, and assessed the relationship between CSP metrics, psychiatric status, and cognition. **Methods:** Participants were 1,266 psychosis spectrum patients (26.9% bipolar with psychotic features, 32.8% schizoaffective disorder, and 40.4% schizophrenia) and 734 healthy controls from the Bipolar Schizophrenia Network on Intermediate Phenotypes 1 and 2 studies who completed a T1 weighted magnetic resonance imaging scan. Mean age was 37 years (50.3% male) for the patient group and 35.5 years (41.8% male) for the healthy group. Five raters, blind to group membership, rated the CSP as present or absent, determined the length of the CSP (number of slices anterior to posterior), and hand-traced the CSP boundaries for volume, all following a reliability process. Chi-Square analysis examined the prevalence of CSP in healthy controls compared to psychosis spectrum patients. Mann-Whitney U tests were used to assess for group differences in CSP length and volume. Cognition was assessed using the Brief Assessment of Cognition in Schizophrenia (BACS). The association of CSP with cognition was examined using analysis of variance with age, sex, and race as covariates and Spearman correlations with false discovery rate correction for multiple comparisons. **Results:** Psychosis spectrum patients did not have a higher prevalence of CSP compared to healthy controls (74.4% of patients; 75.1% of controls) and there was no difference in the proportion of patients with small, intermediate, and large CSP. There were no group differences in CSP length or volume. BACS scores did not differ between participants with and without a CSP or among the length categories, regardless of group. **Conclusion:** In this large sample, no approach to CSP quantification, including binary, categorical size, or absolute size, revealed any difference between psychosis patients compared to healthy controls. In contrast to some prior reports, an abnormally large CSP was not associated with greater cognitive deficits. This suggests that within the idiopathic psychosis population, the presence of a CSP may be a normal variant with limited impact on psychiatric or cognitive outcomes.

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Poster

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Topic: H.13. Schizophrenia

Support: Grant from Takeda Pharmaceuticals Ltd. to Cardiff University

Title: Investigating genetic alleles shared between schizophrenia and blood metabolite concentrations

Authors: *S. E. SMART, A. F. PARDIÑAS, L. HUBBARD, J. T. R. WALTERS;
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Abstract: Introduction
Metabolites are small molecules present in biological samples (e.g. tissue, blood, urine, cerebrospinal fluid). In recent years large metabolite datasets have become available and valuable insights into the genetic architecture of metabolites have emerged. Metabolites have also been investigated as potential biomarkers for schizophrenia and other neuropsychiatric disorders, though to our knowledge no replicated findings have emerged. Our aim in this study was to identify genetic variants that influence both schizophrenia and measurable metabolites.
Methods
Genome-wide association study (GWAS) summary statistics for metabolites were acquired from three studies: TwinsUK/Kora F4 (Shin, et al. 2014; N=7,824), SP2 (Chai, et al. 2019; N=1,954), and VUMC (Dennis, et al. 2021; N=up to 88,107). The schizophrenia summary statistics were acquired from the Psychiatric Genomics Consortium Schizophrenia Wave 3 (SWG of PGC, 2020; N=67,390 cases and N=94,015 controls).
All GWAS summary statistics were aligned to hg19, the MHC region (chr6:28477797-33448354) was removed, as were SNPs with MAF < 1% or INFO < 0.9. We performed exploratory pairwise GWAS comparisons between each metabolite and schizophrenia using GWAS-pw (Pickrell, et al. 2016). Genomic loci were defined using independent blocks of linkage disequilibrium as defined by Berisa and Pickrell (2016). Following the example of Pickrell, et al. 2016 and Giambartolomei, et al (2014), we identified loci where the posterior probability was >90% that the locus containing a genetic variant that influences both the metabolite and schizophrenia.
Results
Of the 801 GWAS summary statistics tested, there were 16 metabolites where the posterior probability of a locus containing a genetic variant that influences both the metabolite and schizophrenia was greater than 90%. Thirteen of these metabolites were identifiable (two monohexosylceramides, vitamin B1, blood platelet volume, monoclonal antibodies, immature granulocytes, glucose, eosinophils, complement C4, urea nitrogen, albumin, fibrinogen cleavage peptide, and indolelactate), while three had not been characterised. For 14 of the metabolites, only one locus reached this probability threshold, but for the other 2 metabolites, two loci passed this threshold (blood platelet volume and complement C4).
Discussion
We have shown co-localisation of genetic association signals for schizophrenia and a number of

metabolites. We intend to conduct further downstream analyses to examine whether these metabolites may be candidate biomarkers or are causally implicated in schizophrenia.

Disclosures: **S.E. Smart:** Other; Employed on a grant from Takeda Pharmaceuticals Ltd. to Cardiff University. **A.F. Pardiñas:** None. **L. Hubbard:** None. **J.T.R. Walters:** Other; An investigator on a grant from Takeda Pharmaceuticals Ltd. to Cardiff University.

Poster

P904. Schizophrenia: Molecular Mechanisms III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P904.09

Topic: H.13. Schizophrenia

Title: Astrocyte-neuronal metabolic interaction: implications for psychiatric disorders

Authors: *Y. JOUROUKHIN[1], M. V. PLETNIKOV[2];
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Abstract: Metabolic alterations have been described in patients with major psychiatric disorders. Astrocytes maintain brain homeostasis and support neuronal metabolism providing nutritional and energy substrates like glucose and lactate and play a role in neurotransmission by uptake excess of glutamate from extracellular space. We studied the role of *NPAS3* (Neuronal PAS domain protein 3), a transcriptional factor highly expressed in astrocytes, in regulation of metabolic interactions between astrocytes and neurons. RNA-seq analyses of hippocampal samples of mice with astrocyte-specific deletion of Npas3 (Npas3 KO) revealed 40-fold decrease in expression of the *Solute Carrier Family 25, Member 18* (*Slc25a18*) and decreased levels of its protein product, the mitochondrial glutamate carrier 2 (GC2). Reduced expression of GC2 resulted in decreased mitochondrial respiration, deficit of glutamate uptake and decreased production of lactate. Mice with astrocyte-specific deletion of *Npas3* have decreased density of parvalbumin-positive (PV+) inhibitory neurons in prefrontal cortex and reduced glutamate decarboxylase (GAD) immunoreactivity in PV+ presynaptic boutons on pyramidal excitatory neurons. Moreover, we found decreased dendritic spine density in layers 2/3 and 5 of cortical pyramidal neurons. Primary astrocytes transfected with a construct expressing GC2 has restored decreased lactate release back to the control levels. Moreover, deficit of cognitive outcomes measured by trace fear conditioning test was ameliorated by intraperitoneal administration of lactate 30 minutes before the training session. Our findings suggest a previously unknown role for *Npas3* in regulation of astrocyte-neuronal metabolic interaction: lactate release and uptake of extracellular glutamate. Excess of extracellular glutamate combined with decreased release of

lactate affect functioning of cortical inhibitory neurons, leading to cognitive deficiency which maybe relevant to major psychiatric disorders.

Disclosures: Y. Jouroukhin: None. **M.V. Pletnikov:** None.

Poster

P987. Methods to Modulate Neural Activity: Ultrasound

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P987.01

Topic: I.08. Methods to Modulate Neural Activity

Support: Natural Science and Engineering Research Council of Canada (RGPIN-2020-04176)

Title: Modulating cerebellar-motor cortex connectivity using transcranial focused ultrasound

Authors: *J.-F. NANKOO[1], A. FOMENKO[1], J. BAARBÉ[1], S. TRAN[1], X. XIA[1], Y. WANG[1], N. RAIES[1], A. M. LOZANO[2], R. CHEN[1];
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Abstract: Although established non-invasive brain stimulation techniques, such as transcranial magnetic stimulation (TMS), can target the cerebellum, they lack the spatial focality to allow detailed investigations of the cerebello-cortical networks. Transcranial low intensity focused ultrasound (TUS) is a novel stimulation technique that uses acoustic waves to alter neuronal excitability. TUS can reach deeper brain structures with greater focality compared to other non-invasive brain stimulation techniques. This makes TUS particularly well-suited to target functional subregions of the cerebellum. Here, we investigated the effects of cerebellar TUS (cbTUS) on motor cortical (M1) excitability.
Hypotheses: 1) Longer TUS duration will induce greater inhibition of M1 excitability; 2) TUS will transiently lower M1 excitability post sonication; 3) cbTUS specifically targeting cerebellar lobule VIII using neuronavigation will lead to greater inhibition and longer lasting effects compared to TMS-based scalp coordinates.
Methods: In Experiment 1, 500 ms of cbTUS was applied at 3cm lateral and 1cm below the inion. Motor evoked potentials (MEPs) were measured using TMS applied to contralateral M1. MEPs were measured at pre-TUS, after 150ms, 300ms, and 450ms of TUS, and 150ms post-TUS. In Experiment 2, the effects of cbTUS when stimulating the same location as Experiment 1, was compared to lobule VIII stimulation. MEP were measured at pre-TUS, after 150ms, and 450ms of TUS, and at 5ms, 50ms, 150ms, 1000ms and 2000ms post-TUS.
Results: In Experiment 1 cortical excitability was significantly reduced 150 ms after 500 ms of TUS (26% decrease in excitability). In Experiment 2, the time point of peak inhibition was dependent on the cerebellar lobule being stimulated. When targeting lobule VIII, the peak

inhibition was at 450 ms after the start of TUS (38% decrease in excitability).
Conclusion: cbTUS can effectively modulate motor cortical excitability. This is the first investigation of cerebellar TUS in humans, and provides the first step towards modulating specific lobules of the cerebellum non-invasively.

Disclosures: J. Nankoo: None. **A. Fomenko:** None. **J. Baarbé:** None. **S. Tran:** None. **X. Xia:** None. **Y. Wang:** None. **N. Raies:** None. **A.M. Lozano:** None. **R. Chen:** None.

Poster

P987. Methods to Modulate Neural Activity: Ultrasound

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P987.02

Topic: I.08. Methods to Modulate Neural Activity

Support: Bertarelli Foundation
Wyss Center for Bio and Neuroengineering

Title: Morphosonic - a morphologically structured intramembrane cavitation model reveals fiber-specific neuromodulation by ultrasound

Authors: *T. LEMAIRE[1], E. VICARI[1], E. NEUFELD[2], S. MICERA[1];
[1]Ecole Polytechnique Federale De Lausanne, Lausanne, Switzerland; [2]Computat. Life Sci., IT'IS Fndn., Zuerich, Switzerland

Abstract: *Objective.* Low-Intensity Focused Ultrasound Stimulation (LIFUS) is emerging as a promising technology for the remote modulation of neuronal activity, but an incomplete mechanistic characterization hinders its clinical maturation. Intramembrane cavitation has been proposed as a candidate mechanism, but it has only been studied in single or two-compartment computational models of brain neurons. Here, we developed a computational framework to investigate this mechanism in multi-compartmental, morphologically structured neuron models, and used it to study ultrasound neuromodulation of peripheral nerve fibers.
Approach. We recast NEURON's internal cable representation to enable the simulation of multi-compartment models with temporally and spatially varying membrane capacitance. This allowed to seamlessly incorporate the multi-Scale Optimized Neuronal Intramembrane Cavitation (SONIC) paradigm across an arbitrary number of connected compartments, while simultaneously ensuring numerical stability and accuracy. Within this framework, we then implemented single-cable models of myelinated and unmyelinated peripheral axons in order to compute their response to spatially-varying pressure fields.
Results. We predict that by engaging mechanoelectrical bilayer coupling, LIFUS can exploit fiber-specific differences in membrane conductance and capacitance to selectively recruit myelinated and/or unmyelinated axons in distinct parametric subspaces. This fiber-specific

neuromodulation is conserved across a wide range of acoustic field distributions and allows to design specific LIFUS protocols to modulate the spiking activity of different fiber types concurrently and independently over physiologically relevant regimes.
Significance. These theoretical results consistently explain recent empirical findings and suggest that LIFUS can simultaneously yet selectively engage multiple neural pathways, opening up opportunities for peripheral neuromodulation currently not addressable by electrical stimulation, such as the specific targeting of nociceptive and sensory fibers. More generally, our framework can be readily applied to other neural targets in order to investigate electrophysiologically relevant LIFUS neuromodulatory effects and guide the development of application-specific LIFUS protocols.

Disclosures: T. Lemaire: None. **E. Vicari:** None. **E. Neufeld:** None. **S. Micera:** None.

Poster

P987. Methods to Modulate Neural Activity: Ultrasound

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Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P987.03

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Grant R01MH116981
NIBIB Grant R01EB027223
NIBIB Grant R01EB030102

Title: Noninvasive, spatially-precise and cell-type specific deep brain neuromodulation using transcranial focused ultrasound

Authors: *Y. YANG[1], C. PACIA[1], D. YE[1], L. ZHU[1], H. BAEK[1], Y. YUE[1], J. YUAN[1], M. MILLER[2], J. CUI[1], J. CULVER[2], M. R. BRUCHAS[3], H. CHEN[1];
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Abstract: Objective*:* Critical advances in the investigation of brain functions and treatment of brain disorders are hindered by our inability to selectively target neurons in a noninvasive manner in the deep brain. This study aimed to develop sonothermogenetics for noninvasive, deep-penetrating, spatially-precise and cell-type-specific neuromodulation by combining a ultrasound sensitive ion channel TRPV1 with focused ultrasound. **Methods:** The sensitivity of TRPV1 to FUS sonication was evaluated *in vitro*. It was followed by *in vivo* assessment of the success rate of sonothermogenetics in the activation of genetically defined neurons in the mouse brain by two-photon microscopic calcium imaging. Behavioral response evoked by sonothermogenetic stimulation at a deep brain target was recorded in freely moving mice that were sonicated by the customized miniature wearable focused ultrasound. **Results/Discussion:**

TRPV1 was found to be a sonothermogenetic actuator. FUS sonication at the mouse brain *in vivo* selectively activated neurons that were genetically modified to express TRPV1 (Fig. 1A). Temporally precise activation of TRPV1-expressing neurons was achieved with its success rate linearly correlated with FUS-controlled mild temperature rise as measured by *in vivo* magnetic resonance thermometry. FUS stimulation of TRPV1-expressing neurons of the Parkinsonian circuit at the striatum repeatedly evoked rotating locomotor behavior in freely moving mice (Fig. 1B). FUS sonication was confirmed to be safe based on inspection of neuronal integrity, inflammation, and apoptosis markers. Sonothermogenetics is a noninvasive and cell-type-specific neuromodulation approach with the capability to target the deep brain. **Scientific Rigor:** The scientific rigor of this research was controlled by the highest-standard experiment design, data analysis, result reporting and reproducibility including but not limited to: 1) Including positive and negative controls; 2) Quantifying the success rate and reporting raw data of the key results; 3) Repeating the key results by the third party.

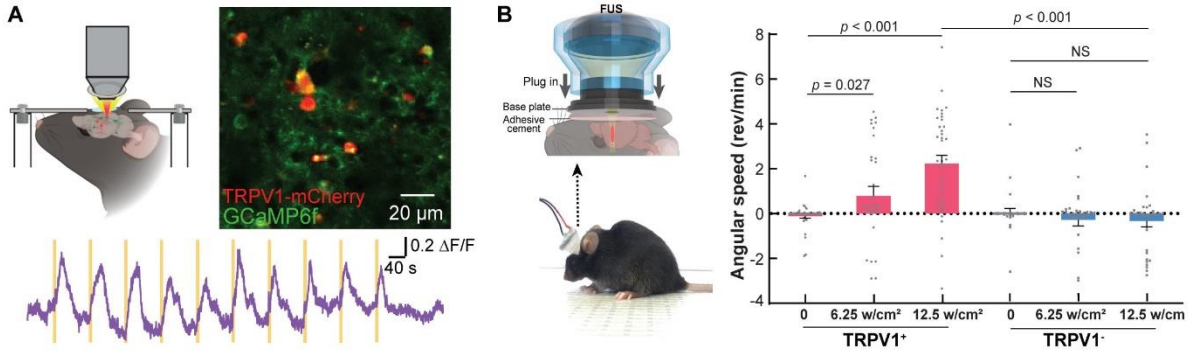


Figure 1. (A) Sonothermogenetics in the mouse brain in vivo. The mouse head was fixed in the stage for simultaneous FUS stimulation and two-photon Ca^{2+} imaging. The coexpression of TRPV1 and GCaMP was observed in vivo. FUS can repeatedly activate neurons expressing TRPV1. The orange bar indicates the FUS on time. (B) Experimental setup for *in vivo* sonothermogenetic stimulation of the striatum in freely moving mice (left panel). The miniaturized wearable FUS transducer was plugged into a base plate adhered on the mouse head. The comparison of the mean angular rotation speed of $TRPV1^+$ mice versus $TRPV1^-$ mice with FUS stimulation at I_{spta} = 0, 6.25, and 12.5 w/cm^2. The positive and negative values indicate rotation in the contralateral and ipsilateral directions, respectively. Error bars represent the standard error of the mean.

Disclosures: Y. Yang: None. **C. Pacia:** None. **D. Ye:** None. **L. Zhu:** None. **H. Baek:** None. **Y. Yue:** None. **J. Yuan:** None. **M. Miller:** None. **J. Cui:** None. **J. Culver:** None. **M.R. Bruchas:** None. **H. Chen:** None.

Poster

P987. Methods to Modulate Neural Activity: Ultrasound

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P987.04

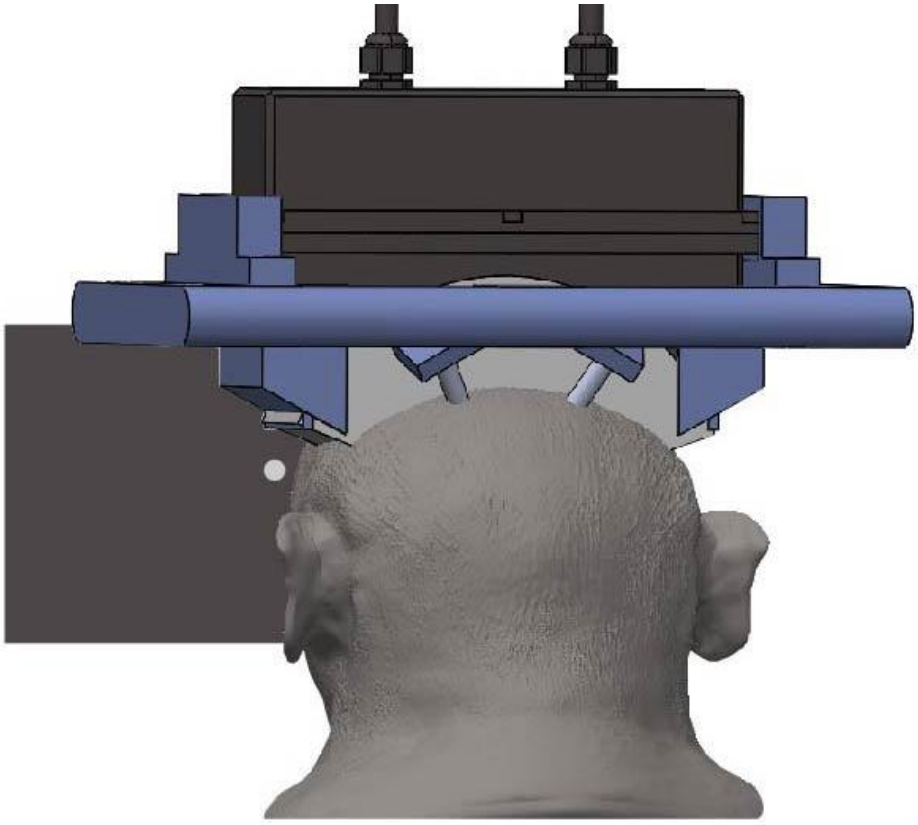
Topic: I.08. Methods to Modulate Neural Activity

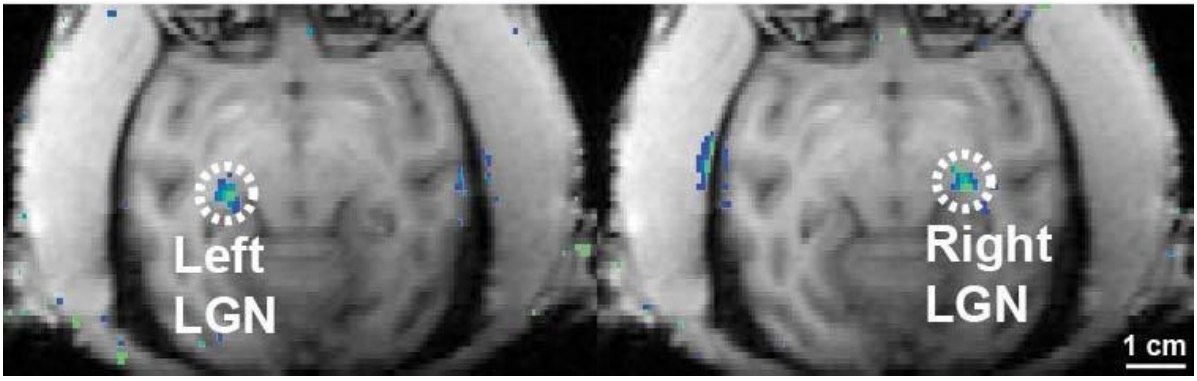
Support: 5F32MH123019
5R00NS100986

Title: Noninvasive, Multifocal Deep Brain Interventions

Authors: *T. WEBB, M. G. WILSON, H. ODÉEN, J. KUBANEK;
Univ. of Utah, Salt Lake City, UT

Abstract: Transcranial focused ultrasound has opened new avenues for noninvasive and spatially specific intervention in deep brain circuits. Low energy interventions include neuromodulation, targeted drug release, and transient disruption of the blood brain barrier. We developed and validated a non-human primate model to test the efficacy and safety of these new applications. The model uses a 256-element transducer to deliver ultrasound into specific deep brain circuit/s on demand, using fast electronic focusing. The electronic focusing allows the device to be fixed with respect to the head and so deliver ultrasound into the desired regions reproducibly across sessions. The device can target multiple deep brain regions simultaneously or in concert. The animal is awake and head-fixed during the ultrasound delivery, thus empowering researchers to quantify the effects of the ultrasonic interventions on behavior in real time. We validated the method by stimulating a deep brain region, the lateral geniculate nucleus (LGN), during a visual discrimination task. Data across multiple sessions show an immediate and reversible ipsilateral bias, thus validating successful and accurate delivery of ultrasound into the deep brain. The task also provides a sensitive measure of the safety of daily ultrasonic neuromodulation since any damage to the LGN would result in a deterioration of the subject's discrimination accuracy. Our data show the opposite effect, with the subject's performance improving across sessions. Thus, our data show that ultrasound delivered into specific deep brain regions can flexibly modulate behavior. We are currently testing the effects of the ultrasound pressure amplitude, frequency, and duty cycle. This investigation will provide the field with parameters that effectively excite or inhibit neurons. This model will provide data on the efficacy and safety of new transcranial applications of ultrasound and thus accelerate their translation into clinics. The developed hardware and software can be applied also for noninvasive and multifocal stimulation of the human brain.





Disclosures: T. Webb: None. **M.G. Wilson:** None. **H. Odéen:** None. **J. Kubanek:** None.

Poster

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Program #/Poster #: P987.05

Topic: I.08. Methods to Modulate Neural Activity

Support: R00 Grant # 59316231

Title: Effective Ultrasound Stimulation in Human Peripheral Nervous System

Authors: *T. RIIS, T. WEBB, J. KUBANEK;
Biomed. Engin., Univ. of Utah, Salt Lake City, UT

Abstract: Low-intensity ultrasound can stimulate excitable cells in a noninvasive and targeted manner, but which ultrasound parameters are effective has remained elusive. Ultrasound frequency is a key parameter: it governs the choice of the transducer, the size of the focal region, and depth of penetration. Yet, whether lower or higher frequencies stimulate the nervous system effectively is unknown. This question could not be addressed in previous studies because changes in frequency lead to large changes in the stimulated volumes, and therefore to large changes in the probability of registering a response. The objective of this study is to control for differences in stimulated tissue volume and evaluate which ultrasound parameters are effective in stimulating excitable cells. We used a single ultrasonic transducer and matched stimulation volumes for different frequencies using a metallic aperture. We applied the ultrasonic stimulation to the peripheral nervous system, receptors and nerve endings of the index finger in particular, in 26 human subjects. We tested the response to ultrasound stimuli while varying the parameters of frequency, pressure level, duration, stimulus kind (continuous stimulation and pulsed stimulation), and duty cycle. Subjects were instructed to report a percept verbally, describing it as either pain, warm, cold, vibration, or tap. We found that low frequencies (300 kHz) are substantially more effective in generating tactile and nociceptive responses in humans compared to high frequencies (900 kHz). The strong effect of ultrasound frequency was observed for all pressures tested, for continuous and pulsed stimuli, and for tactile and nociceptive responses. The prominent effect of frequency may be explained by a mechanical force associated with ultrasound. The effect is not due to heating, which would be weaker at the low frequency. Notably**,** we observed a quadratic dependence of response frequency on the pressure level. This suggests an involvement of a mechanical effect based on ultrasound intensity, most likely candidate of which is the acoustic radiation force. This controlled study reveals that ultrasonic stimulation of excitable cells is stronger at lower frequencies, which guides the choice of transducer hardware for effective ultrasonic stimulation of the peripheral nervous system in humans. The results also inform on the mechanism involved in the ultrasound bioeffects.

Disclosures: T. Riis: None. **T. Webb:** None. **J. Kubanek:** None.

Poster

P987. Methods to Modulate Neural Activity: Ultrasound

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Program #/Poster #: P987.06

Topic: I.08. Methods to Modulate Neural Activity

Support: FDN 154292

ENG 173742
RGPIN-2020-04176
MFE-171317
DFG DA 2374/1-1

Title: Induction of human motor cortex plasticity by theta burst transcranial ultrasound stimulation

Authors: *K. ZENG[1], G. DARMANI[1], A. FOMENKO[1], X. XIA[1,2], S. TRAN[1,3], J.-F. NANKOO[1], Y. SHAMLI OGHLI[1,3], Y. WANG[1,2], A. LOZANO[1,4], R. CHEN[1,5];
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Abstract: Background and Hypothesis: Transcranial focused ultrasound stimulation (TUS) is a novel, non-invasive neuromodulation method that is beginning to be applied in human subjects. We hypothesize that long-term potentiation (LTP)-like plasticity can be induced in the human motor cortex by repetitive low-intensity transcranial focused ultrasound stimulation (TUS). **Materials and Methods**: Three repetitive TUS, theta burst patterned TUS (tbTUS), regularly patterned TUS (rTUS) and sham TUS, were delivered to motor cortex in 15 healthy subjects in separate sessions in random order. The tbTUS and rTUS protocols had same total sonication duration and total time of stimulation. Motor evoked potential (MEP) evoked by single-pulse and paired-pulse TMS was recorded before and at 5 min, 30 min and 60 min after TUS. Short interval intracortical inhibition (SICI) and intracortical facilitation (ICF) were measured for paired-pulse TMS. The effects of motor cortex tbTUS on movement performance in a visuo-motor task and the effects of ipsilateral occipital cortex tbTUS on motor cortex excitability were also assessed. **Results**: MEP amplitudes significantly increased at 5 min (43.4%) and 30 min (27.5%) and returned to baseline level at 60 min after tbTUS. SICI was significantly attenuated at 5 min and 30 min, and ICF was significantly enhanced at 5 min after tbTUS. The movement time on a visuo-motor task was significantly shortened after tbTUS. In contrast, the rTUS, sham TUS and occipital tbTUS had no effect on the MEP amplitudes elicited by both single-pulse and paired-pulse TMS. In addition, the MEP amplitudes elicited by lateral-medial directed TMS were unchanged after tbTUS. **Conclusions**: The tbTUS protocol is capable of inducing LTP-like plasticity in human for at least 30 min. The plasticity occurred at the cortical level and involved both inhibitory and excitatory cortical circuits. This LTP-like plasticity was specific to theta burst patterned TUS and could not be explained by sensory confounds associated with TUS. **Significance:** tbTUS is a novel paradigm to induce cortical plasticity in human and has the potential to be developed for neuromodulation treatment for neurological and psychiatric disorders, and to advance neuroscience research.

Disclosures: K. Zeng: None. **G. Darmani:** None. **A. Fomenko:** None. **X. Xia:** None. **S. Tran:** None. **J. Nankoo:** None. **Y. Shamli Oghli:** None. **Y. Wang:** None. **A. Lozano:** None. **R. Chen:** None.

Poster

P987. Methods to Modulate Neural Activity: Ultrasound

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Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P987.07

Topic: I.08. Methods to Modulate Neural Activity

Support: NSF NRT-UtB DGE-1734815
Institute for Engineering in Medicine

Title: Hearing damage associated with transcranial ultrasound for neuromodulation and blood-brain-barrier opening

Authors: *J. BASILE, G. RODRIGUEZ ORELLANA, H. H. LIM;
Univ. of Minnesota, Minneapolis, MN

Abstract: Ultrasound (US) research has grown rapidly in the past decade for noninvasively modulating brain regions with high spatial resolution and performing targeted drug delivery by transiently opening the blood-brain-barrier (BBB). Studies have also quickly moved into humans citing diagnostic US imaging tissue safety ranges in an FDA guidance document (Docket: FDA-2017-D-5372) with some parameters having additional histological safety data in animal models. Our lab discovered that US applied to the head readily activates the auditory system through vibrations of cerebrospinal fluid that then directly vibrate fluids within the cochlea (Guo et al., Neuron, 2018). We have also demonstrated that neural responses to US stimuli closely resembled those to air-conducted stimuli and that frequency specific activation of auditory regions can be achieved using amplitude-modulated US. Due to the potential applications of US induced auditory activation, our group was interested in characterizing safe levels of US pressure for the hearing system.

In anesthetized guinea pigs, we collected auditory brainstem responses (ABRs) and electrocochleography (ECochG) induced by air-conducted acoustic pure tones (2, 4, 8, 12, 20, and 30 kHz) and broadband noise at varying levels (10-70 dB SPL) before and after US stimulation with specific parameters that have been used in previous studies for either neuromodulation or BBB opening experiments. Control data was also collected for characterizing the stability of the recording protocol and for standard noise-induced hearing loss (NIHL). We assessed ABR and ECochG thresholds, amplitudes, and latencies over time to identify changes that are associated with hearing damage. Many tested US parameters showed neurophysiological changes associated with hearing loss. Threshold shifts were most prevalent in the high frequencies with some more severe cases of US stimuli causing threshold shifts in the middle frequencies as well with a similar pattern to NIHL. Other US parameters that appear to contribute to hearing damage in addition to pressure level are duration, duty cycle, and center frequency. Caution is urged when performing transcranial US stimulation studies in humans,

especially when targeting brain locations closer to the regions of the cochlea or if perception of sound is noted by individuals during stimulation. Future studies will include an in-depth characterization of the US parameters of interest also in large animal models that better mimic the head size in humans.

Disclosures: J. Basile: None. **G. Rodriguez Orellana:** None. **H.H. Lim:** None.

Poster

P987. Methods to Modulate Neural Activity: Ultrasound

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Program #/Poster #: P987.08

Topic: I.08. Methods to Modulate Neural Activity

Support: DARPA BTO HR0011-18-C-0040

Title: UNITE Study (UMN-GE) for COVID-19: a pilot study of the efficacy of ultrasound application to the spleen in the treatment of hospitalized patients

Authors: *M. NEWHOFF[1], *M. N. COLLINS[2], R. S. GRAHAM[2], S. DOORNINK[2], W. SWANSON[2], A. TUMA[2], D. P. ZACHS[3], C. PULEO[6], J. ASHE[6], K. WALLACE[6], R. MADHAVAN[6], A. P. J. OLSON[4], E. J. PETERSON[5], H. H. LIM[3];
[2]Otolaryngology, [3]Biomed. Engin., [4]Pediatrics, [5]Div. of Rheumatic and Autoimmune Dis., [1]Univ. of Minnesota, Minneapolis, MN; [6]Gen. Electric Res., Gen. Electric, Niskayuna, NY

Abstract: Ultrasound stimulation of the spleen is an emerging noninvasive modality for reducing systemic inflammation. It has been shown to attenuate disease symptoms and inflammatory markers of rheumatoid arthritis in humans and mice, as well as in animal models of sepsis. Treatment applications as short as 3 minutes can reduce the production of pro-inflammatory cytokines in LPS-stimulated human blood, while stimulation of only 2 minutes per day across 7 days drives significant recovery from inflammatory arthritis in mice. Splenic ultrasound is believed to exert its anti-inflammatory effects via activation of noradrenergic fibers innervating the spleen that receive input from vagus efferents, thereby activating the cholinergic anti-inflammatory pathway in a manner comparable to vagus nerve stimulation.
Severe complications of SARS-CoV-2, the virus that causes COVID-19, are believed to be caused by elevated levels of cytokines and other inflammatory factors, the so-called “cytokine storm,” that leads to multiorgan dysfunction. We therefore sought to determine whether splenic ultrasound, applied for 9 minutes daily for 7 days, could attenuate levels of inflammatory markers or improve the clinical outcomes of patients hospitalized with COVID-19.
In a pilot study of 30 patients, we found that daily splenic ultrasound yielded improvement in patient outcomes, including a reduction in the number of days hospitalized, a reduction in the duration and volume of oxygen supplementation, and a reduction in mortality rate (zero deaths in

the ultrasound treated group) in patients receiving splenic ultrasound in addition to standard-of-care treatment in comparison to a control cohort of patients receiving standard-of-care treatment alone. A clinically meaningful improvement in symptoms was achieved in about half the time (4 vs 8 days) for ultrasound treated patients compared to the control cohort.
We propose that splenic ultrasound has the potential to reduce inflammation in health conditions associated with acute or chronic inflammatory states, such as in COVID-19, and potentially in a manner comparable to that achieved via implantable vagus nerve stimulators, but without the risks associated with surgery or off-target vagus nerve activation effects.

Disclosures: M.N. Collins: None. **R.S. Graham:** None. **S. Doornink:** None. **W. Swanson:** None. **A. Tuma:** None. **D.P. Zachs:** None. **C. Puleo:** None. **J. Ashe:** None. **K. Wallace:** None. **R. Madhavan:** None. **A.P.J. Olson:** None. **E.J. Peterson:** None. **H.H. Lim:** None.

Poster

P987. Methods to Modulate Neural Activity: Ultrasound

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Program #/Poster #: P987.09

Topic: I.08. Methods to Modulate Neural Activity

Support: DARPA Grant N66001-19-C-4019

Title: Magnetic nanoparticle neuronal stimulation: Avoiding misclassification of optical and vibration induced artifacts

Authors: *E. H. ZHANG[1], P. LIANG[3], M. ABDEL-MOTTALEB[2], B. NAVARRETE[1], Y. AKIN-YILDIRIM[1], M. ALBERTERIS-CAMPOS[1], I. T. SMITH[2], S. CHEN[3], S. KHIZROEV[1]; [1]Electrical and Computer Engin., [2]Biomed. Engin., Univ. of Miami, Coral Gables, FL; [3]Cell. Nanomed, Irvine, CA

Abstract: In recent years, multiple nanoparticle approaches to neuroscience have opened new avenues in brain imaging, interfaces, and treatment. Of particular interest are nanoparticle mediated methods for wireless deep brain stimulation (DBS), which can produce strong localized responses. One of these approaches involves the direct electrical stimulation of neurons with magnetoelectric nanoparticles (MENPs). Magnetoelectric (ME) stimulation of neurons works by using nanoparticles to convert magnetic pulses into localized electric fields, which then generate action potentials in nearby neurons. As the magnetic pulses produce significant artifacts in electrode-based measurements of action potentials, data for neuronal signals typically comes from calcium imaging or related methods. However, imaging approaches are susceptible to a particular artifact related to the mechanical vibrations induced in the magnetic component of the nanoparticles and their optical characteristics. This artifact can produce an apparent neuronal activation through local intensity changes that imitates real neuronal firing. Here we report

differentiating factors between the false activations and true action potentials that we used to successfully isolate the two types. A few critical waveform characteristics are apparent in the false action potentials, which are useful for identifying artificial activity. Differentiating between the two signal types is critical to avoid unintentional misclassification of vibrational effects as neuronal firing.

Disclosures: E.H. Zhang: A. Employment/Salary (full or part-time):; University of Miami. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Cellular Nanomed. **P. Liang:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Cellular Nanomed. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Cellular Nanomed. **M. Abdel-Mottaleb:** A. Employment/Salary (full or part-time):; University of Miami. **B. Navarrete:** A. Employment/Salary (full or part-time):; Cellular Nanomed. **Y. Akin-Yildirim:** A. Employment/Salary (full or part-time):; University of Miami. **M. Alberteris-Campos:** A. Employment/Salary (full or part-time):; University of Miami. **I.T. Smith:** A. Employment/Salary (full or part-time):; University of Miami. **S. Chen:** A. Employment/Salary (full or part-time):; University of Miami. **S. Khizroev:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Cellular Nanomed.

Poster

P987. Methods to Modulate Neural Activity: Ultrasound

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:30 AM - 11:30 AM

Program #/Poster #: P987.10

Topic: I.08. Methods to Modulate Neural Activity

Support: DARPA Grant N66001-19-C-4019

Title: Effect of coercivity on the efficacy of magnetoelectric nanoparticles to modulate neurons

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Abstract: Wireless brain stimulation could facilitate the study of brain circuits more quickly and easily and can treat neurological disorders. Magnetoelectric nanoparticles (MENPs) provide a minutely invasive brain stimulation approach that does not require surgery, by transforming the energy transmitted through remotely applied magnetic fields into local electric fields. Coercivity

(Hc) is a significant property of the MENPs which pertains to its resistance to change in magnetization in the presence of a magnetic field. The coercivity field is an extrinsic parameter which is a fraction of the anisotropy field, an intrinsic parameter of the material of which the magnetostrictive core of the core-shell MENPs is made. In this study we demonstrate the effect of a magnetoelectric nanoparticle's coercivity on its ability to excite nearby neurons. We have varied the coercivity field from below 100 to over 900 Oe during the MENPs synthesis process. We studied the relationship between applied magnetic fields and the coercivity of the MENPs by applying A.C. magnetic fields ranging from below 300 Oe to above 1000 Oe. We found that the magnetically applied field had to exceed the coercivity of the MENPs to maximize the efficiency of neuronal firing. To assess the effect of MENPs with varying coercivities on neurons, we imaged calcium signals in rat hippocampal cell cultures. The results indicate that MENPs that exhibit high coercivity (Hc > 800 Oe) dramatically increase the effect of the MENPs on neuronal modulation, with consistent neuronal firing upon activation of the MENPs with an external A.C. magnetic field. This enhancement to MENPs shows promising potential that could accelerate them to be a new approach for treating neurological diseases.

Disclosures: M. Abdel-Mottaleb: None. **E. Zhang:** None. **B. Navarrete:** None. **Y. Yildirim:** None. **M. Alberteris-Campos:** None. **I.T. Smith:** None. **S. Chen:** None. **P. Liang:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Cellular Nanomed Inc.. **B. Noga:** None. **S. Khizroev:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Cellular Nanomed Inc.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Cellular Nanomed Inc..

Poster

P013. Stem Cell Approaches for Disorders of the Retina and Glaucoma

Location: Virtual Only

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Program #/Poster #: P013.01

Topic: A.03. Stem Cells and Reprogramming

Support: NIH R01 EY024984
NIH R21 EY028687
BrightFocus G2020369
CTSI/SNRI - Sarah Roush Memorial Fellowship in Alzheimer's Disease Research
Glaucoma Research Foundation

Title: Astrocytes contribute to retinal ganglion cell neurodegeneration in a glaucoma patient-specific pluripotent stem cell model

Authors: *C. GOMES[1], K. B. VANDERWALL[2], Y. PAN[1], X. LU[1], S. S. LAVEKAR[2], K.-C. HUANG[2], C. M. FLIGOR[2], C. ZHANG[1], T. R. CUMMINS[2], J. S. MEYER[1];
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Abstract: Retinal ganglion cell (RGC) degeneration is a primary characteristic of glaucoma, although non-cell autonomous mechanisms have also been implicated in RGC dysfunction. Astrocytes associate with RGCs in the nerve fiber layer of the retina and optic nerve, where they can contribute to RGC neurodegeneration. However, the mechanisms by which astrocytes promote neurotoxicity and contribute to glaucoma remain unknown. Human pluripotent stem cells (hPSCs) can serve as powerful tools for the *in vitro* study of neurodegenerative diseases, including neuron-glia interactions. Using hPSC-derived cells from a glaucoma patient with an Optineurin (OPTN) E50K mutation as well as isogenic control cells obtained by CRISPR/Cas9 gene editing, we explored how the OPTN(E50K) mutation affects RGC-astrocyte interactions leading to neurodegeneration. hPSC-derived OPTN(E50K) astrocytes show increased propensity to develop reactive phenotypes and exhibited significant changes in protein expression as well as autophagy dysfunction. Transcriptional analysis by RNA-seq showed upregulation of the inflammatory pathway and downregulated expression of cell cycle genes in OPTN(E50K) astrocytes, suggesting a senescent phenotype exhibited by glaucomatous astrocytes. Subsequently, the contribution of astrocytes to RGC neurodegeneration was analyzed in co-culture experiments. Importantly, OPTN(E50K) astrocytes induced neurodegenerative phenotypes in RGCs, including decreased neurite length and complexity, together with increased excitability. Conversely, healthy isogenic control astrocytes were capable of rescuing degenerative phenotypes observed in OPTN(E50K) RGCs. Additionally, as glaucoma astrocytes secreted reduced levels of growth factors, suggesting a deficit in their neurosupportive role, the supplementation of OPTN(E50K) RGCs with these factors at least partially rescued many of these phenotypes, highlighting the role of astrocyte-derived factors in RGC support. Taken together, these results are the first of its kind to identify a neurotoxic phenotype in hPSC-derived astrocytes from a glaucoma source, as well as to identify a role for astrocytes in the neurodegenerative process associated with glaucoma through non-cell autonomous mechanisms, highlighting these glial cells as novel potential therapeutic targets.

Disclosures: C. Gomes: None. **K.B. VanderWall:** None. **Y. Pan:** None. **X. Lu:** None. **S.S. Lavekar:** None. **K. Huang:** None. **C.M. Fligor:** None. **C. Zhang:** None. **T.R. Cummins:** None. **J.S. Meyer:** None.

Poster

P013. Stem Cell Approaches for Disorders of the Retina and Glaucoma

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Topic: A.03. Stem Cells and Reprogramming

Support: NIH/NEI R01EY024984
NIH/NEI R21EY031120
BrightFocus Foundation G2020369
Glaucoma Research Foundation
NIH UL1TR002529
Sigma Xi Grants in Aid of Research

Title: Autophagy disruption induces mTORC1 hypoactivation and leads to neurodegeneration in glaucoma OPTN(E50K) retinal ganglion cells

Authors: *K.-C. HUANG[1,2], C. GOMES[3,2], K. B. VANDERWALL[1,2], S. S. LAVEKAR[1,2], C. M. FLIGOR[1,2], J. HARKIN[4,2], J. S. MEYER[3,2,4,5];
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Abstract: Optineurin (OPTN) mutations contribute to multiple neurodegenerative diseases including glaucoma, with the more severe forms resulting in normal tension glaucoma due to an E50K mutation in the OPTN gene. Previously, we have derived retinal ganglion cells (RGCs) from human pluripotent stem cells (hPSCs) with an OPTN(E50K) mutation, which exhibited decreased neurite length and complexity when compared with controls as well as a hyperexcitable state. However, the mechanisms connecting the OPTN(E50K) mutation to resulting neurodegenerative phenotypes have not been fully characterized and furthermore, pharmacological intervention to preserve damaged RGCs has been limited. In this study, we derived RGCs with the OPTN(E50K) mutation as well as corresponding isogenic controls from hPSCs to explore mechanisms that induced RGC neurodegeneration. OPTN(E50K) RGCs exhibited a higher number of OPTN puncta along with an overall reduced expression of OPTN protein. Since the OPTN protein plays a primary role as an autophagy receptor, OPTN(E50K) RGCs revealed an accumulation of the autophagosome protein LC3 in a punctal manner that was less likely to be co-localized with OPTN, suggesting that OPTN(E50K) results in a partial loss of function and is unable to recruit LC3. More so, the increased expression of lysosomal proteins was observed in OPTN(E50K) RGCs, indicating that autophagosome accumulation is due to a disruption of autophagosome and lysosome fusion. As this accumulation can activate the stress energy sensor AMP-activated protein kinase (AMPK) and inhibit mTORC1, OPTN(E50K) RGCs demonstrated increased phosphorylated AMPK expression and decreased the mTORC1 effector phosphor-S6 expression, suggesting that reduced mTOR activity may be partially responsible for some neurodegenerative phenotypes. In addition, pharmacological inhibition of mTOR in wild-type RGCs resulted in similar disease-related phenotypes, while activation of the mTOR pathway in OPTN(E50K) RGCs partially rescued neurodegenerative phenotypes. Moreover, these disease-related phenotypes were observed specifically within RGCs but not cortical neurons with an OPTN(E50K) mutation, suggesting that autophagy dysfunction due to

the OPTN(E50K) mutation may act specifically within RGCs to lead to neurodegeneration. Thus, the results of this study demonstrate that autophagy dysfunction can result in profound neurodegenerative phenotypes, and further studies of these pathways may lead to the identification of new targets for therapeutic approaches to glaucomatous neurodegeneration.

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Poster

P013. Stem Cell Approaches for Disorders of the Retina and Glaucoma

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P013.03

Topic: A.03. Stem Cells and Reprogramming

Support: NEI-K08 EY031801
Research to Prevent Blindness: Career Development Award (TVJ) and Unrestricted Grant to the Wilmer Eye Institute
American Glaucoma Society

Title: Understanding and overcoming obstacles to functional integration faced by transplanted human stem cell derived retinal ganglion cells

Authors: *E. A. AGUZZI, K. Y. ZHANG, A. NAGALINGAM, X. CHANG, D. J. ZACK, T. V. JOHNSON, III;
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Abstract: Optic neuropathies, including glaucoma, are characterized by progressive loss of retinalganglion cells (RGCs) and ultimately cause permanent vision loss. Novel neuroprotective approaches to preserving endogenous RGCs following disease onset are emerging. However, because the mammalian retina lacks regenerative capacity, only interventions aimed at RGC replacement and optic nerve regeneration hold potential for reversing blindness. Here, we aim to characterize the mechanisms and barriers to donor human RGC integration following intravitreal transplantation *in vivo*. We demonstrate that under homeostatic physiologicalconditions, the internal limiting membrane (ILM) impedes the migration of transplanted RGCsinto the host retina. To circumvent the ILM, we intravitreally injected the nonspecific protease Pronase-E in immunodepleted mice of both sexes. This enzyme digests the ILM, therebydisrupting the barrier faced by donor RGCs, without affecting the health and survival ofendogenous retinal neurons or glia. Alternatively, intravitreal human RGC transplantation wasperformed in Lama1^{nmf223} mice of both sexes, which harbor a point mutation in the *Lama-α1*gene, and which exhibit developmental ILM defects. Samples sizes included ≥8 mice pergroup and experiments were replicated at least twice, always with observers masked totreatment group. Both proteolytic and

transgenic ILM disruption was associated with an increase in donor RGC survival by ~1.5-fold, though overall survival remained <1%. Somal co-planarity into the host RGC layer also increased by ≥10-fold ($p<0.001$) after both experiments, while dendrite extension into the host inner plexiform layer significantly increased by ≥25-fold after proteolytic treatment ($p<0.0008$) and by ≥100-fold (<0.0001) in the transgenic experiments. Ongoing work aims to characterize the functional connectivity ofdonor RGCs within the host retinal neurocircuitry. Our observations provide advancement inthe understating of human RGC engraftment and synaptogenesis in the adult mammalianretina, and may point towards methods for clinically translating regenerative medicineapproaches to optic neuropathy.

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Poster

P013. Stem Cell Approaches for Disorders of the Retina and Glaucoma

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P013.04

Topic: A.03. Stem Cells and Reprogramming

Support: CIRM Disc 1 (BBT)
CIRM DT1 (MSH)
CIRM TR4-06648 (MJS)
NIH R01EY024045 (HSK)
Bright Focus Foundation (BBT)

Title: Transplants of hESC derived retina organoids alone and with RPE improved vision function in two retinal degenerative rat models

Authors: *B. LIN[1], J. C. M. CAMARILLO[4], D. ZHU[4], D. R. HINTON[4], M. HUMAYUN[4], B. MCLELLAND[2], G. NISTOR[5], R. B. ARAMANT[2], H. KEIRSTEAD[5], B. B. THOMAS[4], M. J. SEILER[3];
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Abstract: Age-related Macular Degeneration (AMD) and Retinitis Pigmentosa (RP) are two major retinal degenerative (RD) conditions that result in irreversible vision loss. Permanent eye damages can also occur in battlefields or due to accidents. To develop a cellular therapy for the above retinal diseases, we tested: 1. Transplant sheets of human embryonic stem cell (hESC) derived retina organoids (RO) into immunodeficient rho S334ter-3 (RD) rats; 2. Transplant sheets of hESC derived RO into immunodeficient Royal College of Surgeon (RCS) rats; 3.

transplant co-grafts of hESC derived RPE and sheets of RO into immunodeficient RCS rats. Vision function and transplant development was tested with different techniques. The H9 hESCs and CSC-14 hESCs (NIH 0284) were differentiated into retina organoids and characterized by immunohistochemistry (IHC), flow cytometry and qPCR. H9 hESC were also cultured and spontaneously differentiated to RPE cells. The hESC-RPE were seeded and cultured around 30 days on ultrathin parylene substrates to produce polarized monolayer RPE implants. For the preparation of hESC-RPE+RO co-grafts, RO sheets were dissected out and combined with RPE implants with the help of suitable bio-adhesive agents, alginate. Growth and development of transplants was evaluated by Optical Coherence Tomography (OCT). Visual function was tested by optokinetic testing (OKT), and electrophysiological recording in the superior colliculus (SC recording). Histological assessments were performed using H&E staining and immunohistochemistry. OCT imaging demonstrated successful surgical transplantation and longtime survival of the ROs and co-grafts. Histological evaluation confirmed the survival and development of the ROs and co-grafts. OKT and SC recording showed significant vision improvement by transplantation of ROs and co-grafts, compared to age-match control and sham. The success rate of rats with response to light in SC recording is higher in co-graft than in RO alone. The study supports the possibility of combining RPE+RO into a co-graft, which can mature and improve vision function after transplanted into the subretinal space. This novel technique can become a new cellular therapy for complete replacement of a degenerated retina and RPE.

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Poster

P013. Stem Cell Approaches for Disorders of the Retina and Glaucoma

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P013.05

Topic: A.03. Stem Cells and Reprogramming

Support: NIH R01EY024984
NIH R21EY031120
BrightFocus G2020369
Glaucoma Research Foundation

Title: Modeling blood-brain barrier phenotypes in glaucoma with human pluripotent stem cells

Authors: *S. LAVEKAR[1], J. HUGHES[2], C. GOMES[3], K.-C. HUANG[1], S. G. CANFIELD[2], J. S. MEYER[4];
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Abstract: Glaucoma is a neurodegenerative disease that affects the retina and optic nerve, resulting in the degeneration of retinal ganglion cells (RGCs) and subsequent loss of vision. While RGCs are the primary cell type affected in the disease state, neighboring cell types are known to influence the disease state as well. In particular, cell types that contribute to the blood brain barrier (BBB), including astrocytes and microvascular endothelial cells, can selectively modulate RGCs to maintain their overall homeostasis and form a tight barrier between the blood and the brain. However, previous studies have demonstrated that these barrier properties may be compromised in the progression of glaucoma, yet the mechanisms by which the barrier is compromised have remained elusive. Thus, the overall goals of this study were to adapt human pluripotent stem cell (hPSC)-based models of the blood-brain barrier to the study of barrier integrity in glaucoma. To achieve this, hPSCs were directed to differentiate into cells of the BBB, including RGCs, astrocytes, and endothelial cells, and then assembled into a model of the BBB using an established transwell assay. The ability of these cell types to contribute to an in vitro barrier model was tested for their ability to recapitulate characteristic properties including transendothelial electrical resistance (TEER), permeability and rhodamine transport. Results revealed the enhancement of barrier properties upon the establishment of the barrier model with RGCs, astrocytes, and endothelial cells compared to endothelial cells grown alone. Conversely, the versatility of this system to model aspects of barrier dysfunction in glaucoma was tested using an hPSC line with a glaucoma-specific Optineurin (E50K) mutation. Upon developing the barrier model with glaucoma-specific cell types, results demonstrated highly compromised barrier integrity, including reduced TEER and increased barrier permeability, indicating BBB dysfunction. To begin to identify factors that could result in barrier dysfunction, results also revealed an increased expression of TGFB2 in OPTN(E50K) astrocytes, indicating a potential role for TGFB2 in disease manifestation. To test this, exogenous TGFB2 was applied to cultures of healthy cells assembled into the barrier model, which resulted in barrier dysfunction similar to those obtained with glaucoma OPTN(E50K) cells. Thus, the repurposing of this barrier model for glaucoma has immense potential for mimicking aspects of diseases affecting RGCs and the BBB, particularly high throughput analyses and drug screening to rescue neurodegenerative phenotypes.

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Poster

P045. Animal, Synaptic, and Cellular Mechanisms

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Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P045.01

Topic: A.07. Developmental Disorders

Support: NIH-NIGMS R35 GM119831
NIH-NIMH MH120513

Title: Pairing transcriptional profiling and stereology to survey the role of Chd8 in the postnatal mouse brain

Authors: *S. A. LOZANO[1], K. CICHEWICZ[1], S. FRANK[2], A. WADE[1], R. ORTIZ[1], J. ZHU[1], J. BENNETT[2], W. AMARAL[2], P. BEAUREGARD[1], P. LAVENEX[3], D. G. AMARAL[2], A. S. NORD[1], C. P. CANALES[1];
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Abstract: *Chd8,* which encodes a chromatin remodeling factor, has emerged as a leading risk gene for neurodevelopmental disorders (NDDs), including Autism Spectrum Disorder (ASD). Mice with heterozygous germline loss-of-function mutations to *Chd8* exhibit genomic, neuroanatomical, and behavioral pathology that aligns with clinical features of ASD. Thus, mapping the transcriptomic landscape of *Chd8* mutants offers a route towards resolving the molecular consequences of *Chd8* haploinsufficiency and better understanding ASD etiology. We used RNA-sequencing in cerebral cortex, hippocampus, and cerebellum from *Chd8* adult heterozygous mutants and wild-type littermates to examine putative differences in gene expression. From these data, we identified differentially-expressed genes (DEGs) that were altered across all three brain regions, as well as region-specific signatures. Through weighted correlation network analysis, we characterized modules that are dysregulated in adult brain and ultimately detected perturbation to neuroinflammatory and metabolic pathways. Parallel unbiased stereological assessment of adult mutant cortices indicated sex-specific differences in cortical volume without significant differences in cell numbers. Interestingly, a module of sex-specific DEGs was identified in bulk RNA-seq data that could corroborate a genotype-by-sex interaction linked to pathology in adult mouse brain. Taken together, our findings suggest that *Chd8* dosage augments gene expression in a brain region-specific and sex-specific manner, but additional resolution is needed to conclude the specific cell types and functional consequences of these effects. Ongoing efforts focus on employing single-cell approaches to investigate the effects of *Chd8* haploinsufficiency with cell-type specificity. This work represents the crucial first steps towards pinpointing molecular pathways that are disrupted in *Chd8* mutants and may reveal new insights to the molecular and cellular origins of severe NDDs.

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Poster

P045. Animal, Synaptic, and Cellular Mechanisms

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Program #/Poster #: P045.02

Topic: A.07. Developmental Disorders

Support: ERANET-NEURON project NeuroTalk
Grant PRE_2020_2_0109

Title: In vivo functional imaging of the Cntnap2 KO model of autism

Authors: *T. SIERRA-ARREGUI, O. PENAGARIKANO;
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Abstract: One of the most accepted theories about the pathogenesis of Autism Spectrum Disorder (ASD) is the alteration in the proportion of excitatory projection neurons (PNs) and inhibitory cortical neurons (cINs). Thus, postmortem studies identify migration abnormalities of PNs, reduced number of cINs and an alteration in glutamatergic and GABAergic receptors, which supposedly lead to associated changes in both excitatory and inhibitory networks. In this work, we characterized the bio-distribution of glutamatergic and GABAergic receptors in the Cntnap2 mouse model of autism, a model characterized by disruption of both PNs and cINs. For this purpose, we performed PET-CT of the mGlu5 and GABA(A) receptors, with ^{18}F-FPEB and ^{11}C-Flumazenil, respectively, in a group of Cntnap2 knockout and wild type mice. We analyzed the following ROIs: olfactory bulb, cortex, hippocampus, dorsal and ventral striatum, thalamus, hypothalamus, amygdala, septum, cerebellum and brain stem. Radioligand quantification was performed using the binding potential (BP_{ND}) for ^{18}F-FPEB, using cerebellum as the reference region, and the total volume distribution (V_t) for ^{11}C-Flumazenil, using in this case heart as reference region. Statistical analyses were performed with Student's t test comparing WT and KO for each region. Our results indicate a normal distribution of mGluR5 in Cntnap2 mice and a tendency towards lower concentration GABA(A) in all areas analyzed, although it did not reach statistical significance. Our results are in agreement with physiological alterations in GABAergic transmission and more precisely, reduced GABA(A)R mediated inhibition, described in the literature for this mouse model. Future research is necessary to determine the contribution of these observations to the pathophysiology of the disorder.

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Poster

P045. Animal, Synaptic, and Cellular Mechanisms

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Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P045.03

Topic: A.07. Developmental Disorders

Support: JSPS Kakenhi 19KK0211

Title: Involvement of nACh-NKCC1-GABAA cascade in the embryonic development of visual predisposition to biological motion in domestic chicks

Authors: *T. MATSUSHIMA[1], M. MIURA[1], N. PATZKE[1], N. TOJI[1], K. WADA[1], Y. OGURA[2], K. J. HOMMA[3], P. SGADÒ[4], G. VALLORTIGARA[4];
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Abstract: Predisposed preference for biological motion (BM) appears early in newly hatched domestic chicks (Vallortigara et al. 2005). It also appears in newborn human babies as an early behavioral characteristic associated with familial risk of ASD (Simion et al. 2008, Di Giorgio et al. 2015). In chicks, the BM preference is induced through non-specific post-hatch experiences such as walking in darkness or visual exposure to non-biological motion pictures (Miura & Matsushima 2016, Miura et al. 2020). The induced BM predisposition would canalize visual attention of chicks, making them selectively attend to living organisms such as the mother hen and siblings. Imprinting also causes influx of thyroid hormone to telencephalon, which makes the sensitive period keep opening until 4-8 days post-hatch (Yamaguchi et al. 2012). Imprinting thus comprises a series of dynamic events leading to robust development of social behaviors. Is the BM preference dissociable from the accompanying memory formation of imprinting? Inspired by the finding that sodium valproate (VPA) impairs social predisposition (Sgadò et al. 2018), we surveyed molecular machineries involved in the canalizing process. Embryonic 14 days eggs received single injection of chemical agents, and a week later, post-hatch chicks were trained and tested. Acute effects of the agents on spontaneous movement of embryo were also recorded. VPA and ketamine suppressed the embryonic motion, but the hatchlings showed distinct behaviors even though both block NMDA receptors; VPA selectively impaired the imprinting memory, whereas ketamine impaired the BM predisposition. Blockade of NMDAR by MK-801 similarly arrested the embryos, but the chicks showed no behavioral impairments. Nipecotic acid (GABA uptake inhibitor) and bicuculline methiodide (GABA-AR blocker) impaired BM but not memory. Finally, tubocurarine (blocker of nicotinic ACh receptor) arrested embryos and selectively impaired BM, and bumetanide (inhibitor of cation-chloride transporter, NKCC1) caused a comparable BM impairment without arresting embryos. VPA reduced neuron/glia ratio of telencephalon and acutely enhanced histone H3K27 acetylation in dissociate neuron culture, while ketamine had no effects. Cascade of nACh, NKCC1 and GABA-A in CNS could thus underlie the BM predisposition, which develop independently from the mechanism for memory formation. Vallortigara et al. (2005) Plos Biol 3, e208; Simion et al. (2008) PNAS 105, 809-813; Di Giorgio et al (2015) Sci Rep 6, 26395; Miura & Matsushima (2016) Anim Behav 116, 171-180; Miura et al. (2020) Anim Cog 23, 169-188; Yamaguchi et al. (2012) Nature Comm 1081; Sgadò et al. (2018) Sci Rep 8, 5919.

Disclosures: T. Matsushima: None. **M. Miura:** None. **N. Patzke:** None. **N. Toji:** None. **K. Wada:** None. **Y. Ogura:** None. **K.J. Homma:** None. **P. Sgadò:** None. **G. Vallortigara:** None.

Poster

P045. Animal, Synaptic, and Cellular Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P045.04

Topic: A.07. Developmental Disorders

Title: Altered cerebellar response to somatosensory stimuli in the Cntnap2 mousemodel of autism

Authors: *M. FERNANDEZ[1], C. SANCHEZ-LEON[3], J. LLORENTE[2], T. SIERRA-ARREGUI[1], J. MARQUEZ-RUIZ[4], O. PEÑAGARIKANO[1];
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Abstract: Atypical sensory processing is currently included within the diagnostic criteria of autism. Thecerebellum is known to integrate sensory inputs of different modalities through its connectivity tothe cerebral cortex. Interestingly, cerebellar malformations are among the most replicated featuresfound in postmortem brain of individuals with autism. We studied cerebellar integration of sensoryinformation in a mouse model of autism, knockout for the Cntnap2 gene. Cntnap2 is widelyexpressed in Purkinje cells and has been recently reported to regulate their morphology. Further,individuals with CNTNAP2 mutations display cerebellar malformations and CNTNAP2 antibodiesare associated with a mild form of cerebellar ataxia. Previous studies in the Cntnap2 mouse modelshow an altered cerebellar sensory learning. However, a physiological analysis of cerebellarfunction has not been performed yet. We studied sensory evoked potentials in cerebellar Crus I/IIregion upon electrical stimulation of the whisker pad in alert mice and found striking differencesbetween WT and Cntnap2 KO mice. In addition, single-cell recordings identified alterations inboth sensory-evoked and spontaneous firing patterns of Purkinje cells. These changes wereaccompanied by altered intrinsic properties and morphological features of these neurons. Together,these results indicate that the Cntnap2 mouse model could provide novel insight into thepathophysiological mechanisms of ASD core sensory deficits.

Disclosures: M. Fernandez: None. **C. Sanchez-Leon:** None. **J. Llorente:** None. **T. Sierra-Arregui:** None. **J. Marquez-Ruiz:** None. **O. Peñagarikano:** None.

Poster

P045. Animal, Synaptic, and Cellular Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P045.05

Topic: A.07. Developmental Disorders

Support: NIH NIDA Grant 5R44DA043954-03

Title: Predicted metabolites from multiple microbiome sequencing datasets modulate Autism Spectrum phenotype in mice

Authors: S. IWAI[1], X. YIN[1], A. PHILLIPS[2], G. E. DEITZLER[3], K. WEST[1], E. RUTHERFORD[1], P. LAL[1], K. DABBAGH[1], T. PALMER[4], D. WALL[4], M. DAVID[2], ***T. DESANTIS**[1];
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Abstract: Introduction: Gut microbiome dysbiosis is associated with central nervous system disorders such as autism spectrum disorder (ASD). The impact of intestinal microbial metabolism in ASD can be estimated from multiple cohorts then tested in vivo. Methods: Ten 16S rRNA amplicon, three shotgun metagenomics and one metatranscriptomics sequencing data from stools of 588 ASD and 518 age- and/or sex-matched neurotypical pediatric subjects were analyzed. Differentially abundant microbial metabolites associated with ASD were predicted, followed by meta-analysis to identify concordance across datasets. Selected metabolites were tested in CNTNAP2 mice for modulation of brain RNA-seq pathways and behavior associated with ASD. Results: We identified two bile acids, Glycodeoxycholate (GDC) and Ulsodeoxycholate (UDC), with others as concordantly decreased in ASD across datasets. Brain RNA-seq of the mice fed the metabolites exhibited enrichment of neuronal system and neurotransmitter receptors and postsynaptic signal transmission pathway gene expressions. Increased habituation exploration and decreased anxiogenic behavior were observed with GDC and UDC fed CNTNAP2 mice, respectively. Conclusion: By applying prediction algorithms and meta-analysis to a large set of microbiome sequencing data, we were able to identify metabolites that modulate behaviors in mice. This approach accelerates the drug discovery process in ASD and can be translated to other disease areas of interest.

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Poster

P045. Animal, Synaptic, and Cellular Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P045.06

Topic: A.07. Developmental Disorders

Support: Landreth Foundation
Nancy Lurie Marks Foundation
BRAIN centre, IOPPN, KCL

Title: Exposure to Autism-specific maternal autoantibodies leads to longitudinal changes in brain structure, neurometabolite profile, and ASD-relevant behaviors in rats

Authors: *M. BRUCE[1], A. COUCH[2], S. GRANT[1], E. KIM[2], D. ROWLAND[1], R. J. MADDOCK[3], M. D. BAUMAN[4], A. VERNON[2], J. VAN DE WATER[1];
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Abstract: Autism Spectrum Disorder (ASD) is a neurodevelopmental disorder characterized by altered social, communicative, and repetitive behaviors, though of unknown etiology. The Van de Water lab has previously identified a set of potentially pathogenic autoantibodies (aAb), specific to ASD, in the plasma of ~20% of mothers of ASD children. We have termed this phenomenon maternal autoantibody-related (MAR)-ASD. However, mechanisms linking these aAb to altered neurodevelopment is still unclear. Therefore, we have created an active immunization model in rats, whereby we induce endogenous aAb production in rat dams to specific epitopes related to ASD diagnosis in the clinical population. To evaluate the impact of gestational MAR-ASD aAb exposure, offspring outcomes were evaluated using behavioral testing, *in vivo* longitudinal neuroimaging, and postmortem brain immunohistochemistry (IHC) along with transcriptional profiling. Using these methods we confirmed that IgG is present, and persists postnatally in the brains of MAR-ASD rat pups; evaluated at postnatal day 2 (P2) using IHC. To examine longitudinal effects of MAR-ASD aAb exposure, we utilized behavioral testing and magnetic resonance imaging and spectroscopy (MRI/S). MAR-ASD rat offspring displayed alterations in communicative, social play, and stereotyped behaviors compared to control animals. Additionally, MRI/S analysis of rat offspring revealed sex-specific volumetric changes in total and regional brain volume in cortical, cerebellar, and midbrain structures. Simultaneous spectroscopic analysis of neurometabolites in the prefrontal cortex (PFC) of rat brains revealed a persistent increase in taurine and decrease in choline in MAR-ASD rat offspring compared to controls which was confirmed using *ex vivo* nuclear magnetic resonance (NMR). Overall these data reveal alterations in brain development and behavior in our MAR-ASD rat model which exhibits substantial translational capacity.

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Poster

P045. Animal, Synaptic, and Cellular Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P045.07

Topic: A.07. Developmental Disorders

Support: ANR CortMem

Title: Mechanisms of atypical sensory information processing within the somatosensory cortex of autism mouse models

Authors: *V. SAKKA[1,2,4], A. A-BHASKARAN[5], T. GAUVRIT[3], A. FRICK[1];
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Abstract: Atypical sensory experience - in particular in the tactile domain - is a key feature of autism spectrum disorder (ASD). Sensory information is processed within the neocortical circuits which are highly amenable to studies via physiological measurements both in humans and mice. Understanding the neurobiological underpinnings of atypical sensory information processing necessitates rigorous analysis in animal models. In mouse somatosensory cortex, we measured in vivo the activity of layer 2/3 neurons - either singly or in populations - evoked by tactile paw stimulation in monogenic mouse modes of ASD. These measurements were combined with those of spontaneous activity and neuronal intrinsic excitability, enabling the identification of mechanisms. Notably, we found complex and nuanced phenotypes across the different mouse models composed of both cellular and circuit alterations. Analysis revealed that most parameters were more variable in the ASD models in comparison with their controls, but also between the different models. Our results mirror the plethora of different phenotypes met on human patients with ASD. Finally, they can be the first step towards a therapeutic approach, suggesting targetable mechanisms at the level of neocortical circuits.

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Poster

P045. Animal, Synaptic, and Cellular Mechanisms

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Program #/Poster #: P045.08

Topic: A.07. Developmental Disorders

Support: Jordan's Guardian Angels

Title: An RNA Targeting Approach for Correction of Pathogenic Nucleotides Implicated in Genetically-Linked Intellectual Disabilities

Authors: *J. L. CARTER[1], J. A. HALMAI[1], J. A. NOLTA[2], K. FINK[1];
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Abstract: Genetically-linked intellectual disabilities (ID) are complex neurodevelopmental disorders. Strategies for targeting ID genes at the DNA level are difficult and potentially dangerous due to DNA repair pathways. ADAR2 is a brain-enriched protein which post-transcriptionally modifies double-stranded RNA (dsRNA) by deaminating adenosine nucleotides to inosine, thereby enabling translational machinery to create a functional adenosine to guanosine edit within the transcript. The CRISPR associated protein 13 (dCas13b) is a catalytically inactive RNA binding protein which achieves site directed RNA editing when fused to the ADAR2 deaminase domain (ADAR2DD). A single guide RNA (sgRNA) sequence forms a dsRNA substrate for dCas13b-ADARDD2 binding at the complementary target while enabling RNA editing at a single base. However, the CRISPR Cas13 system has yet to be employed for RNA editing of G-to-A mutations in patient-derived neuronal cells. We created induced pluripotent stem cell (iPSC) from the most common variant of Jordan's Syndrome, a G-to-A mutation in Exon 5 (E198K) of *PPP2R5D*. Here, we have differentiated isogenic and variant iPSCs to neural stem cells (NSCs) to evaluate RNA editing efficiency and efficacy. Towards this goal, we have designed and screened E198K sgRNA to identify lead gRNA which have high on-target editing and low-moderate off-target editing in the *PPP2R5D* transcript. We demonstrate sgRNA structure is readily amenable to fit sequence constraints within the target transcript, thus improving on-target editing efficiency. We are also using our *in vitro* neurodevelopmental model to begin understanding the consequence of G-to-A mutations on *PPP2R5D* targets at the phosphoproteomic and transcriptomic levels. To our knowledge we are the first to employ the CRISPR RNA editor system to correct G-to-A mutations in a patient-specific neuronal model. These studies support the potential for CRISPR/dCas13 systems to selectively edit mutant transcripts harboring G-to-A mutations while providing an alternative editing technology for neurodevelopmental disorders.

Disclosures: J.L. Carter: None. **J.A. Halmai:** None. **J.A. Nolta:** None. **K. Fink:** None.

Poster

P045. Animal, Synaptic, and Cellular Mechanisms

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Program #/Poster #: P045.09

Topic: A.07. Developmental Disorders

Support: NIMH T32MH112510
Howard Hughes Medical Institute
Hock E. Tan and K. Lisa Yang Center for Autism Research
U54HD090255

Title: Single-cell interrogation of genetically defined neurodevelopmental disorders in human brain

Authors: *C. DIAS[1,2], S. AKULA[2,1], M. TALUKDAR[2,1], K. WALSH[3,1], C. A. WALSH[1,4,2]; [1]Genet. and Genomics, Boston Children's Hosp., Boston, MA; [2]Harvard Med. Sch., Boston, MA; [3]Tufts Univ., Boston, MA; [4]Howard Hughes Med. Inst., Boston, MA

Abstract: Neurodevelopmental disorders (NDDs) such as autism spectrum disorder (ASD) are characterized by significant heterogeneity. The unique features of these conditions (impairments in joint attention or shared enjoyment in ASD for example) necessitate better understanding of these disorders in the human brain specifically. Recent attempts at assaying functional changes on the single cell level in human brain have focused on broad, non-biologically defined categories. We hypothesize that different genetic drivers associated with NDDs have both divergent and shared functional mechanisms in different cellular subpopulations. To test this hypothesis, we are conducting single nuclei RNA sequencing (snRNA-seq) on post-mortem brain from individuals with known genetic disorders associated with NDDs including structural variation (n=6) and Fragile X (n=6) as well as age and sex matched neurotypical controls (n=8). Our preliminary data demonstrates that recurrent structural variation exhibits marked cell-type specific effects in human brain, a finding that has important consequences for identifying critical cell types of interest. We additionally demonstrate for the first time on the single cell level the parallel consequences of both *FMR1* gene deletion and 5' UTR *FMR1* trinucleotide expansion in a variety of neuronal and non-neuronal subtypes. Overall, our data demonstrate the importance of studying genetically defined neurodevelopmental disorders in human brain tissue, and yields novel insight into functional mechanisms which will be critical to understanding the heterogeneity underlying these disorders so apparent on the clinical level. Additionally, findings from this work can be used to inform future rational and personalized therapeutic development for functionally impairing symptoms and co-morbidities.

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Poster

P072. Development of Sensory and Limbic Systems

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P072.01

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Title: Physiological indicators of negative attentional bias in children with prenatal substance exposure are related to caregiver emotional literacy

Authors: *J. E. NORRIS[1], A. MELON[2], J. HEIDARY[1], E. BARD[2], L. E. ETHRIDGE[1]; [1]Psychology, Univ. of Oklahoma, Norman, OK; [2]Pediatrics, Univ. of Oklahoma Hlth. Sci. Ctr., Oklahoma City, OK

Abstract: Prenatal alcohol and drug exposure has a profound effect on development and subsequently social, cognitive, and emotional wellbeing. Previous work has shown that children exposed to adverse childhood experiences (ACEs), including prenatal alcohol and drug exposure, monitor facial expressions of emotion differently from children from settings with low exposure risk. Emotion processing in the child's caregivers can impact the effectiveness of parent-child therapeutic interventions for developmental consequences of ACE exposure but limited work has evaluated physiological relationships between child emotion processing and caregiver emotional literacy. Nine caregiver-child dyads participated in emotion identification tasks using eye-tracking technology to record gaze-point and search strategy data. All children had confirmed or suspected prenatal alcohol or drug exposure. Adult participants [N = 9; Mean age = 41.842 years, SD = 8.34; 9 females] were instructed to search for a specific target emotion from six faces expressing emotions ranging from positive to negative (happy, neutral, sad, anger, fear, and disgust). The child task differed with child participants [N = 10; Mean age = 4.11 years, SD = .99; 5 Females] only required to identify the face expressing a negative emotion, where 5 faces expressed a neutral emotion and 1 expressed either sad, fear, or anger. Areas of interest (AOIs) were predefined to include the eyes, the mouth and nose, and the whole image for each image on a trial. Children were significantly faster to fixate on eyes over mouths for sad faces $t(9) = -2.47$, $p = .036$ but not for other negative emotions. Faster speeds to find sad faces in children were correlated with slower identification speeds for sad and fearful images in their adult caregiver, (p's $< .05$). Children exhibited an initial pattern of attention to eyes over mouth for sad trials, but not other negative emotions. Children's ability to quickly find and fixate on sad images was negatively correlated with their caregiver's speed to find and identify negative emotions, suggesting a potential sensitization or attentional bias to negative emotions, particularly sadness, in children related to reduced negative emotional literacy in their caregiver. These findings suggest that physiologically characterized emotional literacy in the caregiver may be relevant to negative emotion processing in children who have experienced ACEs, and may be informative for optimizing individual outcomes for parent-child-oriented therapeutic interventions.

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Poster

P072. Development of Sensory and Limbic Systems

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Program #/Poster #: P072.02

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Support: NIH K99MH124434
BBRF NARSAD

Title: Social Regulation of Developing Mesolimbic Dopamine Circuitry

Authors: K. PACKARD[1], S. HU[1], J. WANG[2], *M. OPENDAK[2,3,1];
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Abstract: Infants rely on the mother to provide them with the sensory stimulation needed for normal brain development. However, harsh or neglectful care can perturb the mother's ability to regulate the infant brain - an early marker of lasting socioemotional impairments. Recent work has highlighted the mesolimbic dopamine system as a critical interface between early quality care and the ability of the mother to regulate the infant's threat response, but specific mechanisms remain unclear. Furthermore, the lateral habenula(Lhb) has been consistently implicated as a locus of dysfunction in psychopathology in adults. Here, we assessed how rearing experience impacted maternal regulation of core nodes of the mesolimbic dopamine circuit, including the lateral/medial habenula and ventral tegmental area (VTA). To modulate maternal care quality, we employed the Scarcity-Adversity model of low bedding (SAM-LB), in which a dam provided limited bedding resources handles pups more roughly. Following control or SAM-LB rearing from PN8-12, rat pups were exposed to odor-shock conditioning with or without maternal presence at postnatal days 18-20 (PN18, before weaning) and postnatal days 26-28 (PN28, after weaning). 2-Deoxyglucose (2-DG) uptake was used as a proxy measure of regional engagement. In control-reared pups, we observed that maternal presence suppressed 2-DG uptake in the VTA, and lateral habenula. Using chronic VTA recordings in awake-behaving control-reared pups, we observed that maternal cues were associated with increased unit responses, while threat cues were associated with decreased unit responses. The effect of maternal presence was abolished in adversity-reared pups, with early rearing significantly altering patterns of functional connectivity within this circuit. We next assessed the role of this circuit in social behavior using optogenetics. Following virus transduction (ChR2-EYFP, NHpr-EYFP, EYFP control) into Lhb or VTA at PN1, pups underwent control or SAM-LB and then tested on social behavior with the mother (PN18) or peers (PN28). Engagement of this circuit modulated social behavior at both ages. Taken together, these data suggest that lasting social impairments following early adversity may be due to impaired maternal regulation of the mesolimbic dopamine circuit.

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Poster

P072. Development of Sensory and Limbic Systems

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Program #/Poster #: P072.03

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Support: NIH Grant K01MH119241

Title: Impact of trauma exposure and endocannabinoid signaling on frontolimbic white matter pathways in children

Authors: *H. MARUSAK, A. MORALES, B. BORG, J. EVANSKI;
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Abstract: The endocannabinoid system (eCB) is present and functional in early pregnancy and plays a key role in modulating pre- and post-natal brain development, including myelination processes. Recent studies in adolescents and adults link a common variant (C385A) in the gene encoding fatty acid amide hydrolase (FAAH) with higher eCB levels, lower anxiety, and altered frontolimbic development. Frontolimbic white matter pathways demonstrate a protracted maturational course across childhood and adolescence, are associated with anxiety symptoms, and shown to be sensitive to the effects of environmental stress. Here, we examined the impact of the FAAH C385A polymorphism and trauma exposure on anxiety and integrity of frontolimbic white matter pathways in children. We leveraged data collected from the ongoing large-scale NIH Adolescent Brain Cognitive Development (ABCD) study (n = 10,774; $M \pm SD$ age = 9.92 ± 0.62 years; 47.9% female; 54.3% White, 13.8% Black, 19.3% Hispanic, 2.1% Asian, 10.4% Other). Saliva samples were used for genotyping and parents reported on their child's anxiety symptoms and trauma exposure using the Child Behavior Checklist and the Kiddie Schedule for Affective Disorders and Schizophrenia (K-SADS) traumatic events screener, respectively. Fractional anisotropy (FA) was estimated from DTI data for frontolimbic white matter pathways. Overall, 43.4% of youth were FAAH A-allele carriers or homozygotes (52.1% were CC homozygotes), and 35.7% of youth were exposed to trauma (64.3% unexposed). Trauma-exposed youth demonstrated higher anxiety symptoms and higher FA of the right and left uncinate fasciculus and corpus callosum (forceps minor), as compared to trauma-naïve youth. There was no main effect of the FAAH C385A variant or trauma x FAAH interaction on anxiety symptoms. However, there was a main effect of FAAH genotype on FA in the left and right fornix and left parahippocampal cingulum, such that FA was higher in CC homozygotes as compared to A-alleles. There was also a significant trauma x FAAH interaction for FA in the left and right fornix, right uncinate fasciculus, corpus callosum, and right parahippocampal cingulum. Follow-up analyses in gene groups separately demonstrated that the effects of trauma on white matter integrity were significant in the CC (but the not A-allele)

group. In particular, trauma-exposed CC homozygotes demonstrated greater FA than trauma-naïve CC homozygotes. These results highlight the role of trauma exposure and endocannabinoid signaling in modulating frontolimbic development and anxiety risk.

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Poster

P072. Development of Sensory and Limbic Systems

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Program #/Poster #: P072.04

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Title: Developmental predator odor exposure and the expression of innate fear

Authors: *N. COLLINS[1], L. W. AYERS[2], S. FERN[1], S. ROSS[1], T. L. ROTH[1], J. B. ROSEN[1]; [1]Univ. of Delaware, Newark, DE; [2]Widener Univ., Chester, PA

Abstract: Early life stress in both humans and rodents shapes neurodevelopment and later behavior, including higher incidences of psychopathology. Previous work in our laboratory demonstrated the capacity of predator odor exposure (TMT, a synthetic component of fox feces) to induce robust freezing in naïve rodents without prior learning. Further, if this odorant is administered during early development with the presence of a dam the quality of maternal care and later defensive responses of the progeny are altered. More specifically, dams exposed to TMT during the first three weeks of life (PN1-21) engage in significantly longer nursing bouts compared to dams exposed to a noxious control odor (butyric acid, BTA). When tested for freezing to TMT during adolescence (PN30), offspring exposed to TMT during development froze less than rats exposed to BTA (Ayers et al., 2015). The present study sought to extend these findings to another predator odorant, and examine the extent of maternal care quality in modifying defensive freezing in the progeny. First, a log-dose response for PN30 freezing was established by exposing PN30 animals (without developmental odor exposure) to concentrations of BTA, TMT, bobcat urine, and water. Results replicated previous findings in our laboratory, such that animals exposed to TMT at doses of 100 µmole or 300 µmole had significantly higher freezing compared to similarly matched doses of BTA or water. However, a 5 mL dose of bobcat urine was ineffective at inducing freezing behavior compared to control odors. Together, these data suggest that TMT, but not BTA or bobcat urine, elicits a defensive freezing response. Current work is examining the role of maternal care in modifying defensive responses in offspring exposed to predator odor during early development, and exploring concomitant molecular changes in the brain.

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Poster

P072. Development of Sensory and Limbic Systems

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P072.05

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Title: Impact of Prenatal Cannabis Exposure on Childhood Anxiety and Frontolimbic Neurodevelopment

Authors: *M. M. FARAJ, A. MORALES, J. EVANSKI, B. BORG, H. A. MARUSAK; Wayne State Univ., Detroit, MI

Abstract: Cannabis use is common among pregnant women and during the pre-conception period (up to 22%); however, prenatal cannabis use may have detrimental effects on childhood outcomes, including neurodevelopment. Indeed, research shows that constituents of cannabis, including Δ^9-tetrahydrocannabinol (THC), can cross the placenta and alter fetal neurodevelopment, and prenatal cannabis exposure is associated with cognitive, motor, and social deficits that last into the adulthood of offspring. Leveraging the ongoing large-scale NIH Adolescent Brain Cognitive Development (ABCD) study, recent studies also link prenatal cannabis exposure after (but not before) knowledge of pregnancy to lower intracranial volume and white matter volumes. Here. we examine the impact of prenatal cannabis exposure on childhood anxiety symptoms and frontolimbic brain regions involved in emotion regulation and anxiety risk. This is important because anxiety disorders are the most common childhood mental disorders, and the endocannabinoid system — which may be disrupted by cannabis exposure — plays a key role in modulating frontolimbic development and is implicated in anxiety. Neuroimaging data were collected from 11,206 children ($M \pm SD = 9.91 \pm 0.62$ years; 47.7% female; 54.4% White, 13.5% Black, 19.8% Hispanic, 2% Asian, 10.3% other), and prenatal cannabis exposure was assessed based on parents' retrospective report. In this sample, 4.1% of parents reported cannabis use before knowledge of pregnancy (n = 454), and 1.2% reported use after knowledge of pregnancy (n = 135). Regional gray matter volumes and functional activation during an emotional n-back task were estimated for the amygdala, hippocampus, dorsal anterior cingulate cortex (dACC), and insula. Prenatal cannabis exposure during and after knowledge of pregnancy was associated with higher childhood anxiety symptoms. Controlling for whole brain volume, cannabis exposure before, but not after, knowledge of pregnancy was associated with larger volumes of the right insula. During the emotional n-back task, cannabis exposure *after*, but not before, pregnancy was associated with heightened dACC response to emotional faces relative to neutral faces. Cannabis exposure was not associated with volume or activation in other areas. These data indicate that prenatal cannabis exposure may increase child anxiety and alter frontolimbic development. Results of this study may inform educational interventions to discourage pregnant women or those who are contemplating pregnancy from using cannabis.

Health care professionals can use this information and encourage the public to urge women to refrain from cannabis use during pregnancy.

Disclosures: M.M. Faraj: None. **A. Morales:** None. **J. Evanski:** None. **B. Borg:** None. **H.A. Marusak:** None.

Poster

P072. Development of Sensory and Limbic Systems

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P072.06

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Support: Dan and Betty Kahn Foundation
Kids Kicking Cancer

Title: Smaller hippocampal volume predicts improvement in posttraumatic stress symptoms in pediatric cancer patients following a novel martial arts-based intervention

Authors: *Z. OWENS[1], A. IADIPAOLO[1], C. PETERS[1], C. COHEN[2], E. GOLDBERG[2], M. BLUTH[2], J. TAUB[1], F. HARPER[1], C. RABINAK[1], H. MARUSAK[1];
[1]Wayne State Univ., Detroit, MI; [2]Kids Kicking Cancer, Southfield, MI

Abstract: Smaller hippocampal volume predicts improvements in posttraumatic stress symptoms in pediatric cancer patients following a novel martial arts-based intervention
Authors: Zazai Owens, Allesandra Iadipaolo, Craig Peters, Cindy Cohen, Elimelech Goldberg, Martin Bluth, Jeffrey Taub, Felicity Harper, Christine Rabinak, Hilary Marusak
Young people with cancer endure an enormous amount of stress and trauma prior, during and after their cancer treatments. The treatment phase can take a physiological, and emotional toll on youth and many pediatric cancer patients report posttraumatic stress symptoms (PTSS). Our prior research in pediatric cancer populations has demonstrated associations between PTSS and variation in stress-sensitive brain regions, such as the hippocampus. In tandem, prior studies in adults with breast cancer consistently link smaller hippocampal volumes to cancer-related PTSS, particularly re-experiencing PTSS (e.g., flashbacks, nightmares). The present study examines the impact of a novel martial-arts based intervention on hippocampal volume and PTSS in pediatric cancer patients, and whether baseline (i.e., pre-intervention) hippocampal volumes predict changes in PTSS over time. Eighteen pediatric cancer patients or survivors (ages 5-17 years, 8 female) participated in this 4-week prospective study. Participants underwent structural magnetic resonance imaging (MRI) scans at baseline and then completed 4 weekly Kids Kicking Cancer classes. Kids Kicking Cancer is a martial arts-based therapy that integrates meditation, breathing, and physical movements, and has been shown to lower pain and distress in pediatric cancer and other populations (e.g., sickle cell, schoolchildren, adults with opioid use disorder). Before and

after the 4-week program, participants self-reported on cancer-related PTSS. Analyses examined whether PTSS and hippocampal volumes changed over time, and whether hippocampal volume at baseline predicted change in PTSS. Overall, PTSS symptoms and hippocampal volumes did not change significantly over the 4-week period. However, youth with smaller left (but not right) hippocampus at baseline demonstrated greater reductions in PTSS over time. Exploratory analyses suggests that the association between hippocampal volumes and change in PTSS was driven by re-experiencing PTSS. Results from this pilot study suggest that smaller hippocampal volumes may predict reductions in cancer-related PTSS following a martial arts-based intervention. They also extend prior studies suggesting that a martial arts-based approach may be effective for reducing pain and distress in pediatric cancer populations.

Disclosures: Z. Owens: None. **A. Iadipaolo:** None. **C. Peters:** None. **C. Cohen:** None. **E. Goldberg:** None. **M. Bluth:** None. **J. Taub:** None. **F. Harper:** None. **C. Rabinak:** None. **H. Marusak:** None.

Poster

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Program #/Poster #: P072.07

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Support: University of Minnesota UROP grants

Title: Association between spatial acuity and empathy in typically developing young children

Authors: *R. LLOYD, J.-B. QUILLIEN, A. BYGD, R. MILLER;
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Abstract: Raine et al (2002) report that persistently antisocial individuals demonstrated spatial but not verbal cognitive deficits at three years of age. Verbal deficits did not emerge until eleven years of age. The authors posit an “early starter” model in which early spatial deficits are a marker for right hemisphere impairments in affect which then results in antisocial behavior, possibly including mother-infant bonding. The subsequent verbal deficits are interpreted as resulting from these social deficits. We examined the relationship between spatial acuity and the positive social quality, empathy, in 29 typically developing 3-5 year olds (17 males; 12 females) from The Children’s Place at the University of Minnesota Duluth campus, a facility for children whose parents are faculty and staff at the University. Spatial acuity was assessed by measuring the speed and accuracy of maneuvering in two, successively difficult, physical mazes. Since the validity of instruments measuring empathy in children this young have been called into question, we developed our own instrument in which 16 very short vignettes were read to the child. After each vignette the child was asked if they were more like the empathic character or more like the

non-empathic character. Empathic responses were then totaled. When empathy scores were regressed against the total number of wrong turns in the two mazes, a significant quadratic relationship ($p<0.048$) was found with an R-square of 0.223. When the data were submitted to a quadratic hierarchical regression analysis (empathy as the dependent variable), with age entered into the equation first, followed by total number of wrong turns, the R-square value increased significantly from 0.181 to 0.324. Thus, 14.3% of the variability in empathy scores can be associated with accuracy in the two mazes independent of the age of the child. Male and female children did not differ in total number of wrong turns ($p=0.889$), speed in the first (smaller) maze ($p=0.662$), speed in the larger maze ($p=0.557$), or Empathy score ($p=0.274$, $r^2=0.048$). Care must be taken in the interpretation of these data. For any given age, children will develop at different rates. The relationship between empathy and spatial acuity, after controlling for age, may merely reflect general neurological development. Data in additional cognitive/neurological domains are now being collected to determine if this relationship between empathy and spatial acuity in very young typically developing children is specific in nature, independent of other levels of cognitive development, per Raine et al. We are also attempting to widen our reach of participants to a larger, more generalizable population.

Disclosures: R. Lloyd: None. **J. Quillien:** None. **A. Bygd:** None. **R. Miller:** None.

Poster

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Program #/Poster #: P072.08

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Support: NIH Grant GM122657

Title: Offspring of Calorically Restricted Mothers Exhibited Neuroanatomical and Behavioral Alterations

Authors: *K. BERTA, S. LUTCHMAN, J. BRUMBERG;
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Abstract: Calorie consumption during prenatal development has been shown to impact postnatal health, and inadequate prenatal caloric intake in particular has been linked with later physical and mental health issues. To better study this, adult female CD-1 mice were placed on a calorically restricted diet before and throughout pregnancy. Pups birthed to calorically restricted mothers were assayed for their behavior, microglial and neuronal morphology along with perineuronal net density in the barrel cortex was assessed to gauge the impact of a restricted prenatal diet. An open field paradigm was conducted to examine anxiety levels in the resulting pups through analysis of locomotor activity. While no dramatic differences to behavior were

found, the oldest group of control mice spent the least amount of time in the outer arena during the open field paradigm. Female prenatally restricted pups also expressed less willingness to explore in the open field compared to control pups. Mice that were calorically restricted in utero had significantly fewer perineuronal nets compared to non-restricted mice. There was also a sex difference with female offspring being more affected by maternal caloric restriction than male offspring. The microglia of prenatally restricted mice were more rammified compared to non-restricted mice. Mice that had been calorically restricted in utero had significantly smaller neuron somata, a difference which was specific to certain layers of the barrel field. From studying the impact of food restriction in mice, this data can be used to better understand the effects of inadequate caloric intake on children born to mothers during times of food shortage.

Disclosures: K. Berta: None. **S. Lutchman:** None. **J. Brumberg:** None.

Poster

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Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Support: NSERC RGPIN-2018-04908
Bertha Rosenstadt endowment fund

Title: Developmental trajectory of the trigeminal nerve: a diffusion tensor imaging study

Authors: *E. E. OSOKIN, B. KAYA, I. CIOFFI, M. MOAYEDI;
Fac. of Dent., Univ. of Toronto, Toronto, ON, Canada

Abstract: The trigeminal nerve is the largest cranial nerve and is a mixed nerve. It innervates the craniofacial region, including skin on the face, teeth, oral mucosa, cranial vessels, the temporomandibular joint, and the muscles of mastication. Most of these structures undergo significant growth and change during development through adolescence. Although there is evidence that brain regions and the white matter connecting them undergo structural changes with age, it is not known whether the trigeminal nerve undergoes developmental changes. Diffusion-weighted imaging (DWI), and structural T1-weighted imaging allow the study of neural microstructure and microstructure, respectively. Microstructural characteristics of the trigeminal nerve can be investigated by fractional anisotropy (FA), and the cross-sectional area of the nerve can be measured based on structural T1 images. Here, we aimed to determine whether the microstructural and macrostructural features of the trigeminal nerve change with age in a large cohort of children and young adults. In a second aim, we tested whether there were sex differences in the developmental trajectory of the trigeminal nerve. We obtained approval for the study from the University of Toronto Research Ethics Board. We acquired data from 202 MRI

participants (99 F, 103 M; aged between 6-15 years), from the Child Mind Institute database. We first delineated both trigeminal nerves, and extracted FA values at the root-entry zone (REZ), and measured cross-sectional areas at two locations: REZ and cisternal segment. We performed two multivariate analysis of covariances—one for each outcome measure—and tested whether our metrics of trigeminal nerve structure (FA and nerve cross-sectional area) are affected by age, sex, and the interaction age-by-sex, while controlling for the data collection site. We found that FA and cross-sectional area significantly increased with age for the right nerve (both $p<0.005$), and that the cross-sectional area of the left nerve was significantly positively related to age ($p<0.05$). Neither sex nor age-by-sex interaction had significant effects on either outcome measures (all $p>0.05$). Our study provides novel normative data on the structural development of the trigeminal nerve and lays the groundwork for future studies aimed at investigating whether abnormal nerve development contributes to chronic craniofacial pain conditions.

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Poster

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Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Support: This work was supported by the Elizabeth H. Solomon Center for Neurodevelopmental Research

Title: Electroencephalographic measures of rapid auditory processing at pre-experience baseline predict infant behavior during an interactive acoustic experience

Authors: *K. WOLFERT, T. REALPE-BONILLA, C. ROESLER, A. A. BENASICH;
Ctr. for Mol. & Behavioral Neurosci., Rutgers Univ., Newark, NJ

Abstract: Interactive acoustic experience during infancy has been shown to impact neural measures of rapid auditory processing on a group level, enhancing markers of acoustic discrimination and generalizing to novel auditory stimuli. However, individual differences observed in training performance may capture variance in cortical maturation and neural plasticity. To explore how the baseline state of the auditory cortex impacts learning, we examined neural measures of auditory processing in 4 month-old infants (N = 18) and behavioral measures of their performance during auditory training sessions. Before training, we recorded dense-array electroencephalographic (EEG) data as infants passively listened to tone pairs containing rapid spectrotemporal changes presented in an oddball paradigm. Event-related potential peaks were characterized at frontal electrode clusters in both left and right hemispheres. We also examined demographic and behavioral predictors: measures of maternal age and

education, ratings of infant-caregiver interactions during play (Parent/Caregiver Involvement Scale), and standardized assessments of cognitive, language, and motor development. After the pre-training sessions, infants received six eyetracker-based training sessions in which they learned to discriminate sound pairs at increasingly fast inter-stimulus intervals (ISIs). We hypothesized that a combination of factors, particularly those related to acoustic discrimination, would predict three metrics of training performance: time engaged in the task, discrimination index (d-prime), and fastest ISI reliably detected. Elastic net regularized regression models were used to evaluate the relationship between baseline measures and training behavior, identifying the best predictors in the presence of multicollinearity. Nested cross-validation was used to select regularization parameters and assess prediction error. Predictive models were identified for all three metrics (engagement $R^2 = 0.99$, d-prime $R^2 = 0.49$, fastest ISI $R^2 = 0.78$). For metrics most directly related to acoustic discrimination (fastest ISI and d-prime), EEG measures associated with more mature auditory processing (i.e. faster latencies of N2 and P2 peaks) predicted better training performance. Maternal age and cognitive/motor skills also predicted fastest ISI and d-prime, whereas maternal education was predictive of engagement. These results help us understand the factors influencing experience-dependent plasticity during development and suggest that future protocols might be optimized by adjusting the timing of training to coincide with peak receptiveness to acoustic experience.

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Poster

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Program #/Poster #: P072.11

Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Support: DGE-1845298

Title: Topographic maps in the human neonate

Authors: *M. A. BARNETT, M. J. ARCARO;
Psychology Dept., Univ. of Pennsylvania, Philadelphia, PA

Abstract: Topographic maps of the sensory periphery are a fundamental feature of information processing in the adult primate brain. The adult visual system comprises several topographic maps of visual space spanning occipital, temporal, and parietal cortices. The adult somatomotor system comprises several maps of the body spanning parietal and frontal cortices. Here, we asked whether topographic representations of visual space and the body are present at birth in humans. We performed seed-based correlation analyses on resting-state fMRI data in 34

neonates at gestation weeks 38-42 from the developing human connectome project (developingconnectome.org). Primary visual and somatomotor cortices were identified anatomically based on the location of the calcarine and central sulci, respectively. Each of these anatomical regions were segmented into 5 bands along their respective long axes and correspond to representations of either different parts of retinal eccentricity or different parts of the body in adults. Pearson correlations were computed between the activity in each seed band and all voxels in the brain. Occipital, temporal and posterior parietal cortices were most strongly correlated with visual seeds. Anterior parietal and frontal regions were most strongly correlated with somatomotor seeds. There was a clear topography of correlations along the cortical surface for both visual and somatomotor seeds with neighboring cortical regions most strongly correlated with neighboring seeds. These topographic gradients were symmetric between hemispheres and matched the large-scale organization of visual and somatomotor maps found in adults. A secondary seed-based analysis using a probabilistic atlas of face- and scene- areas in temporal cortex revealed a lateral-medial distinction consistent with the eccentricity organization in adults with little evidence for domain-specific correlations. The somatomotor topography is consistent with recent studies that showed anatomically distinct representations of the body within primary somatomotor cortex in neonates (Dall'Orso et al. 2018; Meltzoff et al. 2019). In addition, topographic correlations were found both anterior and posterior to primary somatomotor cortex as well as in the lateral sulcus likely corresponding to the SII and PV maps. We also found evidence for a topography of thalamo-cortical correlations though these results varied across subjects, possibly indicating immaturity of these pathways. Overall, our results provide evidence that a proto-organization of topographic maps is established prenatally which can provide a scaffold for later modifications through experience.

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Poster

P072. Development of Sensory and Limbic Systems

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Topic: A.08. Development of Motor, Sensory, and Limbic Systems

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Title: Eeg spectral activity and its association with sensory-motor coordination in blind and sighted children

Authors: *H. VITALI[1], C. CAMPUS[1], S. SIGNORINI[2], V. DE GIORGIS[2], F. MORELLI[2], G. PAPALIA[2], M. FASCE[2], M. GORI[1];
[1]U-VIP, Italian Inst. of Technol., Genova, Italy; [2]IRCCS Mondino Fndn., Pavia, Italy

Abstract: Vision acquires an increasingly important role in the first years of age, both for the maturation of neural processing and the interaction with the environment. Literature data show a different spectral composition of brain activity in blind than sighted adults, particularly an absence or a reduction of the alpha activity. Moreover, the lack of vision compromises the ability to reconstruct spatial information leading, on one side, to a reduced exploration of the external environment, and on the other side, worst motor skills. This result suggests a greater difficulty in sensory-motor coordination. Despite this, few studies are investigating the development of alpha activity in the first years of life. Here we explore, from EEG retrospective data, the spectral power of resting-state EEG to reconstruct the developmental trajectory of alpha activity and its association with motor coordination impairment indices in blind and sighted children between 0 and 11 years of age.The results show that background activity has two peaks in both sighted and blind subjects, progressively merging during development. The first one shifts with age, while the second corresponds to the classical alpha band observed in typical adults. However, blind subjects show weaker and slower maturing posterior alpha activity, with a first significant differentiation between 3 and 6 years of age. Starting from this period, reduced alpha activity also increases the probability of motor coordination impairment in blind subjects, suggesting that in blind children, some processes could fail. To investigate if the alpha activity could have a role in the sensory-motor coordination in the first three years of life, we indagated the modulation of the non-nutritive sucking (NNS) motor behaviour and its relationship with cortical rhythms. NNS is one of the first coordinated muscular activities and represents an excellent marker of neural system functioning. The results showed that NNS frequency correlates with the baseline alpha activity in somatosensory and SMA areas in blind and sighted children and acquires a different role during the sucking behaviour. Specifically, only in sighted children, also a beta band correlation was found along the visuomotor pathway. These results suggest that visual experience mediates the neural mechanisms generating neural oscillations during the first years of life. Moreover, they indicate that alpha and beta oscillations have a role in the optimal development of the neural network involved during sensory-motor coordination.

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Poster

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Topic: A.08. Development of Motor, Sensory, and Limbic Systems

Support: NIH R01 MH114994

Title: Oxytocin-paired orofacial stimulation in neonatal mice

Authors: *K. R. DAY, C. SMITH, E. A. HAMMOCK;
Psychology, Florida State Univ., Tallahassee, FL

Abstract: Oxytocin (OXT) is an essential neuropeptide involved in social behaviors such as mother-infant attachment. Milk and saliva contain OXT produced by the mother that may be shared with the infant during early life care, such as grooming or breastfeeding. We have previously identified oxytocin receptor (OXTR) in the orofacial region of neonatal mice, which may be sensitive to maternal (exogenous) OXT cues during the neonatal period. We propose that exogenous OXT may cue the infants' oxytocin receptors (OXTR) to label maternal touch as "social" since OXT is not available from non-social tactile stimuli. To begin to investigate this potential role, we hypothesize that OXT co-applied with tactile stimulation will modulate acute infant behavior responses to orofacial touch. Neonatal male and female C57BL/6j wildtype mice were habituated to a socially isolated environment for two hours on the day of birth (P0). After habituation, tactile stimulation was applied to the whisker pads and external mouth of neonates for 30 seconds with a paintbrush coated in either saline, a low dose (1.56pg/uL) of OXT in saline, or a high dose (1.25ng/uL) of OXT in saline. Neonates were observed for an additional 60 seconds after stimulation. This experiment establishes an ethogram for investigating motor behavior of neonatal mice on the day of birth. Our results demonstrate changes in neonatal behavior in response to the presentation of tactile orofacial stimulation paired with exogenous OXT exposure in a socially isolated setting. We also observed behavioral differences between neonates with and without prior nursing experience. Exogenous OXT paired with somatosensory stimulation may facilitate optimal nursing and infant attachment in early social development.

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Poster

P117. Structural Plasticity: Dendritic Spines

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Topic: B.07. Synaptic Plasticity

Support: NIH R01-NS-062736
NIH T32-GM099608

Title: Saturation of structural plasticity at individual CA1 dendritic spines

Authors: *J. FLORES, K. ZITO;
Ctr. for Neurosci., Univ. of California, Davis, Davis, CA

Abstract: Learning is crucial for survival. One intriguing aspect of learning in humans is that efficiency of learning can be improved when there are breaks between episodes of learning. Long-term potentiation (LTP) of synaptic strength has been proposed as a cellular basis of learning and memory. Notably, it has been shown that LTP can saturate in the hippocampus of rodents and that this saturation can lead to deficits in behavioral learning tasks. LTP saturation has also been shown to occur in hippocampal slices following a single theta burst stimulus (TBS). Intriguingly, this saturation is released over time. Thus, it is possible that release from saturation of LTP in the hippocampus contributes to the increased efficacy of spaced learning over massed learning paradigms. We propose that LTP saturates at individual dendritic spine synapses. In the hippocampus, most excitatory synapses occur at small protrusions on dendrites called dendritic spines. Spines are dynamic and have been shown to grow and stabilize during learning in vivo and in response to high-frequency stimulation in vitro. The size of a spine is strongly correlated with the strength and size of its associated synapse. Here, we use 2-photon glutamate uncaging and time-lapse imaging to show that structural LTP can saturate at individual dendritic spines after a single LTP induction. Additionally, this structural saturation is accompanied by a saturation of AMPAR insertion, thus spine size and synaptic strength both saturate in response to a single glutamatergic stimulus. We also show that saturation of LTP does not spread to spines on other dendrites on the same cell. Notably, if the intensity of the second stimulus is increased, spines can once again exhibit structural LTP, although the magnitude is reduced compared to size-matched control spines. Furthermore, individual spines are fully recovered from saturation and can undergo further structural LTP if the second stimulus is administered 60 minutes after the initial stimulus. Using 2-photon fluorescence lifetime imaging (2p-FLIM) of a genetically-encoded CaMKII FRET probe, we show that 30 minutes following structural LTP induction at single spines, CaMKII activation in response to high-frequency stimulation is significantly reduced. We are currently investigating the mechanisms that lead to this alteration in CaMKII signaling and postsynaptic saturation of LTP at single spine synapses. This work will further our understanding of the signaling mechanisms that underlie learning.

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Poster

P117. Structural Plasticity: Dendritic Spines

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Topic: B.07. Synaptic Plasticity

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Title: Learning binds previously unrepresented inputs into functional synaptic clusters via spinogenesis

Authors: *N. HEDRICK[1,2,3], Z. LU[7], E. A. BUSHONG[4], S. SINGHI[5], P. NGUYEN[5], B. LIM[6], M. H. ELLISMAN[5], T. KOMIYAMA[5,2,3];
[2]Ctr. for Neural Circuits and Behavior, [3]Halıcıoğlu Data Sci. Inst., [4]Ctr. for Res. in Biol. Systems, [5]Neurobio. Section, Div. of Biol. Sci., [6]Biol. Sci., [1]Univ. of California San Diego, La Jolla, CA; [7]Neurobio. Section, Div. of Biol. Sci., UCSD, La Jolla, CA

Abstract: Plasticity in the motor cortex is thought to underlie the generation of novel skilled motor patterns. Such motor learning is accompanied by the formation of new dendritic spines in the motor cortex, but how new spines modify the existing circuit is unknown. Using longitudinal in vivo 2-photon imaging the glutamate reporter, iGluSNFR, we monitored the spatial and functional context of spinogenesis on apical dendrites of layer 2/3 excitatory neurons in the motor cortex during motor learning. We found that new spines primarily form in dendritic subdomains showing clustered, task-related spines during previous learning sessions. A subset of these spines show hallmarks of synaptic potentiation leading up to the spinogenesis event, suggesting that the strengthening of select inputs facilitates nearby new spine formation. By characterizing the glutamatergic signals at new spines, we found that they exhibit strong co-activity with nearby task-related spines, and thus functionally engage with their parent clusters. This co-activity is a behaviorally relevant signal, as it is selective to movement periods, and signals movements that are similar to the learned movement pattern. To understand the microstructural details of new spines and their environment, we performed correlated light- and electron microscopy (CLEM) on dendrites imaged in vivo. We found evidence supporting a filopodial search-and-capture model of spinogenesis, whereby hotspots of nascent filopodia sample the surrounding neuropil, forming synapses with pre-existing axonal boutons not associated with the imaged dendrite. Functional data suggests that the new successful connections are then filtered based on their co-activity with nearby spines; those exhibiting functional clustering survive, while those that do not are eliminated. These data support a model in which select dendritic sub-domains randomly sample potential new inputs while simultaneously ensuring that only those that are functionally related are incorporated into the dendritic repertoire. Finally, we used CLEM to reconstruct the axonal inputs to imaged dendrites, allowing us to ask whether the strong co-activity of new spines with nearby task-related spines could be explained by sharing of the same inputs. Surprisingly, most new spines do not share an axon with any other spine on the imaged region of dendrite, an observation supported by functional measurements showing that new spines do not show sufficiently high co-activity levels to be consistent with axon sharing. Thus, learning-related new spine formation utilizes local dendritic mechanisms to bind previously unrepresented inputs into the dendritic representation.

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Poster

P117. Structural Plasticity: Dendritic Spines

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Program #/Poster #: P117.03

Topic: B.07. Synaptic Plasticity

Support: HHMI Gilliam Fellowship for Advanced Study
NIH R01 NS06736
T32 MH11250

Title: CaMKII plays a non-enzymatic role in activity dependent new spine stabilization

Authors: *N. CLAIBORNE, ***N. CLAIBORNE**, *N. CLAIBORNE, K. ZITO;
Univ. of California Davis, Davis, CA

Abstract: The spontaneous outgrowth of new dendritic spines and their ability to stabilize and persist for hours and even years, are linked to learning and lasting memory. Most new spines are transient, and only a subset need to stabilize to support lasting memory. Our lab has shown that glutamatergic stimulation mimicking synaptic input onto a new spine enhances its survivorship. This enhanced stabilization requires the interaction between the GluN2B subunit of the NMDAR and CaMKII, implicating a role for CaMKII in this phenomenon. CaMKII performs both enzymatic and structural/scaffolding roles at dendritic spines, however, it is unknown which of these roles is needed to enhance survivorship. My goal is to determine which role CaMKII is playing in new spine stabilization.
I first used a genetic approach to determine if CaMKII kinase activity is required for activity-dependent new spine stabilization. I expressed GFP-tagged kinase-dead mutant, K42R, with DsRed Express in organotypic hippocampal slice cultures. New spines were identified using 2-photon time-lapse imaging and a single new spine per cell was stimulated with high-frequency glutamate uncaging. The survivorship of stimulated and unstimulated control new spines was monitored for 70 min. Glutamate uncaging onto a new spine on cells expressing kinase-dead CaMKII-K42R enhanced spine stability at 70 min compared to unstimulated new spines, suggesting CaMKII kinase activity is not required to enhance new spine stability.
To independently determine if new spine stabilization requires CaMKII kinase activity and avoid overexpression effects, I used a pharmacological approach. Hippocampal slices were incubated for 30 min staurosporine (Sta), a CaMKII kinase inhibitor that retains GluN2B-CaMKII binding, or vehicle control. Stimulated new spines incubated in either vehicle or Sta were more stable than unstimulated new spines. This supports that CaMKII kinase activity within the GluN2B-CaMKII complex is not required to enhance new spine stability following glutamatergic

stimulation.
CaMKII kinase activity plays a known role in synaptic plasticity, however my results suggest that it may be dispensable for new spine stability. To determine if CaMKII is required in a structural capacity for new spine stabilization, I knocked down CaMKII in hippocampal slice cultures. CaMKII knock-down blocked activity-dependent new spine survivorship. Rescuing CaMKII with GFP-tagged shRNA-resistant CaMKII restored activity-dependent new spine stabilization. Together, these results indicate that CaMKII is required to enhance new spine stability downstream of glutamatergic stimulation in a kinase-independent manner.

Disclosures: N. Claiborne: None. **K. Zito:** None.

Poster

P117. Structural Plasticity: Dendritic Spines

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P117.04

Topic: B.07. Synaptic Plasticity

Support: NIH Grant 1F31NS122477-01
NIH grant NS089578
DOD/USAMRAA grant: W81XWH1810388

Title: Par3-mediated regulation of dendritic spine plasticity in vivo

Authors: *M. VOGLEWEDE[1], E. N. OZSEN[2], M. SUN[1], H. ZHANG[1];
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Abstract: Dendritic spines are small, highly polarized protrusions on excitatory neurons serving as sites of postsynaptic input. Plasticity of dendritic spines is necessary for learning, while stable dendritic spines are thought to encode long-term memories. The polarized nature of dendritic spines suggests their plasticity and stability may be mediated by polarity proteins. The polarity protein Partitioning defective 3 (Par3) regulates mature dendritic spine formation in vitro, and several single nucleotide polymorphisms (SNPs) and copy number variation (CNV) of *Pard3*, which encodes Par3, are associated with intelligence, schizophrenia, and autism spectrum disorder (ASD). Together, these data implicate Par3 in mature dendritic spine stabilization, which may play a role in cognition and social interaction. However, the mechanisms of Par3 in dendritic spine plasticity and cognition *in vivo* remains completely unknown. We established a novel Par3 mouse model to conditionally knockout Par3 in postnatal forebrain pyramidal neurons. We found that loss of Par3 *in vivo* increases immature dendritic spines and increases overall dendritic spine density in the CA1 region of the hippocampus. In addition, loss of Par3 *in vivo* alters spatial learning in the Morris water maze, phosphorylation of cytoskeletal regulating

proteins, and levels of synaptic proteins. Together, our data suggest that Par3 plays a role in regulation of mature dendritic spine stability and cognitive functions.

Disclosures: M. Voglewede: None. **E.N. Ozsen:** None. **M. Sun:** None. **H. Zhang:** None.

Poster

P117. Structural Plasticity: Dendritic Spines

Location: Virtual Only

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Program #/Poster #: P117.05

Topic: B.07. Synaptic Plasticity

Title: Plastic changes to dendritic spines in the cerebellar and prefrontal cortices underlie the decline in motor coordination and working memory during successful aging

Authors: *M. VÁZQUEZ HERNÁNDEZ[1], N. I. MARTÍNEZ-TORRES[2,3], I. GONZALEZ-BURGOS[4];
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Abstract: Old age is the last stage of life and by taking a multidimensional view of aging, Neuroscientists have been able to characterize pathological or successful aging. Psychomotor and cognitive performance are recognized as two major domains of successful aging, with a loss of motor coordination and working memory deficits two of the most characteristic features of elderly people. Dendritic spines in both the cerebellar and prefrontal cortices diminish in aging, yet the plastic changes in dendritic spines have not been related to behavioral performance neither the changes in the cerebellar or prefrontal cortices. As such, motor coordination and visuospatial working memory (vsWM) was evaluated here in aged, 22-month-old rats, calculating the density of spines and the proportion of the different types of spines. These animals performed erratically and slowly in a motor coordination related paradigm, and the vsWM was resolved deficiently. Spine density was reduced in aged animals, and the proportional density of each of the spine types studied diminished in both the brain regions studied. The loss of dendritic spines and particularly, the changes in the proportional density of the different spine types could underlie, at least in part, the behavioral deficits observed during aging. This study shows the plastic changes in different dendritic spine types that might underlie the behavioral alterations in motor and cognitive abilities associated with aging. Further neurochemical and molecular studies will help better understand the functional significance of the plastic changes to dendritic spines in both successful and pathological aging.

Disclosures: M. Vázquez Hernández: None. **N.I. Martínez-Torres:** None. **I. Gonzalez-Burgos:** None.

Poster

P117. Structural Plasticity: Dendritic Spines

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Program #/Poster #: P117.06

Topic: B.07. Synaptic Plasticity

Support: RCMI BRAIN Core (NIMHD U54MD007579)
R15 MH101700 from the National Institutes of Health
RCMI 5U54MD007579-39
MD007579

Title: Plasticity of GluN2B at Ventral Hippocampal Synapses in the Infralimbic Cortex

Authors: *Y. CASTILLO-OCAMPO, A. HERNANDEZ, M. COLON, P. LOPEZ, J. T. PORTER;
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Abstract: Our laboratory recently found an increase of GluN1 subunit expression at ventral hippocampus(vHPC)-to-infralimbic cortex (IL) synapses after auditory fear conditioning (AFC) suggestingthat AFC increases the number of NMDA receptors at this synapse. In addition to changes in thenumber of NMDA receptors, synaptic transmission can also be altered by changes in theexpression of GluN2 subunits. GluN2B has larger single-channel conductance and slower decaykinetics which alters the basal state of synapses and affects the induction of synaptic plasticity.Since increases in GluN2B expression contributes to various forms of synaptic plasticity andmemory formation, we hypothesized that the expression of GluN2B would increase after AFC.Ventral hippocampal synapses were labeled with channelrhodopsin-EYFP in male and femalerats. On the first day, animals were exposed to AFC with paired tones and foot shocks orpseudoconditioning with unpaired tones and shocks (PSEUDO). The next day animals wereexposed to two tones to measure fear recall. Then animals were sacrificed and synaptosomeswere isolated from IL and GluN2B was labeled with fluorescent antibody. Then, we used FACSto identify double-labeled (EYFP+/GluN2B+) synaptosomes which represent presynaptic vHPCinputs and postsynaptic, membrane-bound GluN2B. We found increased GluN2B expression onvHPC-to-IL synapses in the males exposed to AFC compared to the PSEUDO group. In contrast,the female AFC and PSEUDO groups expressed similar levels of GluN2B on the vHPC-to-ILsynapses. In conclusion, the data suggest that AFC induces sex-dependent changes in theexpression of GluN2B subunit on vHPC-to-IL with increased GluN2B expression in the males.These changes in NMDA synaptic communication may contribute to altered fear expression afteran aversive event.

Disclosures: Y. Castillo-Ocampo: None. **A. Hernandez:** None. **M. Colon:** None. **P. Lopez:** None. **J.T. Porter:** None.

Poster

P117. Structural Plasticity: Dendritic Spines

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Program #/Poster #: P117.07

Topic: B.07. Synaptic Plasticity

Title: Structural changes in dendritic spines of Fos-expressing neuronal ensembles in the nucleus accumbens during novel context and cue-induced cocaine craving

Authors: *D. E. OLIVARES[1], F. RUBIO[1], E. M. HILAIRE[1], A. HENRY[1], L. KANE[1], C. MEJIAS-APONTE[2], B. T. HOPE[1];
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Abstract: Learned associations between discrete cues (or contexts) and drug effects are thought to be encoded by sparsely distributed patterns of neurons called “neuronal ensembles”. Here, rats undergoing cocaine self-administration were exposed to two different stimuli after a period of abstinence: novel context or cocaine-related cue. We determined whether neuronal activation induced specific changes in dendritic spines of activated neurons within the nucleus accumbens core (NAcc), an area where Fos-expressing ensembles have been associated with cue-induced cocaine seeking. We trained rats to self-administer cocaine (0.75 mg/kg/infusion, days 1-6; 0.375 mg/kg/infusion, days 7-12). We trained rats to lever press for cocaine infusions paired with 3.5-s light cue. After training, rats were injected in NAcc with a cocktail of AAVs encoding Cre recombinase and Cre-dependent GFP for sparse neuronal labeling. After 3 weeks of abstinence in their home cages, rats were exposed for 30 min to either a novel context (non-drug related) or cocaine-cue light associated with drug intake (relapse test). A third group of rats were not exposed to any stimulus. Rats were sacrificed at 60 min and brains were processed for Fos immunohistochemistry, endogenous GFP, and clearing tissue prior to confocal imaging. Morphological analysis of GFP-labeled spines on NAcc Fos-positive neurons and Fos-negative neurons were performed using 3D reconstruction of images. Dendritic tree complexity, spine density and spine head and neck measurements from the proximal (<50 um from soma) and distal (>70 um from soma) were performed using Neurolucida software. In proximal dendritic segments, Fos-positive neurons had significantly larger spine head diameter than Fos-negative neurons for both relapse and novel context groups. This can be explained by an increased proportion of mushroom-like spines. The distal dendritic segments responded differently. After novel context exposure, Fos-positive neurons had larger spine head diameters compared to Fos-negative neurons. In contrast, after cue-induced cocaine, Fos-negative neurons had larger spine

heads relative to Fos-positive neurons. This increase in the relapse group can be explained by larger spine head diameters in mushroom spines in the Fos-negative neurons as well as alterations in levels of the mushroom and filopodia/long thin spines. We hypothesize that distal dendritic segments of NAcc medium spiny neurons respond differently to novelty or cocaine-associated cues, and different mushroom spine adaptations in Fos-positive (compared to Fos-negative) neurons are induced during reactivation of a cocaine-cue learned-association.

Disclosures: D.E. Olivares: None. **F. Rubio:** None. **E.M. Hilaire:** None. **A. Henry:** None. **L. Kane:** None. **C. Mejias-Aponte:** None. **B.T. Hope:** None.

Poster

P117. Structural Plasticity: Dendritic Spines

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Program #/Poster #: P117.08

Topic: B.07. Synaptic Plasticity

Support: Fondo de Investigación en Salud (project number: FIS/IMSS/PROT/G15/1483).

Title: Ibotenic acid induced lesions impairs the modulation of dendritic spines plasticity in the prefrontal cortex and amygdala, phenomenon that underlies working memory and social behavior.

Authors: *N. MARTINEZ TORRES[1,2], M. N. VÁZQUEZ HERNÁNDEZ[3], F. MARTÍN AMAYA BARAJAS[4], M. FLORES-SOTO[5], I. GONZALEZ-BURGOS[6];
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Abstract: The lesion induced by Ibotenic acid (IA) emulate some of the symptoms associated with schizophrenia, such as impaired working memory that is predominantly organized by the medial prefrontal cortex (mPFC), or difficulties in social interactions that are mainly organized by the amygdala (AMG). The plastic capacity of dendritic spines in neurons of the mPFC and AMG is modulated by molecules that participate in the known deterioration of working memory, although the influence of these on the socialization of schizophrenic patients is unknown. Therefore, the effect of a neonatal IA induced lesion on social behavior and working memory was evaluated in adult rats, along with the changes in cytoarchitecture of dendritic spines and their protein content, specifically the postsynaptic density protein 95 (PSD-95), Synaptophysin (Syn), AMPA receptors, and brain-derived neurotrophic factor (BDNF). Both working memory and social behavior were impaired, and the density of the spines, as well as their PSD-95, Syn,

AMPA receptor and BDNF content was lower in IA lesioned animals. The proportional density of thin, mushroom, stubby and wide spines resulted in plastic changes that suggest the activation of compensatory processes in the face of the adverse effects of the lesion. In addition, the reduction in the levels of the modulating factors also suggests that the signaling pathways in which such factors are implicated would be altered in the brains of patients with schizophrenia. Accordingly, the experimental study of such signaling pathways is likely to aid the development of more effective pharmacological strategies for the treatment of schizophrenia.

Disclosures: N. Martinez Torres: None. **M.N. Vázquez Hernández:** None. **F. Martín Amaya Barajas:** None. **M. Flores-Soto:** None. **I. Gonzalez-Burgos:** None.

Poster

P117. Structural Plasticity: Dendritic Spines

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Program #/Poster #: P117.09

Topic: B.07. Synaptic Plasticity

Support: NIH Grant R01NS117585
NIH Grant R01NS123154

Title: Microglia eliminate immature synapses in autism associated Nav1.2-deficient mice during development

Authors: *J. WU[1], J. ZHANG[2], M. EATON[1], X. CHEN[2], Z. QUE[3], M. OLIVERO-ACOSTA[1], Y. YANG[1];
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Abstract: Microglia play a key role in regulating synaptic remodeling. Multiple studies have showed that synaptic abnormalities in neurological diseases such as autism and schizophrenia are mediated by microglia. Our recent studies suggested that severe deficiency in *Scn2a*, which encodes voltage-gated sodium channel Nav1.2, result in major behavioral deficits and increased neuronal excitability in adult mice. Microglia can sense the neuronal hyperactivity and be recruited to the synapses. Microglia is also known to prune immature spines. However, the roles of microglia in Nav1.2-deficinet mice are unclear. Here we report that the dendritic spine density in adulthood is decreased, accompanied by the changed microglia morphology in hippocampal CA1 region of Nav1.2 deficient mice. We also observe a strong accumulation of engulfed post-synapse (PSD95) within microglia lysosome, suggesting elevated pruning of microglia. As microglia synaptic pruning during development is critical for spine density in adulthood, we further studying the spine density in the early stage of development. We observe extensive immature spines of Nav1.2-deficient neurons. Interestingly, the post-synapse loss and increased

microglial synaptic engulfment occurred at P30, but not P4 or P10. Synapse elimination mediated by the activation of classical complement pathway has emerged as an important process during the development of brain. C1q and C3 localize on “weak” synapses as the “eat me” signal for microglia during the synaptic pruning. Thus, we studied the involvement of complement signals in our mouse model. We find that the density of C1q and C3 in Nav1.2-deficient neurons are significantly increased at P30 compared to these of the WT mice. Our results indicate that complement-mediated microglial synapse elimination might play a key role in the impaired neuronal plasticity, revealing a potential cellular mechanism underlying *Scn2a* deficiency-related neurodevelopmental disorders.

Disclosures: J. Wu: None. **J. Zhang:** None. **M. Eaton:** None. **X. Chen:** None. **Z. Que:** None. **M. Olivero-Acosta:** None. **Y. Yang:** None.

Poster

P117. Structural Plasticity: Dendritic Spines

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Program #/Poster #: P117.10

Topic: B.07. Synaptic Plasticity

Support: NIH R01 NS062736

Title: Molecular mechanisms of heterosynaptic spine shrinkage

Authors: *J. JANG, L. K. PARAJULI, K. ZITO;
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Abstract: Competitive interaction between activated synapses and adjacent inactive synapses is thought to play an important role in balancing excitability of dendritic segments, cells, and the neuronal network. In a previous study, we showed that activity-dependent synaptic potentiation in multiple spines drives spine shrinkage in inactive neighboring spines (Oh et al., 2015). This heterosynaptic spine shrinkage requires activation of calcineurin, IP_3Rs, and mGluRs. However, much remains unknown about the molecular mechanisms of heterosynaptic spine shrinkage. Here, we examined the cellular and molecular mechanisms that regulate heterosynaptic plasticity of spines on local dendritic segments using two-photon glutamate uncaging and time-lapse imaging in CA1 hippocampal pyramidal neurons. We found that blocking ion flux through NMDA receptors with L-689,560, a competitive inhibitor of the NMDAR glycine binding site, converted long-term spine growth of clustered stimulated spines to long-term spine shrinkage, as expected. Notably, shrinkage of neighboring unstimulated spines was converted from long-term to short-term, suggesting that the signaling for heterosynaptic shrinkage is only partially intact. In contrast, when glutamate binding to NMDARs was blocked by CPP, the result was complete inhibition of homosynaptic and heterosynaptic structural plasticity. Interestingly, protein

synthesis inhibition did not suppress enlargement of stimulated spines, but completely blocked short-term and long-term heterosynaptic spine shrinkage. Furthermore, when the stimulated spine cluster is small, the unstimulated spine showed transient or two-phase shrinkage; only with robust activation of multiple stimulated spines did consistent long-term spine shrinkage ensue. Our results support a model in which: 1) induction of long-term heterosynaptic spine shrinkage depends on Ca^{2+} influx, 2) for both short-term and long-term shrinkage, newly synthesized plasticity proteins are required, and 3) at least two different groups of signaling molecules (for short-term and long-term shrinkage) participate in heterosynaptic spine shrinkage.

Disclosures: J. Jang: None. **L.K. Parajuli:** None. **K. Zito:** None.

Poster

P117. Structural Plasticity: Dendritic Spines

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Program #/Poster #: P117.11

Topic: B.06. Synaptic Transmission

Support: Ministry of Research and University grant PRIN 2017HPTFFC

Title: Cartography of synaptic memory engrams using activity-dependent, genetically encoded reporters of synapse potentiation.

Authors: *A. JACOB[1], F. LATINI[1], A. FARAONE[1], M. DI CAPRIO[1], M. MAINARDI[2,1], A. CATTANEO[1];
[1]Lab. of Biol. "Bio@SNS", Scuola Normale Superiore, Pisa, Italy; [2]Neurosci. Inst., Natl. Res. Council, Pisa, Italy

Abstract: The encoding and retrieval of memories occur via the recruitment of specific neuronal ensembles, termed “cellular engrams” (Josselyn et al., 2020, Science). Engram formation correlates with synaptic plasticity processes, such as long-term potentiation (LTP), which has been proposed as a cellular mechanism supporting learning and memory (Takeuchi et. al, 2014, Phil. Trans.; Ryan et al., 2015, Science). However, neuronal activation during the encoding phase of memory recruits specific subsets of the synapse repertoire on the activated neurons. This paradigm shift of spatial scale, from whole-cell to synaptic memory engrams, prompted us to develop SynActive (Gobbo et al., 2017, Nat Comm), a genetic toolbox based on regulatory sequences from Arc mRNA and synaptic-targeting proteins sequences, that allows the expression of any protein of interest specifically at potentiated synapses. Here, we have exploited SynActive to control the expression of mVenus to label potentiated dendritic spines recruited by learning and memory processes. Temporal control over the expression of SynActive-mVenus was obtained via the TetON, doxycycline-dependent system, and AAV vectors were used to introduce the constructs into the mouse brain. Then, mice were exposed to contextual fear

conditioning (CFC) to obtain the synaptic map supporting the encoding of this specific form of associative learning. In addition to mapping the whole array of CFC-linked potentiated synapses, we combined SynActive with GFP Reconstitution Across Synaptic Partners (GRASP), to characterize the potentiated synapse pattern between structurally connected brain regions (Choi et al., 2018, Science). In SynActive-GRASP, one split-GFP fragment was expressed constitutively by presynaptic neurons, while the postsynaptic half was synthesized in a SynActive-dependent fashion at potentiated spines. After extensive validation of SynActive-GRASP in cultured neurons, we stereotaxically injected AAVs for the pre-and post-synaptic components in the CA3 and CA1 regions of the mouse hippocampus, respectively. Comprehensive analysis of reconstituted GFP at the CA3-CA1 potentiated synapses following CFC, revealed a spatially nonuniform and clustered pattern, providing an imaging basis to the observations that the same neuron could encode different memories in different synaptic subsets (Abdou et al., 2018, Science; Kastellakis et al., 2019, Cell Rep). The SynActive toolbox represents a new approach to map and characterize synaptic memory engrams in a variety of memory tasks, both in physiological and pathological (neurodevelopmental syndromes and neurodegeneration) - contexts.

Disclosures: A. Jacob: None. **F. Latini:** None. **A. Faraone:** None. **M. Di Caprio:** None. **M. Mainardi:** None. **A. Cattaneo:** None.

Poster

P137. Oscillations and Synchrony: LFP Studies II

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Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P137.01

Topic: B.09. Network interactions

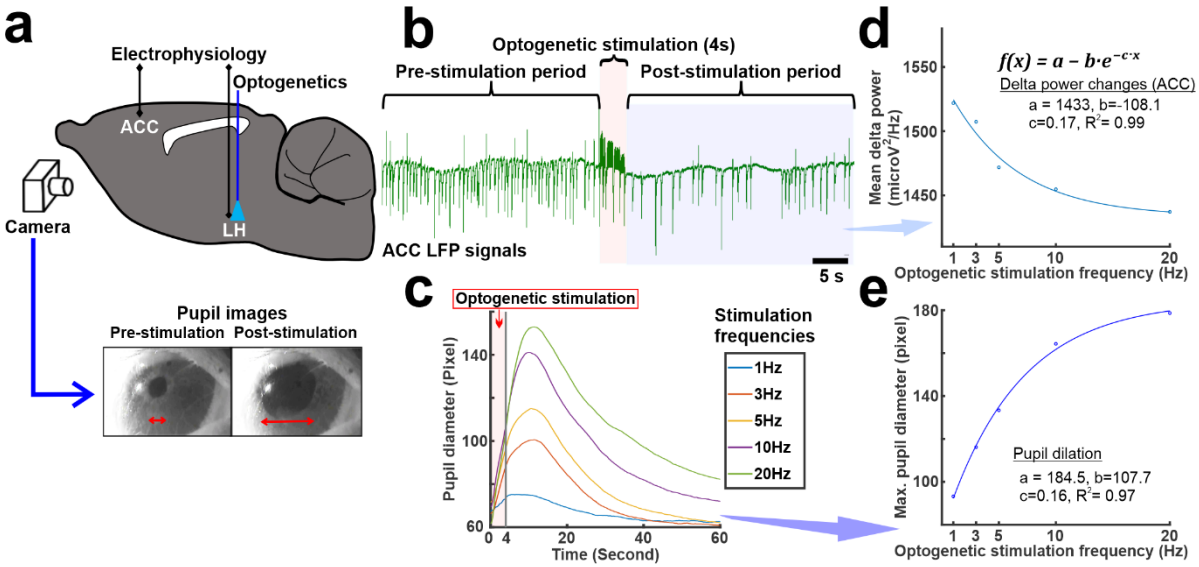
Support: NIH grant RF1NS113278
DFG grant Yu215/3-1
BMBF 01GQ1702

Title: Investigating the brain state and pupil dynamic modulation by frequency-dependent optogenetic regulation of the lateral hypothalamus

Authors: *K. TAKAHASHI[1,2], F. SOBCZAK[1,2], X. YU[3];
[1]Max Planck Inst. for Biol. Cybernetics, Tübingen, Germany; [2]GTC Neurosci., Univ. of Tübingen, Tübingen, Germany; [3]MGH/HST Martinos Ctr. for Biomed. Imaging and Harvard Med. Sch., Charlestown, MA

Abstract: ***Introduction***. Lateral hypothalamus (LH) plays a crucial role in regulating the brain state, which is reflected in pupil dynamics that serve as a specific arousal indicator. The activation of awake promoting neurons in the LH leads to brain state changes that depend on

stimulation frequencies. Animal behavior also varies depending on optogenetic stimulation frequencies of neurons in the LH. Although LH activation modulates both brain states and pupil dynamics, the influence of frequency-specific LH activation on brain state and pupil size regulations is under-examined. Since the LH also regulates the autonomic system, it is important to examine the relationship between LH-mediated brain state and pupil dynamic changes, which will help elucidate the dual regulatory roles played by the LH. Here, we activated CaMKII+ neurons in the LH of anesthetized rats and recorded pupil size changes and local field potentials (LFP) in the anterior cingulate cortex (ACC). Optogenetic activation of the LH induced stimulation frequency-dependent pupil dilation and cortical LFP power changes. These results suggest that the LH can modulate both brain state and pupil dynamics with similar frequency-dependent regulatory mechanisms. ***Methods***. We performed optogenetic stimulation on CaMKII+ neurons of the LH in five adult Sprague-Dawley rats under anesthesia and concurrently recorded LFP signals in the ACC and pupil size (**Fig. 1a**). All acquired data were processed with Matlab and DeepLabCut. ***Results***. LH activation evoked robust LFP changes in the ACC (**Fig. 1b**) and pupil dilation (**Fig. 1c**). In particular, we observed LFP delta (1-4Hz) power reduction in the ACC with an exponential decay feature (**Fig. 1d**) and logarithmic growth of the maximum pupil diameter (**Fig. 1e**) as a function of optogenetic stimulation frequencies from 1 to 20Hz at the LH. ***Conclusion***. This work shows that frequency-dependent optogenetic activation of the LH can induce pupil dilation and brain state modulation simultaneously in a similar manner, suggesting a distinctive up-regulatory role of the LH on both brain state and pupil dynamics.



Disclosures: K. Takahashi: None. **F. Sobczak:** None. **X. Yu:** None.

Poster

P137. Oscillations and Synchrony: LFP Studies II

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Program #/Poster #: P137.02

Topic: B.09. Network interactions

Support: NSF 1634445
NIH 5P50NS098573

Title: A closed-loop controller for inducing clusters in neuronal populations in real-time

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Abstract: Symptoms of Parkinson's disease and essential tremor are associated with pathological synchronization of neurons. Therapeutic effect of the conventional treatment, high frequency Deep Brain Stimulation (DBS), is therefore attributed to chaotic desynchronization of neurons with electrical pulses. A promising new technique, called Coordinated Reset (CR), has more persistent therapeutic effects than conventional DBS. In CR, multiple electrodes deliver periodic and phase-locked stimuli to different regions of the brain. Theoretical studies suggest that CR may increase synaptic plasticity by inducing clusters in neuronal populations. As an alternative approach, we tested a closed-loop controller called the Input of Maximal Instantaneous Efficiency (IMIE) that induces clusters with a uniform input signal. Here we present an experimental test of IMIE in hippocampal slices with fluorescence imaging to measure neuronal oscillations and electrical stimulation to control them. We modeled pathological synchronization in brain slices by bath perfusion of a cholinomimetic drug, carbachol. Then we divided the CA1 region of the hippocampus to 4 arbitrary regions of interest (ROIs). The phase of each ROI was estimated from the average fluorescence data analyzed with a new discrete Fourier transform algorithm called the Sliding Windowed Infinite Fourier Transform (SWIFT). We estimated the phase response curve (PRC) by stimulating the slice and measuring the change in the oscillation phase. Using the estimated phase in real-time and the PRC, IMIE determined the stimuli to disperse the oscillation phases. Stimulation was delivered when the population was most vulnerable to cluster dispersal. We observed a significant difference in the average phase difference between ROIs before and after modulation. Interestingly, the desynchronization persisted beyond the modulation for seconds. Our work is the first experimental demonstration of a real-time closed-loop control of clustering in neuronal populations in brain slices with a single electrode stimulus. We are extending our work to in vivo modulation of neuronal activities in real-time.

Disclosures: S. Faramarzi Ganj Abad: None. **T. Netoff:** None.

Poster

P137. Oscillations and Synchrony: LFP Studies II

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Program #/Poster #: P137.03

Topic: B.09. Network interactions

Support: NIH P20GM109025

Title: Impaired GABAergic signaling on glia alters hippocampal and cortical network activity and long term contextual memory.

Authors: *R. A. WIRT[1], A. M. LEISGANG[2], E. FLORES[1], A. PLATT[3], L. A. CREW[1], J. M. HYMAN[3], J. W. KINNEY[2];
[1]Neurosci., [2]Brain Hlth., [3]Psychology, Univ. of Nevada Las Vegas, Las Vegas, NV

Abstract: Contextual processing and recall are reliant on coordinated activity between the hippocampus (HC) and frontal areas of the brain and involve coordination of excitation and inhibition. While glia support neurons interacting in these areas, their role in systems communication is still unclear. To better understand how glial cells support normal brain function, we developed a mouse with significantly decreased GABAB receptors on glia (GABABflox X CX3CR1cre-er; GABAB). Subjects were implanted with a 64 channel device to record local field potentials (LFPs) and neuronal spiking activity in the HC and anterior cingulate cortex (ACC). A conditioned place preference (CPP) recall task was used to measure long-term memory (21 days after conditioning) and we found that GABAB mice remote recall was impaired, but their recent contextual memories appeared intact. Local field potentials revealed multiple possible sources of these long-term memory deficiencies. We found that GABA mice had significantly decreased gamma power (30 - 120 Hz) in the HC and impaired theta-gamma coupling in the ACC. We also found hypersynchrony between the ACC and HC in the theta (5 - 12 Hz) and delta (1-4 Hz) bands. Most notably, we found GABAB mice had significantly fewer hippocampal sharp-wave ripples (SWR), which are thought to underlie memory consolidation. The impairments found in these electrophysiological signals suggest glia, in particular GABAergic signaling on glia, may have a more active role in neural systems than previously thought. Any of these effects could potentially impair long-term memory formation, consolidation, or recall, but the combined effect could be catastrophic for memory function.

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Poster

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Program #/Poster #: P137.04

Topic: B.09. Network interactions

Title: Anterior cingulate cortical hippocampal desynchronization linked to cilia dysfunction in PCM1 knockout mice

Authors: *L. A. CREW[1], R. A. WIRT[1], C. CHANG[2], A. CHENG[2], E. FLORES[2], K. N. CALVIN[1], E. C. OH[2], J. M. HYMAN[3];
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Abstract: Network interactions between the hippocampus (HC) and anterior cingulate cortex (ACC) are critical for efficient memory performance and impaired ACC-HC interactions are thought to contribute to the cognitive deficits seen in schizophrenia (SZ). Although many genes have been identified to contribute to the development of SZ, underlying genetic processes, such as primary cilia functioning, have been less studied and may provide better insight into factors influencing cognitive symptoms. Cilia are rod-like organelles that extend from cell surfaces, providing motility and sensory functions and act as signaling devices during development. A known gene associated with primary cilia dysfunction, pericentriolar material 1 (PCM1), has shown to affect neurogenesis and leads to behavioral phenotypes, such as memory deficits, related to SZ. Recent research has revealed that localized cilia in the HC and ACC are altered in genetic knockouts of PCM1. We hypothesized that dysregulated cilia functioning leads to disruption in network oscillatory activity in the ACC and HC, thus, compromising cognitive performance. To test this hypothesis, we recorded from PCM1 knockout mice during spatial exploration and found substantial alterations in ACC and HC electrophysiology. Notably, spectral changes showed decreased theta power, irregular theta peak frequencies, and increased delta power in PCM1 -/- mice in both brain regions. This increase in delta power resulted in an overall decrease in the theta/delta ratio, a metric designed to show proportional instances of theta and delta that has been linked to human SZ cognitive symptomatology. We found significantly lower ACC-HC theta coherence in PCM1 -/- animals, which is surprisingly consistent with the desynchronization hypothesis of SZ. All together, we found remarkable changes in HC and ACC network activity in a knockout mouse associated with SZ that are coherent with a leading SZ mechanistic explanation, thus, these findings help to link genetic, behavioral, theoretical, and electrophysiological mechanisms that lead to cognitive deficits observed in SZ.

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Poster

P137. Oscillations and Synchrony: LFP Studies II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P137.05

Topic: B.09. Network interactions

Support: NIH Grant R01GM124023

Title: Anesthetic-induced spontaneous state transition synchrony and oscillatory activity correspondence are region-dependent

Authors: *E. B. BLACKWOOD[1], B. P. SHORTAL[1], A. PROEKT[2];
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Abstract: At fixed concentrations of general anesthesia, the brain spontaneously transitions through discrete states of arousal that are readily identifiable from electrophysiological recordings. These transitions are induced by subcortical signals, and under the prevailing model, they are synchronous throughout the cortex. However, asynchronous local transitions between sleep states have been observed in species such as cetaceans, reptiles, and possibly sleep-deprived rats. Given that sleep states can be local, it is worth investigating whether anesthetic state transitions can also happen asynchronously. If signals to transition first arrive at spatially restricted cortical regions and then spread to connected regions, we should observe transient asynchrony of states and their transitions. Furthermore, if transition synchrony between pairs of regions reflects functional connectivity, the distinct activity patterns of each electrode in pairs with more synchronized transitions should be related by a more consistent correspondence. We recorded local field potential (LFP) from rats held under 1−1.2% isoflurane in oxygen, either from the right primary visual (V1) and motor (M1) cortices (n=4) or from left and right V1 (n=4). We computed time-frequency spectrograms of LFP at each electrode and used non-negative matrix factorization (NMF) to project them onto compact bases of spectral components. We then identified discrete activity states and transition times for each electrode from the relative power over time of each of its NMF components. In both sets of recordings, the correspondence (normalized mutual information) of discrete states and the synchrony of state transitions were higher for within-region than cross-region electrode pairs, indicating that oscillatory activity and transition timing differed on a region-to-region basis. To measure activity correspondence without imposing discrete states, we also used canonical correlation analysis to compare series of NMF component scores; correspondence was again higher for within-region than across-region pairs. Mean transition synchrony was also higher for left/right V1 pairs than for M1/V1 pairs (bootstrap, $p < 0.001$), but this contrast was not reflected in activity correspondence differences. These results support a model of decentralized transition induction but suggest that regions with more synchronized transitions may not be more synchronized in general. Future research should record from a greater number of regions and test more specific models of how weak, nonspecific inter-region coupling can consistently give rise to transitions that appear global macroscopically.

Disclosures: E.B. Blackwood: None. **B.P. Shortal:** None. **A. Proekt:** None.

Poster

P137. Oscillations and Synchrony: LFP Studies II

Location: Virtual Only

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Program #/Poster #: P137.06

Topic: B.09. Network interactions

Title: Using multiple evoked and functional network metrics to measure and interpret cortical connectivity patterns in humans

Authors: *L. H. LEVINSON[1], S. SUN[2], C. J. PASCHALL[3], A. KO[4], J. A. HERRON[4], K. E. WEAVER[5], S. I. PERLMUTTER[6], J. G. OJEMANN[4];
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Abstract: Electrical stimulation of the cortex can be used to change patterns of neural activity, a technique that, in the future, may be used in neuro-rehabilitative therapies. For instance, electrical stimulation could induce and enhance neuroplasticity after a stroke event, altering cortical connectivity to bypass injured areas and promote functional recovery. To accurately understand how activity can be modulated, we need robust, interpretable metrics of functional and effective cortical connectivity. Many groups use cortico-cortical evoked potentials (CCEPs) to measure cortical responses to stimulation and use changes in CCEP magnitude as a proxy for changes in effective connectivity between sites; however, the neural bases of CCEPs are incompletely understood. This makes it difficult to interpret CCEPs, limiting the information we can use while developing protocols to induce or enhance connectivity changes. We may improve our ability to interpret CCEP results by combining CCEPs with metrics of activity with well-established ties to behaviors and neural outputs, such as resting state functional connectivity (rsFC) measures. This will allow us to functionally refine plasticity-inducing stimulation protocols. While previous studies have compared CCEP networks to various rsFC measures, results have been variable and, to our knowledge, no study has systematically compared CCEP results to rsFC networks built using a wide variety of computational techniques, including amplitude-amplitude (e.g. correlation, coherence), phase-phase (e.g. phase locking), phase-amplitude (e.g. phase-amplitude coupling), directed (e.g. Granger causality), and graph theoretical measures. Here, we perform such comparisons in stimulation and resting datasets from individual human subjects. By examining the relationships between CCEP and rsFC networks within and across subjects, we will build a robust, multifaceted metric of cortical connectivity that can be used to measure and interpret changes in brain activity after plasticity-inducing brain stimulation.

Disclosures: L.H. Levinson: None. **S. Sun:** None. **C.J. Paschall:** None. **A. Ko:** None. **J.A. Herron:** None. **K.E. Weaver:** None. **S.I. Perlmutter:** None. **J.G. Ojemann:** None.

Poster

P137. Oscillations and Synchrony: LFP Studies II

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Program #/Poster #: P137.07

Topic: B.09. Network interactions

Support: NASA TRISH Grant NNX16AO604A (RAD0103)
Eunice Kennedy Shriver National Institute of Child Health & Human Development NIH Award P50HD103555

Title: Low-dose radiation impairs neurogenesis in human forebrain organoids

Authors: *L. PARKITNY[1], S. OSENBERG[1], L. BRONK[2], A. LAGRONE[1], F. GUAN[3], M. W. GABER[5], D. GROSSHANS[4], M. MALETIC-SAVATIC[1];
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Abstract: Exposure to galactic cosmic ray (GCR) and solar particle event (SPE) radiation during interplanetary spaceflight may exert adverse effects on the adult human brain. Particularly susceptible to these effects are hippocampal neuroprogenitor cells which generate newborn neurons throughout life. Neurogenesis plays a critical role in brain plasticity and has been linked to cognition and mood control. Here, our main aim was to generate a relevant in vitro model to test the effects of GCR/SPE-like radiation on the human brain and, in particular, NPCs, neurons, and astroglia. We predicted that exposure to spaceflight-like conditions would result in anti-neurogenic effects such as increased cell apoptosis and reduced NPC proliferation. First, we developed a method for delivering GCR/SPE-mimicked doses of proton radiation to tissues contained in a culture plate, using the Geant4 Monte Carlo software package. Dose validation was performed via ETB3 film and Advanced Markus Chamber measurements. Then, to test the radiation effect on neurogenesis and relevant cell types, we generated human forebrain organoids which, in their early stages, are principally composed of NPCs but become increasingly heterogeneous as they mature. Organoids were exposed to a single or multi-dose course of low (4 keV/µm) or high (15 keV/µm) linear energy transfer (LET) proton radiation at a cumulative dose of 0.5 Gy to simulate long-term GCR/SPE-like exposure. We tested the radiation effects at multiple time points during the subsequent organoid maturation. We examined cell proliferation and apoptosis using a combination of imaging, immunohistology, flow cytometry, and transcriptomic methods. Radiation was associated with abnormal cell function; expression of activated caspases was elevated acutely and subacutely by as much as 40% of that in control organoids. On day 3 following radiation, the proliferation marker Ki-67 was reduced by 25%, and subsequent growth of irradiated organoids was impaired; this was associated with a reduced expression of Pax6, an NPC marker. Our data show that low-dose proton radiation induces cell apoptosis and impairs cell growth. These data are relevant to space flight as well as for clinical contexts where proton therapy is employed. Having developed this model, we will now test several newly developed pharmacological and non-pharmacological agents that might attenuate these anti-neurogenic effects of radiation.

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Poster

P141. Epileptogenesis and Seizure Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P141.01

Topic: B.10. Epilepsy

Support: F3213126557

Title: Cellular-resolution monitoring of pilocarpine-induced epileptogenesis in the mouse hippocampus

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Abstract: Epilepsy is a devastating neurological condition affecting 3 million adults in the U.S. alone. Most epileptic seizures initiate focally in the temporal lobe (TLE) and ~30% of these cases are unaffected by medical treatment. Refractory TLE usually starts with an initial seizure, followed by a seizure-free latent period, before chronic seizures appear. The unpredictable nature of the disease imposes a major challenge for clinicians: only some patients develop chronic seizures, which can emerge anytime in the days to years after the initial episode. Thus, the disease course is unknown during latency and immediate treatment cannot be prescribed. This work aims to identify activity changes in hippocampal neurons during latency that could predict the development and severity of chronic seizures in the pilocarpine model of TLE. Transgenic mice expressing the red genetically-encoded calcium indicator jRGECO1a in excitatory projection neurons were implanted with 1) an optical window providing access to the hippocampus and 2) EEG electrodes to monitor hippocampal and cortical seizures. We used two-photon microscopy to record from CA1 and DG neurons before and after the administration of pilocarpine hydrochloride, documented the severity of the resulting seizures according to the Racine scale, and detected the appearance of chronic stages using the EEG recordings. Analyses of calcium transients recorded from jRGECO1a-labeled neurons in DG and CA1 reveal a rapid transformation of the activity of CA1 neurons from a normal to pro-epileptic state upon pilocarpine treatment. Starting on the first day after the pilocarpine treatment, we identified a significant increase in CA1 firing rate, as well as a substantial increase in synchronous firing across CA1 pyramidal cells. These changes persisted throughout the latency period. Increased synchronicity of DG neuronal firing was identified several weeks after pilocarpine administration, suggesting a gradual failure of its gating properties. Preliminary analyses suggest these changes correspond to the severity of chronic seizures. These data begin to unveil the pro-

epileptic hippocampal transformation during the latent phase of pilocarpine-induced epilepsy. The development of significant synchronous firing events may serve as a biomarker for predicting the long-term outcome of chronic seizure development in pilocarpine-treated mice.

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Poster

P141. Epileptogenesis and Seizure Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P141.02

Topic: B.10. Epilepsy

Support: NIH/NINDS R01NS090645
NSF 1350654

Title: Investigating the effects of reduced GABA levels in gad1b null zebrafish on neural activity in the optic tectum

Authors: C. R. DUFFY[1], Y. LIU[2], Y. CHEN[3], P. MA[3], P. KNER[2], *J. D. LAUDERDALE[1]; [1]Cell. Biol., [2]Sch. of Electrical and Computer Engin., [3]Statistics, Univ. of Georgia, Athens, GA

Abstract: Reduced levels of gamma aminobutyric acid (GABA), the main inhibitory neurotransmitter, is connected to health issues such as epilepsy, stiff person syndrome and anxiety. GABA is formed by the decarboxylation of glutamate by glutamic acid decarboxylase (GAD). While most vertebrates have two *GAD* genes, known as *GAD1* and *GAD2*, zebrafish have three *gad* genes: *gad1a*, *gad1b*, and *gad2*. Using whole mount RNA *in situ* hybridization, we show that in the in the 1 dpf zebrafish spinal cord, the paralogs *gad1a* and *gad1b* are both expressed, though they are expressed in different neurons. In the 5 dpf zebrafish optic tectum, both are expressed, but exact localization needs to be investigated further. While the mice model of null *Gad1* causes neonatal fatality, we've created a *gad1b* null zebrafish which has reduced GABA levels, investigated using HPLC, and displays hyperactivity in the brain as measured by electrophysiology. This provides a unique model to study the effects reduced GABAergic signaling has on neural development. Using an *elavl3:Gcamp5G* expressing fish, and light sheet microscopy, we imaged activity in the 5 dpf zebrafish optic tectum of ten fish for each of the following three conditions: *gad1b*-null, wild type treated with pentylenetetrazol (PTZ-a drug that blocks the $GABA_A$ receptor), and wildtype with no treatment. For each larva, five recordings were made of two minutes in length. Patterns of activity were compared both within each fish and between fish. We found wild type activity is localized to the specific layers of the tectal neuropil, and PTZ treated fish activity is seen broadly throughout the whole tectum, as expected. *gad1b*-null larvae show an increase in the area of activity in the neuropil as well as in the cell

body layer. The increased activity in *gad1b*-null larvae is consistent with activity-dependent failure of surround inhibition of the tectal micro-circuits mediating visual input, and is providing insight into the impact of reduced GABA on neural development and function.

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Poster

P141. Epileptogenesis and Seizure Mechanisms

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Program #/Poster #: P141.03

Topic: B.10. Epilepsy

Support: the European Union's Horizon 2020 framework program for research and innovation under grant agreement No. 765549

Title: Diurnal and Multidien Rhythm of Interictal Epileptic Discharges and Epileptic Seizures in an Animal Model of Temporal Lobe Epilepsy Using Tetanus Toxin

Authors: *M. NAKATANI[1,2], A. GHESTEM[1], A. IVANOV[1], C. BERNARD[1];
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Abstract: Attempts to predict the likelihood of epileptic seizures based on the circadian and multidien rhythms have received increasing attention not only to improve the quality of life, but also to avoid the secondary injuries including sudden unexpected death in epilepsy. However, whether such rhythms exist in the beginning process of the acquisition of epileptogenicity in an epileptic animal model is still unexplored well. In this study, we aimed to investigate the occurrence patterns of interictal epileptic discharges (IEDs) and epileptic seizures using an animal model of temporal lobe epilepsy. We enrolled 10 male Sprague-Dawley rats and 4 rats were treated with tetanus toxin (TeNT) to develop spontaneous focal epileptic seizures (Jefferys et al., 1995, Nilsen et al., 2006). A 30 ng TeNT dissolved in phosphate buffered saline (Sigma-Aldrich, USA) with 0.2% bovine serum albumin (Sigma-Aldrich, USA) were stereotactically injected to the CA3 region of the hippocampus. An insulated tungsten electrode was inserted to the same coordinate to record the electroencephalogram with sampling rate 1000 Hz and DC setting for two months. We evaluated the occurrence pattern of the IEDs more than 1000 μV and epileptic seizures during continuous recording periods. The seizure onset pattern in each seizure was additionally identified based on the classifications previously proposed (Jirsa et al., 2014). Three out of 4 rats developed IEDs and spontaneous seizures. Regarding the IEDs, they were often observed during the daytime, which is a sleep period for rats. Although the total number of IEDs dramatically fluctuates over time, there seemed a diurnal variation in which the number of

IEDs go up and down every several days. Ictal seizures also tended to be more frequent during the daytime. Two to four onset patterns were observed in each rat, and the onset pattern changed from SN onset pattern, which is characterized by the sudden baseline shifts, to SNIC or SupH pattern over time. No clear regularity was detected in the onset pattern. Interestingly, epileptic seizures appeared more frequently one week before the period of frequent IEDs. As previously reported, both IEDs and seizures almost disappeared approximately 50 days after the drug administration. We conclude that some circadian rhythm of IEDs and epileptic seizures appear even at the beginning of epileptogenesis.

Disclosures: M. Nakatani: None. **A. Ghestem:** None. **A. Ivanov:** None. **C. Bernard:** None.

Poster

P141. Epileptogenesis and Seizure Mechanisms

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Program #/Poster #: P141.04

Topic: B.10. Epilepsy

Support: NSERC-RGPIN-2019-06378

Title: Roles of pannexin1 in epilepsy

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Abstract: The molecular determinants of excitation-inhibition imbalances promoting seizure generation in epilepsy patients are not fully understood. Experimental evidence suggests that Pannexin1 (*Panx1*), an ATP release channel, modulates excitability of the brain. Here, we use zebrafish larvae with Panx1a and Panx1b channels genetically knocked out or pharmacologically inhibited to evaluate the consequences of targeting Panx1 for antiepileptic drug therapies. Pentylenetetrazole was used to chemically induce seizures during *in vivo* recordings of local field potentials and for behavioral and molecular phenotyping. We find that loss-of-function *panx1a* gene mutations, or pharmacological blockade of both channels significantly reduces ictal-like events and seizure-related locomotion. Loss of *panx1a* also improves survival rates and transcriptome data demonstrate altered metabolic and cell signaling states. The pro- and anticonvulsant activities of both Panx1 channels affect ATP release and the purinergic receptor *P2rx7*. In summary, the present study highlights the importance of *panx1a*$^{-/-}$ as the primary source for pro-convulsant activities in the zebrafish. We present models suitable for targeting different neuronal circuitries of the zebrafish, which will allow dissecting *in vivo* the mechanisms of how the brain is guarded from excessive excitability, and which functions require *Panx1*.

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Poster

P141. Epileptogenesis and Seizure Mechanisms

Location: Virtual Only

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Program #/Poster #: P141.05

Topic: B.10. Epilepsy

Support: RO1 NS110767

Title: Layer- and pathway-specific disruption of perisomatic inhibition in the somatosensory cortex of the stargazer model of absence epilepsy

Authors: *S.-Y. SONG, A. MAHESHWARI, J. L. NOEBELS, X. JIANG;
Baylor Col. of Med., Houston, TX

Abstract: Absence seizure is a special type of epilepsy in children featured by generalized spike-wave (SW) electroencephalography (EEG) change. While earlier studies attribute the origin of SW discharges of absence seizure to the thalamus, there is now growing evidence that the cortex exerts a more prominent role in the generation and expression of SW activity, and their focal onset zone may reside in the somatosensory cortex (S1). To provide more insight into the minimal circuit elements necessary for the induction of SW discharge, we take advantage of a monogenic mouse model of absence epilepsy, stargazer, to examine what circuit defect may be specifically developed in the S1 as a result of mutations in *Cacng2*, a gene encoding an AMPA receptor trafficking protein stargazin (*stg*). We examined spontaneous excitatory postsynaptic currents (sEPSCs) of both excitatory and parvalbumin-expressing (PV^+) inhibitory neurons in S1 across the layers from homozygous stg (*stg*/*stg*) mutant mice and then compared to their wildtype littermates. In addition, simultaneous eight-cell patch recordings were performed on S1 neurons across the layers to examine cell-type specific connectivity change in *stg*/*stg* to further dissect the specific cell types and pathways impacted by the genetic lesions. The amplitude and frequency of sEPSCs recorded from PV^+ interneurons located in the S1 deep layers (Layer V and VI) were significantly decreased in *stg*/*stg* compared with wildtype littermates, while the PV^+ interneurons in Layer II/III and Layer IV showed no significant difference between genotypes. These effects appear to be cell type-specific as sEPSCs recorded from excitatory neurons across all layers in S1 did not change significantly in *stg*/*stg*. Consistent with this, our connectivity mapping indicates that connectivity from excitatory neurons to PV^+ interneurons in S1 deep layers was severely disrupted in *stg*/*stg*, while their connectivity in superficial layers and Layer IV remained intact. In addition, the connectivity from PV^+ interneurons to excitatory neurons and the connectivity between excitatory neurons across all layers showed no significant change in *stg*/*stg*, indicating the circuit changes in *stg*/*stg* is both layer-specific and pathway-specific.

This layer-specific and pathway-specific disruption of perisomatic inhibition as a result of single-gene mutations in absence seizure not only provide new insight on the circuit mechanisms underlying absence seizure, but also help to direct the field toward the development of innovative circuit-based interventions for absence epilepsy and its related comorbidities.

Disclosures: S. Song: None. **A. Maheshwari:** None. **J.L. Noebels:** None. **X. Jiang:** None.

Poster

P141. Epileptogenesis and Seizure Mechanisms

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Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P141.06

Topic: B.10. Epilepsy

Support: NIH grant R01 NS104368
CIHR MOP 136969

Title: Sustained network activation prevents post-traumatic epilepsy

Authors: *O. C. GONZALEZ[1], S. SOLTANI[3], S. CHAUVETTE[4], B. MARSH[2], I. V. TIMOFEEV[5], M. V. BAZHENOV[1];
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Abstract: Traumatic brain injury remains one of the most common factors leading to acquired epilepsy. Post-traumatic epilepsy (PTE) is characterized by a prolonged epileptogenic period which has been reported to extend up to 15 years after the occurrence of the brain trauma. Additionally, an age-dependent effect on the development of PTE has been reported. Recent *in vivo* studies demonstrated that older animals were more susceptible to the development of epilepsy following cortical undercut as compared to younger animals. The mechanism that gives rise to PTE remains to be fully understood but may involve misregulation of synaptic weights through homeostatic synaptic scaling. In response to brain trauma, there is a reduction of network activity within and near the traumatized as thus deafferented brain area. This triggers homeostatic up-regulation of synaptic strength in an attempt to recover normal levels of network activity. If trauma is severe, homeostatic scaling may overcompensate and increase synaptic weights such that the network is primed for transitions to seizure states. In this new study, we tested the hypothesis sustained network activity following cortical deafferentation could prevent homeostatic up-scaling and PTE. Using a detailed biophysical model of the neocortex, we found that a sustained depolarization was capable of preventing up-scaling of synaptic weights to a pathological state and thereby preventing occurrence of spontaneous recurrent seizures. In contrast, a sustained hyperpolarization of the traumatized network resulted in increased

homeostatic up-scaling, triggering a severe pathological state characterized by the occurrence of frequent spontaneous recurrent seizures. Furthermore, our analysis demonstrates that pathological increases in synaptic weights drives seizure generation by perturbing extracellular potassium concentration dynamics and initiating a positive feedback loop between extracellular potassium concentration and neuron firing rates. This feedback loop drives increased excitability and hypersynchrony eventually leading to spontaneous seizure onset. These findings from the computational model were validated with our *in vivo* experiments in mice where cortical undercut was followed by activation of DREADDs (hM3DGq or hM4DGi) to alter baseline network activity around the undercut area. hM3DGq prevented and hM4DGi exaggerated seizure activities. Together, these results provide evidence for the role of homeostatic synaptic scaling in the development of post-traumatic epilepsy and may provide new insights into novel treatments or preventative measures for trauma-induced epilepsy.

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Poster

P141. Epileptogenesis and Seizure Mechanisms

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Program #/Poster #: P141.07

Topic: B.10. Epilepsy

Support: NIH Grant NS096092

Title: Epileptiform activity triggered by KCC2 blockade in subicular circuits

Authors: M. ANSTOETZ, M. P. FISKE, *G. MACCAFERRI;
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Abstract: Depolarizing excitatory GABAergic signaling following the reduced expression of the KCC2 membrane transporter has been observed in subicular tissue of patients suffering from temporal lobe epilepsy. However, as many other alterations are inherently present in epileptic tissue, it is difficult to assess unequivocally whether impaired KCC2 function alone is epileptogenic or other epilepsy-related changes are also necessary. Here, we have addressed this question in a reduced model in vitro, and have studied the mechanistic involvement of a specific class of interneuron, i.e. the parvalbumin-expressing cell. Pharmacological blockade of KCC2 by the selective antagonist VU0463271 applied to isolated subicular slices from non-epileptic mice was sufficient to generate hypersynchronous population discharges in simultaneously patch-clamp recorded pyramidal neurons. Spontaneous events displayed the same pharmacological dependency on $GABA_A$ and AMPA postsynaptic receptors, as described for interictal events in human epileptic tissue in vitro. Calcium imaging of slices exposed to VU0463271 revealed

clusters of spontaneously co-active subicular neurons, whose estimated area was comparable to the axonal spread of single parvalbumin-expressing interneurons. Light-evoked activation of channelrhodopsin conditionally expressed in this class of cells was able to trigger interictal-like discharges, similar to VU0463271-induced spontaneous events. In contrast, light stimulation of archaerhodopsin expressed in the same neurons reduced the frequency of spontaneous interictal-like activity. Lastly, preliminary data suggest that this type of epileptiform synchronization may not be reversible upon washout of VU0463271.In summary, our results indicate that the loss of KCC2 function in subicular networks is sufficient to induce interictal-like events via altered GABAergic signaling originated by parvalbumin cells and that no other changes, commonly present in epileptic tissue, are necessary. In addition, this type of activity may trigger maladaptive changes preventing the return of the network to the original state. These results corroborate the idea that impaired KCC2 function may act as a key epileptogenic step in the progression of temporal lobe epilepsy.

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Poster

P264. Neuroprotective Mechanisms II

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Program #/Poster #: P264.01

Topic: C.03. Parkinson’s Disease

Support: Davis Phinney Foundation
TeCK Fund
Kent State University EHHS Seed Grant

Title: Optimization of the SMART dynamic bike to improve motor function in Parkinson's disease

Authors: *A. RIDGEL[1], P. GATES[2], R. MELCZAK[3];
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Abstract: High-cadence dynamic cycling improves functional motor scores in individuals with Parkinson's disease (PD). However, variability in responses among individuals suggests that an adaptive controller could help to decrease that variability. **PURPOSE:** To develop a Speed Manipulated Adaptive Rehabilitation Therapy (SMART) model for control of a custom designed motorized dynamic cycle and to evaluate the model accuracy for predicting motor function changes and physical effort. **METHODS:** Individuals with PD (N=16) completed three sessions of dynamic cycling under three different acceleration and velocity settings. Outcome measures included the pre and post UPDRS scores, cadence, and effort. Effort was defined as percent

positive power over the length of the session, indicating the ability to keep up with the motor. Demographic data included body mass index (BMI), age, gender, disease duration, medication dosage, most-affected side, and exercise history. Multiple linear regression models were built to examined how baseline UPDRS III scores, medication dosage, cycling cadence, entropy of cycling cadence (variability) and rider effort (power) affected the change in UPDRS III scores after a single bout of high-cadence cycling. **RESULTS:** This model showed that effort (p=0.04) and baseline UPDRS (p=0.01) significantly contributed to the variance in the change of UPDRS III after high-cadence dynamic cycling. Age (p=0.06) and daily medication intake (p=0.09) were not significant contributors. It is important to note because the testing and exercise were done with individuals 'on' medication. The second model showed that effort during high-cadence cycling explained 64% of the variance (p = 0.04) and that BMI (p=0.006) and age (p=0.04) were significant contributors. **CONCLUSION:** These findings suggest that symptom presentation, rider effort, age and BMI are important predictors of UPDRS III improvement after high-cadence dynamic cycling. These models will be further tested in a broad range of individuals with PD (different ages, BMI, baseline UPDRS, medication dosage) to optimize the SMART controller.

Disclosures: A. Ridgel: E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Patent holder. **P. Gates:** None. **R. Melczak:** None.

Poster

P264. Neuroprotective Mechanisms II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P264.02

Topic: C.03. Parkinson’s Disease

Support: State Committee of Sciences, Armenia

Title: Ratio of expiatory & inhibitory synaptic process in antinociceptive raphe magnus nucleus, activated by periaqueductal gray matter, on a model of Parkinson's disease when protection with hydrocortison

Authors: *N. BEHNAM DEHKORDI, Professor[1], V. H. SARKISIAN, Professor[2], M. HOVSEPYAN, Jr[3], M. V. POGHOSYAN, Sr[4], J. S. SARKISSIAN, Professor[5];
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Abstract: Ratio of excitatory and inhibitory synaptic processes in anti-nociceptive *raphe magnus* nucleus, activated by *periaqueductal gray matter*, on a model of Parkinson's disease when protection with hydrocortison

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Objective: In neurodegenerative diseases, most often Parkinson's disease (PD), antinociceptive centers are involved in neurodegeneration, which is accompanied by constant pain that cannot be relieved by drugs. We tried to find out the mechanisms of this phenomenon.**Methods:** In 3 series of experiments on 13 Albino rats, the impulse activity of 390 single neurons of Raphe Magnus (RMg) nucleus was analyzed during high-frequency stimulation (HFS) of Periaqueductal gray matter (PAG) in normal conditions, on a rotenone model of PD and when protection with Hydrocortisone. **Results: T**he number of neurons responding with depressor reactions decreased many times, with an increase in those responding with excitatory ones. This is the result of excitotoxicity, which is indicative of neurodegenerative damage to RMG neurons. Under conditions of exposure to Hydrocortisone, depressive reactions sharply increased with a significant increase in the number of neurons involved in them. In the PD model, in RMG neurons on the PAG HFS, a pronounced increase in the prestimulus frequency of activity preceded by post-stimulus depressor effects was shown. With hydrocortisone, a significant decrease in the prestimulus frequency preceded by excitatory post-stimulus manifestations of activity was obtained, which clearly indicates in favor of protection. The post-stimulus frequency of activity of RMG neurons, in the PD model, accompanied by excitatory effects much more exceeded the norm, also indicating tremendous excitotoxicity. Under conditions of protection, there was a powerful decrease in the post-stimulus frequency in the PD model accompanied by excitatory effects. **Conclusion:** The successful protective effect of Hydrocortisone is documented. The PD model provides evidence of the involvement of the important antinociceptive structure RMG in the neurodegenerative process. Reciprocal interaction with PAG, and the possibility of therapeutic provision with hydrocortisone is revealed. The PD model provides evidence of the involvement of the important antinociceptive structure RMG in the neurodegenerative process.
Keywords: *Parkinson's disease; antinociceptive Raphe magnus nucleus, Periaqueductal gray matter, Single spike activity, Hydrocortisone*.

Disclosures: N. Behnam Dehkordi: None. **V.H. Sarkisian:** None. **M. Hovsepyan:** None. **M.V. Poghosyan:** None. **J.S. Sarkissian:** None.

Poster

P264. Neuroprotective Mechanisms II

Location: Virtual Only

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Program #/Poster #: P264.03

Topic: C.03. Parkinson’s Disease

Support: NIH R01NS110694

Biomedical Research Awards for Interdisciplinary New Science (BRAINS)
NIH R21NS116260

Title: Huntington's disease: between HTT poly-glutamine expansion and alpha-synucleinopathy

Authors: D. YU[1], N. ZARATE[1], T. BROWN[1], R. MANSKY[1], A. WHITE[1], W. TSAI[1], C. NANCLARES[1], C. TOMAS-ZAPICO[2], Y. ZHANG[1], J. J. LUCAS[3], A. ARAQUE[1], M. CVETANOVIC[1], ***R. GOMEZ-PASTOR**[4];
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Abstract: Huntington's Disease (HD) is a neurodegenerative disorder caused by a CAG trinucleotide repeat expansion in the Htt gene for which no therapies are available. This mutation causes HTT protein misfolding and aggregation, preferentially affecting medium spiny neurons (MSNs) of the basal ganglia. Transcriptional perturbations in synaptic genes and neuroinflammation are key processes that precede MSN dysfunction and motor symptom onset. Understanding the interplay between these processes is crucial to develop effective therapeutic strategies to treat HD. We investigated whether protein kinase CK2α', a kinase upregulated in MSNs in HD and previously associated with Parkinson's disease (PD), participates in the regulation of neuroinflammation and synaptic function during HD progression. We used the heterozygous knock-in zQ175 HD mouse model that lacks one allele of CK2α' and performed cytokine proteome profiling, RNA-seq, electrophysiological recordings, and behavioral analyses. We showed that CK2α' haploinsufficiency in zQ175 mice ameliorated neuroinflammation, HTT aggregation, excitatory synaptic transmission deficits, and motor dysfunction. Our RNA-seq analyses also revealed a connection between α-syn, a protein associated with PD, and the transcriptional perturbations mediated by CK2α' in HD. We found that CK2α' increased α-syn serine 129 phosphorylation (pS129-α-syn), a post-translational modification linked to α-synucleinopathy, and its accumulation in the nucleus of MSNs in zQ175 mice and in patients with HD. Our data demonstrated that CK2α' negatively contributes to HD by promoting striatal synucleinopathy and neuroinflammation.

Disclosures: D. Yu: None. **N. Zarate:** None. **T. Brown:** None. **R. Mansky:** None. **A. White:** None. **W. Tsai:** None. **C. Nanclares:** None. **C. Tomas-Zapico:** None. **Y. Zhang:** None. **J.J. Lucas:** None. **A. Araque:** None. **M. Cvetanovic:** None. **R. Gomez-Pastor:** None.

Poster

P264. Neuroprotective Mechanisms II

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Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P264.04

Topic: C.03. Parkinson's Disease

Support: NIH Grant R01NS115809

Title: Neuroprotective roles of Estrogen and Cytisine in a 6-OHDA Mouse Model of Parkinson's Disease

Authors: *S. ZARATE, G. PANDEY, A. GONZALEZ, E. ONYEWUENYI, J. ABSHIRE, K. WILLIAMS, S. CHILUKURI, M. HOOK, R. SRINIVASAN;
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Abstract: Parkinson's disease (PD) is a neurodegenerative disorder caused by the loss of midbrain dopaminergic (DA) neurons with an incidence rate of 2:1 in men versus women. PD will affect over 12 million people by 2040 underscoring a need for neuroprotective treatments. Unfortunately, current treatments for PD are symptomatic, cause severe side effects, and lose efficacy over time. Previously, we showed that the smoking cessation drug, cytisine exerts neuroprotective effects only in female mice by attenuating the endoplasmic reticulum (ER) stress response pathway in combination with17-β-estradiol. Here, we further characterize the role of 17-β-estradiol in female parkinsonian mice. We induced parkinsonism in four groups of mice by unilateral injection of 6-OHDA in the dorsolateral striatum. The four groups included: 1) ovariectomized (OVX) female mice treated with alternate day i.p. injections of 0.2 mg/kg cytisine, 2) sham-operated female mice treated with alternate day injections of cytisine, 3) sham-operated female mice treated with alternate day injections of saline, and 4) OVX female mice treated with alternate day i.p. injections of saline. PD related behaviors in all four groups were assessed using apomorphine rotations, pole test, and gait analysis with the Digigait system. We found that cytisine attenuation of apomorphine rotations was lost in OVX cytisine treated females. During pole test, we found no change in time to descend between all four groups after 6-OHDA lesion. Interestingly, all mice displayed spontaneous rotational behavior during pole descent with saline treated mice showing 2-fold more rotations than either OVX or intact cytisine treated mice. Gait analysis during a forced walking test showed that the left hindpaw was turned outward in sham operated saline treated female mice but not in either OVX or intact cytisine treated mice. These data suggest either a potential role for local brain derived 17-β-estradiol in the neuroprotective effect of cytisine or mechanisms related to sex specific differences in cytisine pharmacology.

Disclosures: S. Zarate: None. **G. Pandey:** None. **A. Gonzalez:** None. **E. Onyewuenyi:** None. **J. Abshire:** None. **K. Williams:** None. **S. Chilukuri:** None. **M. Hook:** None. **R. Srinivasan:** None.

Poster

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Program #/Poster #: P264.05

Topic: C.03. Parkinson's Disease

Support: NIH Grant R21AG068607
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NIH Grant 1R01DC018516

Title: Vesicular glutamate transporter expression mediates mitochondrial oxidative stress related to dopamine neurodegeneration

Authors: *S. A. BUCK[1], T. KUNKHYEN[2], S. A. RUBIN[1], C. E. J. CHEETHAM[2], Z. FREYBERG[3];
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Abstract: Parkinson's disease (PD) is characterized by dopamine (DA) neurodegeneration, but some DA neurons are relatively protected compared to others. Recent evidence suggests DA neuron expression of vesicular glutamate transporter 2 (VGLUT2) plays a role in mediating this selective vulnerability. To date, however, the mechanism behind this protection conferred by VGLUT2 is still unknown. Considering the role of mitochondrial dysfunction in PD etiology, we hypothesized VGLUT2 expression may improve mitochondrial resilience to oxidative stress observed in DA neurons in animal models of PD and human PD. To test this hypothesis, we employed the genetically tractable *Drosophila melanogaster* model. We employed a tyrosine hydroxylase (TH)-Gal4 driver to induce DA neuron-specific RNA interference (RNAi)-mediated knockdown of *Drosophila* VGLUT (dVGLUT), which caused a 44.0%-63.6% decrease in TH^+/$dVGLUT^+$ neuron density, a 51.3%-69.0% decrease in percent of TH^+ neurons that are $dVGLUT^+$, and a 36.1%-51.4% decrease in average number of dVGLUT mRNA grains in TH^+ neurons compared to control genotypes. To test if dVGLUT's neuroprotective properties are linked to its ability to lower potentially neurotoxic reactive oxygen species (ROS), we analyzed the impact of DA neuron dVGLUT RNAi on mitochondrial oxidative stress using flies that express MitoTimer, a mitochondrially-localized DsRed mutant that shifts fluorescence from green to red when oxidized, in DA neurons. Using paraquat (10 mM, 5 days), a pesticide model of PD that induces oxidative stress in DA neurons, we performed multiphoton imaging of brains from exposed MitoTimer flies and measured mitochondrial oxidative stress in distinct DAergic projection areas. We found an intriguing sex difference, as control females had lower mitochondrial ROS in all DAergic regions analyzed compared to control males ($P<0.05$). Conversely, DA neuron dVGLUT RNAi increased ROS in females ($P<0.01$) but not males, suggesting dVGLUT plays a role in mediating sex differences in mitochondrial oxidative stress and may provide a new mechanism for greater protection from DA neurodegeneration in females versus males. Finally, paraquat increased mitochondrial ROS levels in the DAergic fan-shaped body of all genotypes ($P<0.05$) with the greatest increase observed in the male dVGLUT RNAi group ($P<0.05$). Together, these findings uncover a novel mechanism of dVGLUT-mediated

protection, highlighting its role in mediating mitochondrial oxidative stress in DA neurons in the paraquat model of PD.

Disclosures: S.A. Buck: None. **T. Kunkhyen:** None. **S.A. Rubin:** None. **C.E.J. Cheetham:** None. **Z. Freyberg:** None.

Poster

P264. Neuroprotective Mechanisms II

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Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P264.06

Topic: C.03. Parkinson's Disease

Support: Dept of Defense Grant W81XWH-19-0757

Title: Exercise-related motor recovery in the rat 6-OHDA hemi-parkinson's model without increased striatal dopamine biosynthesis: a glimpse into exercise impact in human Parkinson's disease?

Authors: E. A. KASANGA[1], M. SHIFFLET[2], ***M. SALVATORE**[2];
[1]Inst. of Healthy Aging, Univ. of North Texas Hlth. Sci. Ctr., Fort Worth, TX; [2]Univ. of North Texas Hlth. Sci. Ctr., Ft Worth, TX

Abstract: Exercise can mitigate the progression of motor impairment in Parkinson's disease (PD), which strongly infers that there is a critical neurobiological response to exercise that diminishes the rate of nigrostriatal dopamine (DA) neuron loss or function. Identifying the mechanisms of this exercise response in rodent PD models arguably points to the most germane pharmacological or gene-based approach to treat motor impairment in PD. However, for optimal translation of these mechanisms to human PD, the exercise modalities used in rodents must be practicable for the human PD patient. Moreover, the identification of these exercise mechanisms, once the practical considerations of exercise are engaged in a rodent PD model, may necessitate investigation in areas outside of striatum, given more than 80% loss of DA is already present at onset of motor impairment. To begin to address these gaps, we trained four to five month old Sprague-Dawley rats to exercise on a treadmill prior to induction of nigrostriatal lesion. Seven days after lesion, motor impairment was established by a minimum of 25% reduction in the use of the forepaw associated with lesion. Rats were then segregated into the non-exercise (NE) and exercise (EX) groups with similar impairment. Treadmill exercise was initiated for three times per week, 40 minutes each session, for three weeks, with motor assessments done the day following the last exercise session each week. The treadmill exercise regimen produced highly significant recovery (~50%) in forepaw use with nine exercise sessions over the three-week period of exercise. Notably, this motor recovery was not associated with any recovery of striatal tyrosine hydroxylase (TH) protein or DA tissue content loss. Moreover, there was a significant

decrease of TH protein and DA tissue content in the striatum, contralateral to lesion, in the EX group. Taken together, our results show that an exercise regimen in an established rat PD model that matches the intensity, frequency, and duration used in exercise programs in PD patients does induce recovery of motor function, demonstrably lost prior to exercise initiation. Moreover, the results also indicate that exercise-mediated motor recovery may not require any increase in striatal dopamine biosynthesis.

Disclosures: E.A. Kasanga: None. **M. Shifflet:** None. **M. Salvatore:** None.

Poster

P264. Neuroprotective Mechanisms II

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Program #/Poster #: P264.07

Topic: C.03. Parkinson's Disease

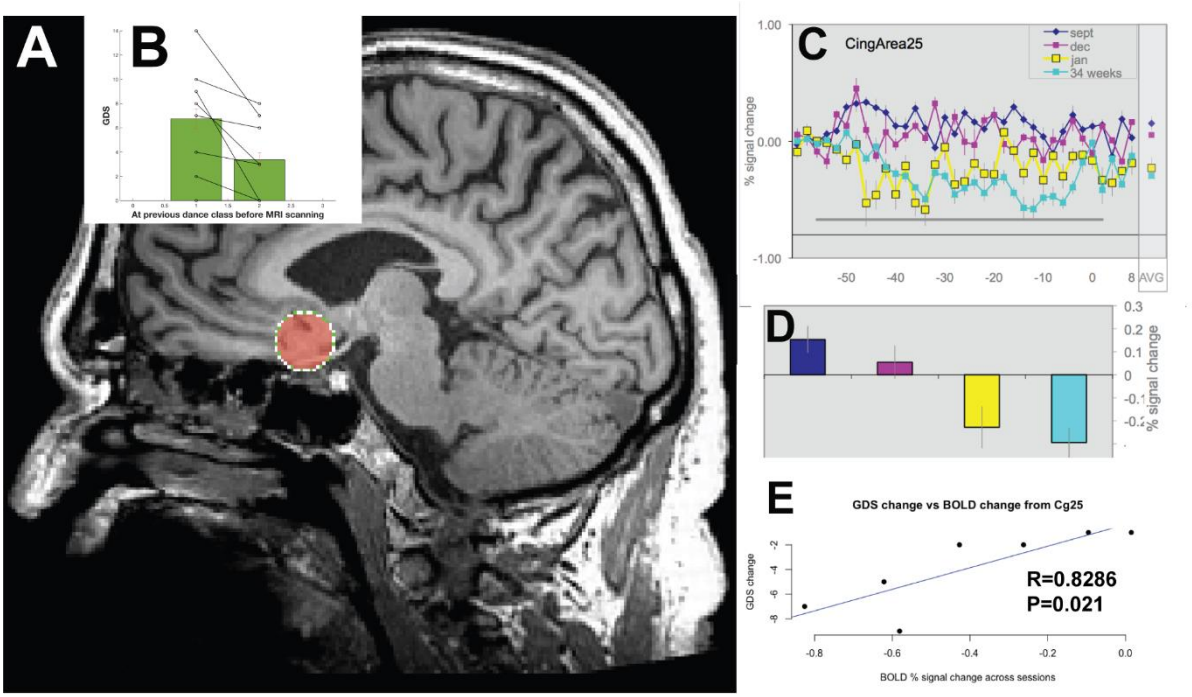
Support: Parkinson's Society Canada
NSERC Discovery Grant

Title: Non-invasive neurosurgery: Dance participation modulates brain signal in subgenual cingulate gyrus (Cg25) while decreasing depressive symptoms for people with Parkinson's Disease over 8-months

Authors: R. E. BARNSTAPLE[1], K. BEARSS[1], A. M. PAGLIUSO[1], G. R. LEVKOV[1], R. J. BAR[1], P. M. DI NOTA[1], *J. DESOUZA[2];
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Abstract: As global incidence of neurodegeneration is on the rise, recent studies have investigated the potential for dance to foster adaptive plasticity in these conditions. Results have shown that dance participation leads to brain-based changes in expert dancers (Bar & DeSouza 2016) and older adults, and can modify disease progression for people with Parkinson's Disease (PwPD; Bearss & DeSouza, 2021). While there are indications of improvements in motor and psychological symptoms as a result of participation in dance, the neural mechanisms underlying these are currently unclear. Our current study follows participants in a Dance for PD classes over 8 months and correlates outcomes on the Geriatric Depression Scale (GDS) with a subset of 8 participants' functional imaging changes. Using an anatomical seed in subgenual cingulate gyrus (Cg25, a node of the depression network) and surrounding area, we probed BOLD signals while subjects visualized learned choreography accompanied by the music they danced to in the studio. GDS scores of PwPD were collected in the dance studio before and after class and showed a significant decrease ($p<0.05$, paired t-test), indicating a reduction in depression. BOLD signals were averaged during visualization during 4 scanning sessions (September, December, January

and April) for all 10 PwPD (28-scans in total; fig C). We ran a correlation on GDS and BOLD signal data pre/post for all subjects with two or more scanning sessions and found a significant correlation between reduced BOLD signal from Cg25 and reduced GDS scores across our dataset (Pearson Correlation of 0.83 of $t(5)=-3.3099$, $p<0.025$, Fig E). Cg25 is a traditional target for deep brain stimulation in treatment-resistant depression; changes in signal strength from this area correlated with improvements in depression scores suggest dance-based learning accompanied by music may be facilitating network reconfiguration. This is the first study to identify a neural substrate for changes associated with dance and contributes to our understanding of mechanisms involved in adaptive plasticity.



Disclosures: R.E. Barnstaple: None. **K. Bearss:** None. **A.M. Pagliuso:** None. **G.R. Levkov:** None. **R.J. Bar:** None. **P.M. Di Nota:** None. **J. DeSouza:** None.

Poster

P275. Huntington's Disease: Preclinical Interventions

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P275.01

Topic: C.04. Movement Disorders other than Parkinson's Disease

Support: Drexel Coulter foundation award

Title: Development of a novel neuroprotective agent to treat Huntington’s disease

Authors: *S. KORTAGERE, S. DAS;
Drexel Univ. Col. of Med., Philadelphia, PA

Abstract: Huntington's disease (HD) is a fatal neurodegenerative genetic disorder caused by increased CAG repeats in exon 1 of the Huntingtin gene. Huntington's Disease is characterized by uncoordinated movement (Chorea), psychomotor impairment, cognitive impairment and neuropsychiatric abnormalities in addition to other somatic symptoms. The CAG repeat generates a mutant form of huntingtin protein (HTT) which is toxic to brain cells. The HTT protein can interfere with a variety of cellular functions including packaging and release of neurotransmitters and mitochondrial functions and protein degradation. This HTT protein activity also causes aberrant changes to glutamatergic neurotransmission causing an excess glutamate release in various regions of the brain leading to excitotoxicity and cell death. In addition, the major glutamate transporter also called excitatory amino acid transporter 2 (EAAT2) present predominantly on astrocytes is downregulated in several neurodegenerative diseases including HD causing a significant accumulation of synaptic glutamate. We have recently developed a novel small molecule activator of EAAT2 and characterized its ability to increase the uptake of glutamate in MDCK-hEAAT2 TRANSFLEX plate assay. Results from the assay showed that GTS467 increased glutamate uptake with an EC50 value of 35.3nM. Further, GTS467 was tested for in vivo bioavailability with three modes of administration (i.p, i.v and p.o). The oral bioavailability was determined to be 80.34% and the compound has a half life of 1.81h in the brain. Based on these good physicochemical properties, GTS467 was tested in a transgenic mouse model of HD developed in R6/2 mice expressing the human HTT 128Q repeats for locomotor and cognitive effects. Results from the study showed that the HTT mice had significant deficits in locomotor and cognitive phenotypes measured using open field task and novel object recognition tasks and treatment with GTS467 dose dependently improved the performance of mice in these tasks. Current studies are aimed at understanding the mechanism by which GTS467 confers protection in HD.

Disclosures: S. Kortagere: None. **S. Das:** None.

Poster

P275. Huntington's Disease: Preclinical Interventions

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P275.02

Topic: C.04. Movement Disorders other than Parkinson's Disease

Title: Evaluation of a small molecule splicing modulator in a humanized HTT intron 49 mouse

Authors: *D. MAGNANI[1], M. VISSER[2], D. MOTA[1], C. DAVIE[1], M. HERVA MOYANO[1], P. MITCHELL[1], D. FISCHER[1], G. MCALLISTER[3], A. BHATTACHARYYA[4], I. MUNOZ-SANJUAN[3], R. CHEN,[3];

[1]Charles River Labs., Saffron Walden, United Kingdom; [2]Charles River Labs., Leiden, United Kingdom; [3]CHDI, Los Angeles, CA; [4]3PTC Therapeut., South Plainfield, NJ

Abstract: Lowering the expression of the pathogenic mutant huntingtin (mHTT) mRNA and protein is a leading therapeutic approach for the treatment of Huntington's Disease (HD). Clinical trials involving the use of HTT -lowering antisense oligonucleotides (ASO) have shown promising pharmacodynamic potential data although they have halted poor pharmacokinetic and safety margins. Notably, ASOs are normally delivered intrathecally or directly into the brain via invasive methods. Recently, small molecules directly targeting HTT mRNA, such as Branaplam (LMI070), have been discovered. These small molecules provide the advantage of greater distribution and non-invasive delivery methods. Branaplam targets splicing of the exon junction 49-50 of HTT pre-mRNA, promoting retention of a pseudo-exon within intron 49 which contains a premature stop codon, subsequently resulting in nonsense mediated decay (NMD)-dependent mRNA and consequential protein lowering. However, Branaplam and other HTT mRNA targeting compounds do not modulate endogenous rodent Htt splicing due to differences in the target sequence, precluding pre-clinical efficacy and tolerability studies in standard rodent models. To overcome this, CHDI has generated and CRL is characterizing a new mouse model i49 Exon Q175, containing the humanized exon 49-50 junction on an existing Q175 knock-in model. The Q175 knock-in mouse contains human exon 1 mHTT with an expanded CAG repeat (~179 repeats) within the native mouse huntingtin gene. Thus, this animal model is representative of HD genetics and consistently displays associated motor symptoms and behavioural deficits, accompanied by brain atrophy and metabolite changes. This makes it a suitable model for preclinical safety and efficacy testing. In the novel i49 Exon Q175 mouse model the mouse intron 49 genomic region has been replaced with the human one, including the Branaplam target sequence, via Crispr-Cas9. In addition, the wt mouse allele (Q7) has been targeted to contain the human sequence in intron 49, which enables us to also affect expression of the non-expanded allele in mice. Here, we validate these models and demonstrate significant human intron 49 retention and disruption of the endogenous 49-50 junction upon acute treatment with LMI070, in both periphery and CNS. Moreover, we investigate mutant and wt Htt protein reduction following 21 days sub-chronic treatment with LMI070. Further work will be aimed at studying compound mediated effects on mutant and wt Htt protein isoforms levels, and on behavioral and brain phenotypes.

Disclosures: D. Magnani: None. **M. Visser:** None. **D. Mota:** None. **C. Davie:** None. **M. Herva Moyano:** None. **P. Mitchell:** None. **D. Fischer:** None. **G. McAllister:** None. **A. Bhattacharyya:** None. **I. Munoz-Sanjuan:** None. **R. Chen,:** None.

Poster

P275. Huntington's Disease: Preclinical Interventions

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P275.03

Topic: C.04. Movement Disorders other than Parkinson's Disease

Title: Profiling of a novel orally active and brain penetrant splicing modulator small molecule demonstrates CNS and systemic mHTT lowering in BACHD mice

Authors: D. MAGNANI[1], K. MALAGU[1], S. CLIFTON[1], H. ATTON[1], D. MOTA[1], C. DAVIE[1], M. STEBBEDS[1], A. BARNARD[1], I. MANCINI[2], M. VISSER[2], D. TODD[1], ***M. HERVA MOYANO**[1], P. MITCHELL[1], M. CHAMBERS[1], G. MCALLISTER[3], L. LIU[3], V. KHETARPAL[3], D. MACDONALD[3], C. DOMINGUEZ[3], I. MUNOZ-SANJUAN[3];
[1]Charles River, Saffron Walden, United Kingdom; [2]Charles River, Leiden, Netherlands; [3]CHDI Mgmt. Inc./CHDI Fndn., Los Angeles, CA

Abstract: Several drug modalities aimed at lowering levels of mutant HTT (mHTT) are currently in development for the treatment of Huntington's Disease (HD). Importantly, mHTT mRNA is increasingly being considered an important therapeutic target, and HTT lowering pharmacodynamic data have been recently shown in clinical trials with the use of antisense oligonucleotide (ASO) targeting HTT mRNA. However, those trials have recently been stopped for insufficient safety over therapeutic margin.
Notably, ASOs are delivered intrathecally into the spinal cord or directly into the striatum due to their poor distribution. Small molecules aimed at the same target will provide the potential advantages of greater distribution, non-invasive delivery methods and higher patient accessibility. In this context, a growing number of small molecules targeting RNA are being identified and this class of molecules are progressing into the clinic. One such molecule is Branaplam (LMI070) targeting directly HTT mRNA and is now moving forward to clinical trials for HD. Branaplam was originally designed to target splicing of SMN2 exon 7, but also targets splicing of the exon junction 49-50 of HTT pre-mRNA, promoting retention of a pseudo-exon within intron 49b which contains a premature stop codon, subsequently resulting in nonsense-mediated decay (NMD)-dependent mRNA and consequential protein lowering.
CHDI, with Charles River Early Discovery, has discovered a novel compound (Compound Y) targeting the same mechanism. Herein, we describe the *in vitro* and *in vivo* profile of Compound Y. Potency was assessed in an HD-relevant human stem cell model and demonstrated concentration-dependent splicing modulation and HTT protein-lowering. Moreover, Compound Y exhibited favourable ADME and PK properties, as well as significant HTT mRNA splicing and protein-lowering *in vivo* in brain and peripheral tissues of an HD rodent model.

Disclosures: D. Magnani: None. **K. Malagu:** None. **S. Clifton:** None. **H. Atton:** None. **D. Mota:** None. **C. Davie:** None. **M. Stebbeds:** None. **A. Barnard:** None. **I. Mancini:** None. **M. Visser:** None. **D. Todd:** None. **M. Herva Moyano:** None. **P. Mitchell:** None. **M. Chambers:** None. **G. McAllister:** None. **L. Liu:** None. **V. Khetarpal:** None. **D. Macdonald:** None. **C. Dominguez:** None. **I. Munoz-Sanjuan:** None.

Poster

P275. Huntington's Disease: Preclinical Interventions

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Program #/Poster #: P275.04

Topic: C.04. Movement Disorders other than Parkinson's Disease

Support: NIH RO1 grants NS113612
Hereditary Disease Foundation

Title: Fan1 genetic reduction in a murine Huntingtin knockin mouse model modifies CAG repeat instability and striatal neuropathology

Authors: *L. DENG[1,2], J. RICHMAN[1], N. WANG[1], K. T. TAMAI[2], P. LANGFELDER[1], F. GAO[1,2], G. COPPOLA[1,2], C. S. COLWELL[2], X. W. YANG[1,2];
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Abstract: In Huntington's Disease (HD), the mutant Huntingtin (mHtt) CAG repeat length is inversely correlated with the age of onset of motor symptoms. However, this CAG repeat length does not account for all of the variance in HD onset, suggesting other genetic and environmental factors may be involved. Recent Genome Wide Association Studies (GWAS) for genetic modifiers of HD motor symptom onset revealed several genome-wide genetic variants that are significantly associated with modifying HD age of motor onset. One of the most significant loci identified was on chromosome 15 adjacent to a gene, FAN1, which has a known function in DNA repair. Specifically, FAN1 has been shown to be involved in the repair of certain DNA lesions (i.e. interstrand cross-links) and may also be involved in the mismatch repair (MMR) pathway. In order to pursue unbiased molecular analyses of the impact of the Fan1 mutation on the mutant huntingtin-induced molecular pathogenesis in vivo, we crossed homozygous Fan1 hypomorphic knockdown (Fan1-KD) alleles onto mHtt knock-in mice with 140 CAG repeats (mHtt-KI-Q140 vs. Q140). We found a significant increase in the somatic CAG repeat instability in Fan1-KD/Q140 mice compare to Q140 at 6m. Moreover, we found neuropathological evidence of increased disease severity in Q140 mice with a reduction of Fan1 expression. We also performed transcriptomic analysis of these mice at 12m, and identified selective changes in Q140 brains with reduced Fan1 expression compared to Q140 controls. Together, our study provides novel insights into how a Fan1 deficiency could modify molecular and cellular pathogenesis in a full-length mHtt knock-in mouse model.

Disclosures: L. Deng: None. **J. Richman:** None. **N. Wang:** None. **K.T. Tamai:** None. **P. Langfelder:** None. **F. Gao:** None. **G. Coppola:** None. **C.S. Colwell:** None. **X.W. Yang:** None.

Poster

P275. Huntington's Disease: Preclinical Interventions

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P275.05

Topic: C.04. Movement Disorders other than Parkinson's Disease

Support: Contracted research - Wave Life Sciences

Title: Exploration of the effects of Huntingtin lowering on stress resistance in aged Huntington disease neurons

Authors: *E. MACHIELA, N. POLTURI, R. JELOKA, J. AQUINO, Y. XIE, A. L. SOUTHWELL;
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Abstract: Huntington’s disease (HD) is an autosomal dominant disease caused by a CAG repeat expansion in exon 1 of the huntingtin gene. Cellular stress is a consequence of biological aging and plays a role in the pathogenesis of HD. Wildtype Huntingtin (wtHTT) acts in cellular stress responses, and mutant HTT (mtHTT) confers both a toxic gain-of-function, contributing to cellular stress, and a loss-of-function, leading to deficient stress responses. HTT lowering has shown preclinical therapeutic benefit, and multiple strategies are in clinical development. Because of the critical role of HTT in the stress response, loss of wtHTT may affect the stress resilience of cells. Preclinical HTT lowering studies have used cells and animals free from exogenous stress, thus the effect of HTT lowering on stress susceptibility has not been evaluated. In an effort to better model the cellular effects of HTT lowering in aging adults subject to various stressors, we treated mature primary neurons from HD mice and WT littermates with progerin, a truncated form of the Lamin A protein that causes Hutchinson-Gilford progeria syndrome, a disease of rapid aging. Subsequently, we used antisense oligonucleotides (ASOs) to suppress HTT either selectively or non-selectively, followed by induced oxidative stress. We found that in WT neurons, non-selective HTT lowering increased ROS and DNA damage. However, in induced-aged , stressed HD neurons, both pan-HTT lowering and allele-selective mtHTT lowering prevented stress-induced ROS and DNA damage. This suggests that the benefit of mtHTT suppression may outweigh the detriment of HTT loss in the disease state. However, pan-HTT lowering also increased susceptibility to age and stress-induced cell death. Together, these data suggest that allele-selective HTT suppression may provide enhanced tolerability in aged and stressed conditions. Considering the mechanistic overlap in mtHTT toxicity and HTT deficiency toxicity, this may translate to enhanced efficacy of selective suppression approaches.

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Poster

P275. Huntington's Disease: Preclinical Interventions

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Team KJ

Title: Use of a split AAV delivery platform for the evaluation of dxCas9 for allele-specific silencing in Huntington's disease

Authors: *J. WALDO, J. A. HALMAI, D. CAMERON, P. DENG, K. FINK;
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Abstract: Huntington's disease (HD) is a rare, autosomal dominant neurodegenerative disorder caused by a trinucleotide expansion in exon 1 of the Huntingtin gene (HTT), which leads to neuronal dysfunction and cell death. Healthy HTT is implicated in axonal trafficking and trophic factor regulation, making targeted mutant allele-specific reduction critically important. This study aimed to target single nucleotide polymorphisms (SNPs) in HD patient-derived cells to reduce expression of mutant HTT (muHTT) using CRISPR epigenome editing. Heterozygous SNPs near regulatory regions of the HTT promoter were confirmed in HD patient cells, allowing for the design of allele-specific sgRNAs. Our novel vector expressing dxiCas9 containing repressive effector domains has higher binding specificity and broader PAM site requirements compared to SpdCas9. CRISPR constructs were introduced to a HD patient fibroblast line and knockdown of total HTT transcript levels was assessed to screen for sgRNA selection. Knockdown was assessed at multiple loci in the HTT gene and significant downregulation was achieved using several of our allele-specific sgRNAs. Targeting SNP rs762855 or rs3856973 resulted in significant downregulation compared to a non-treated control. A sgRNA targeting rs762855 also showed significant downregulation in primary cortical neurons derived from YAC128 mice. The delivery of large proteins to the brain continues to be a barrier to success when using these gene-editing tools. By using a split AAV system, large proteins such as Cas9 can be split into two domains fused to inteins that are able to recombine and create a full-length Cas9 in target cells. Previously established sgRNAs targeting muHTT were cloned into the C terminus of the split AAV vector system containing dxiCas9 fused to KRAB. These AAV were injected into the striatum of YAC128 mice containing the full-length human HTT locus. Knockdown of HTT was assessed via qPCR, western blot, and immunohistochemistry (IHC). These studies support the potential of targeted allele-specific strategies paired with split AAV delivery for CNS disorders.

Disclosures: J. Waldo: None. **J.A. Halmai:** None. **D. Cameron:** None. **P. Deng:** None. **K. Fink:** None.

Poster

P275. Huntington's Disease: Preclinical Interventions

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Program #/Poster #: P275.07

Topic: C.04. Movement Disorders other than Parkinson's Disease

Title: Pharmacological modulation of UBE2K mitigates toxicity in Huntington's Disease patient-derived fibroblasts.

Authors: *K. M. ROSEN, J. RANJAN, A. REBELLO, C. S. LEE, V. K. VISHNUDAS, N. R. NARAIN, S. GESTA, R. SARANGARAJAN;
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Abstract: Huntington’s disease (HD) is an autosomal dominant, progressive, neurodegenerative disorder caused by a polyglutamine (poly(Q)) triplet repeat expansion in the gene encoding the Huntingtin (Htt) protein. Huntingtin protein with pathologic poly(Q) expansion leads to a significant loss of neurons in the striatum, by mechanisms that are not clearly defined. The poly(Q) repeat sequence is localized to the N-terminal region of the very large (3144 amino acids) Htt protein. The N-terminal region of Htt is characterized by a high incidence of post-translational modifications and is the source of modified fragments that are commonly found as intracellular and intranuclear aggregates. Htt is known to be ubiquitinated in its N-terminal region and suggested to play a role in multiple sub-cellular systems and pathways. Htt interacts with many proteins but one of particular interest is the E2 ubiquitin-conjugating enzyme UBE2K, which had previously been identified through a yeast two-hybrid screen as Huntingtin-interacting protein-2 (Hip2) and shown to be involved in the ubiquitination of Htt. The ubiquitin proteasome system plays a critical role in protein quality control and is frequently disrupted in neurodegenerative disorders such as Huntington’s disease. HD patient derived fibroblasts display increased sensitivity to known chemical inducers of cellular stress via their actions on the proteasome and the endoplasmic reticulum. Interestingly, small molecule modulators allosterically targeting the function and activity of UBE2K demonstrate dose dependent decreases in the induction of chemical stressors. In addition, these UBE2K targeting compounds improve survival in differentiated neuroblastoma cell cultures exposed to the same chemical stressors in the absence of mutant Htt. The disease modifying role of UBE2K in HD and the therapeutic application of small molecule modulators to improve cell function and survival through in vitro and in vivo PoC studies is under active investigation.

Disclosures: K.M. Rosen: A. Employment/Salary (full or part-time):; Berg. **J. Ranjan:** A. Employment/Salary (full or part-time):; Berg. **A. Rebello:** A. Employment/Salary (full or part-

time):; Berg. **C.S. Lee:** A. Employment/Salary (full or part-time):; Berg. **V.K. Vishnudas:** A. Employment/Salary (full or part-time):; Berg. **N.R. Narain:** A. Employment/Salary (full or part-time):; Berg. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Berg. **S. Gesta:** A. Employment/Salary (full or part-time):; Berg. **R. Sarangarajan:** A. Employment/Salary (full or part-time):; Berg.

Poster

P275. Huntington's Disease: Preclinical Interventions

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Topic: C.04. Movement Disorders other than Parkinson's Disease

Support: Bev Hartig Huntington's disease Foundation to (W.D)

Title: Elimination of Huntington disease-related phenotypes by expanded-CAG RNA-targeting CRISPR/Cas13d in HD models

Authors: *Q. WU[1,4], K. MORELLI[5], M. GOSZTYLA[5], C. ZHANG[1], H. LIU[1], J. CHEN[4], R. MARINA[5], K. LEE[5], K. L. JONES[5], G. W. YEO[5], W. DUAN[1,2,3];
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Abstract: Currently incurable, Huntington's disease (HD) is a fatal, autosomal dominant neurodegenerative disorder caused by a CAG trinucleotide expansion (CAG^{EX}) in the first exon of the huntingtin (*HTT*) gene. Although the etiology of HD remains complex, many proposed mechanisms arise from the transcription and subsequent RAN translation of CAG-expanded HTT, ultimately causing disease through a toxic gain-of-function mechanism. Thus, therapeutic approaches that suppress mutant *HTT* at the RNA level are actively being pursued. We recently pioneered the repurposing of the Cas9 system to target and eliminate toxic repeat RNAs *in vitro* and *in vivo*, but for efficient *in vivo* delivery and clinical translation, Cas13d proteins are compact systems that can be packaged into a single adeno-associated viral vector (AAV9). Here, we engineered a AAV9 Cas13-based CAG^{EX} RNA-targeting CRISPR system to selectively target mutant *HTT* in patient iPSC-derived striatal neurons with minimal off-target effects from RNA-seq experiments. Intrastriatal delivery of CAG^{EX}/Cas13d in zQ175 mice, an established murine model of HD that expresses one copy of full-length human mutant *HTT* also mediates a significant and selective reduction of mutant HTT, and a sustained reversal of HD phenotypes, including a reduction of mutant HTT protein aggregates, attenuated striatal atrophy, and improved motor coordination in CAG^{EX}/Cas13d-treated zQ175 mice compared to control cohorts. Importantly, phenotypic improvements were durable for at least 8 months after

treatment without gross behavioral adverse effects, with minimal off-target interactions of CAG^{EX}/Cas13d in the mouse transcriptome. Future directions will be to determine potential off-target effects in the human transcriptome. Taken together, we demonstrate for the first time the proof-of-principle of CAG^{EX} RNA-targeting CRISPR/Cas13d as a potential therapeutic approach for HD, a strategy with broad implications for the treatment of other dominantly-inherited neurodegenerative disorders.

Disclosures: **Q. Wu:** Other; Co-first authors, School of Traditional Chinese Medicine, Beijing University of Chinese Medicine. **K. Morelli:** Other; Co-first authors. **M. Gosztyla:** None. **C. Zhang:** None. **H. Liu:** None. **J. Chen:** None. **R. Marina:** None. **K. Lee:** None. **K.L. Jones:** None. **G.W. Yeo:** Other; Co-Director, Bioinformatics and Systems biology Graduate Program, Associate Director, Genetics T32 Training Grant, Deputy Chair, Sanford Consortium for Regenerative Medicine, Return-to-learn, Steering group member, Co-Director, EXCITE lab, Scientific Director, SCRM Genomics Core, Chair, SCRM Space Committee. **W. Duan:** A. Employment/Salary (full or part-time):; Department of Neuroscience, Johns Hopkins University School of Medicine, Program in Cellular and Molecular Medicine, Johns Hopkins University School of Medicine.

Poster

P275. Huntington's Disease: Preclinical Interventions

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P275.09

Topic: C.04. Movement Disorders other than Parkinson's Disease

Title: Avocado oil prevents neurological and oxidative damage in a model of neurodegeneration induced by quinolinic acid.

Authors: *E. GONZÁLEZ-GUEVARA[1], L. TRISTÁN-LÓPEZ[2], P. MARTÍNEZ-GOPAR[2], Q. D. ÁNGELES-LÓPEZ[2], D. LÓPEZ-OROZCO[2], M. EL-HAFIDI[3], M. ALCARAZ-ZUBELDIA[2], A. SÁNCHEZ-MENDOZA[3], A. MORALES-MARTÍNEZ[4], F. PÉREZ-SEVERIANO[2];
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Abstract: Avocado oil prevents neurological and oxidative damage in a model of neurodegeneration induced by quinolinic acid.
González-Guevara Edith, Tristán-López Luis, Martínez-Gopar Pablo, Ángeles-López Quetzalli Dennise, López-Orozco Diego, El-Hafidi Mohammed, Alcaraz-Zubeldia Mireya, Sánchez-Mendoza Alicia, Morales-Martinez Adriana, Pérez-Severiano Francisca.

The intake of essential fatty acids (EFA) in both Huntington's disease (HD) patients and transgenic models has shown motor and cognitive improvement. Avocado oil contains different EFA and 60% is oleic acid, and its use through diet exerts important effects on the lipid composition of the neuronal cell membrane. The aim of this work was to study the effect of a fatty acid-rich diet included in avocado oil on the damage induced by quinolínic acid (QA). Male Wistar rats (250-280 g) were fed individually for 20 days with avocado oil (15% weight / weight). At the end of the diet, rats from the different groups were injured intrastriatally with QA (240 nmol/μL) and 2 hours after, striatal oxidative damage were measured through lipid peroxidation and the reactive oxygen species. Neurological changes were observed by counting of rotational turns in each animal after 3 days of QA lesion. We found that avocado oil diet for 20 days did not alter following parameters: the daily food intake, the body weight, and the biochemical parameters in blood. Interestingly avocado diet, diminished the ipsilateral turns (89%), and recovered the GABA levels (70%) and also the strital oxidative damage was prevented. Our data confirm the neuroprotective effect of a fatty acid-rich diet included in oil avocado.

Disclosures: E. González-Guevara: None. **L. Tristán-López:** None. **P. Martínez-Gopar:** None. **Q.D. Ángeles-López:** None. **D. López-Orozco:** None. **M. El-Hafidi:** None. **M. Alcaraz-Zubeldia:** None. **A. Sánchez-Mendoza:** None. **A. Morales-Martínez:** None. **F. Pérez-Severiano:** None.

Poster

P303. Neuroinflammation: Beyond Microglia

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Program #/Poster #: P303.01

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Title: Neural dysfunction following Francisella tularensis infection

Authors: *A. TENA, R. HIGHTOWER, M. HOLGUIN, M. SANCHEZ, C. SPENCER; Univ. of Texas at El Paso, El Paso, TX

Abstract: *Francisella tularensis* is a gram-negative intracellular coccobacillus that is the causative agent of tularemia. Local, acute inflammation is beneficial since it can clear bacterial infections, remove cellular damage and promote the healing of tissue. However, *F. tularensis* is capable of bypassing the immune system and causing an overactivation of inflammation, known as a “cytokine storm”. This cytokine storm increases host cell damage, nutrient availability and serves an immune distraction allowing the bacterium to replicate. Other infections that cause cytokine storms include Ebola, pandemic influenza, hantavirus, and HIV, all BSL3 pathogens. The routes of transmission for *F. tularensis* are many and intertwined. Infection could be the result of inhaling air droplets containing infectious bacteria, coming into direct contact with an

infected animal or being bitten by an infected arthropod such as a tick, flea or mosquito. Eating and drinking contaminated meat and water can also lead to infection and different disease manifestations occur depending on the route of transmission.
While extensive research has documented infection of the spleen, liver and lung, there hasn't been any reports of *F. tularensis* productively infecting the brain, until now. This work focuses on characterizing the effects of *F. tularensis* infection of the brain. We hypothesize that damage caused by *F. tularensis* neural infection will have a negative effect on behavior. This work aims to identify these negative effects based on the anatomical location of the bacteria foci, regions containing numerous bacteria. With the use of brain mapping and behavioral assays, we seek to determine the impact of *F. tularensis* infection on behavior and cognition.

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Poster

P303. Neuroinflammation: Beyond Microglia

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Program #/Poster #: P303.02

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: F31 NS116924
R01 NS103212

Title: Interactions with distinct MHC Class I molecules on brain endothelium differentially modulates CD8+ T cell entry and brain pathology during experimental cerebral malaria

Authors: *C. FAIN[1], K. AYASOUFI[1], J. ZHENG[2], F. JIN[1], R. H. KHADKA[1], Z. P. TRITZ[1], A. J. JOHNSON[1];
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Abstract: Malaria is a mosquito-borne disease caused by *Plasmodium* parasite that specifically infects red blood cells. *Plasmodium falciparu*m is the deadliest species of plasmodium in humans, namely due to its ability to cause severe encephalopathy; referred to as cerebral malaria. The mechanisms underlying brain involvement and subsequent fatality are not entirely understood. Using the animal model of experimental cerebral malaria (ECM), we sought to elaborate upon mechanisms of pathogenesis. We employ T1 and T2-weighted MRI as a novel diagnostic tool which confirms recapitulation of human data. In this etiology we confirm increased adhesion molecule expression, increased antigen (Ag)-presentation molecules and with two-photon we show extensive T cell interactions at the brain vasculature, resulting in CD8 T cell infiltration into brain parenchyma, brain pathology and neuronal death. While CD8 T cell interaction with MHC class I on cerebrovascular endothelial cells (CECs) has been put forward

as a putative mechanism for ECM, it has not been directly confirmed *in vivo*. We therefore hypothesized that removing Ag presentation by either H-2K^b or H-2D^b class I molecules specifically on brain endothelium would result in controlled modulation of activation and entry of parasite-specific CD8 T cells; attenuating pathology. To test this, we crossed endothelium-specific cre, CDH5-Cre mice with novel H-2K^b LoxP and H-2D^b LoxP mice to create endothelium-specific cKO of either or both class I molecules. In these mice we show differential class I dependent CD8 T cell infiltration and activation as well as subsequent changes in antigenic drainage and cervical lymph node involvement. Importantly, T cell interactions with cerebral vessels were modulated, and vascular permeability at the blood-brain barrier was attenuated, resulting in differentially improved survivals between the cKO strains. Together these data demonstrate that parasite-specific CD8 T cells undergo differential Ag-dependent engagement with H-2K^b or H-2D^b class I molecules on CECs in order to coordinate activation, entry and ECM development. Therefore, these studies indicate that specific modulation of these interactions may hold therapeutic benefit and reduce mortality following this severe complication. This work also provides much needed mechanistic insights into how this pathology develops.

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Poster

P303. Neuroinflammation: Beyond Microglia

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Program #/Poster #: P303.03

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Title: In vitro approaches to modulate neuroinflammatory astrocytes

Authors: N. HAGAN, ***F. RAPINO***, L. WOODWORTH, A. BLAZIER, J. SANCHO, D. OFENGEIM;
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Abstract: In vitro approaches to modulate neuroinflammatory astrocytes

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Glia cells, such as microglia and astrocyte, are critical regulators of inflammation in the brain. Neuroinflammation can be a protective mechanism that helps maintain CNS homeostasis. However, sustained and prolonged inflammation is detrimental and contributes to the pathology of neurodegenerative diseases such as Alzheimer's Disease (AD), Parkinson's Disease (PD), and Amyotrophic Lateral Sclerosis (ALS). Because of the dual role that astrocytes play in

neuroprotection and neurotoxicity, a characterization of astrocyte states is needed to generate in vitro models to develop effective anti-inflammatory therapies. To characterize the molecular identity of reactive astrocytes, we performed bulk RNA sequencing on primary human astrocytes treated with A1 cocktail (C1q, IL1-α, and TNF- α) and compared them to untreated astrocytes. In addition to gene expression, we examined changes in cell surface markers using a panel of 361 antibodies in resting or activated conditions. We then used this system to investigate whether we could alter the transition of astrocytes into a pro-inflammatory phenotype using both a pharmacological and genetic approach. Finally, to mimic the complexity of the brain and to generate a robust system to study the effects of candidate targets in disease-relevant human cells, we implemented and characterized a novel triculture system using iPSCs derived neurons, microglia, and astrocytes. By using the tools we have generated, we aim to better understand the dynamic state of astrocytes with the ultimate goal of developing therapies to modulate their phenotypes to ameliorate neuroinflammation in neurodegenerative diseases.

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Poster

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Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: NIH R01 NS101167 (to NHV)
CURE Epilepsy (to NHV)

Title: The temporal dynamics of brain invading monocytes following status epilepticus

Authors: *L. SHAPIRO, S. HUNTER-CHANG, R. DINGLEDINE, N. H. VARVEL;
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Abstract: Status epilepticus (SE) is a medical emergency that triggers a cascade of pathological events including neuron loss, blood-brain barrier breakdown, activation of brain-resident microglia, and infiltration of blood-borne monocytes into the brain, culminating in the development of epilepsy and cognitive decline. It is of great interest to develop novel therapeutic strategies to prevent the onset of epilepsy and cognitive comorbidities following SE. Previous work in our laboratory identified brain invading CCR2+ monocytes as pathogenic following SE in mouse models. Blocking monocyte brain invasion after SE via global knockout of *Ccr2* enhances weight regain, reduces neuron loss, prevents BBB erosion, and attenuates neuroinflammation, highlighting the potential for CCR2+ monocytes as a novel disease-modifying target. However, it remains to be determined how long these CCR2+ monocytes

remain in the brain following SE. To address this, we crossed mice expressing inducible CreER^{T2} under the CCR2 promoter to Ai9 reporter mice to allow for permanent fluorescent tagging of CCR2-expressing cells following tamoxifen administration. Tamoxifen (2mg, i.p.) was administered to male mice 8-10 weeks of age 48 and 24 hours prior to kainic acid-induced SE (30 mg/kg, i.p.) and the presence of CCR2+ cells in the brain was evaluated on a time course following SE. We observed peak engraftment of CCR2+ cells in the hippocampus and amygdala at 7 days post-SE (n=7), with virtually no cells encountered by day 14 (n=3). Ongoing work is being conducted to further immunophenotype the brain-invading cells and examine their inflammatory profile at each time point in order to better understand how they may contribute to the pathology observed following SE. Altogether, our results provide insight into the temporal dynamics of monocyte invasion into the brain following SE, which contributes to our understanding of how these cells may be used as a target for intervention in clinical populations.

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Poster

P303. Neuroinflammation: Beyond Microglia

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Title: A novel model for paired epigenetic and transcriptomic studies of Müller glia with age related macular degeneration

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Abstract: Aging is the major risk factor for age-related macular degeneration (AMD) but how ‘normal’ aging contributes to AMD pathogenesis is unclear. Epigenetic mechanisms, such as the DNA modifications of methylcytosine (mC) and hydroxymethylcytosine (hmC), affect DNA accessibility, regulate genomic organization and gene expression, and are altered with aging.

DNA methylation analyses of AMD patient blood and retinas suggest differential methylation resulting in gene expression changes during development/progression of AMD. In addition, Müller glia are activated in AMD patient retinas. The aim of this study is the validation of a cre/ERT2-NuTRAP mouse model for the parallel interrogation of differential mC/hmC genome-wide and transcriptome specifically in Müller glia with retinal aging. Female NuTRAP $^{flox/flox}$ were bred with male Aldh1l1-cre/ERT2$^{+/wt}$ to generate Aldh1l1-cre/ERT2$^{+/wt}$; NuTRAP$^{flox/wt}$ (Aldh1l1-cre/ERT2^{+}; NuTRAP^{+}). At 5 months old, female and male mice received intraperitoneal tamoxifen treatment or left untreated as controls. Mice were euthanized and their retinas harvested and processed for: 1) Immunohistochemistry of mCherry, EGFP, GS, and GFAP expressions on retina sagittal sections via confocal microscopy; 2) Ribosome bound RNA isolation via TRAP protocol, qPCR validation and stranded RNA-Seq profile, and 3) nuclei suspension preparation and DNA isolation via INTACT method for bisulfite amplicon sequencing (BSAS). A cohort of mice was subjected to the optic nerve crush model (ONC) to study the transcriptomic response of Müller glia at 72h post procedure. Specific co-localization of EGFP, mCherry, and the Müller glia marker GS was observed in the Aldh1l1-cre/ERT2^{+}; NuTRAP^{+} retinas. RNA-seq bioinformatic analysis revealed enrichment of Müller marker genes and pathways and depletion of other cell type markers in the positive TRAP fraction of Aldh1l1-cre/ERT2^{+}; NuTRAP^{+} retinas, compared to input profiles. qPCR validated enrichment and depletion of selected transcript expressions. BSAS showed a decrease of mC in the promoter region of selected Müller glia-specific genes in INTACT-DNA positive fraction relative to whole retina input. Over-representation analysis of differentially expressed genes in the ONC vs control group identified 7 biological processes related to inflammation with significant enrichment ratio. The selective recombination in Müller glia, enrichment of Müller glia genes, and the epigenetic findings support the Aldh1l1-cre/ERT2; NUTRAP model is suitable for the study of Müller glia-specific contributions to the transcriptome and epigenome of the retina in both acute stress and chronic aging settings.

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Poster

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Program #/Poster #: P303.06

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: Automating Biomedical Data Analysis R15GM139115

Title: Changes to Astrocyte Activation Markers at Different Stages of Cuprizone Treatment

Authors: *L. FRANKLE, K. LI, C. LU, Q. GUAN, E. J. FREEMAN, R. CLEMENTS;
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Abstract: Cuprizone is a copper chelator that demyelinates neurons and is therefore frequently used to model multiple sclerosis as well as other demyelinating and degenerative diseases. Our previous work examining astrocyte dynamics using glial fibrillary acidic protein, or GFAP staining, confirmed a pattern of activation including increased process number and branching after one, two, and four weeks of cuprizone treatment. The current phase of the project aims to determine whether similar patterns are observed at one or two weeks of cuprizone treatment as well as when such changes begin. Animals were fed 0.3% cuprizone chow ad libitum for one or two weeks, sacrificed, dissected, and their brains halved with one half stained using GFAP and DAPI and imaged using confocal microscopy with astrocytes in the corpus callosum assessed for activation, and one half processed through Western blot. We used Western blots to measure levels of C3 or complement component 3, and Emp1 or epithelial membrane protein 1, in the whole-cell lysate of homogenized samples taken from the corpus callosum and/or cortex of control and experimental animals. C3 levels are measured because C3 is known to be upregulated in astrocytes that have an A1 reactive phenotype as well as by other cell types such as mast cells and macrophages in times of stress. Emp1 levels are assessed because Emp1 is upregulated in another type of activation phenotype less detrimental, A2. To confirm that the presence assessed by Western blot was representative of astrocyte protein levels of C3 and Emp1, immunofluorescence was done using these proteins on fixed tissue slides taken at one, two and four weeks of cuprizone treatment. The relative levels of A1 and A2 activation would therefore be relevant to the progression of cuprizone toxicity.

Disclosures: L. Frankle: None. **K. Li:** None. **C. Lu:** None. **Q. Guan:** None. **E.J. Freeman:** None. **R. Clements:** None.

Poster

P303. Neuroinflammation: Beyond Microglia

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P303.07

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: Mohn Foundation
Intramural Research Program, NIEHS/NIH
IIRG, Alzheimer's Association

Title: The role of astrocytic α7 nicotinic acetylcholine receptors in neuroinflammation-mediated cognitive impairment

Authors: *E. ISHIMWE[1], I. CORTEZ[1], I. E. CISNEROS[2], C. M. HERNANDEZ[3], ***K. T. DINELEY**[1];
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Abstract: Background: An important component of Alzheimer's disease (AD) is neuroinflammation. In the periphery, α7 nAChR activation inhibits macrophage production of pro-inflammatory cytokines. In the CNS, α7 nAChRs are enriched in brain regions affected early in AD. However, α7 nAChR expression is not restricted to neurons; these receptors are highly enriched within the CNS immune system. We have previously reported important roles for astrocytic α7 nicotinic acetylcholine receptor (nAChR)-Aβ interaction with subsequent effects on cognition. To refine these studies, we sought to test the hypothesis that astrocytic α7 nAChRs serve an analogous anti-inflammatory role to that in the periphery.Method: To test our hypothesis, we generated mice in which the fourth exon of the α7 nAChR gene (Chrna7) is flanked by loxP sites (α7nAChR^{flox}). We then crossed α7nAChR^{flox} mice with mice expressing Cre recombinase driven by the glial acidic fibrillary protein (GFAP) and studied neuroinflammatory responses and cognition following intracerebroventricular (ICV) injection of LPS (10 ng); a dose that induces transient neuroinflammatory responses without measurable peripheral inflammation. We studied hemizygous astrocytic α7nAChR (GFAP-A7$^{+/-}$) knockdown mice compared to α7nAChR$^{flox+/-}$ littermates.Result: Whereas sham and LPS ICV-injected α7nAChR$^{flox+/-}$ littermates performed similarly in hippocampus-dependent associative learning and memory, GFAP-A7$^{+/-}$ mice were impaired. Bio-Plex analysis of serum, hippocampus and cholinergic basal forebrain revealed that α7nAChR$^{flox+/-}$ littermates mounted both a pro- and anti-inflammatory response to ICV LPS; GFAP-A7$^{+/-}$ mice did not. However, GFAP-A7$^{+/-}$ mice exhibited pronounced microglia activation and tau hyperphosphorylation in hippocampal structures. Morphometric analysis showed that GFAP-A7$^{+/-}$ mice presented with blunted astrocyte response to LPS challenge. We used cultured astrocytes from hemizygous GFAP-Cre α7nAChR knockdown mice and α7nAChR^{flox} littermates to decipher mechanistic details and report that similar dysregulated inflammatory responses to LPS are detected in GFAP-A7$^{+/-}$ astrocyte cultures.Conclusion: Blunted astrocyte response, absence of an anti-inflammatory response, microglia activation and tau pathology resulting from ICV LPS injection suggests that knockdown of α7 nAChRs in astrocytes triggers a feedforward pro-inflammatory response to elicit AD-like cognitive deficits. Thus, similar to their role in the periphery, astrocytic α7 nAChRs serve an anti-inflammatory role in the CNS. This has significant implications for AD since Aβ binds α7 nAChRs to silence these receptors.

Disclosures: E. Ishimwe: None. **I. Cortez:** None. **I.E. Cisneros:** None. **C.M. Hernandez:** None. **K.T. Dineley:** None.

Poster

P303. Neuroinflammation: Beyond Microglia

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P303.08

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: 5R01NS115126-02

Title: Inhibition of the pentose phosphate pathway in anti-neuronal cytotoxic T lymphocytes preserves neuronal function and ameliorates neuroinflammatory disease

Authors: *E. GRUND[1], B. CLARKSON[2], C. L. HOWE[2];
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Abstract: Despite the presence of physical barriers and developmental immune tolerization, the CNS acts throughout life as a source of antigens that can induce aberrant auto-reactivity against neuronal and glial proteins. In diseases such as multiple sclerosis and the autoimmune limbic encephalitides, activated $CD8^+$ cytotoxic T cells infiltrate the brain and are found in clonally expanded clusters indicative of antigen-specific recognition. At present, the pathophysiological role of these cells is poorly understood and therapeutic strategies for neutralizing these cells are limited. T cells exhibit profound metabolic reprogramming in support of the energetic and proliferative demands of activation against autoantigens. Key metabolic pathways in this reprogramming include glycolysis and the pentose phosphate pathway (PPP). We have demonstrated that pharmacologic inhibition of the PPP reduces the acquisition of an effector phenotype and suppresses the secretion of proinflammatory cytokines in $CD8^+$ T cells. We have also found that limiting the metabolic flux of glucose-derived metabolites through the PPP rescues neurons from $CD8^+$ T cell-mediated cytotoxicity and preserves the electrophysiological function of primary neurons under stress. Finally, in the EAE preclinical model of multiple sclerosis, we found that pharmacologic inhibition of the PPP when administered at the onset of symptoms is sufficient to delay and ameliorate motor disability. These results demonstrate the potential of immunometabolic therapies to limit autoinflammatory and autoimmune injury to the CNS and to disrupt the progression and accumulation of neurologic deficits in patients with immune-mediated neurologic disease.

Disclosures: E. Grund: None. **B. Clarkson:** None. **C.L. Howe:** None.

Poster

P303. Neuroinflammation: Beyond Microglia

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P303.09

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Support: Transpharmation Ltd
MRL MSD Pharmaceuticals, London UK and Merck & Co., Inc., Kenilworth, N.J., U.S.A

Title: Investigation of lipopolysaccharide (LPS) administration by intraperitoneal (i.p.) injection and intracerebroventricular (i.c.v) infusion as mouse models of neuroinflammation

Authors: L. R. DAVISON[1], D. ZWILLING[2], H. WOLFE[3], R. WILLIS[3], M. J. HAFEY[2], T. XIONG[2], E. STOJEK[1], M. MITSOGIANNIS[1], S. BOGEN[2], J. BROWNLEES[3], *J. PRENDERVILLE[1];
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Abstract: Dysregulated immune signalling is implicated in disease and neuroinflammation has been linked to the pathogenesis of multiple neurodegenerative and psychiatric disorders. The establishment of effective and high-throughput preclinical models of neuroinflammation is required to advance drug development. LPS, a gram-negative bacteria outer membrane component, administered by peripheral (i.p.) or central (i.c.v.) injection has been used to model neuroinflammation, yet few studies have compared peripheral and central cytokine expression. Male C57BL/6J mice (n=24) received an i.p. injection of saline or one of 3 LPS doses (100, 250, 500 ug/kg). A second group of mice (n=16) received a stereotaxic unilateral i.c.v. infusion of artificial cerebrospinal fluid or one of 3 LPS doses (1, 5, 20 ug/mouse). In a third group of mice (n=50), the effect of minocycline (acute and sub-chronic, 100 mg/kg, i.p.) and dexamethasone (30 mg/kg, p.o.) administration prior to LPS (250 ug/kg, i.p.) was investigated. Plasma and brain were collected 4 hours after LPS administration and cytokines (TNF-α, IL-6, IL-1β) and chemokines (CCL2, CCL3, CCL4) were analysed using Meso Scale Diagnostics multiplexed assays.LPS (i.p.) increased all cytokines and chemokines analysed in plasma. In brain tissue, LPS (i.p.) increased IL-6, IL-1β and CCL2 in a dose-dependent manner. LPS (i.c.v.) increased all cytokines and chemokines in plasma except for CCL2. In brain tissue, 20 ug LPS (i.c.v.) induced an increase in all cytokines and chemokines analysed. TNF-α and IL-1β were increased in brain by 1 and 5 ug LPS (i.c.v.). Dexamethasone but not minocycline reduced all three cytokines and CCL4 expression in plasma and brain.Both i.p. and i.c.v. administration of LPS induced a robust peripheral inflammatory response. Similarly, both routes of administration induced neuroinflammation, although the specific cytokines and chemokines that increased in brain tissue differed between models. These data demonstrate that i.p. administration of LPS is sufficient to induce a model of neuroinflammation, responsive to anti-inflammatory drugs, and shows advantages over the i.c.v. model requiring surgical infusion. Neuroinflammation induced by i.c.v. administration was not contained within the brain at the doses tested here.

Disclosures: L.R. Davison: A. Employment/Salary (full or part-time):; Transpharmation Ireland Ltd. **D. Zwilling:** A. Employment/Salary (full or part-time):; MRL MSD Pharmaceuticals, London UK and Merck & Co., Inc., Kenilworth, N.J., U.S.A and. **H. Wolfe:** A. Employment/Salary (full or part-time):; MRL MSD Pharmaceuticals, London UK and Merck & Co., Inc., Kenilworth, N.J., U.S.A and. **R. Willis:** A. Employment/Salary (full or part-time):;

MRL MSD Pharmaceuticals, London UK and Merck & Co., Inc., Kenilworth, N.J., U.S.A and. **M.J. Hafey:** A. Employment/Salary (full or part-time):; MRL MSD Pharmaceuticals, London UK and Merck & Co., Inc., Kenilworth, N.J., U.S.A. **T. Xiong:** A. Employment/Salary (full or part-time):; MRL MSD Pharmaceuticals, London UK and Merck & Co., Inc., Kenilworth, N.J., U.S.A. **E. Stojek:** A. Employment/Salary (full or part-time):; Transpharmation Ireland Ltd. **M. Mitsogiannis:** A. Employment/Salary (full or part-time):; Transpharmation Ireland Ltd. **S. Bogen:** A. Employment/Salary (full or part-time):; MRL MSD Pharmaceuticals, London UK and Merck & Co., Inc., Kenilworth, N.J., U.S.A. **J. Brownlees:** A. Employment/Salary (full or part-time):; MRL MSD Pharmaceuticals, London UK and Merck & Co., Inc., Kenilworth, N.J., U.S.A. **J. Prenderville:** A. Employment/Salary (full or part-time):; Transpharmation Ireland Ltd.

Poster

P351. Traumatic Brain Injury: Human Studies: Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P351.01

Topic: C.10. Brain Injury and Trauma

Title: Corticothalamic Functional Connectivity Post Traumatic Brain Injury

Authors: *H. KARBASFOROUSHAN[1], S. SIDDIQI[2], L.-L. WU[3], G. SWAMINATH[4], R. TOLL[5], J. L. TAYLOR[1], O. A. HARRIS[1], M. M. ADAMSON[6];
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Abstract: Previous animal and human studies in traumatic brain injury (TBI) have frequently reported the thalamus' abnormalities post injury, as well as its role in recovery. There is a topographically organized pattern of functional connectivity between human cortex and thalamus, indicating parallel pathways linking distinct cortical areas to specific thalamic nuclei. However, the changes to corticothalamic connectivity network post traumatic brain injury is still unknown. The goal of this study was therefore to use advanced functional connectivity analysis methods to investigate the alterations in corticothalamic connectivity network in patients with traumatic brain injury. Thirty-one patients with TBI and 30 age-match healthy controls were recruited to this study. T1-weighted anatomical and functional MRI scans were collected on a 3T MRI scanner. T1-weighted images were acquired with isotropic resolution = 0.9 mm, flip angle = 12 degrees, and acquisition matrix = 256 × 256. The functional imaging scans had the following parameters: 42 axial slices, 2.9mm isotropic resolution, TR = 2s, TE = 30ms. Preprocessing and statistical analysis of the functional imaging data were performed using SPM12 software. Preprocessing steps included head motion correction, co-registration of the functional data to the T1-weighted scan, spatial normalization to MNI space, and smoothing with

a 2 mm kernel. The cortex was divided into 6 nonoverlapping regions of interest: the prefrontal cortex, motor cortex and supplementary motor area, somatosensory cortex, posterior parietal cortex, temporal lobe, and occipital lobe. The resting state preprocessed images were then used to create functional connectivity maps for each participant using Conn toolbox. The mean BOLD time series from each ROI was entered as a predictor in a multiple regression general linear model at each voxel. The variance related to head motion, the global gray matter signal, white matter and CSF were removed. Statistical analysis of functional connectivity proceeded by entering the individual subject connectivity maps into a two-sample t-test to create between group functional connectivity statistical parametric maps. All statistical maps for each ROI were then thresholded at $p<0.01$, cluster size corrected $p = 0.05$. Our results indicated increased functional connectivity between cerebral cortex and thalamus in patients with TBI, compared to healthy controls. Especially, the prefronto-thalamic functional connectivity had the greatest increase in these patients. These findings are aligned with the previous reports of increased global connectivity of thalamus post TBI.

Disclosures: H. Karbasforoushan: None. **S. Siddiqi:** None. **L. Wu:** None. **G. Swaminath:** None. **R. Toll:** None. **J.L. Taylor:** None. **O.A. Harris:** None. **M.M. Adamson:** None.

Poster

P351. Traumatic Brain Injury: Human Studies: Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P351.02

Topic: C.10. Brain Injury and Trauma

Support: National Collegiate Athletic Association
Department of Defense
Office of the Assistant Secretary of Defense for Health Affairs
Combat Casualty Care Research Program – Psychological Health and Traumatic Brain Injury Program under Award No. W81XWH1420151

Title: Head Impact Exposure May Influence Post Concussion Acute Clinical Outcomes in NCAA Division I Football Players: Findings from the NCAA-DOD CARE Consortium

Authors: J. SEIFERT[1], A. SHAH[2], J. HAREZLAK[3], S. ROWSON[4], J. MIHALIK[5], L. RIGGEN[3], S. DUMA[4], A. BROOKS[6], K. L. CAMERON[7], C. C. GIZA[8], J. GOLDMAN[8], K. GUSKIEWICZ[5], M. N. HOUSTON[7], J. C. JACKSON[9], G. MCGINTY[9], P. PASQUINA[10], S. BROGLIO[11], T. MCALLISTER[12], M. A. MCCREA[2], *B. D. STEMPER[1,13];
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CA; [9]United States Air Force Acad., Colorado Springs, CO; [10]Uniformed Services Univ., Bethesda, MD; [11]Univ. of Michigan, Ann Arbor, MI; [12]Indiana Sch. of Med., Indianapolis, IN; [13]Clement J. Zablocki Veterans Affairs Med. Ctr., Milwaukee, WI

Abstract: Different clinical assessments are used to identify specific post-concussion changes in athletes. The Standardized Assessment of Concussion (SAC) is a cognitive screening tool used to assess concussion. It was hypothesized that increased severity of concussive head impacts, and preinjury head impact exposure (HIE) can influence concussion severity, which may be reflected by a greater change in SAC score from baseline. We hypothesized that greater HIE and concussive impact accelerations would lead to decreased SAC assessment scores at the 24-48 hour timepoint in concussed athletes.

During the 2015-2017 NCAA Division I football competition seasons, athletes enrolled from 6 institutions were equipped with helmet-mounted accelerometer arrays that recorded each head impact exceeding a 10-g threshold and completed baseline clinical assessments, including the SAC. The SAC was scored out of a maximum of 30 points, which indicated no questions missed. HIE was compared between cohorts with decreased or increased/no change in SAC score at the 24-48 hr post-injury time point compared to baseline. HIE was assessed as either the total number of head impacts or risk weighted exposure (RWE), a cumulative injury risk function based on peak linear and rotational accelerations of repetitive impacts.

46 clinically diagnosed concussions were sustained by 41 players. As a group, athletes had significantly lower SAC scores at the 24-48 hour post injury timepoint when compared to baseline (U_{base}= 27, $U_{24\text{-}48hr}$= 25.7, p = 0.03). However, some athletes tested below (n= 28) and others tested at or above (n = 18) their baseline scores. Athletes that tested below their baseline SAC score sustained a higher number of head impacts on the injury date ($median_{below}$= 13 (95% CI [10.7,17.3])) compared to those that tested at or above their baseline scores ($median_{above}$= 8 (95%CI [6.13,14.3]), p = 0.095). This was also true for the two (p =0.03), four (p=0.08), and six (p =0.02) calendar days prior to injury plus the injury date. No difference was evident for RWE over any time prior to concussion. Between the two groups, there was no difference between linear (p =0.66) and rotational (p =0.19) accelerations of the concussive impacts.

These findings demonstrated that athletes with negative post-injury change in SAC score (i.e., decreased cognitive processing) had greater HIE leading up to concussion. Although injury date HIE was not statistically significant, HIE during the 3, 5, and 7 days leading up to concussion was significantly greater in athletes with negative SAC change. Therefore, accumulation of head impacts over time may be an important factor in producing post-concussion cognitive changes.

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Poster

P351. Traumatic Brain Injury: Human Studies: Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P351.03

Topic: C.10. Brain Injury and Trauma

Support: NIH NIGMS Grant # 2RL5GM118963-06
DoD USAMRAA W81XWH-16-D-0024

Title: Hyponatremia in TBI Patients

Authors: *A. RANO[1], S. UNDERWOOD[2];
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Abstract: Background: Hyponatremia is frequently seen in patients with traumatic brain injuries (TBI). Hyponatremia (HN) is an electrolyte imbalance seen often in hospitalized patients. It occurs notably more frequently in patients who have suffered TBI (15%-20%). HN is defined as sodium levels less than 135 mmol/L. When TBI patients develop HN it can lead to serious neurological issues. Moderate to severe HN can increase the risk of death. This study looks at data in TBI patients who develop hyponatremia and variables such as medications, type of injury, demographics, etc. Methods: A retrospective analysis of 1232 patients treated at a level 1 trauma center from 2017 to 2020 was completed. Data was obtained from the local trauma registry and electronic medical records. Inclusion criteria were an injury severity score > 8, known age, and documented TBI. Patients with HN were identified as having serum sodium levels less than 135 mmol/L.Descriptive statistics were used to analyze the data. A p-value of < 0.05 was used to assign significance. Results: Of 1232 patients, 13.2% developed HN. The data was split into two groups, patients who developed HN and those who did not. There was no significant difference in admission vital signs. Patients who developed HN were older and had a lower GCS on admission. HN patients had significantly lower sodium levels on admission (136 mmol vs 140 mmol) and were more likely to be admitted to the ICU (92.1% vs. 79.3%). There was no difference in OR visits (5.3% vs. 4.6%). Patients in the HN group were more likely to receive saline-based fluids, especially 3% hypertonic saline (41.7% vs 8.3%) and 7% hypertonic saline (3.7% vs. 0.8%). Type of TBI did not appear to correlate with developing HN. Conclusion: Patients who develop HN are likely to have a lower GCS and sodium level on admission, be older, and receive more hypertonic saline. However, the type of TBI does not correlate with HN. Further studies are required to determine a specific profile of patients who may be at higher risk of developing HN after suffering a TBI.

Disclosures: A. Rano: None. **S. Underwood:** None.

Poster

P351. Traumatic Brain Injury: Human Studies: Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P351.04

Topic: C.10. Brain Injury and Trauma

Title: Binocular Coordination of Horizontal Saccades in mild TBI

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Abstract: Mild traumatic brain injury (mTBI) can result in significant problems affecting vision and oculomotor function (Armstrong, 2018; Mani et al., 2018; Stuart et al., 2020), vergence eye movements (Magone et al., 2014; Suhr et al., 2015), saccades (Samadani et al., 2015; Hunfalvay et al., 2019), spatial orientation, movement, and balance (Wallace and Lifshitz, 2016; Hoffer et al., 2010). These issues can result in problems with gaze control, eye-head-coordination, and visual-motor transformations underlying goal-directed movements. This can occur after multiple head trauma events, and in some cases after a single mild TBI event. Also, symptoms can persist years after the original trauma (Danna-Dos-Santos et al., 2018) and can become progressively worse over time. Effects of the natural aging processes probably interact with the pathophysiology resulting from TBI. The general aims of this study are to characterize the coordinated movement of the two eyes during changes in gaze in response to movement of a visual target and to relate the velocity trajectories of the two eyes to vergence dysfunction in mTBI. For this presentation, the horizontal position and velocity of the left eye versus the right eye were analyzed for the data from 18 control participants and 23 participants with mTBI. Saccade targets were presented 5 to 25 degrees left/right of center. Plots of left versus right horizontal eye velocity were quantified with polynomial regressions of eye velocity toward and away from zero velocity. Discriminant analyses of the regression coefficients identified those subjects with abnormal velocity trajectories. Some patterns included the following. For large saccade amplitudes there could be different velocities for the adducting eye versus the abducting eye in mTBI subjects who have convergence insufficiency or convergence excess. Furthermore, there were differences between the acceleration and deceleration phases of the saccades, with the acceleration phase of horizontal saccades being significantly different in mTBI compared to controls. Simulations of the velocity and acceleration profiles suggest deficits in the motor commands for adduction versus abduction movements. These results have identified eye velocity patterns and possible motor command deficits that show promise for characterizing binocular eye movements in mTBI. Further work will evaluate vertical and diagonal saccades and gaze in three dimensions where there are changes in vergence. The results could provide further insight into underlying the pathophysiology affecting the control of gaze in mTBI and suggest possibilities for vision therapy.

Disclosures: J.H. Anderson: None.

Poster

P351. Traumatic Brain Injury: Human Studies: Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P351.05

Topic: C.10. Brain Injury and Trauma

Title: Sensory functions and their relation to balance metrics: A secondary analysis of the LIMBIC-CENC multicenter cohort.

Authors: *S. M. VAN DER VEEN[1], R. PERERA[2], P. C. FINO[4], L. MANNING-FRANKE[2], A. A. AGYEMANG[2], K. SKOP[5], E. A. WILDE[6], S. R. SPONHEIM[7], A. STAMENKOVIC[1], J. S. THOMAS[1], W. C. WALKER[3];
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Abstract: Among patients with traumatic brain injury (TBI), symptoms of dizziness, nausea, and postural instability often persist. Besides balance impairments, blast related TBI has been associated with a loss of hearing (19%), vision (34%) or both (32%). When both hearing and vision were impaired, independent of TBI severity, poorer functional progress was found. The ability to regain premorbid balance and independent gait may be dictated by the ability to process, interpret, and combine, sensory information. Secondary analysis was performed on the complete CENC/LIMBI data set (N=1550 ,1346 male, mean 40±STD 10 age, 1248 TBI). Five imputed datasets were created using a fully condition specification. The estimates were then combined, and standard errors adjusted to account for the uncertainty due to missingness. Hierarchical regressions were performed, for each outcome on the separate items of sensory tests grouped in the following 5 steps: 1) the number of TBIs suffered, age, and sex, 2) the separate HHIA items, 3) separate BVMT items, 4) visual acuity, and 5) items of the SCAN3. Separate regression analyses were carried out for each of the balance assessment (i.e., different scores on the CDP). Sensory measures were removed from the regression equations when multicollinearity was found (VIF>10). Statistical significance determined using a Bejamin Hochberg correction. All sensory measures combined (demographics, hearing, reported by a hearing handicap questionnaire, visual memory, visual acuity, and auditory processing) accounted between 10.9% and 14.4% of the variability in the balance measures. Number of TBI was significantly related to CDP scores 2-6 ($p=.001$, $p<.001$, $p=.018$, $p=.005$, $p=.042$ respectively). When hearing (item on HHIA) was impaired lower scored on CDP 3, 5, and 6 were found ($p=.020$, $p=.003$, $p=.005$ respectively). Declines in auditory processing (SCAN3, competing words) was associated with worse CDP1-3 ($p=.001$, $p<.001$, $p=.021$ respectively). Better visual processing (SCAN3) was related to changes in CDP 2,5,6 scores (SCAN 3 Recall, sores CDP2 $p=.011$, Better CDP 5 and 6, $p=.010$, $p=.009$, Learning worse CDP 2 $p=.014$ and DCP6 $p=.013$, and Delayed recall better

CDP2 p=.003). TBI patients score as well as controls on static balance, but balance deficits may be unmasked when adjustments in weighting of theses sensory inputs are required due to various experimental perturbations, CDP2-6. In combination with very little associations with visual acuity and hearing deficits but stronger associations with sensory processing measures it is likely that processing of sensory information affects balance negatively and may increase fall risk in TBI patients.

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Poster

P351. Traumatic Brain Injury: Human Studies: Mechanisms

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Program #/Poster #: P351.06

Topic: C.10. Brain Injury and Trauma

Support: R01-AG049607-02
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S10OD011928
Football Players Health Study at Harvard University, funded by the NFL Players Association

Title: Tau aggregation in American-style football players exposed to repetitive head injury using positron emission tomography.

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Abstract: Background: American-style football players are likely to have experienced exposure to repetitive head impacts. Several studies suggested an association between traumatic brain injury (TBI) and risk of developing misfolded p-Tau protein identified in chronic traumatic encephalopathy (CTE) and Alzheimer's disease (AD). *In vivo* p-Tau positron emission tomography (PET) tracers with high binding affinity and selectivity were developed recently. This study's goal is to assess the ability of PET imaging to detect early stages of neurodegenerative disease in a population at increased risk of head injury. **Methods:** We recruited 28 former professional American-style football players who played from 1960 to 2019 and were between 33 and 60 years old at the time of testing. Each participant underwent imaging on GE Discovery MI PET/CT scanner using [^{11}C]-PiB for Amyloid-β and [^{18}F]-FTP for p-Tau. Reference region-based analysis was performed with in-house software quantifying [^{11}C]-PiB binding by distribution volume ratios (DVr) and [^{18}F]-FTP by standardized uptake value ratios (SUVr). Structural magnetic resonance imaging (MRI) was performed on a 3T Tim Trio Siemens and then processed with FreeSurfer. We compared the PET and MRI results of this population with 23 age-matched control subjects with no TBI exposure who underwent [^{11}C]-PiB and [^{18}F]-FTP scans on the same camera. **Results:** All subjects showed negative Amyloid-β burden (DVr<1.2) and no significant [^{18}F]-FTP uptake in CTE-related brain areas. However, we saw a notable uptake of [^{18}F]-FTP among football players compared to the control group in areas known for early AD stages (e.g., parahippocampal SUVr=1.01+/-0.03 vs 0.97+/-0.03; $p<0.01$ two-tailed t-test) with a significant age effect ($p<0.05$) for the football players. MRI showed decreased cortical thickness and increased white matter hyperintensities for football players with higher self-reported exposure to concussion related event. Cortical thinning could reduce the apparent uptake of PET tracers due to finite resolution of the camera, thus our findings of increased [^{18}F]-FTP binding cannot be explained by partial volume effects. **Conclusion:** p-Tau PET imaging with [^{18}F]-FTP may be useful in the investigation of repeated sport related concussion patients for early AD-like pathology. Further research is necessary to better characterize the potential and limitations of the technique.

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Poster

P351. Traumatic Brain Injury: Human Studies: Mechanisms

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P351.07

Topic: C.10. Brain Injury and Trauma

Support: Blythedale Children's Hospital

NIH Grant: P41 EB018783
Stratton Albany VA Medical Center

Title: What about the children? Cognitive motor dissociation following pediatric brain injury

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Abstract: Neuroimaging studies in adults have revealed that as many as 16% of those with severe brain injury retain high-level cognitive capacities not reflected in their clinical assessment; this has been designated cognitive motor dissociation (CMD). Pediatric medicine lacks both practice guidelines for identifying covert cognition, and epidemiologic data regarding the prevalence of CMD. To quantify cognitive function in children and adolescents diagnosed with disorders of consciousness (DoC), we developed a novel, hierarchical, pediatric-feasible, battery of quantitative neuroimaging assessments.
We evaluated cognitive function in two adolescents remaining chronically in DoC following severe brain injury. MRI and FDG-PET studies evaluated brain integrity. We assessed command-following using fMRI and EEG. We used EEG to assess selective attention to, and semantic processing of, auditory stimuli including spoken language.
Subjects were aged 15 and 18 (injured at 9 and 13 years respectively). Regional parenchymal volume loss and decreased resting cerebral metabolism were noted. Both subjects' EEG, and one subject's fMRI, provided evidence of the capacity to understand commands, make voluntary decisions to follow them, and sustain attention to the task. Both subjects' EEG demonstrated semantic processing of the meaning of words in context, and selective attention of narrative language.
Converging lines of evidence lead us to conclude that both subjects' cognitive function was dissociated from their motor output. The crucial gap in current clinical practice is vividly highlighted by the fact that, throughout the 5 years or more in which the subjects had remained in DoC, all attempts at communication and therapy had remained uninformed by any objective evidence of their cognitive abilities. Proper diagnosis of cognitive-motor dissociation is an ethical imperative. Without this, children who may have covert consciousness risk being abandoned to a lifetime of isolation. Emerging therapies and neuroprosthetics could re-establish self-determination, foster social integration, and improve interaction with caregivers to promote recovery.

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Poster

P351. Traumatic Brain Injury: Human Studies: Mechanisms

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Program #/Poster #: P351.08

Topic: C.10. Brain Injury and Trauma

Support: Blythedale Children's Hospital
NIH Grant: P41 EB018783
Stratton Albany VA Medical Center

Title: Event-related EEG markers of cognitive recovery following pediatric brain injury

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Abstract: Following brain injury, assessment of residual and emerging cognitive function is difficult and error-prone. In large part, this is because cognitive testing depends on motor output, which may be comorbidly impaired. In extreme cases, high-level cognitive function may be present but undetectable by conventional clinical testing—the term "cognitive motor dissociation" (CMD) has been used to denote such states. Identification of CMD has been associated with better clinical outcomes.
Accordingly, recent clinical practice guidelines recommend objective neuroimaging and neurophysiological measures to support diagnosis in adults. However, equivalent measures are lacking in pediatrics—an especially great challenge due to the combined heterogeneity of both brain injury and pediatric development. To address this gap, we aim to establish quantitative, objective, clinically practicable measures of cognitive function following pediatric brain injury. We recruited participants aged 8-18 with and without brain injury. Participants with brain injury were clinically classified according to cognitive recovery state: N=8 in disorders of consciousness (DoC), N=7 in confusional state (CS), and N=19 cognitively impaired (CI). In addition, N=13 typically-developing (TD) uninjured controls were included. We prospectively measured electroencephalographic (EEG) markers of sensory processing and attention in an auditory "oddball" paradigm, and of covert movement attempts in a command-following paradigm.
In three DoC participants, EEG markers of active attempted command-following revealed cognitive function that clinical assessment had failed to detect. These three individuals could also be distinguished from the rest of their group by two event-related potentials (ERPs): an auditory

evoked potential reflecting passive sensory processing, and the P3 reflecting rudimentary attention mechanisms. Across the whole cohort, the noise-corrected magnitudes of these two ERPs reflected cognitive recovery state (ANOVA: each $p < 0.001$); viewed jointly, they cleanly delineated the groups.
We conclude that objective EEG markers can be good indicators of individual cognitive recovery, even in a cohort in which there was great heterogeneity of both etiology and brain development. These markers could be measured at the bedside in a practicable way without requiring participants to make overt motor responses. They allowed us to identify, we believe for the first time in pediatrics, three individuals who meet the criteria for cognitive motor dissociation. To diagnose, prognose and track cognitive recovery accurately, such markers should be employed in pediatrics.

Disclosures: J. Hill: None. **N. Kim:** None. **W.D. Watson:** None. **E. Caliendo:** None. **S. Nowak:** None. **N.D. Schiff:** None. **S.A. Shah:** None.

Poster

P453. Retina: Photoreceptors

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Topic: D.07. Vision

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Title: Extracellular lactate as an alternative energy source for retinal bipolar cells

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Abstract: Purpose: Metabolism is a fundamental issue for cell function. Interestingly, Warburg´s studies from 1924 pointed out that tumors cells and retina are the main tissues that use aerobic glycolysis, converting almost the 90% of consumed glucose into lactate. The aim of this study is a better understanding of energy metabolism in the inner retina and a possible consumption of lactate as an alternative energy source for rod bipolar cells (RBCs). **Methods:** We used immunofluorescence staining to determine the retinal expression of enzymes involved

in energy metabolism. Retinal explants obtained from P9 wild-type C57BL/6 and C3H mice were cultured and treated with selective drugs to alter energy metabolism. The percentages of TUNEL-positive cells in the INL was quantified. Finally, retinal slices from P30 mice were used to test the role of extracellular lactate in the neuronal activity in INL cells through calcium imaging and whole-cell patch-clamp. **Results:** The results showed that photoreceptors express MCT1 in the inner segment, and in the outer plexiform layer. Immunofluorescence indicated that MCT2 was exclusively expressed in cones and abundantly expressed in the INL. Co-immunostaining in the INL indicated that MCT2 colocalizes with RBCs markers, and with amacrine cells. Other key enzymes involved in aerobic glycolysis, such as PKM2 and LDHa, were also expressed in photoreceptors. Retinal explants treated with SR-13800 (MCT1 inhibitor), AR-C155858 (MCT1 and MCT2 inhibitor) and shikonin (PKM2 inhibitor) showed increased cell death in the INL. Additionally, under the three different treatments, the RBC number decreased significantly. Interestingly, the exposure of retinal slices to these drugs decreased neuronal activity. Specifically, the replacement of glucose by lactate maintained the membrane potential in RBCs, and the amplitude in both outward current and calcium currents. However, the replacement of glucose by mannitol and the treatment with MCTs inhibitor depolarized RBCs and decreased the amplitude of the currents. **Conclusions:** These results suggest that photoreceptors and cells in the INL have different expression patterns of the key enzymes involved in aerobic glycolysis. Our data support the notion that RBCs can consume a portion of the extracellular lactate liberated by photoreceptors to meet their physiological activity. This lactate can be used to produce energy through OXPHOS.

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Poster

P453. Retina: Photoreceptors

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Program #/Poster #: P453.02

Topic: D.07. Vision

Support: Canadian Institutes of Health Research (PJT-156029)

Title: G protein-coupled receptor 55 modulates the murine retinal function

Authors: *I. BACHAND, S. RAMDANE, B. CÉCYRE, J.-F. BOUCHARD;
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Abstract: The G protein-coupled receptor 55 (GPR55) is mostly activated by lysophosphatidylinositols (LPIs), but also by endogenous and exogenous cannabinoids. The endocannabinoid ligands and enzymes, as well as the cannabinoid receptor type 2 (CB2) have

been shown to play a role in the mediation of murine retinal function and visual acuity. Moreover, GPR55 expression and effect on the retinal cell response have already been investigated in vervet monkeys. However, the expression and effects of the endocannabinoid system were shown to be very different in rodents and primates. To investigate the role of GPR55 in the mouse vision process, we evaluated the differences between knockout mice for the *gpr55* gene (*gpr55* KO) and their age-matched wild type controls (*gpr55* WT). We used a full field electroretinogram (ERG) of adult mice (n=20 mice, 3 to 4 months old) in scotopic and photopic conditions to reveal a significative difference between groups (mixed effect two-way ANOVA) for several of its graphical components. Scotopic a-wave latency and amplitude, scotopic b-wave and photopic a-wave amplitudes were significatively lower for *gpr55* KO mice. The first three main oscillatory potentials (OPs) were also lower for *gpr55* KO mice, as well as the sum of the four main OPs. Visual acuity differences during development from P15 to P50 (n=17 mice) were studied using the minimal spatial frequency capable of triggering an optomotor response. The spatial frequency threshold was also measured for adult mice between three and four months old. For both developing and adult mice, no significative differences between *gpr55* WT and KO groups were brought out. In summary, because the different components of the ERG are caused by different cell types depending on the experimental conditions, the ERG results show expression of GPR55 in a great diversity of retina cells in mice. As for the lack of effects on visual acuity, it opens the opportunity for the use of a GPR55 agonist to treat chronic photophobia without affecting vision quality otherwise.

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Poster

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Topic: D.07. Vision

Support: NIH Grant EY026817
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Title: Unraveling the roles of $Ca_v1.4$ as an organizer of photoreceptor synapses.

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Abstract: Photoreceptors capture and convert photon energy into a neuronal signal that is transferred to second-order neurons. The transfer of this visual information relies on the voltage-gated Ca^{2+} (Ca_v) channel $Ca_v1.4$ to trigger Ca^{2+}-dependent neurotransmitter release from highly

specialized ribbon synapses of rods and cones in the outer plexiform layer (OPL) of the retina. In mice that lack $Ca_v1.4$ expression, photoreceptor ribbons and synapse structure fail to develop, but whether this is due to the absence of $Ca_v1.4$ Ca^{2+} signaling or lack of $Ca_v1.4$ protein remains unknown. To dissect the synaptogenic requirements of $Ca_v1.4$, we generated mice that express a non-conducting $Ca_v1.4$ mutant (G369i KI). Remarkably, ribbons and the molecular composition of rod and cone synapses of these mice are largely intact; however, horizontal and rod bipolar neurites fail to invaginate into rod and cone terminals. Moreover, rod synapses of G369i KI mice are occasionally localized with horizontal and rod bipolar neurites that sprout into the outer nuclear layer (ONL). Although misshapen, cone photoreceptor terminals of G369i KI mice are properly localized in the OPL with little sprouting of cone bipolar cells into the ONL. Thus, while dispensable for the initial development and molecular organization of the photoreceptor synapse, $Ca_v1.4$ Ca^{2+} signaling enables the appropriate arrangement of postsynaptic partners and is differentially required for the localization of rod and cone synapses in the OPL. Our results support the hypothesis that $Ca_v1.4$ channels promote the development and function of the first synapse of the visual pathway by operating as both structural organizers and as conduits of Ca^{2+} ions.

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Poster

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Topic: D.07. Vision

Support: NIH R01 EY012146
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F31 EY031962
INBRE P20 GM103408

Title: Gene expression in cone photoreceptors of adult zebrafish is plastic to external hormone treatment

Authors: *A. FARRE[1], J. HUANG[2], A. DUNCAN[1], R. POULSEN[3], D. L. STENKAMP[1]; [1]Univ. of Idaho, Moscow, ID; [2]Univ. of Washington Sch. of Med., Spokane, WA; [3]BYU-Idaho, Rexburg, ID

Abstract: Vertebrate color vision requires the differential expression of specific cone opsins in separate cone populations. One model for the regulation of the human long and medium wavelength sensitive (*LWS/MWS*) opsin tandem array suggests an upstream regulatory region interacts with replicated opsin genes at random, resulting in mutually exclusive expression of a

specific opsin. However, our prior investigations into the orthologous long wavelength sensitive (*lws1/lws2*) array in zebrafish suggest that thyroid hormone (TH) and retinoic acid serve as trans regulators of this gene array in larvae/juveniles (Mitchell et al., 2015, *PLOS Genetics*; Mackin et al., 2019, *PNAS*). This study investigates whether cone opsin expression remains plastic to TH treatment in adult zebrafish, where cone distribution is considered stable. Adult zebrafish were treated with NaOH (0.01%, control) or TH (386 nM) for 1 or 5 days. qRT-PCR was performed on homogenized eyes, and whole retinas and cryosections were analyzed by confocal imaging. In adult zebrafish, exogenous TH drastically increased *lws1* expression in both 1 and 5 day-treated groups ($p<0.001$, 0.01, respectively) while decreasing *lws2* expression ($p<0.01$, 0.001). Other phototransduction-related transcripts (*gngt2b, rh2-1*) also demonstrated expression changes following TH treatment. Transgenic *lws* reporter fish revealed a clear switch from *lws2* to *lws1* in dorsal retina, consistent with the qPCR data. Exogenous TH induced a drastic shift from *lws2 to lws1* in adult zebrafish, consistent with previous studies of larvae and juveniles. This shift occurs as rapidly as 1 day when exposed to TH, which shows that cones remain highly plastic even into adulthood. Plasticity in spectral sensitivity (to be sensitive to higher wavelengths) in response to TH suggest a role in visual system function well into adulthood.

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Poster

P453. Retina: Photoreceptors

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McPherson Eye Research Institute (David and Nancy Walsh Family Professorship in Vision Research)

Title: Human stem cell derived retinal organoid cone photoreceptors demonstrate robust intrinsic light responses mimicking primate foveal cones.

Authors: *A. SAHA[1,3], E. CAPOWSKI[2], M. A. F. ZEPEDA[2], D. M. GAMM[2,3,4], R. SINHA[1,4,3]; [1]Dept. of Neurosci., [2]Waisman Ctr., Univ. of Wisconsin, Madison, Madison, WI; [3]McPherson

Eye Res. Inst., [4]Dept. of Ophthalmology and Visual Sci., Univ. of Wisconsin Madison, Madison, WI

Abstract: Title: Human stem cell derived retinal organoid cone photoreceptors demonstrate robust intrinsic light responses mimicking primate foveal cones.
Abstract: In human and non-human primates, the high definition central vision originates at a highly specialized, tightly packed, dense pixel array of cone photoreceptors, called the fovea. Death of the foveal cone photoreceptors is the ultimate cause of vision loss in retinal degenerative diseases like age related macular degeneration – a leading cause of blindness in the world. Although there are no current treatment options available, 3D retinal organoids (ROs) derived from human pluripotent stem cells (hPSCs) show much promise to model and treat such diseases. However, RO photoreceptors haven't been able to elicit light evoked electrical responses comparable to primate cone photoreceptors to date. Here we used patch clamp electrophysiology to record light evoked responses from mature stage 3 organoid cones (OCs, differentiated 240-270 days in vivo) and compared their responses and membrane properties to primate foveal cones (FCs). 35% of OCs elicited robust, graded, wavelength-sensitive light-evoked responses across several hPSC derived organoids without the addition of any external chromophore. RO cones of different spectral sensitivities could be identified, with most of them being mid-wavelength sensitive. Light-evoked voltage responses of a RO cone displayed the key features of a typical photoresponse measured from primate FCs – hyperpolarization in response to onset of light stimulus and repolarization at the cessation of the stimulus. The OCs were also able to demonstrate adaptation at higher background luminance as well as to increasing intensities of light flashes, comparable to a primate FC. The size and kinetics of light evoked responses from OCs show variability within and across organoids we recorded from. OCs also replicate some of the membrane properties of primate FCs and express HCN channels in their outer segments, crucial for shaping the light evoked responses. Overall, our results provide evidence of intrinsic light responses of OCs at physiological light levels which helps to establish the hPSC OCs as important sources of functional human cone photoreceptors for use in stem cell-based transplantation therapies and as model system to study retinal degenerative disorders.

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Poster

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Title: The role of microRNA-150 (miR-150)-ETS transcription factor ELK1 (Elk1) axis in the diabetic retina

Authors: F. YU, *G. KO;
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Abstract: Diabetic retinopathy (DR) is a leading cause of blindness in US adults, and retinal inflammation is observed in the development of DR. As microRNAs (miRs) represent a set of regulators that impact metabolism and inflammation and serve as possible biomarkers for DR, it is not clear how miRs might be involved in the pathogenesis of DR. Among the miRs, decreased miR-150 is observed in the diabetic patients and animals, and miR-150 is known as an endogenous inhibitor of inflammation and angiogenesis. We previously showed that deletion of miR-150 further exacerbates retinal inflammation and microvascular degeneration in obesity-associated type 2 DR. However, the link between miR-150 and retinal inflammation under diabetic insults is not clear. In this study, we used the 661W cells (a mouse photoreceptor cell line) and miR-150 knockout (miR-$150^{-/-}$) mice to decipher the downstream target of miR-150 in obesity-associated T2D. The mice were fed with a high fat diet (HFD) to induce T2D, and the 661W cells were treated with palmitic acid (PA) to mimic the HFD condition in mice. In cultured 661W cells, we found that knocking down miR-150 exacerbated the PA-induced increase of phosphorylated P65 (pP65), an inflammation marker. Using a functional assay with 661W cells, we found that ETS transcription factor ELK1 (Elk1), a bona fide target of miR-150, was consistently elevated while miR-150 was decreased in PA-treated cells, but other known miR-150 targets were not. Knocking down Elk1 alleviated the PA-induced increase of pP65. In miR-$150^{-/-}$ mice with HFD-induced T2DR, they had a significantly higher retinal inflammation that correlated with a higher retinal expression of pELK1 compared to the wild type (WT) mice with HFD-induced T2DR. Thus, decreased miR-150 exacerbates diabetes-induced retinal inflammation by upregulating Elk1.
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Poster

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Title: Cultured hiPSC-RPE cells exhibit increased resistance to oxidative stress triggers throughout differentiation and maturation

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Abstract: Human induced pluripotent stem cell-derived retinal pigment epithelium (hiPSC-RPE) cells more closely exhibit the human RPE than existing RPE-derived cell lines, making them a promising tool for drug discovery and development for ophthalmic conditions affecting the RPE, including age-related macular degeneration and geographic atrophy. However, there is a lack of rigorous scientific data regarding the response of hiPSC-RPE cells to oxidative stress triggers. The purpose of this study was to determine the susceptibility of hiPSC-RPE cells to exogenously applied oxidative stress (*tert*-butyl hydroperoxide; *t*BHP) and an oxidizing agent (sodium iodate; $NaIO_3$) that triggers intracellular oxidative stress and to investigate their effects on hiPSC-RPE differentiation and maturation.HiPSC-RPE cells were seeded on Matrigel®-coated 96-well cell culture plates and differentiated according to the manufacturer's instructions. After varying amounts of differentiation time *in vitro* (days *in vitro*, DIV), cells were challenged with either *t*BHP (0.1 to 1.0 mM) or $NaIO_3$ (0 to 18 mM). Cell viability was assessed by resazurin assay and lactate dehydrogenase (LDH) release assay. Reactive oxygen species (ROS) generation was quantified by chloromethyl 2',7'-dichlorodihydrofluorescein diacetate (CM-H_2DCFDA) assay and vascular endothelial growth factor (VEGF) release was quantified by ELISA. HiPSC-RPE cells exhibited a dose-response to increasing doses of *t*BHP and $NaIO_3$. In *t*BHP experiments, IC_{50} values for tBHP from resazurin assay were ~0.4 mM from DIV13 to DIV19, ~0.7 at DIV26 and ~0.9 mM at DIV28. LDH release showed a similar shift (EC_{50} values for *t*BHP: range 0.4 mM to 0.9 mM). ROS generation was significantly attenuated at DIV26 and DIV28, which was positively associated with VEGF release. Similarly, $NaIO_3$ induced a dose-response in the cells with IC_{50} (resazurin assay) and EC_{50} (LDH assay) values of 7.8 mM. $NaIO_3$ elicited a significant 26-fold increase in ROS generation at the lowest dose of 2 mM $NaIO_3$, which slowly decreased with higher doses, likely attributable to a dose-dependent toxicity. Our data suggest that hiPSC-RPE cells continue to mature over time *in vitro*, manifesting as increasing resistance to exogenously-applied oxidative stress. Differences in the correlation between insult (tBHP *vs*. $NaIO_3$) and cell viability are likely due to mechanistic differences in the generation of intracellular *vs*. exogenously-applied oxidative stress. HiPSC-RPE cells exhibit a strong endogenous antioxidant system and potent VEGF response, making them a useful model for *in vitro* applications.

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investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; AcuiSee, Inc.. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Experimentica Ltd., K&P Scientific. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Experimentica Ltd., K&P Scientific LLC, eyeNOS, Inc. F. Consulting Fees (e.g., advisory boards); Experimentica Ltd..

Poster

P453. Retina: Photoreceptors

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P453.08

Topic: D.07. Vision

Title: Morphological homologies in diurnal and nocturnal Lepidopteran eyes

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Abstract: The number of photons available during the day versus the night varies by orders of magnitude. It is therefore reasonable to hypothesize that there are evolved differences in eye structure of species active at different times of day. Ommatidial diameter, interommatidial angle, and resolving power have been measured in some species of bees and parasitic wasps. It was found that there was a square root relationship between the height of the eye and the diameter of the average ommatidium, and the ratio of the interommatidial angle to resolving power was between 0.4 and 1 in the regions of highest acuity in the eye. However, these finding only relate to diurnal species. We use different species of moths and butterflies that can either be grouped as diurnal, nocturnal, or crepuscular to determine if these metrics match those found in wasps and bees and whether they differ among species in different light environments. Surprisingly, using synchrotron x-ray data, we found that the relationship of the height of the eye to the ommatidial diameter matched the trend seen in wasps and bees except for diurnal species. We also found that the interommatidial angle to resolving power ratio was significantly greater than 1, regardless of the level of acuity, and the metrics we measured did not differ significantly between groups. These findings suggest the values measured are optimal for all Lepidopterans and that the correlation structure of natural scenes might set this general eye structure to support effective vision independent of light levels.

Disclosures: D. Paukner: None. **G. Wildenberg:** None. **G. Badalamente:** None. **P. Littlewood:** None. **N.B. Kasthuri:** None. **S.E. Palmer:** None.

Poster

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Topic: D.07. Vision

Support: NIH Grant EY015573
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Title: Vamp1 immunoreactivity form ribbon-like structures in photoreceptor terminals in mouse retina

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Abstract: The cellular mechanisms and synaptic protein isoforms underlying synaptic vesicle release from the photoreceptor ribbon synapse remain incompletely understood. The mammalian photoreceptor releases the neurotransmitter glutamate in a continuous fashion onto bipolar and horizontal cells. The synaptic vesicle protein, VAMP1/synaptobrevin, forms part of the core SNARE complex that mediates vesicle fusion with the plasma membrane. The expression of VAMP1 was studied in lightly fixed mouse retina using immunohistochemistry, super-resolution confocal microscopy, conical EM tomography, and RNAscope *in situ* hybridization. VAMP1 immunolabeling formed crescents in the outer plexiform layer, where the photoreceptor synaptic terminals are located. The VAMP1 immunolabeling was observed in both rod and cone photoreceptor terminals. Double labeling with antibodies to PSD-95 confirmed that the ribbon-like VAMP1 immunolabeling occurred within photoreceptor terminals. Double labeling with CtBP2/ribeye antibodies showed that the VAMP1 label formed a nested crescent inside the arch of the synaptic ribbon. Double labeling with bassoon antibodies, which is thought to label the base of the ribbon, demonstrated that there was a significant overlap between VAMP1 and bassoon immunolabeling. To assess if VAMP1 co-localized with horizontal cells, double immunolabeling for calbindin indicated that VAMP1-immunoreactive structures were closely apposed to horizontal cell endings, but extended beyond the horizontal cell endings. Consistent with VAMP1 localization to photoreceptors, VAMP1 mRNA is expressed in the outer nuclear layer. Conical EM tomography of mouse rod photoreceptor terminals revealed a row of synaptic vesicles at the base of the synaptic ribbon. Taken together, we propose that the VAMP1 immunolabeling observed may correspond to this pool of synaptic vesicles at the base of the synaptic ribbon.

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Poster

P453. Retina: Photoreceptors

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Program #/Poster #: P453.10

Topic: C.07. Neurotoxicity, Inflammation, and Neuroprotection

Title: Disruption of retinal layers by laser photocoagulation affects Visuospatial memory and visual functions.

Authors: *P. MEHRA[1], P. BALI[3], J. SINGH[4], P. K. SAHA[2], A. ANAND[1];
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Abstract: Disruption of retinal layers by laser photocoagulation affects Visuospatial memory and visual functions. Priya Mehra[1, 2], Parul Bali [3], Jagtar Singh[1], Pradip Kumar Saha[4], Akshay Anand[2*1]Department of Biotechnology, Panjab University, Chandigarh, 160014, INDIA[2]Neuroscience Research Lab, Department of Neurology, Post Graduate Institute of Medical Education & Research, Chandigarh, 160012, INDIA[3]Department of Biological Science, Indian Institute of Science Education and Research (IISER), Mohali, INDIA[4]Department of Obstetrics and Gynecology, Post Graduate Institute of Medical Education and Research, Chandigarh-160012, INDIA
AbstractVisual pathway involves neuronal activity between presynaptic and post synaptic pathway to generate action potential to amplify the signal resulting in visual image formation. Although, various studies have been carried out to reverse the injury but these have not clinically translated successfully. This study was conducted to evaluate the effect of injury on visual spatial memory by establishing a laser injury model and performing neurobehavioral tests (Morris Water Maze and Passive Avoidance). *C57BL/6J* male mice were used for the study. The laser spots with defined parameters were given to the mice retina around the optic disc using laser photocoagulation that disrupted the RPE layer of the retina. Fundus Fluorescein Angiography confirmed the establishment of animal model. Subsequently, visual-spatial memory was analysed by neurobehavioral tests and short-term memory by using Morris Water Maze (MWM) and Passive Avoidance. In neurobehavioral experiments, we found that escape latency time in both healthy and eight laser groups was comparable. In the passive avoidance test, the latency time was non-significant between the healthy and laser groups. This shows that visuospatial memory may not get affected by retinal injury induced by laser photocoagulation. It may depend upon the intensity of the laser and the duration of the laser for

which additional experiments are required. **Ethical Statement-** Study was conducted after taking ethical approval from Institute Animal Ethical committee. The ethical approval number is 105/IAEC/720.**Conflict of Interest-** None

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Disclosures: P. Mehra: None. **P. Bali:** None. **J. Singh:** None. **P.K. Saha:** None. **A. Anand:** None.

Poster

P487. Representation of Faces and Bodies III

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CEA PE Bottom Up 2020 (20P28)

Title: Fmri-guided electrophysiological characterization of macaque orbitofrontal face patches

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Abstract: fMRI studies in primates revealed face-category selective activations in prefrontal areas [Tsao et al., 2008]. Electrophysiological studies targeting orbitofrontal cortex reported neurons responding more to faces compared to objects. The latter face-selective neurons often convey social and emotional information [Barat et al., 2018]. However, the relationship of these orbitofrontal face selective neurons with respect to fMRI-defined face patches is not clear. Using fMRI-guided laminar recordings in macaques, we characterized the neuronal responses of 2 face patches within the orbitofrontal sulcus, i.e., face patch POa which is located rostro-lateral relative to a second orbitofrontal face patch POp. Extensive sets of images of bodies, animals, objects, and faces varying in position, viewpoint, and facial expression were presented to two

fixating rhesus monkeys. In both orbitofrontal face patches of both monkeys, we found on average only 1 out of 3 visually driven cells to be face selective (79/100 out of 217/265 cells in POp and 163/120 out of 504/406 cells in POa for monkey F and L, respectively). Surprisingly and contrary to previous studies in which face cells were recorded in orbitofrontal cortex without fMRI guidance [Barat et al., 2018], the face cells in POa and POp were not modulated by the emotional content of the images. However, many of the face cells in both areas were sensitive to the viewpoint of the faces. The selectivity of these neurons also showed a high degree of tolerance to changes in position of the faces. On population level the receptive fields of face neurons in both face patches covered large eccentricities, yet with a strong foveal bias, and a relatively small contralateral bias. Overall, our data provide a more comprehensive view on the properties of face-selective neurons within fMRI-defined orbitofrontal face patches POa and POp.

Disclosures: E. Mergan: None. **Q. Zhu:** None. **X. Li:** None. **R. Vogels:** None. **W. Vanduffel:** None.

Poster

P487. Representation of Faces and Bodies III

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Title: Mapping Dynamic Body Patches in Macaque Inferotemporal Cortex

Authors: M. BEGHELLA BARTOLI[1,2], ***R. RAMAN**[1,2], N. TAUBERT[3], Y. ZAFIROVA[1,2], B. DE GELDER[4], M. GIESE[3], R. VOGELS[1,2];
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Abstract: Visual information from body movements is an imperative element of non-verbal social communication and action recognition. Since previous fMRI studies employed static images to identify patches in the macaque inferotemporal (IT) cortex that were activated more strongly by bodies compared to faces and objects, the organization of patches that are activated by dynamic bodies is still unclear. In this fMRI study, we investigated the neural substrate underlying the visual processing of dynamic bodies in the macaque's brain. We measured the response to a set of 1 s long videos of dynamic monkey bodies, dynamic monkey faces, and

dynamic artificial objects. The body videos displayed natural actions of a monkey with its head blurred, while in the face videos a monkey displayed various expressions or turned its head. The actions and objects were presented on a dynamic white noise background. In addition, we presented mosaic scrambled versions of these videos, keeping the local motion of the full video displays. For scanning, we employed a block design during passive fixation. Dynamic body patches (bodies – objects) were present in the dorsal bank, fundus and upper bank of the superior temporal sulcus (STS), and ventral to the rostral STS. The dynamic body and face (faces – objects) patches were close to each other. The most posterior body patch was located in or close to the Fundus of the Superior Temporal (FST) area. More anterior ones coincided with the patches previously defined with static images of bodies: MSB and ASB in the ventral bank of STS (Kumar et al., Cerebral Cortex, 2019), and one in ventral IT, dorsal to face patch AM (Bao et al., Nature, 2020). In addition, several body patches were present in the fundus and dorsal bank of the rostral STS, neighboring face patches MF, MD and AF. Dynamic body patches were also observed in the frontal cortex and in 2/4 animals weakly in the parietal cortex. In some monkeys, weak activations to the scrambled body videos were present in the dynamic body patches, suggesting responses to local features. This fMRI mapping study provides a roadmap of the network of dynamic body patches in the macaque brain, which are currently targeted with single-unit recordings.

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Poster

P487. Representation of Faces and Bodies III

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Topic: D.07. Vision

Support: ERC Grant 856495

Title: Hemisphere-specific coding of left vs. right viewpoint for perception of the human body in lingual gyrus.

Authors: *G. MARRAZZO[1], M. VAESSEN[1], F. DE MARTINO[1,2], A. LAGE-CASTELLANOS[1,3], B. DE GELDER[1,4];
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Abstract: Visual information from body movements conveys crucial information about emotions, intentions and actions of conspecifics. Our current understanding of body posture and movement shows a gap between processing of low-level features and high-level semantic categories (de Gelder & Poyo Solanas 2021). An important intermediate level processing stage in models of object perception is viewpoint (in)tolerance. Viewpoint sensitivity is of special importance for body perception because self vs. other oriented body images may have a very different behavioral impact on the viewer which stresses the importance of maintaining viewpoint tolerant representation. Previous studies showed that EBA, but not the FBA, is sensitive to the orientation of the body (Lawson et al. 2009; Kumar et al. 2019). In this fMRI study we investigated the brain representation of differences in viewpoint on body perception. Stimuli were generated using a variational autoencoder (VAE), via a random sampling of the latent space parameters, depicting 3D rendered body poses. We used 108 randomly generated unique poses from 3 different viewpoints (frontal view, 45° rotation to the right and to the left respectively), for a total of 324 stimuli. In a behavioral experiment participants (N=54) evaluated (using a Multi-arrangement method, RSA and features survey) the similarities between all stimuli including triplets (same stimulus from 3 viewpoints). For the fMRI experiment participants were scanned using a 7T (T2*-weighted Multi-Band accelerated EPI 2D BOLD sequence, MB = 3, voxel size = 1.6 mm^3, TR = 1000 ms, TE = 20 ms) in a fast event-related design over 12 separate runs. Each run consisted of 54 unique stimuli (18 unique poses x 3 viewpoints) which appeared on the screen for 750 ms. Participants were asked to fixate and attention was controlled with a one-back task (6 trials per run) in which a button press was required if following a specific visual cue, the current stimulus was the same as in the previous trial. For the univariate analysis, data were combined in a fixed effect GLM in which 3 regressors of interest were generated representing the 3 viewpoints conditions. The contrast between opposite viewpoints (-45 vs +45) showed activation in the lingual gyrus with opposite pattern in left and right hemisphere as higher activation was found for the left rotated stimuli from the observer point of view in the right hemisphere whereas higher activation for the right rotated stimuli from the observer point of view in the left hemisphere. These results have implications for understanding the importance of viewpoint specific perception of human bodies and its special role in hierarchical models of object perception.

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Poster

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Title: Physiologically-inspired neural model for the visual recognition of dynamic bodies

Authors: P. KUMAR[1], N. TAUBERT[1], R. RAMAN[2,3], R. VOGELS[2,3], B. DE GELDER[4,5], *M. A. GIESE[1];
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Abstract: The recognition of bodies and body movements is essential for the survival of many social species. The detailed underlying neural circuitry is not yet fully understood. We present a physiologically-inspired hierarchical neural model for the recognition of body movements for comparison with electrophysiological data from monkeys and human fMRI data. METHODS: The model combines a feed-forward deep neural network (VGG-19) with a neurodynamical model that reproduces responses at the single-cell level in the superior temporal sulcus (STS) and premotor cortex (Giese and Poggio, 2003). The initial parts of the visual pathway were modeled by the layers up to the MaxPool4 layer of VGG-19 (pre-trained on the ImageNet database). Output features were reduced by eliminating features with low variance and applying PCA. The reduced feature detector responses provided input to RBF units (snapshot neurons) which were trained with individual keyframes of the actions, and which were embedded in sequence-selective recurrent neural networks (neural fields) with asymmetric lateral connections (Giese and Poggio, 2003; Fleischer et al., 2013). Neural fields encoding different actions mutually inhibit each other, resulting in a competition between the different learned actions. The outputs of the individual neural fields are summated by motion pattern neurons that are activated by only one of the learned actions. We tested the model using videos of macaques and humans performing different types of actions. Similar movies of macaques are presently being used in physiological experiments and fMRI experiments on body motion encoding. RESULTS: The model successfully recognizes the actions from real videos. The snapshot neurons show a traveling pulse of activity that encodes the corresponding pattern. The motion pattern neuron responses clearly differentiate between the different encoded actions. Consistent with the behavior of real neurons in the inferior temporal cortex (IT), the model trained with naturalistic stimulus movies generalizes spontaneously to silhouette stimuli. The model makes predictions about the responses of different neuron classes along the hierarchy, which are presently being compared to recordings from the macaque visual cortex and to human fMRI data.

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Poster

P487. Representation of Faces and Bodies III

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Program #/Poster #: P487.05

Topic: D.07. Vision

Title: Investigating the mental representation of observed grasp movements through similarity judgements

Authors: *L. J. ETTENSOHN, K. WOODARD, L. G. UNGERLEIDER, C. I. BAKER, M. VAZIRI PASHKAM;
Lab. of Brain and Cognition, NIH, Bethesda, MD

Abstract: Hand grasp movements constitute a large portion of human's everyday activities. In addition to performing hand grasp movements ourselves, we regularly observe others grasping objects and can judge these grasps by their similarity. What are the core features relevant for similarity judgements of observed grasp movements? Do people's similarity judgements of grasp movements match objective grasp similarities based on kinematics of the movement? To answer these questions, we first conducted a hand grasp movement experiment. We placed 16 sensors on each participant's (n=14) hand and measured their movements in 3 dimensions as they grasped a set of 58 naturalistic 3D printed objects (e.g. spoon, teapot, teddy bear). Using these kinematic data, we calculated the similarity between grasp movements and constructed a similarity matrix based on objective measurements. Next, we used the kinematic data to create point-light videos to serve as stimuli in our second experiment. Our second experiment was an odd-one-out similarity judgement task. Participants on Amazon Mechanical Turk identified the most distinct grasp among triplets of the grasp videos (130 data points were collected for each pair). These judgements were used to construct a subjective similarity matrix. Both the objective and subjective similarity matrices had high reliabilities ($r^2 > 0.7$). Surprisingly, the correlation of these two matrices was ~0.37: significantly above zero ($p < 0.001$) but also significantly below the noise ceiling ($p < 0.001$). These results demonstrate that humans rely on systematic features when judging the similarity of grasp movements, but these subjective judgements do not mirror the objective similarity of grasp kinematics. These results suggest that distinct neural representations may support action observation and action execution.

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Poster

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Program #/Poster #: P487.06

Topic: D.07. Vision

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Title: Dynamic Body Feature Processing and species-specific body patches in the human brain

Authors: B. LI[1], M. POYO SOLANAS[1], G. MARRAZZO[1], R. RAMAN[2,3], R. VOGELS[3,2], N. TAUBERT[4], M. A. GIESE[4], *B. DE GELDER[1,5];
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Abstract: Like face perception, body recognition is a fundamental social skill for many animals including primates. While several body-selective regions have already been identified in both humans and nonhuman primates, there is no comprehensive picture of which regions are specifically involved in dynamic body perception and whether they are species specific. Interestingly, homologous face and body patches mainly along the superior temporal sulcus (STS) have been observed in monkeys (Tsao et al. 2003) leaving open questions about species specificity of social signals (Qi et al. 2012). This high-field fMRI 7T ($1.6mm^3$ isotropic, TR = 1s) study investigated the neural correlates of dynamic human and monkey bodies and faces in human participants. Stimuli consisted of 1-s videos of dynamic faces and bodies of both monkeys and humans (neutral, face information removed), as well as dynamic artificial objects (e.g. machine parts). A scrambled version of each category was used to control for motion and low-level visual features. The experiment used a block design and a fixation change based oddball task. Several body patches were found when contrasting human bodies to objects including the extrastriate body area, STS, fusiform body area, occipito- and temporal-parietal junction, premotor cortex, inferior frontal cortex and anterior cingulate extending previous findings (de Gelder & Poyo Solanas 2021). The STS patches were clearly separated into anterior, middle and posterior parts. A contrast between human faces and objects revealed a large overlap between the STS body patches and face patches, especially for posterior and middle parts. Human bodies compared to monkey bodies elicited more activity in the ventral and dorsal premotor cortex, inferior frontal cortex, extrastriate body area and fusiform body area. In addition, we saw higher sensitivity to human bodies in the middle and posterior STS, even after controlling for low-level features. The STS patches corresponded to those found in monkey studies (Beghella Bartoli et al. 2021) suggesting a species continuity in the representation of social stimuli in the brain. However, here, the STS body patches were more selective for human bodies than monkey bodies indicating a species-specific coding in those areas.

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Poster

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Title: Face neurons see Waldo

Authors: M. J. ARCARO[1], *M. LIVINGSTONE[2];
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Abstract: Inferotemporal cortex (IT) contains neurons that respond selectively to complex visual images. These neurons are thought to be critical for our ability to detect and classify things (e.g., faces) in our environment. The speed and accuracy with which we recognize faces is impaired when faces are embedded within cluttered environments. Here, we explored the impact of clutter on response properties of IT neurons.
We recorded from microelectrode arrays implanted in fMRI-defined posterior, middle, and anterior lateral face patches across four adult monkeys. We probed responses to faces embedded within visually cluttered and uncluttered scenes. Cluttered scenes comprised either non-faces (e.g., a face within a pile of rocks) or other faces (e.g., a monkey face within an array of human faces). To measure spatially specific responses across each scene, we presented each image centered at different positions relative to each neuron's receptive field such that the neuron's receptive field sampled all parts of each image across the experiment. We assessed perceptual awareness of the cluttered faces with a preferential looking paradigm. Neurons responded strongly when faces in both cluttered and uncluttered scenes were presented within their receptive fields. Responses were spatially focal to faces across a variety of retinal sizes and even when presented briefly (32-100ms). Only when faces were embedded within a dense array of other faces did responses become erratic. Together, these results demonstrate that cluttered environments had a minimal impact on the spatial and feature specificity of these neurons. In contrast to the neural recordings, the monkeys did not appear to be aware of faces in clutter. When presented in isolation or in uncluttered scenes, the monkeys preferentially looked at faces. In contrast, the monkeys did not look at faces in cluttered scenes where the clutter comprised non-faces, even for presentations several times longer in duration (500ms) than the neural

recordings. Only when scenes were presented for even longer epochs (2000ms) where they were able to scan the image, did the monkeys look at the cluttered faces. These results indicate that cluttered environments impaired the monkeys' ability to perceive faces without substantially affecting the response properties of face-selective neurons in IT. The spatial and feature tuning of IT neurons under visual clutter illustrates the strong tie of these neurons to the retinal input. Thus, while IT neurons may be necessary for and modulated by the perception and recognition of faces and objects, our results indicate that downstream areas that receive input from IT may be the 'secret sauce' to perception.

Disclosures: M.J. Arcaro: None. **M. Livingstone:** None.

Poster

P487. Representation of Faces and Bodies III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P487.08

Topic: D.07. Vision

Support: NIH Intramural Program ZIA MH002898

Title: Tuning for Absolute Size in Macaque Anterior Fundus Face Patch

Authors: *A. P. KHANDHADIA[1], A. P. MURPHY[2], K. W. KOYANO[3], E. ESCH[4], D. A. LEOPOLD[4];
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Abstract: Primates use vision to guide their interactions with objects and conspecifics in the three-dimensional physical world. However, study of object related areas of the ventral visual pathway of the cerebral cortex have focused on questions regarding spatially invariant object recognition. Relatively few studies have asked how neurons in object-specialized areas are affected by geometric properties of a scene, including the absolute sizes and distances of objects. Here, we asked to what extent the brain is sensitive to the 'real-world' 3D geometry of objects, as opposed to their 2D retinal subtense. We recorded from isolated neurons in the macaque anterior fundus face patch while stereoscopically presenting 3D faces and objects, which permitted systematic variation of virtual object size and distance. The results revealed neural sensitivity to properties of real-world geometry, while retinal subtense accounted for relatively less variation in neural response. Neurons were tuned for the absolute size of heads. Surprisingly, while real macaque heads are typically ~13 cm in width, 45.5% of face patch neurons responded most strongly to most extreme sized heads, which in our stimulus set were very small (~1cm) or

very large (~26 cm). Across the population, 70.5% of neurons responded most strongly to faces in the largest or smallest quartile. This sensitivity to for extreme sizes was not unique to faces but also observed for a variety of non-face objects. Together, these results demonstrate that object-selective neurons in ventral visual cortex are highly sensitive to the 3D geometry of objects within an environment. The findings suggest that high-level visual object representations in the brain are not abstracted from physical space but integrated with it, rendering them directly useful for a range of actions and judgements.

Disclosures: A.P. Khandhadia: None. **A.P. Murphy:** None. **K.W. Koyano:** None. **E. Esch:** None. **D.A. Leopold:** None.

Poster

P525. Hand Control: Physiology and Pathology

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P525.01

Topic: E.04. Voluntary Movements

Support: Eunice Kennedy Shriver National Institute of Child Health and Human Development, HD090126

Title: Assessing finger individuation abilities in cerebral palsy using a 3-dimensional force device

Authors: *B. CRABTREE[1], O. A. KHAN[1], S. V. WILHOITE[1], J. PASCAL[1], P. R. IYER[1], G. COLQUITT[2], C. M. MODLESKY[1], J. XU[1];
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Abstract: Children with Cerebral palsy (CP) often demonstrate impairments in fine motor ability that are associated with poor selective motor control (SMC). Existing clinical assessments of SMC in CP are often subjective, require highly experienced examiners, and lack the granularity of functional kinematics/kinetics to precisely identify fine motor control deficits. Here we used a novel device, Hand Actuation Neuro-training Device (HAND, JHU reference #C14603), that tracks subtle isometric forces at all five fingertips in three dimensions (3D). We tested CP and typically developed (TD) children's finger individuation, the ability to move one finger without moving others. The purpose of the study was to accurately assess individuation deficits for children with CP.
Sixteen children with CP (ages 5 -16) and seven age-matched TD children participated in the study. Participants placed their hand in the device (more affected for CP, dominant for TD) in a comfortable, pronated, resting posture. They were instructed to control a dot's movement along each axis of a virtual 3D space on a computer screen by exerting isometric forces with one finger

(max=10N) while keeping all the other fingers immobile. Movement of the dot along the virtual XYZ axes reflected forces exerted by metacarpophalangeal (MCP) ab/adduction, proximal interphalangeal joint (PIP) flexion/extension, and MCP flexion/extension, respectively. Force trajectories were recorded from all five fingers during the task. From these trajectories, Individuation Indices were derived as the -log(slope) of the function relating non-instructed finger forces and forces in the instructed finger's target direction.
Linear mixed effect models were used to test the difference across CP and TD groups. Overall, we found that the average Individuation Index was higher for TD children (M = 0.16, SD = 1.08) than CP children (M = -0.42, SD = 1.14) ($\chi 2$ = 24.38, p = 7.9e-07). The Individuation Index among all participants was the highest in digit F1 (M = 0.384, SD = 1.08) and the lowest in F4 (M = -0.87, SD = 0.86); this had an interactive effect with the patient group ($\chi 2$ = 141.81, p = 2.2e-16). Individuation in movement across participants was the highest in finger flexion (M = -0.10, SD = 1.17) and the lowest in finger ab/adduction (M = -0.55, SD = 0.94); movement direction also interacted with group ($\chi 2$ = 83.21, p = 2.2e-16). These results suggest the device and paradigm are highly sensitive in detecting the distinct impairment patterns of finger individuation in CP children. This approach has the promise of providing detailed and precise assessment of the hand function in children with CP to inform future evidence-based targeted treatment.

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Poster

P525. Hand Control: Physiology and Pathology

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Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P525.02

Topic: E.04. Voluntary Movements

Support: PSC-CUNY 62793-00 50

Title: Identifying mechanical biomarkers of peripheral neuropathy via machine learning method in carpal tunnel syndrome applications

Authors: *W. ZHANG[1], Y. WEI[3], F. GU[2];
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Abstract: Peripheral neuropathy that interrupts sensorimotor integration in the motor control process will lead to hand and upper limb motor function deficits in daily life. However, behavioral biomechanics and motor functions have never been considered in available diagnosis and clinical evaluation. Previous studies that investigate behavioral biomechanics to delineate

specific peripheral neuropathy and its severity have shown evidences that certain biomechanical parameters have the potential to be identified as biomarkers for the detection of the neuropathy from an early stage. Nevertheless, datasets formed by behavioral biomechanical parameters are often characterized by high dimensionality, small sample size, and high redundancy, which brings us challenges for making binary classification between patients and healthy controls. In this study, we propose a two-step machine learning protocol using random forests for early variable screening and (K)PCA-SVM system for final variable interpretation and classification prediction. We suggest determining the hierarchy between feature groups by analyzing the distribution of the variable importance for early elimination of redundancy. The result of the application of our protocol in the carpal tunnel syndrome dataset show better rationality in the interpretation of variable selection and higher accuracy in prediction when compared to a single application of random forests. The final prediction accuracy reaches 90.3% when 6 variables are selected. The choice of variables implies that kinetic variables from all three-dimensional directions possess an aspect of importance in the reflection of disease. It also suggests that digits should be grouped for future studies and analysis according to their major function in grasping or lifting. Further experiments and tests should be designed with specifications to collect supplement biomechanical parameters so that better classification could be made for the detection of diseases in an early stage. The result of its application in our example dataset provides us guidelines and directions for future clinical test designs in the aim of optimizing disease diagnosis.

Disclosures: W. Zhang: None. **Y. Wei:** None. **F. Gu:** None.

Poster

P525. Hand Control: Physiology and Pathology

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Program #/Poster #: P525.03

Topic: E.04. Voluntary Movements

Support: NIH C-STAR, NICHD Grant Number P2CHD101899

Title: A Novel Method for the Assessment of Precision Force Control in Children

Authors: *V. L. ROSE[1], A. R. AJOY[1], G. R. GOGOLA[2], P. J. PARIKH[1];
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Abstract: Hand sensorimotor function depends on intact sensory and motor systems, but also on sensorimotor integration to successfully handle objects. Sensorimotor integration is responsible for critical aspects of manual function such as precise control of force magnitude and direction exerted by each digit on an object. This is important during daily tasks which require dexterity.

Clinically, the measurement of hand function identifies children with developmental pathology, guides treatments, and quantifies post treatment progress. Current tests are time-based measures and thus assess only a subset of functional abilities needed for manual activities. Therefore, these tests might not be sensitive enough to detect subtle changes in dexterity following an intervention. There is a need for a test designed specifically for children to measure the quality and control of movement (i.e., precision force control). To meet this challenge, we developed a novel device to comprehensively assess hand sensorimotor function by considering precision control of digit forces (i.e. total force) exerted on the object in addition to a time-based measure (trial duration). Children are asked to move a small object from one location to another and perform this task under three difficulty conditions: low, medium, and high. Our aim is to assess interrater reliability of the device. We have recruited 9 typically developing children aged 4-15 years. To assess interrater reliability, two independent researchers analyzed the output of the new device, and a third researcher calculated intraclass correlation coefficients (ICC). For the trial duration measure, we found that the ICC (2,1) between the 2 raters (absolute agreement) was 0.97 for the low difficulty condition, 0.99 for the medium difficulty condition, and 0.99 for the high difficulty condition. For the total force measure, we found that the ICC (2,1) between the 2 raters was 0.99 for the low difficulty condition, 0.99 for the medium difficulty condition, and 0.98 for the high difficulty condition. Our findings provide preliminary evidence of high interrater reliability for our new device that measures precision force control for a wide age range of children and adolescents.

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Poster

P525. Hand Control: Physiology and Pathology

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Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P525.04

Topic: E.04. Voluntary Movements

Title: Assessing the control of residual muscles in children with congenital upper limb deficiencies

Authors: *J. FITZGERALD[1], M. BATTRAW[2], A. BAGLEY[3], M. JAMES[3], J. SCHOFIELD[2], W. M. JOINER[1];
[1]Neurobiology, Physiology, and Behavior, [2]Mechanical and Aerospace Engin., UC Davis, Davis, CA; [3]Shriners Hosp. for Children Northern California, Sacramento, CA

Abstract: Recently, ultrasound imaging coupled with machine learning has been used to quantify spatiotemporal tissue deformation patterns of the residual muscles in adults with acquired below-the-elbow limb loss. However, the pediatric population is more likely to present with a congenital upper limb deficiency (ULD). Currently, it is unknown to what extent these

methods can be applied to the pediatric population. Furthermore, there is a lack of understanding on the extent to which pediatric patients can volitionally contract residual muscles to a desired state, at will and reproducibly. This includes both static states (e.g., power grasp or index pinch) and proportional control where the hand is partially closed. Here, we investigated the sensorimotor control capabilities of residuum muscles in children with unilateral congenital below-the-elbow ULDs. We examined whether their control was sufficiently robust and reliable for a machine learning algorithm to accurately classify multiple tissue deformation patterns when children attempted to perform distinct hand or wrist movements. Patients performed a minimum of 5 trials of six hand or wrist motions with both their affected and unaffected limb. A K nearest neighbors (KNN) algorithm was used to classify the spatiotemporal tissue deformation patterns. We found that all patients were able to execute unique tissue deformation patterns with some reliability (classification accuracy > 60%) when simultaneously classifying 4 grasp patterns, and some were able to reliably perform all six motions. To further study the extent of patients' motor control, we examined the intra-grasp variability by comparing the similarity of spatiotemporal tissue deformation patterns within each motion. This enabled us to examine in depth the variability of executed tissue deformation patterns. Interestingly, we found that intra-grasp variance did not correlate with grasp classification accuracy in most patients. Collectively, these results suggest that children with congenital below-the-elbow ULDs have significant control of their residual muscles. Future systems that fully utilize this control could provide prostheses with greater functionality than current pediatric interfaces.

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Poster

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Program #/Poster #: P525.05

Topic: E.04. Voluntary Movements

Support: NSF Grant IIS-1219231
NSF Grant BCS-1455866

Title: Classifying hand grip and release events in object grasping with Granger Causality features extracted from scalp electroencephalography

Authors: *A. Y. PAEK, J. L. CONTRERAS-VIDAL;
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Abstract: Objective: Brain machine interfaces (BMIs) are systems that allow users to control assistive devices through motor intent detected from the brain. Such systems can be used to help

restore hand-based motor function to paralyzed individuals. Most BMI studies have utilized scalp electroencephalography (EEG) to predict motor intent with spectral features that are extracted from individual sensors. Few studies have explored predicting motor intent with functional connectivity, a metric that estimates the magnitude of communication between different cortical areas. This intercortical communication is a fundamental aspect of cortical activity that is found to be modulated in motor tasks and shows potential to be used as a feature for predicting motor intent. In this work, we assessed if Granger Causality, a functional connectivity measure, can be used to predict when able-bodied participants held or released a grasped object. Methods: Eleven able-bodied volunteers participated in an experiment where they grasped and lifted a sensorized object. The object contained load cells that measured the grip forces exerted by the thumb and index finger. EEG signals were recorded simultaneously with the grip force recordings. Utilizing the force trajectories, we found time regions in trials when subjects were either holding the object or not touching the object. These "hold" and "release" periods were designated as the classes we aimed to predict with EEG signals. 5-fold cross validation was used to train and test a classifier that used Gaussian Mixture Models to model the classes and Local Fisher Discriminant Analysis to reduce the dimensionality of the data. For predictive features, we tested lowpass filtered delta band potentials (<1 Hz), spectral power in six frequency bands (4-8 Hz, 8-13 Hz, 13-30 Hz, 30-50 Hz, 70-110 Hz, 13-200 Hz), and Granger Causality. The features were extracted from 8 bipolar EEG sensors that were mostly centered on sensorimotor areas. Results: While classification accuracies were modest (median balanced accuracies ranged from 52%-58%), we found that Granger Causality features performed significantly better than the other features used in the study. The results demonstrate that functional connectivity may be a promising feature that can improve future EEG-based BMIs. Acknowledgement: We note that this work is new analysis on data that was previously collected and published in collaboration with Dr. Pranav Parikh, Dr. Alycia Gailey, and Dr. Marco Santello from Arizona State University.

Disclosures: A.Y. Paek: None. **J.L. Contreras-Vidal:** None.

Poster

P525. Hand Control: Physiology and Pathology

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Topic: E.04. Voluntary Movements

Support: NSF Grant IIS-1219231
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University of Houston, Division of Research High Priority Area Research Seed Grant

Title: Influence of grip force magnitude on temporal structure of electrocortical variability

Authors: *N. RAO[1], A. PAEK[2], J. L. CONTRERAS-VIDAL[2], P. J. PARIKH[1];
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Abstract: Fine grip force adjustments in routine tasks such as holding a coffee-mug allows one to adapt to task requirements. The temporal structure of grip force variability that underlies such adjustments is known to be modulated with the magnitude of force required to hold an object. Furthermore, neuroimaging evidence suggests that spatial activation in fronto-parietal brain regions is also driven by the grip force magnitude. However, it remains unclear whether the temporal structure of neural variability is influenced by the magnitude of grip force exerted on a hand-held object. To this end, we hypothesized that the temporal structure in neural variability assessed using electroencephalography (EEG) over sensorimotor regions would modulate with the force magnitude during a moment-to-moment grip force control task. We recruited 11 right-handed healthy young adults (age range: 18-40 years) and recorded their cortical activity using 64-channel EEG during performance of an isometric force production task. For each trial, the subject was cued to exert 5, 10, or 15% of their maximum voluntary contraction (MVC) for 8 sec. Subject's grip force was displayed on a computer screen and each subject performed 100 trials during the session with ~33 trials per force level. We used sample entropy to assess temporal structure in the EEG signal variability. Consistent with previous reports, our preliminary findings (n=7) show an increase in sample entropy of the grip force with the increase in grip force magnitude. Interestingly, sensor-level analysis of EEG channels depicts a modulation in sample entropy of the electrocortical activity with grip force magnitude. The sample entropy of EEG time series recorded using a channel over the hand sensorimotor cortical regions was lower for 10% versus 5% MVC. There was a trend for an increase in sample entropy of EEG signal between 10% and 15% of MVC. However, there was negligible difference in sample entropy of the EEG signal between 5% and 15% MVC. Such a modulation in sample entropy was not observed in the EEG signals captured from an electrode placed closer to the midline. These findings are suggestive of non-linear and spatially constrained modulation in the temporal structure of electrocortical variability with grip force magnitude. Findings from this study could contribute to mechanistic underpinnings of moment-to-moment force control with applications in neurorehabilitation.

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Poster

P525. Hand Control: Physiology and Pathology

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Topic: E.04. Voluntary Movements

Support: JSPS KAKENHI Grant Number 21J20955
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Title: Pre-movement interhemispheric synchronicity of sensorimotor activities is associated with the sustained anti-phase bimanual tapping

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[1]Grad. Sch. of Sci. and Technol., [2]Dept. of Biosci. and Informatics, Keio Univ., Kanagawa, Japan; [3]Dept. of Neurosurg., Osaka Univ., Osaka, Japan

Abstract: Coherent coupling of neuronal oscillation represents perception, sensorimotor control and ongoing actions. However, its impact on subsequent behavior remains to be explored. Here, to test whether the coherence of scalp EEG over bilateral sensorimotor cortices (SMC) at pre-movement period is associated with subsequent unstable behavior, we developed experiment setup that cued in participants in a brain-state dependent manner. Because the anti-phase bimanual tapping spontaneously alters into in-phase putatively due to the interaction of interhemispheric sensorimotor activities (Aramaki et al., 2006), we hypothesized the initiation of finger tapping at the trough of spontaneous fluctuation of functional connectivity between bilateral SMCs prolongs the duration and vice versa.Fifteen participants participated in the study. Each participant underwent 96 trials of anti-phase tapping task using bimanual index and middle fingers during recording whole-head scalp EEG. The first 16 trials were served as "No-triggered" session in which cues of motor task were given independent from EEG. Parameters for online estimation of coherence of bilateral SMC were determined from the data; For EEG signals around bilateral SMC that exhibited sensorimotor event-related desynchronization (ERD) in beta frequency band (14-30 Hz), phase-locking value (PLV) was predicted using autoregressive model in the following trials. During ready period (1 ± 0.5 s before movement onset), if online calculated PLV exceed or inferior to the 75/25 percentile of distribution from resting-state EEG, the auditory cue of finger tapping was presented to the participant (High and Low PLV condition, respectively). The duration of anti-phase bimanual tapping was quantified from the online acquired keyboard data. The pre-movement PLV and duration were subjected to Spearman's rank correlation test unless the normality of both variables were confirmed by the Shapiro-Wilk test.We found significant negative correlation between PLV in beta-band and duration (Spearman's rank correlation test, $p < 0.001$, $\rho = -0.70$), while the difference of the magnitude of bilateral ERD was not confirmed between high and low conditions (Two-way repeated measure ANOVA, $p = 0.61$, 0.59 main effect for "Condition" and "Hemisphere", respectively). Moreover, the correlation between the ERD magnitude and duration was not observed for the bilateral hemisphere (Right: $p = 0.95$, Left: $p= 0.81$, respectively). Collectively, not the local excitability but the inter-regional coupling would be the substrate to execute the bimanual asynchronous behavior, that underlies our sophisticated performance such as piano playing.

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Poster

P525. Hand Control: Physiology and Pathology

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Program #/Poster #: P525.08

Topic: E.04. Voluntary Movements

Title: Neuromuscular control of smartphone manipulation - selfie strain?

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Abstract: Taking 'selfies' has become common behavior among smartphone users. Such self-portrait photography requires individuals to hold their arm either in a fully extended position or slightly flexed at an elbow compared to the regular texting position. One study has shown that smartphone users maintain such posture frequently until a good photo is taken (Khanal et al. 2019). Such repetitive arm position for selfies could put excessive loading on the upper limb muscles and strain the elbow and wrist joint. The primary goal of this study was to understand the muscle loading generated when a smartphone was held and manipulated at two comfortable arm heights: shoulder and abdomen, by recording the 14 muscles of the upper limb. Participants pressed instructed keys in these two arm positions (heights) and moved at self-defined walking speeds (movement conditions). This study examined interaction effects among movement conditions and arm height on the muscle activity of the upper limb involved in this smartphone manipulation. It was hypothesized that when manipulating a smartphone device at the shoulder, there would be an increase in arm muscle activity to stabilize the arm and overcome the moment. Likewise, a significant increase in hand muscle activity when the device was manipulated at the shoulder compared to the abdomen was expected. Lastly, walking was expected to result in greater muscular activation patterns across both hand and arm muscles compared to standing when the device was held at the shoulder compared to the abdomen. We found that there was an effect of arm height on the muscle activity of the upper limb. The mean amplitude of surface EMG recorded for ECU, EPL, ED, BIC, TRI, ADEL, MDEL, PDEL, and RTRAP increased significantly when the hand was held at the shoulder position. In contrast, FDI, FCR, and LTRAP had higher amplitudes at the abdomen position. There was an absence of significant interaction effects of arm height and movement conditions for twelve muscles out of fourteen recorded muscles except for ECU and TRI muscles. From our observations, we concluded that the neuromuscular system generates a 'simple motor control strategy' specifying the necessary muscle activity to accomplish smartphone manipulation.

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Poster

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Program #/Poster #: P525.09

Topic: E.04. Voluntary Movements

Support: Starting grant from Swedish Research Council (2019-01909)
Ake Wibergs Stiftelse (M20-0038)

Title: The positive dimension of schizotypy is associated with both an impaired attenuation and discrimination of self-generated touch

Authors: *E. ASIMAKIDOU, X. JOB, K. KILTENI;
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Abstract: Self-generated touch feels weaker compared to externally generated touch of identical intensity (Bays and Wolpert 2008; Kilteni, Engeler, and Ehrsson 2020), and prevalent theories of motor control posit that attenuation arises because the brain predicts the sensory consequences of our voluntary movements (McNamee and Wolpert 2019; Wolpert and Flanagan 2001). Although somatosensory attenuation has been shown in 98% of healthy adults in a population-based cohort (Wolpe et al. 2016), it appears greatly reduced in patients with schizophrenia (Blakemore et al. 2000; Shergill et al. 2005, 2014) suggesting a failure to accurately predict self-generated touch. Here, we tested whether somatosensory attenuation in non-clinical individuals is related to schizotypy, a premorbid phenotype associated with increased liability for schizophrenia (Ettinger et al. 2014). One hundred volunteers participated in a psychophysics force discrimination task that quantified the perceived magnitude and discrimination ability of self-generated and externally generated tactile stimuli applied on their left index finger. Afterwards, participants completed the Schizotypal Personality Questionnaire (SPQ) (Raine 1991).
A significant negative relationship was found between the participants' somatosensory attenuation and their total SPQ score (Spearman's rho = −0.207, $p = 0.039$). This association was driven by the Cognitive-Perceptual SPQ factor (Spearman's rho = −0.256, $p = 0.010$), a factor that represents the positive dimension of schizotypy. Moreover, the Cognitive-Perceptual SPQ factor was associated with a reduced sensitivity for discriminating self-generated touches from externally generated touches (Spearman's rho = 0.331, $p < 0.001$). Furthermore, individuals with high scores in the Cognitive-Perceptual factor displayed significantly reduced somatosensory attenuation (Mann-Whitney test, $W = 775.5$, $p = 0.030$) and sensitivity for their self-generated touches (Mann-Whitney test, $W = 763.5$, $p = 0.042$) compared to those with low scores in the Cognitive-Perceptual factor.
Our results suggest that the Cognitive-Perceptual factor and by extension the positive dimension of schizotypy is associated with a reduced ability to attenuate and discriminate self-generated tactile stimuli compared to externally generated stimuli. Given that the positive dimension of schizotypy represents the analogue of positive psychotic symptoms, deficits in processing self-

generated tactile information could potentially serve as a bio-cognitive marker for early diagnosis and prevention of schizophrenia.

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Poster

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Topic: E.04. Voluntary Movements

Support: WUOT Young Investigator Pilot Award 93230A
NCATS UL1 TR000448

Title: Failure to Compensate: Nerve Injury Patients Use Their Injured Dominant Hand, Even When Their Non Dominant is More Dexterous

Authors: *B. A. PHILIP[1], M. R. THOMPSON[2], N. A. BAUNE[4], M. HYDE[1], S. E. MACKINNON[3];
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Abstract: Compensation with the non-dominant hand (NDH) is critical for patients with irreversible chronic impairment of the dominant hand (DH) due to nerve injury, orthopedic injury, or stroke. However, it remains unknown how performance and behavior change after unilateral impairment of the DH, even among individuals with a healthy brain (i.e. peripheral nerve injury). We hypothesized that compensatory use of the NDH would depend on performance of the DH rather than the NDH, because current rehabilitation is not designed to encourage compensation. We addressed this question by studying adults with unilateral upper extremity peripheral nerve injury to the DH (n = 22, 15 female). This sample was recruited from all individuals with significant injury-related disability (Quick Disabilities of the Arm, Shoulder, and Hand score ≥ 18) in one outpatient rehabilitation clinic and one nerve surgery clinic. We measured hand usage (% of actions with each hand) via an unconstrained reach-to-grasp block building task, and dexterity via four subtests of the Jebsen-Taylor Hand Function Test. Most participants were more dexterous with their uninjured NDH than their injured DH (77%; mean laterality index -0.10 ± 0.26), but hand usage patterns did not differ from typical adults (p = 0.08). Compensatory use of the NDH was associated only with DH dexterity; not NDH dexterity, nor any other factor including rehabilitation, sex, injured nerve, or injury location. In other words, we confirmed our hypothesis that compensation would depend only on performance of DH, not NDH. Specifically, patients only compensated with their NDH if their DH was severely

injured, and compensation did not depend on whether their NDH was their more dexterous hand post-injury. Overall, most peripheral nerve injury patients with DH injury did not engage in compensatory use of the NDH, even when they might benefit from compensation, i.e. because their NDH became more dexterous. This demonstrates a persistent influence of hand preference despite peripheral nerve injury, and indicates the need to understand (and support) the neural mechanisms that could potentially allow people to compensate with the NDH.

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Poster

P525. Hand Control: Physiology and Pathology

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P525.11

Topic: E.04. Voluntary Movements

Support: FAPERJ, E26/010002474/2016, CNE202.785/2018
Centro de Pesquisas, difusão e inovação em Neuromatemática- NeuroMat (FAPESP 2013/07699-0)
CNPq 309560/2017-9
FINEP PROINFRA 2010 0.1.12.0308.00

Title: Bilateral reorganization of upper limb kinematics after traumatic brachial plexus injury

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Abstract: Traumatic brachial plexus injury (TBPI) causes a sensorimotor deficit in upper limb (UL) movements, usually compromising shoulder muscle force. We hypothesized that the hand kinematics of both the injured UL and uninjured UL would be disturbed through changes in the arm-forearm intersegmental coordination. Individuals with TBPI (n=12) and controls (n=9) matched in age, gender, and anthropometric characteristics were recruited. Participants kept a standing position and held an empty cup with the UL relaxed beside the body. The movement of transport of the cup to the mouth and return to the starting position was recorded with the Vicon System. Reflexive markers were positioned in the shoulder, elbow, wrist and index finger. Hand kinematics was calculated based on index finger movement, and the arm-forearm dynamics (angular displacement and angular velocity) was measured by extracting the polar phase angle. Intersegmental coordination was measured as the relative phase (RP) between the arm and the forearm phase angles. Five TBPI participants scoring 3/5 in MRC in the injured UL biceps were

able to execute the task. TBPI participants exhibited a longer movement duration for both UL ($p<0.05$) and a lower peak velocity for uninjured UL when compared to controls ($p<0.05$). Shorter time to peak velocity was observed during transport in comparison to return ($p<0.01$). Area under the arm phase angle curve presented a difference for the injured UL during transport ($p<0.001$), and for the uninjured UL during return ($p<0.05$). We analyzed the RP in three different time windows based on hand movement (start, peak velocity, ending). RP analysis indicated more forearm use during the transport to mouth phase in the hand peak velocity for all groups ($p<0.001$). Furthermore, a greater arm use was found in the injured UL as compared to controls and to the uninjured UL ($p<0.001$). During the transport deceleration phase, a negative correlation was found between the time to peak velocity and the area under arm phase angle curve for the injured UL ($p<0.05$), contrasting to controls and the uninjured UL, in which time to peak velocity correlated with that of the forearm phase angle curve ($p<0.001$). In the return deceleration, the area under arm phase angle curve of the uninjured UL correlated positively with movement duration ($p<0.05$) and negatively with the peak velocity ($p<0.05$). These results suggest a more active control of the arm in the injured UL, modifying the natural UL coordination and causing a disturbance in the control of hand kinematics. Furthermore, the uninjured UL presented modifications in hand kinematics and arm dynamics, indicating that motor control of this limb is also affected by TBPI.

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Poster

P525. Hand Control: Physiology and Pathology

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Program #/Poster #: P525.12

Topic: E.04. Voluntary Movements

Support: NSF Graduate Research Fellowship Grant No. DGE-1610403
Robert J. Kleberg, Jr. and Helen C. Kleberg Foundation Medical Research grant
National Eye Institute of the National Institutes of Health under Award Number R01EY028746

Title: A common template for neural reinforcement of finger individuation

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Abstract: Stroke is the leading cause of serious long-term disability and often results in lasting deficiencies in the strength and control of individual fingers. Functional MRI neurofeedback may

provide a useful tool to guide neuroplastic changes in the brain after injury to restore fine motor function. However, identifying an individualized neural training target in the presence of a neurological injury may not be possible. To address this issue, we have investigated the use of functional alignment, or *hyperalignment*, of brain data across healthy individuals to create an idealized neural template that can be used as a surrogate training target for new participants. We conducted an offline study of 17 healthy young adults who were engaged in a finger individuation task during an fMRI localizer session. For each individual, a support vector machine classifier was trained on the functionally aligned data of the 16 other participants. We found that the classification accuracy of individual finger presses significantly improved ($p < 0.001$) when group data was aligned based on function (88.8%) rather than conventional anatomy (46.0%). Furthermore, we have demonstrated that our functionally aligned template is robust to impaired finger presses. If a participant has a deficit in their ability to individuate a movement for one or two fingers, it is possible to infer the proper neural representation for those fingers from the surrogate template. To assess this, the data for one or two fingers was completely removed when aligning the left-out individual to the neural template. The neural representations for the removed finger(s) were then obtained by applying the transformation matrices from subject space to the common template space to the entire dataset with all fingers included. We found that the classification accuracy when one finger (80.6%) or two fingers (66.5%) were removed during functional alignment was still significantly better ($p < 0.001$) than anatomical alignment with all fingers included (46.0%). These results indicate that our functionally aligned template can provide an effective substitute neurofeedback training target for patients following neurological injury.

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Poster

P547. Neurophysiology: Implanted Electrodes and Other Direct Interactions with Neurons III

Location: Virtual Only

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Program #/Poster #: P547.01

Topic: E.05. Brain-Machine Interface

Support: Dutch Epilepsy Foundation 17-07
R01MH122258

Title: The development of efficient communication in the human connectome

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Abstract: The development of rapid communication between different brain regions is essential for brain function. Post-mortem studies have shown that the myelination of white matter pathways starts in the late prenatal period and continues in late adolescence. MRI measurements have shown that pathways develop at different rates in different areas, often reaching a plateau around 30-40 years of age. These indirect measurements do not give quantitative information about how the transmission speed develops throughout development. Quantitative neurophysiological measurements of transmission speed in the developing human brain are essential to understand the maturation process of efficient communication that is necessary for brain functions like cognition and behavior. We collected data in 75 subjects, age 4 to 51 years, who were implanted with intracranial electrodes (electrocorticography - ECoG) to evaluate epilepsy surgery. The electrodes were positioned in MNI space and categorized in either frontal, parietal, temporal or sensorimotor sites. Adjacent electrode pairs were stimulated with ten consecutive pulses (1ms, 4-8mA, 0.2Hz), and we measured cortico-cortical evoked potentials (CCEPs) throughout the other electrodes. These CCEPs expose the underlying physiological networks. We calculated the latency of the initial negative potential deflection (N1-response) that occurs within 100ms after stimulation. We fitted distributions of latency as a function of age for all 16 combinations of these four brain regions. We found that the latency of the N1 decreased significantly with age, from ~40ms at age 4 years to ~25ms in adulthood. More detailed fits indicated that the transmission delay between cortical areas decreases with 0.23-0.88ms each year, until it plateaus around age 15-29 years old. These increases in speed were observed in both long and short range connections between and within frontal, parietal and temporal areas. The decreases in the N1 latency reveal that transmission speed in the human brain connectome increases about two-fold from childhood to adulthood. These measurements provide quantitative estimates of the development of efficient communication and can be used in large scale models of the human brain that bridge structural and functional development. With this study, we facilitate the construction of growth curves with velocities in the brain. This could be helpful in diagnosing abnormal development in cognition and behavior.

Disclosures: D. van Blooijs: None. **J.F. van der Aar:** None. **G.J.M. Huiskamp:** None. **G. Castegnaro:** None. **M. Demuru:** None. **W.J.E.M. Zweiphenning:** None. **N.E.C. van Klink:** None. **P. van Eijsden:** None. **K.J. Miller:** None. **F.S.S. Leijten:** None. **D. Hermes:** None.

Poster

P547. Neurophysiology: Implanted Electrodes and Other Direct Interactions with Neurons III

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Program #/Poster #: P547.02

Topic: E.05. Brain-Machine Interface

Support: NIH Grant 1K99GM140215
Department of Anesthesiology, Perioperative and Pain Medicine, Stanford University School of Medicine

Title: Comparison of intracranial and scalp EEG complexity measures during emergence from anesthesia

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Abstract: Uncovering the neural basis of anesthesia calls for direct brain recordings. However, the most commonly used tool, electroencephalography, is largely driven by synchrony in widespread ensembles and may miss different types of local cortical activity. Simultaneous recordings from both scalp (EEG) and intracranial (iEEG) electrodes could provide a unique opportunity to understand combined local and ensemble neural activity underlying anesthesia. This opportunity presents itself in epilepsy patients undergoing invasive monitoring for seizure localization. Simultaneous EEG and iEEG activity was captured from 13 epileptic patients as they emerged from anesthesia. Spectral and complexity measures were calculated and compared before, during, and after emergence from anesthesia in both EEG and iEEG signals. Topological maps of spectral and complexity changes demonstrate the extent and timing by which different brain areas exhibit changes in activity during emergence from anesthesia. These findings provide a unique signature of the neural basis of anesthesia and may offer insights into mechanisms of human consciousness.

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Poster

P547. Neurophysiology: Implanted Electrodes and Other Direct Interactions with Neurons III

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Topic: E.05. Brain-Machine Interface

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Title: Adaptive Neurostimulation with Multi-Stage Control Loop: Application to Drug Resistant Epilepsy

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Abstract: Responsive neural stimulation (RNS) delivers electrical stimulation in a brain region in response to brain activity to treat neurological disorders. RNS directed at seizures and abnormal epileptiform activity proved to be successful in reducing seizures in drug resistant epilepsy, but efficacy optimization can be slow. There is ongoing interest in systems where stimulation paradigms and network targets are optimized adaptatively over time. Here we present a system where a cascade of detectors in implanted neural sensing and stimulation (INSS) device and a handheld device enabling machine learning seizure detection algorithms to adaptively control stimulation parameters and brain targets. We developed a system for multi-stage control over adaptive neurostimulation paradigms. The system utilizes an INSS with continuous real-time intracranial EEG (iEEG) recording and bi-directional communication with a handheld for off-device programmable control. The adaptive RNS settings on the INSS device engage multiple different network targets. In this application, the initial electrical stimulation targets detected hippocampal seizures. If the seizure activity continues despite the responsive stimulation for a specified time interval, the algorithm adaptively switches to a second network target with different stimulation parameters. The detected events are further evaluated by detection algorithms running on the handheld device and can alter the stimulation paradigm. We tested the detector cascade system in benchtop settings on 200 seizures and 6 interictal days using hippocampal iEEG previously recorded from 6 patients undergoing evaluation for epilepsy surgery. We measured performance of the second cascade detector which is engaged after initiation of the first detector and achieved 96% sensitivity and 99.5% specificity with 10.62 seconds average detection lag from seizure onset. The off-device detector was developed on large dataset from the NeuroVista study (5 dogs and 10 humans with more than 10 years of data). The detector was tested using data from 4 patients (181 seizures over 496 days in total). Our off-device detector improved the specificity up to 99.9% which enables more control over adaptive network stimulation. We developed and tested the RNS system with multi-stage control loop for

adaptive network stimulation. The system enables hypersensitive RNS targeting abnormal epileptiform activity that runs concurrently with a second INSS cascade detector for detecting seizures of a specified length and triggers RNS of a different network target. Deployment of the system to an INSS might prove useful in disrupting epileptogenic networks.

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Poster

P547. Neurophysiology: Implanted Electrodes and Other Direct Interactions with Neurons III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P547.04

Topic: E.05. Brain-Machine Interface

Support: Mayo Clinic

Title: Cortec brain interchange in freely behaving canine

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Abstract: ***Objective:*** We report our initial experience with the CorTec Brain Interchange, a fully implantable bi-directional system for chronic nervous system tracking and interaction. Because

canines are large enough to accommodate devices designed for humans, they are a good model system for the preclinical research required to develop an implantable bi-directional system for tracking electrophysiology, implementing algorithms, and adaptive stimulation.
Methods: A healthy 24-month-old beagle (*canis familiaris*) named Laika was implanted with the CorTec Brain Interchange system. The CorTec Brain Interchange device features up to 32 channels for local field potential recording and electrical stimulation. Two grids (20 total channels) were implanted through bilateral craniotomies into the subdural space spanning frontal and parietal canine cortex. The subdural grid electrode tails were tunneled to the shoulder, adjacent to the scapula, and connected to the internal electronics unit. The internal electronics unit in the shoulder inductively couples via a telemetry wand to an external interchange box for data streaming onto a computer using BCI2000 (1kHz Sampling Rate, Dynamic Range 0.5 - 450 Hz). Local field potentials were obtained in three different activities as ascertained by simultaneous video: 1) quiet wakefulness 2) movement and 3) sleep. The power spectral density in the three activities were analyzed. The LFP signals were segmented into 10-second-long epochs. The data were then filtered by a 0.5-200 Hz band-pass filter, and power spectral density (PSD) was estimated using Welch's method for each 10-second-long epoch.
Results: The CorTec Brain Interchange system provided continuous data streaming that was only limited by the ability to maintain a physical connection between the magnet in the implant and the magnet in the coupling device of the telemetry wand. Recordings of up to 30 minutes in duration were obtained. The PSD for the 3 activities showed spectral changes enabling differentiation of wake, movement, and slow-wave sleep.
Conclusions: These preliminary results demonstrate the feasibility of continuous streaming of cortical electrophysiology in a behaving canine with the CorTec Brain Interchange. Simple spectral measures were shown to identify behavioral state, wake, and sleep. Future plans include further engineering refinements for reliable coupling of the device, reduction of artifact, and the ability to stimulate. All of these are required for a clinical brain computer interface trial in humans, and this technical experience represents a meaningful step toward this.

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Poster

P547. Neurophysiology: Implanted Electrodes and Other Direct Interactions with Neurons III

Location: Virtual Only

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Program #/Poster #: P547.05

Topic: E.05. Brain-Machine Interface

Support: NIH UH3 NS095495

Title: A case study for tracking verbal memory performance during anterior nucleus of the thalamus stimulation in patients with epilepsy

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Abstract: Verbal memory complaints are common in patients with temporal lobe epilepsy (TLE). The impact of therapeutic electrical brain stimulation on memory can be difficult to disentangle from seizures. Verbal memory encoding and recall abilities can be probed with a simple free recall task and can provide objective measures of memory function. The purpose of this study was to investigate the changes in human verbal memory performance while receiving chronic electrical stimulation to bilateral anterior nucleus of the thalamus (ANT) as a therapy to reduce seizures in a patient with drug resistant TLE. Recordings were obtained from a patient with drug resistant epilepsy implanted with an investigational Medtronic Summit RC+S™ sensing and stimulation device. As patient seizure diaries are notoriously inaccurate, the patient's intracranial electroencephalography (iEEG) was analyzed for seizures to create a gold standard seizure diary. The verbal memory tasks were performed by the ambulatory subject living in their home environment with iEEG and behavioral data streamed to a handheld device and cloud repository. The participant was presented with lists of words on a handheld tablet screen for a delayed test of free recall. Lists were composed of 12 randomly chosen nouns. After a distractor task, the participant was asked to recall as many of the words as possible. The participant completed 33 independent sessions over 18 months with 15 lists per session for 5940 total word presentations. We were then able to compare changes in verbal memory score to the patient's seizure frequency changes during therapy. Using Pearson's Correlation Coefficient, we found that memory performance and seizure frequency had a significant negative correlation (Pearson's Correlation Coefficient $r = -0.4396$, $p = 0.0105$). While these results are for one patient, they are promising, and they demonstrate the feasibility of dense cognitive tracking in ambulatory patients. Emerging devices for electrical stimulation of the brain enabling smart sensing and continuous streaming of the iEEG data enable long-term investigation of underlying brain dynamics during cognitive tasks. This case study shows that a simple verbal memory task may provide a suitable measure for tracking a therapy's impact on memory and it's relationship with seizures.

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Poster

P547. Neurophysiology: Implanted Electrodes and Other Direct Interactions with Neurons III

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Program #/Poster #: P547.06

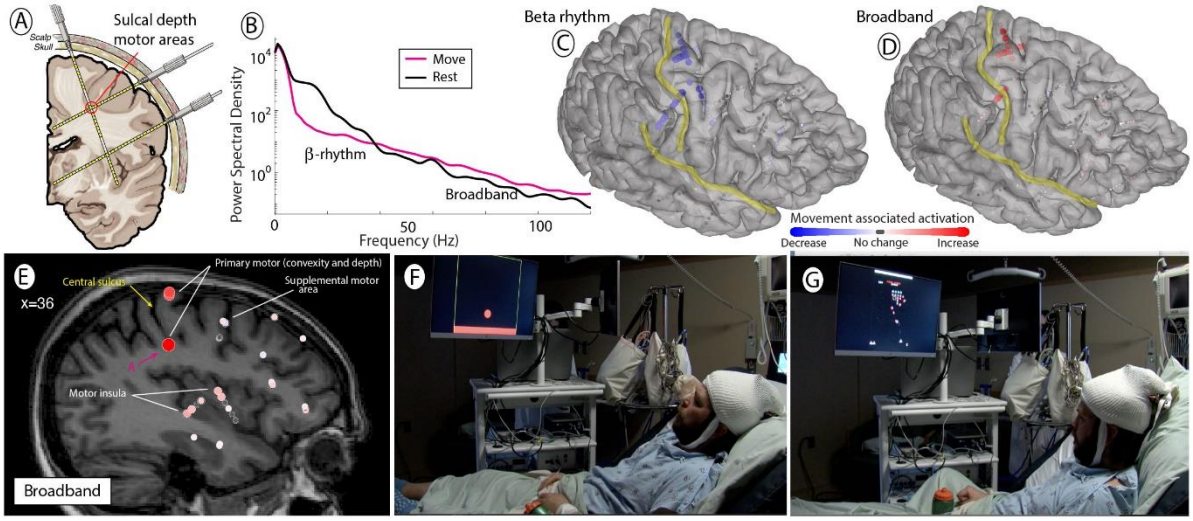
Topic: E.05. Brain-Machine Interface

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Title: New Depths in Human Brain-Computer Interfacing

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Abstract: Implanted brain-computer interfaces (BCIs) have, to date, measured from the brain's convex surface to identify control signals. However, the circuitry of cognition and behavior is distributed throughout the brain volume. Nuanced applications for BCI will need to engage circuit nodes from the depths of the brain where they are located. Intracranial implanted electrode monitoring for epilepsy with penetrating stereoelectroencephalography (SEEG) has enabled our team to study the electrophysiology of behavior and BCI from the brain's depths. We recorded SEEG electrophysiology during movement and movement imagery of the tongue, foot, and hand of 5 epilepsy patients, aged 6 to 36 (2 females). Movement and imagery associated broadband spectral changes from the SEEG measurements were used for simple ball-target BCI feedback and imagery-based application control of the video game Galaga. Robust maps of movement and imagery-based brain activity were identified in primary, premotor, insular, and supplementary motor cortical areas. Recordings from the sulcal depths were used for robust control of the BCI feedback task (peak accuracies 92-100%) and the Galaga video game.



A) SEEG measures from motor sites at sulcal depths. **B)** Power spectrum from the sulcal base of primary motor cortex (pink arrow in E) during hand movement and rest. **C&D)** Low frequency beta (12-20Hz) power decreases (C) and high frequency broadband power increases (D). **E)** Sagittal slice with electrodes within 6 mm of this slice allows viewing broadband power increases on the surface and at depth. Activation maps in C-E for movement are shown in the central colorbar (signed r^2, scaled to maximum). **F&G)** Patient playing an online (control) cursor task (F) and the video game Galaga (G).

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Poster

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Topic: E.05. Brain-Machine Interface

Support: NIH UH3-NS095495
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Darpa - HR0011-20-2-0028

Title: Network Excitability in Human Mesial Temporal Lobe Epilepsy

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Abstract: Objective: Seizures are a consequence of abnormal brain network excitability. Two fundamental electrophysiologic biomarkers of epilepsy and network excitability, interictal epileptiform spikes (IES) and seizures, show circadian patterns supporting behavioral state modulation of brain excitability. The circadian patterns of seizures and IES in mesial temporal lobe epilepsy (mTLE), however, differ as seizures show daytime peaks and IES show nocturnal peaks in occurrence. Methods: We investigated this paradox in mTLE brain excitability using a chronically implanted device enabling continuous intracranial EEG (iEEG), wireless data streaming, and electrical stimulation in four patients with electrodes targeting two key mTLE network nodes, the hippocampus (Hc) and anterior nucleus of the thalamus (ANT). Automated sleep staging, IES rates, and seizure detection iEEG classifiers provided dense behavioral state and brain excitability tracking. The Hc evoked responses to continuous ANT electrical stimulation were used to track ANT-Hc network excitability over multiple months in ambulatory subjects living in their natural environments. Results: Consistent with prior literature, seizure probability was highest during the day and average IES rates were highest at night. In particular, seizures had a daytime bimodal peak, increased in the morning and evening whereas IES had a unimodal nocturnal peak during non-REM slow wave sleep. Using continuous behavioral state, IES, seizure, and ANT-Hc evoked potential tracking, we show that mTLE seizures are increased in wakefulness, while IES are increased in slow-wave sleep. Tracking ANT-Hc evoked potentials we show that the ANT-Hc evoked responses have shorter latency in wakefulness relative to slow wave sleep, suggesting that limbic network excitability and connectivity are reduced in slow-wave sleep. Conclusion: We hypothesize that increased local Hc excitability in slow-wave sleep, as demonstrated by increased spike rates, is not associated with increased seizure probability due to reduced long-range limbic (ANT-Hc) effective connectivity. This reduced limbic effective connectivity in slow wave sleep may suppress the broader network activation required for initiation and propagation of mTLE seizures. These results help clarify a standing paradox of Hc excitability and seizure occurrence in mTLE and provide insights to the mechanism of therapeutic electrical brain stimulation. Furthermore, the ability to effectively track brain network connectivity and excitability in behaving humans may prove useful for adaptive electrical brain stimulation to improve electrical brain stimulation efficacy.

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Study (EPAS), Neuropace RNS System Responsive Stimulation for Adolescents with Epilepsy (RES. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); named inventor for intellectual property developed at Mayo Clinic and licensed to Cadence Neuroscience Inc. BNL waived contractual rights to royalties.. **P. Nejedly:** None. **T. Pal Attia:** None. **D. Crepeau:** None. **E. St. Louis:** None. **D. Hermes:** None. **S. Messina:** None. **T. Denison:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Shares of Bioinduction. **K. Miller:** None. **J. Van Gompel:** E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); named inventor for intellectual property developed at Mayo Clinic and licensed to Cadence Neuroscience Inc. BNL waived contractual rights to royalties. **B.H. Brinkmann:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; investigators for UNEEG Inc. investigational device trial.. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Mayo Clinic has received research support and consulting fees on behalf of BB from UNEEG, NeuroOne, Epiminder, Medtronic, and Philips Neuro.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); named inventor for intellectual property developed at Mayo Clinic and licensed to Cadence Neuroscience Inc. **G.A. Worrell:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; GW, BB and NG are investigators for UNEEG Inc. investigational device trial.. C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Mayo Clinic has received research support and consulting fees on behalf of GW, BNL and BB from UNEEG, NeuroOne, Epiminder, Medtronic, and Philips Neuro.. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); GW, BB, JVG and BNL are named inventors for intellectual property developed at Mayo Clinic and licensed to Cadence Neuroscience Inc. BNL waived contractual rights to royalties., GW has licensed intellectual property to NeuroOne Inc..

Poster

P547. Neurophysiology: Implanted Electrodes and Other Direct Interactions with Neurons III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P547.08

Topic: E.05. Brain-Machine Interface

Support: NIH - NIMH, R01 MH122258

Title: Signatures of electrical stimulation driven network interactions in the human limbic system

Authors: *A. OJEDA VALENCIA[1], N. M. GREGG[2], G. A. WORRELL[3], B. N. LUNDSTROM[2], B. H. BRINKMANN[2], J. J. VAN GOMPEL[4], K. J. MILLER[4], D. HERMES[1]; [1]Physiol. and Biomed. Engin., [2]Neurol., [4]Neurologic Surgery, [3]Mayo Clin., Rochester, MN

Abstract: The human limbic network is involved in emotional and memory processing. Understanding its dynamics is important to better understand several clinical conditions, such as epilepsy and related psychiatric co-morbidities, and it is often targeted with therapeutic brain stimulation. To further understand the basic dynamics in this network, single-pulse electrical stimulation (SPES) can be delivered to various limbic substructures, and cortico-cortical evoked potentials (CCEPs) can be measured in other regions to assess network effective connectivity. Typical CCEP responses have been described as either an early surface negative deflection within 10-50 ms (N1) or as a later slow-wave within 80-250 ms (N2). We investigated whether the limbic network evoked responses after SPES follow the same profile. Seven patients with drug-resistant focal epilepsy who had stereotactically-targeted invasive EEG electrodes implanted for clinical purposes participated in the study. SPES was delivered through pairs of adjacent electrodes to limbic network substructures (hippocampus, amygdala, thalamus, posterior and anterior cingulate, and parahippocampal gyrus) ~10 times every 3-5 sec with 200 microsec square wave pulses of 6 mA. The electrodes studied were not involved in spontaneous seizure generation. Robust CCEPs were observed throughout the limbic system. Many evoked potentials had profiles outside the typically reported N1/N2 deflections. Stimulation of hippocampus and amygdala elicited prominent CCEPs in the posterior cingulate gyrus (PCG) across all patients, and responses in the PCG were more often observed and had larger amplitudes compared to responses in the anterior cingulate gyrus. However, these responses in the PCG did not follow typical N1/N2 shape, but showed a unique electrophysiological signature of 3 to 4 repetitive deflections within ~500-600 milliseconds. These data demonstrate that limbic network CCEPs cannot be fully described by amplitude and latency of one particular feature, but that different networks may show complex response patterns and interactions. Characterizing these different CCEPs waveforms can be relevant to better understand the dynamics of the limbic system and its implication in clinical states such as epilepsy.

Disclosures: A. Ojeda Valencia: None. **N.M. Gregg:** None. **G.A. Worrell:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Medtronic. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Cadence. **B.N. Lundstrom:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Medtronic. **B.H. Brinkmann:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Medtronic. E. Ownership Interest (stock, stock options, royalty, receipt of

intellectual property rights/patent holder, excluding diversified mutual funds); Cadence. **J.J. Van Gompel:** None. **K.J. Miller:** None. **D. Hermes:** None.

Poster

P547. Neurophysiology: Implanted Electrodes and Other Direct Interactions with Neurons III

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P547.09

Topic: E.05. Brain-Machine Interface

Title: A framework to characterize sources of stereotactic error in neurosurgical electrode implantation

Authors: *M. JENSEN[1], K. J. MILLER[2];
[2]Neurosurg., [1]Mayo Clin., Rochester, MN

Abstract: Title: A framework to characterize sources of stereotactic error in neurosurgical electrode implantation Michael A. Jensen and Kai J. Miller **Introduction:** Understanding the sources of error in stereotactic systems is paramount to the progression of functional neurosurgery. A centralized framework is a necessary component to identify these sources. **Objectives:** In order to define sources of error, we identified common sources of error, categorized them, and applied them to the different stereotactic systems used by modern neurosurgeons. That is, each system has a unique set of sources of error derived from this framework, but not each source of error will apply to each system. No such framework currently exists. **Methods:** We created a list of targeting systems reported in the recent neurosurgical literature: ROSA®, Mazor Renaissance™, Starfix (FHC) ™, Nexframe®, Clearpoint®, and Leksell®. We reduced each system to its most basic description (such as entry versus target deviations), and subsequently identified common sources of error (table 1). Within this framework, sources of error were categorized as stochastic or systematic. For systematic errors, we attempted to describe the source of error and how this error will manifest intraoperatively and in post-operative imaging. **Results:** We describe and illustrate the details of this framework applied to each of the stereotactic system. Each system is individually characterized and we discuss the unique framework-based score held by each. This framework describes each error type, and we discuss what might be done by surgeons to limit this error in their practice. We discuss why a generalized framework is crucial to driving both innovation in these systems' designs and improvement in stereotactic accuracy. **Conclusion:** This framework provides a simple rubric by which to analyze the sources of error for each of the most prominent stereotactic systems in use. This allows for ease and continuity of analysis between systems, across surgical centers, as well as a shared means to describe stereotactic error.

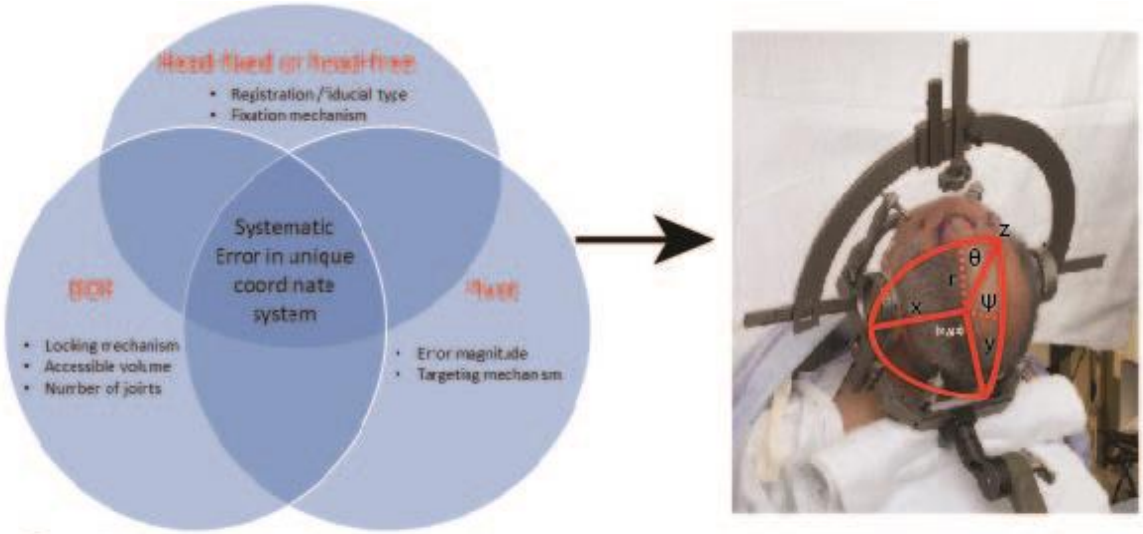


Figure 1. Left: Ven diagram demonstrating the three fundamental questions of our framework along with which fundamental characterstics that can contribute directly to systematic error. Answering all three qestions leads to a coordinate system(s) of best fit for the system being analyzed. Right: Leksell frame with superimposed Cartesian and Spherical Coordinate Systems.

Disclosures: M. Jensen: None. **K.J. Miller:** None.

Poster

P599. Stress and Emotional Regulation

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Program #/Poster #: P599.01

Topic: F.04. Stress and the Brain

Support: This project is funded by the NEUROSCIENCE RESEARCH OPPORTUNITY TO INCREASE DIVERSITY program (NEURO-ID-UPR RIO PIEDRAS NIH-ENDURE R25-NS080268706).
This research was supported by NIDA, Grant# 1R15 DA044500-01A1 to CS Maldonado-Vlaar. No financial interests or potential conflicts of interests have biased these studies.

Title: The interplay between oxytocin and CB1 receptors within the mesolimbic and limbic brain structures of rats followed by the effects of chronic oxytocin intranasal administration in acute exercise and stress

Authors: *P. MUÑOZ-RODRÍGUEZ, J. M. PADILLA-ESCALONA, L. RODRÍGUEZ-SANTOS, A. P. RAMOS-ROLÓN, W. NORZE, V. M. SCHLEIER, M. C. APONTE-REYES, A. I. RODRÍGUEZ-LEÓN, A. ACEVEDO-JETTER, C. S. MALDONADO-VLAAR;
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Abstract: Oxytocin (OT) is a neuropeptide associated with social behaviors, stress responses, and drug addiction. A previous study showed a cross-talk between the cannabinoid Type 1 receptor (CB1R) of the endocannabinoid system (ECS) and oxytocin receptors (OTR) within the mesolimbic system to modulate anxiety-like behaviors. Several studies support that intranasal oxytocin administration and voluntary wheel running are treatments endowed with anxiolytic properties. Other studies demonstrated that ECS is crucial for voluntary wheel running performance. OTR and CB1R within the limbic, mesolimbic, and endocrine systems are associated with anxiolytic responses. The purpose of this study was to examine whether OT intranasal pretreatment enhances voluntary wheel running behaviors and potentiates the anxiolytic properties of exercise in Sprague-Dawley male rats. Animals were divided into two groups, the first group received intranasal infusions of OT 0, 1 μg, 20 μl before being exposed to the running wheel two hours daily for 10 consecutive days. The second group received the same dose of OT or vehicle and was not exposed to the running wheel. The results indicated that the rats treated with OT exercised more than those that received the vehicle. Using the Light and dark box as a behavioral paradigm to measure responses related to anxiety, we found that rats treated with OT and acute exercise showed a decrease in anxiety-like behavior. We measure the OTR and CB1R expression within the paraventricular nucleus (PVN), amygdala (AMG), nucleus accumbens (NAc), and the ventral tegmental area (VTA) in order to examine the potential cross-talk between oxytocinergic and the endocannabinoid systems as mediators of the anxiolytic response and exercise performance. Biochemical assays showed a co-expression between the CB1 and OT receptors within the NAc. Additionally, the trend of the CB1Rs expression in each group remained the same for all the areas we examined (AMG, VTA, NAc, and PVN). Studying the physiological and behavioral changes of each of these groups allows us to have a better understanding of which would be the most effective anxiolytic treatment for people with a sedentary or physically active lifestyle. Future biochemical studies are required to examine the potential cross-talk between OT and the ECS as mediators of the anxiolytic response and exercise performance and, the implications it can have as a treatment for drug addiction, sedentary lifestyle, and anxiety disorders.

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Poster

P599. Stress and Emotional Regulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P599.02

Topic: F.04. Stress and the Brain

Title: Swimming exercise counteracts acute stress-induced neurobehavioral impairments via hippocampal FNDC5/BDNF pathway

Authors: *M. JODEIRI FARSHBAF[1], A. KUMAR MONDAL[1], T. IYER[2], K. GOLDINAK[2], P. KEYEL[1], K. ALVINA[2];
[1]Texas Tech. Univ., Lubbock, TX; [2]Univ. of Florida, Gainesville, FL

Abstract: Stress is a powerful stimulus that can change the structure, function, and activity of the brain. Depending on the type of stress—acute or chronic—the CNS shows a wide range of behavioral and physiological changes. The hippocampus is one of the most vulnerable regions of the brain to stress, as hippocampal functions such as emotion, spatial navigation, and memory are heavily influenced by stress. On the contrary, physical exercise has wide beneficial effects on physical and mental health, including minimizing the symptoms of neurological diseases such as Alzheimer's and Parkinson's diseases (AD and PD). Myokines, which are secreted from skeletal muscles during exercise, are thought to strengthen the muscle-brain axis. One example is Fibronectin Type III Domain Containing 5 (FNDC5), myokine that is abundantly expressed in skeletal muscle and brain, and that induces brain derived neurotrophic factor (BDNF) expression in the hippocampus after exercise. BDNF has crucial role in memory formation and decreased by psychological stress. Based on our previously published data showing that FNDC5/Irisin can counteract stress, we now tested the hypothesis that swimming exercise can suppress acute stress-induced neurobehavioral impairment by impacting hippocampal FNDC5/BDNF pathway. To test our hypothesis, we exposed male mice to daily swimming exercise for 20 days. 24h after last swimming session, mice were subjected to acute restraint stress for 3h while being probed in our combined OFT/NOR test. Our analysis showed that acute stress had no impact on anxiety-like behavior and short-term memory in exercised male mice, while the sedentary group showed increased anxiety-like behavior and short-term memory impairment after acute stress. We also found that swimming exercise increased FNDC5 expression in skeletal muscle and hippocampus. Furthermore, BDNF as downstream target of FNDC5, showed higher expression in the hippocampus of exercised mice. We also revealed an important connection with the Akt pathway in the hippocampus, all changes that were not observed in the prefrontal cortex. Taken together our results showed swimming exercise can promote resilience against acute stress in male mice. Also, our result support the idea that exercise has the potential to stimulate the maintenance of healthy neural function.

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Poster

P599. Stress and Emotional Regulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P599.03

Topic: F.04. Stress and the Brain

Support: Valley Baptist Legacy Foundation Institute for Neuroscience Grant

Title: Differential cortical monoamines release during exercise in rats chronically implanted with microdialysis probes

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Abstract: Physical exercise is known to positively influence mood, to reduce anxiety and to improve reaction to stress. Cerebral monoaminergic systems are thought to underly the neurochemical influence of exercise on mood and behavior. We hypothesized that beneficial effects of exercise can be shown by variations of neurotransmitters release in the cerebral cortex. We aimed at demonstrating that there is a temporal relationship between release of plasma and cerebral monoamines (A, NA, 5-HT, DA) and spontaneous running exercise in rats.Ten Spague Dawley rats (250 g) were chronically implanted (for up to 39 days) with a left common carotid cannula and a bilateral fronto-parietal transverse microdialysis probe. Blood and microdialysate were sampled 7 and 15 days after surgery. Sampling occurred for a total 180 minutes (divided in 20 minutes samples) during a one hour each period: Rest, treadmill exercise (80%V_{O2}max), and post-exercise rest. One animal was used for *in vivo* microdialysis recovery study. Seven animals were used for monoamines studies. Results showed: (1) that the development of the rat head device allowed sampling in an unrestrained running rat; (2) the permeability and functionality of carotid cannulas and microdialysis probes for up to 39 days in chronic implantation; (3) NA release increase at the end of exercise (100-120 min) and during the post-exercise rest period (120 to 180 min); (4) a significant DOPAC increase of 36% during exercise and of 58.3% during post-exercise rest when compared to rest. Post-exercise DOPAC increase was of 15.5% when compared to exercise; (5) that 5-HIAA did not vary at any time point. These findings indicate that spontaneous exercise specific stress induces DOPAC extracellular increase resulting from DA neuronal terminal release in layers IV and V of the fronto-parietal terminal field cortex. Differences between DA, NA, 5-HT release might be due to synaptic arrangements or to differences in the actual release of NT and in neural paths evoked during acute exercise. Abbreviations: A: Adrenaline, NA: Noradrenaline, DA: Dopamine, DOPAC: 3,4-Dihydroxyphenylacetic acid, 5-HT: 5-Hydroxytryptamine or Serotonin, 5-HIAA: 5-hydroxyindoleacetic acid, NT: Neurotransmitter. Acknowledgement: Author thanks Dr. L. Peyrin for advising this experimentation.

Disclosures: C.G. Gerin: None.

Poster

P599. Stress and Emotional Regulation

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Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P599.04

Topic: F.04. Stress and the Brain

Title: Effects of environmental enrichment on emotional regulation in C57BL/6J mice

Authors: *A. C. BASU, M. E. CRONIN, L. AMEZCUA, E. R. MONTANARI, R. M. PRICE, V. D. VU;
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Abstract: Etiological studies of neuropsychiatric disorders depend on the use of animal model systems to experimentally test the effects of putative risk factors. Environmental Enrichment (EE) affects behavior, cognition, neuroanatomy, and physiology in laboratory rodents, a fact that has implications for the design and interpretation of studies, and the ability to detect subtle effects of genetic manipulations. Previous findings by our research group (Hendershott et al., 2016) suggested that EE reduces anxiety-like behavior in mice, but did not conclusively rule out confounding effects on locomotor activity. In this study, we attempt to further characterize the effects of EE on emotional regulation in 72 male and female C57BL/6J mice by administering a battery of behavioral tests that implicate neural systems associated with emotional response and for which there are benzodiazepine-validated anxiety-like outcome measures: open field, grooming assay, marble bury, stress-induced hyperthermia, social ethogram, and novelty suppressed feeding. There were no significant effects of sex or housing in the benzodiazepine-validated outcome measures in the open field, grooming, or marble bury tests. There was a sex difference in novelty suppressed feeding, with females exhibiting significantly shorter latencies to feed than males. The stress-induced hyperthermia assay revealed a significant effect of sex with males exhibiting high hyperthermic responses to females, and a significant effect of housing with mice reared with EE exhibiting high hyperthermic responses relative to standard housed mice, suggesting that HPA axis regulation is affected by environmental enrichment. The interpretation of this result as an anxiety-like outcome would not be consistent with the negative results of the tests of anxiety-like behavior in the same animals. Analysis of the social ethogram is still underway.

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Poster

P599. Stress and Emotional Regulation

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Program #/Poster #: P599.05

Topic: F.04. Stress and the Brain

Support: Research Society Grants

Title: Effect of Imipramine on various components of depression in learned helplessness model in C57/BL6 mice

Authors: *P. PATANKAR[1], A. NAIR[3], A. MANE[2], H. HAVALDAR[3], R. SALVI[3], D. KOKATE[4];
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Abstract: Introduction: For treatment of Major depressive disorder (MDD), though imipramine is commonly used, its effectson various components of MDD are not well understood. Imipramine has shown to enhanceneuroplasticity , seen by increase in BDNF levels. We studied the effect of imipramine on variouscomponents of MDD using a battery of behavioural tests for each component viz.i) Apathy: Splash test: duration of groomingii) Despair: Tail suspension test: Ratio of immobile time during 5 miniii) Anxiety: Elevated Plus maze: Time spent in closed armsiv) Psychomotor agitation or retardation: Open-field test: total distance travelledv) Abnormalities of eating behaviour: Change weight of the miceWe are also studying the effect of imipramine on neuroplasticity.**Objectives:**To observe the effect of imipramine on brain BDNF levels analysed through Western Blot techniqueand through the battery of behavioural tests assessing the potential antidepressant effect on eachcomponent.**Materials and Methods:**The methodology is described in figure 1. Assessment by following criteria is done and then animalsare divided as:**CharacteristicsCriteria**1. Complete failure of animals to move despite 10 seconds**Learned HelplessnessFailure** on continuous shock on day 3**(Failure in >7 trials out of**2. Escape latency of >7 seconds out of 10 seconds**10)No.Name of the GroupNumber ofDosage givenAnimalsintraperitoneal(i.p.)**1.Vehicle Control (No induction)130.5 ml NS i.p.2.Disease Control (Induction130.5 ml NS i.p.with vehicle)3.Imipramine1330 mg/kg in 0.5ml NS i.p.**Results:** Analysis was done using repeated measures ANOVA for quantitative data and chi square test forqualitative data. On 7th and 10th day post imipramine dosing, despair component markedly decreasedwhile anxiety component markedly increased. There was significant increase in distance travelledwhile no change was observed in other parameters. Western blot analysis is yet to be done.

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Poster

P599. Stress and Emotional Regulation

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Program #/Poster #: P599.06

Topic: F.04. Stress and the Brain

Support: CONACyT

Title: Cortical thickness in patients with Obsessive Compulsive Disorder: a study by Magnetic Resonance Imaging

Authors: *C. ROSALES MONDRAGÓN, E. H. PASAYE ALCARÁZ, S. ALCAUTER SOLORZANO, A. REYES AGUILAR;
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Abstract: Introduction: Traditionally, OCD has been described as a pathological condition in which the patient experiences intrusive, repetitive and uncomfortable thoughts that can become disabling (obsessions). Later, they resort to certain behaviors or rituals that tend to reduce the feeling of anxiety caused by these thoughts (compulsions). Worldwide, they have found alterations in the amount of gray matter of different structures of patients with obsessive compulsive disorder. The present study aims to measure the cortical thickness and the morphometry of the gray matter, focusing mainly on the cortico-striato-thalamus-cortical circuit in OCD patients with washout and revision dimension and a control group. **Material and methods:** A comparative study was carried out between 23 patients with OCD (10 scrubbers and 13 reviewers) and 23 HC. The images were acquired by means of a G.E. 3.0 T magnetic resonator. In order to carry out the measurement of cortical thickness and the statistical comparison, FreeSurfer and its statistical tool QDEC were used. To perform the morphometry, the structural data were analyzed with FSL-VBM. For the analysis of the volumetric the statistical program IBM SPSS Statistics was used. The comparison of means was carried out by means of the one-way ANOVA, subsequently, a Pearson correlation test was used, in order to identify a linear relationship between the severity index and the volume variable in the different structures under study. **Results:** We found a statistically significant reduction in the density of gray matter in the ventral face of the occipital lobe was found bilaterally, decrease in the density of the anterior lobe of the cerebellum at the level of the predominantly left bilateral quadrangular lobule. A significant increase in the density limited to the medial aspect of the left postcentral gyrus was also found. Also we found a statistically significant linear association was moderately and inversely proportional ($r_P = -0.513$, $p < 0.05$) between the score of the severity index of the patients and the volume gray matter of the left globe pallidus. **Conclusions:** The present results suggest an important role of the cerebellum in the pathophysiology of OCD. In addition, it would link the condition of the globe pallidus within the cortico-striato-thalamus-cortical circuit with the severity of the symptoms, forming part of the anatomical abnormalities involved in the pathophysiology of OCD. However, studies with larger samples are still needed to corroborate these findings. **Acknowledgments:** to the CONACyT, to the Institute of Neurobiology, to the National Laboratory of Magnetic Resonance Imaging (LANIREM).

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Poster

P599. Stress and Emotional Regulation

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Program #/Poster #: P599.07

Topic: F.04. Stress and the Brain

Support: Arkansas Biosciences Institute Grant
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Title: Interaction between the hypothalamic pituitary adrenal (HPA) and thyroid (HPT) axes during immobilization stress

Authors: H. J. KADHIM[1], **P. J. STRAIGHT**[2], *W. J. KUENZEL[2];
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Abstract: Food deprivation (FD) and immobilization (IM) stress have been utilized to determine the sequence of gene activation within the avian HPA axis that includes the septal nucleus of the hippocampal commissure (NHpC), hypothalamic paraventricular nucleus (PVN), anterior pituitary (APit), and adrenal gland. FD or IM stress resulted in an upregulation of corticotropin releasing hormone receptor 2 (CRHR2), located on thyrotropes within the chicken APit, suggesting that stressors not only activated the HPA axis, but stimulated the thyroid axis as well. Therefore, an experiment was designed to study the effect of an IM stressor on gene expression of thyrotropin-releasing hormone (TRH) and its receptors (TRHR1 and TRHR3), urocortin (URO3), deiodinase 2 (DIO2), and a recently discovered second type of CRH named CRH 2 within the neural component of the avian HPA axis. Additionally, TRHR1 and TRHR3, TSHβ and DIO2 mRNA expression were determined in the APit. Male broiler chicks, 5 weeks of age, were divided into six groups, n = 10/trt. Group one was unstressed controls, while 5 groups formed stress groups immobilized for different time periods: 15m (m=minutes), 30m, 60m, 90m, and 120m. Immediately after IM, blood, brain and APit were sampled. Brains were cryosectioned, and the NHpC and PVN were dissected. Total RNA was extracted from the NHpC, PVN, and APit, and cDNA was synthesized for qPCR. Results showed significantly increased CORT levels in all stressed groups. TRH gene expression data displayed significant increases at 90m and 120m of IM in the PVN and at 120m of IM in the NHpC. Of interest, URO3 mRNA showed no significant increase in the PVN, however, a 3-fold increase occurred at 15m in the NHpC. Importantly, CRH2 mRNA showed a 3-fold increase in the NHpC at 60m, 90m, and 120m of IM while the PVN displayed a modest increase only at 120m. TRHR1 mRNA at 15m and TRHR3 mRNA at 60m and 90m were significantly elevated in the NHpC with no significant increases displayed by either receptor in the PVN. DIO2 mRNA was significantly upregulated solely at 60m of IM in the NHpC. At the APit level, TSHβ, TRHR1 and TRHR3

gene expression were significantly upregulated from 15m to 120m treatments of IM stress. Results suggest that an activation of the avian HPA axis using IM stress also stimulates the HPT axis based primarily upon significantly increased gene expression of TSHβ in the APit. However, it appears not due to increased TRH mRNA from two neural structures examined: the NHpC nor PVN. A prime neuropeptide is CRH that showed a significant increase in gene expression following IM that was associated with increased CRHR2 expression within the APit. Other candidates included CRH2 and URO3 peptidergic neurons.

Disclosures: H.J. Kadhim: None. **P.J. Straight:** None. **W.J. Kuenzel:** None.

Poster

P599. Stress and Emotional Regulation

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P599.08

Topic: F.04. Stress and the Brain

Support: GIF Grant I-270-421.10-2016

Title: The neural circuit of enhanced post-traumatic threat detection

Authors: *A. RITTER, O. KLAVIR;
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Abstract: Anxiety disorders in general and posttraumatic stress disorder in particular are associated with heightened levels of arousal. The current work is aimed at investigating the neural networks which regulate threat detection, and their malfunctioning after traumatic experiences. Specifically, we are interested in the neural activity in the cortico-basal ganglia related circuits which may facilitate threat detection by inhibiting the nigro-tectal pathway including the substantia nigra and its connection to the superior colliculus (SC). The striatum as the main input of the basal-ganglia receives dense projections from the medial prefrontal cortex (mPFC). Trauma-related mPFC activity potentially allows the introduction of affective information into the basal-ganglia to modulate the threat detection system and the behavioral responses to threat. Thus, we are investigating alterations in threat detection top-down control as executed by the mPFC, both as it projects directly onto the SC, and indirectly via the striatum and the nigro-tectal pathway. We combine in-vivo electrophysiology and optogenetics together with a behavioral assessment of threat detection in a mouse, 7 days and 21 days after an inescapable electric foot shock. We use a newly developed task, involving a randomly moving robotic bug (robo-beetle), creating incidental interactions with the mouse in a testing arena. In this exploratory investigation, we found, significant differences in fear related behaviors, such as altered timing and distance of avoidance and escaping onset, higher fleeing velocities following an anticipated contact with the robo-beetle and overall, more instances of fear related actions

following the inescapable foot shock. This was measured before the shock, 7 days, and 21 days post shock. Most SC recorded neurons (n=1730) show a firing pattern change which precede fleeing onsets by a few milliseconds, suggesting direct involvement. This change is correlated with both the magnitude and temporal nature of fleeing behavior. Additionally, SC neurons, receiving the different routes of mPFC inputs, respond to different types of fear behavior, suggesting a complex circuit of control and balance, involved in changing threat detection and provoked action after a traumatic event.

Disclosures: A. Ritter: None. **O. Klavir:** None.

Poster

P602. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: Virtual Only

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Program #/Poster #: P602.01

Topic: F.04. Stress and the Brain

Support: NIEHS ES028202
NICHD HD097093
NIMH MH 104184
NIMH MH 108286

Title: Stress-mediated cellular mechanisms of allostasis regulate mitochondrial bioenergetics

Authors: *N. KANYUCH[1], J. CHAN[3], C. P. MORGAN[6], C. MANNELLA[4], N. YADAVA[5], T. L. BALE[2];
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Abstract: Reproductive and developmental outcomes are impacted by parental lifetime experiences including stress, trauma, and infection. In males, cellular reprogramming of reproductive tissues in response to stress poses an intriguing model to examine cellular allostatic mechanisms contributing to lasting post-exposure effects. At the epididymis, sperm receive essential maturation signals via extracellular vesicles produced by epididymal epithelial cells (EECs). Our previous work in mice demonstrates that the content of these signals is altered following chronic stress and that these persisting changes are causal in post-conception neurodevelopmental outcomes. However, the mechanism by which past stress experiences alter the basal EEC set point to maintain these lasting changes is unclear. The glucocorticoid receptor (GR) is a key regulator of the stress response and known target involved in allostasis. To test the hypothesis that stress-dependent EEC programming involves GR, we exposed mice with EEC-

targeted reduction of GR (GR^{Het}) to chronic stress followed by 12 weeks of recovery and evaluated the intergenerational transmission of stress by assessing offspring. In fact, GR^{Het} expression in sire EECs eliminated the offspring neurodevelopmental phenotype. Transcriptomically, genes differentially expressed between wildtype and stress GR^{Het} EECs accounted for two very large clusters of co-regulated genes (over 400 and 1200 genes, respectively) related to chromatin-modifying and mitochondrial processes identified by functional annotation analysis. To further investigate the impact of stress on allostasis, we utilized corticosterone exposure in an *in vitro* EEC model of stress. We identified significant transcriptional downregulation by RNA-sequencing six days after recovery from stress treatment suggesting the involvement of a repressive mark. Using CUT&RUN sequencing, a high efficiency epigenetic profiling approach, we probed the distribution of the transcriptional repressor, H3K27me3. Indeed, we found a robust increase in H3K27me3 at over 7000 peaks following stress treatment. Gene set enrichment analysis of the associated genes revealed associations with mitochondria. Given these data, we assessed the capacity of GR to program basal mitochondrial respiration by cell-based respirometry. While prior stress treatment decreased basal respiration after recovery, GR reduction by targeted shRNA protected against this change. Together, these studies demonstrate a vital role for GR at the intersection of the epigenetic and metabolic consequences of stress to regulate a sustained change in the cellular allostatic set point.

Disclosures: N. Kanyuch: None. **J. Chan:** None. **C.P. Morgan:** None. **C. Mannella:** None. **N. Yadava:** None. **T.L. Bale:** None.

Poster

P602. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: Virtual Only

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Program #/Poster #: P602.02

Topic: F.04. Stress and the Brain

Support: UofSC ASPIRE 1
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UofSC Magellan Mini-Grant

Title: Programming the postnatal mitochondrion: long-term effects of prenatal stress on brain metabolism and behavior

Authors: *B. ROBERTSON[1], E. GORMAN-SANDLER[1], A. CROCKETT[1], J. CRAWFORD[1], D. JOYNER[1], F. HOLLIS[1,2];
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Abstract: Mitochondria are dynamic, energy-supplying organelles that utilize 20% of the body's energy to support critical brain processes. Thus, deficits in mitochondrial energy could have detrimental effects on an individual's health. Indeed, mitochondrial deficits have been associated with several neuropsychiatric disorders including depression and anxiety. Furthermore, we have previously identified a link between individual differences in adult brain mitochondrial function, trait anxiety, and social behavior. However, it is unclear how these individual differences in brain mitochondrial function develop. We hypothesized that experiences in early life might induce long-term alterations in brain mitochondrial function that drive behavioral responses in adulthood. To test this hypothesis, we investigated whether early life experiences can program mitochondrial brain function through exposure to prenatal stress. We exposed pregnant outbred Wistar rats to chronic mild unpredictable stress or control conditions during gestational days 10 through 20. Following birth, we assessed offspring for social, anxiety- and depressive-like behaviors at different developmental timepoints beginning at postnatal day 4 through adulthood. Using high-resolution respirometry, we measured mitochondrial function in the cortex immediately following behavioral assays such as ultrasonic vocalizations, play behavior, and the forced swim test. Our findings indicate that exposure to prenatal stress had notable and sex-specific effects on the postnatal mitochondrion that correlate with differences in offspring behavior, highlighting interactions between prenatal experiences and long-term mitochondrial development.

Disclosures: B. Robertson: None. **E. Gorman-Sandler:** None. **A. Crockett:** None. **J. Crawford:** None. **D. Joyner:** None. **F. Hollis:** None.

Poster

P602. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P602.03

Topic: F.04. Stress and the Brain

Title: Prenatal stress: comparing three models on embryonic cortical interneuron outcomes

Authors: *S. V. MAURER, B. A. KELVINGTON, B. HING, H. E. STEVENS;
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Abstract: Background: Prenatal stress is associated with increased risk of autism and schizophrenia, disorders in which abnormal development and function of cortical interneurons are implicated. Prenatal restraint stress as well as other rodent models of stress lead to broad activation of maternal stress physiology and are translatable to human psychosocial stress. However, consistency across models in impacts on cortical interneurons, which would be expected from common mechanistic pathways, has not been demonstrated. The current work compares three prenatal stress models and their effects on cortical interneuron development.

Methods: CD1 female mice were mated with GAD67-GFP+ CD1 males. Dams were randomly assigned to four experimental groups: control without stress (Ctrl), restraint stress (RS), chronic unpredictable stress (CUS), or foot shock stress (FS). Stress was administered for 45 minutes, 3-4 hours apart, three times daily on embryonic days 12 and 13 (E12 & E13), and once more on the morning of E14. Embryo tissues were collected on E14 one hour after the last stress session. One male and one female per litter was assessed for each outcome. GFP+ offspring brains were fixed, sectioned, immunostained for GFP, and assessed with digital microscopy blind to condition. GAD67GFP+ interneuron progenitor migration was measured as the length of the GFP+ cell migratory path in the cortical pre-plate as a proportion of total cortical surface length. RNA was isolated from GFP- ventral forebrains, cDNA was synthesized, and qPCR performed for genes related to interneuron development. Results: General effects on E14 placental and fetal weight differed by stress model; CUS decreased placental and fetal weight while FS increased placental and fetal weight across both sexes ($p < .01$ for both placental and fetal weight). Interestingly, only RS and CUS decreased E14 interneuron migration across both sexes (86.0% in RS, 87.3% in CUS versus 92.2% in Ctrl, $p < .05$). Surprisingly, in female ventral forebrain, CUS increased expression of *Erbb4*, a gene for a critical receptor underlying chemoattraction of interneurons to the cortex (fold change = 5.1, $p < .05$). Differential effects of these three stress models on other interneuron developmental outcomes will also be reported. Conclusion: Different models of stress lead to some similar and some distinct offspring outcomes, relevant to understanding the underlying mechanisms of risk for neurodevelopmental disorders. Further work is required to assess the underpinnings of these differences.

Disclosures: S.V. Maurer: None. **B.A. Kelvington:** None. **B. Hing:** None. **H.E. Stevens:** None.

Poster

P602. Early-Life Stress: Molecular Mechanisms and Cellular Effects

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Program #/Poster #: P602.04

Topic: F.04. Stress and the Brain

Support: F32HD101302
ES028202
HD097093
MH104184
MH108286

Title: Preconception stress impacts metabolic signaling in maternal extracellular vesicles and sex-specific offspring neurodevelopment

Authors: *Y. CISSE[1], W. HUANG[2], M. A. KANE[2], T. BALE[1];
[1]Univ. of Maryland Sch. of Med., Baltimore, MD; [2]Univ. of Maryland Sch. of Pharm., Baltimore, MD

Abstract: Preconception lifetime adversity is a strong predictor of adverse perinatal outcomes and offspring neurodevelopment. Cumulative stressful life experiences precipitate a state of allostatic load; however, little is known about the biological mechanisms involved in storing and perpetuating this signal across generations. In our mouse model of maternal preconception stress (MPS), stress exposure prior to conception programs offspring-sex specific sensitivity to stress and metabolic insults in adulthood. We hypothesize that preconception allostatic load interacts with the incredible energetic demands of pregnancy, unmasking latent programming affecting highly metabolic tissues such as the placenta, and ultimately affecting offspring development. We first investigated changes in the cargo of circulating extracellular vesicles (EVs) in MPS females. EVs are secreted lipid-bound nano particles that mediate intercellular communication via transport of bioactive compounds, the composition of which is sensitive to cellular stress. Proteomic analysis revealed increased abundance of proteins involved in cellular metabolism in circulating EVs from females 4 weeks post-stress. During pregnancy, control females robustly increased abundance of proteins involved in cellular metabolism, a rise absent in MPS dams suggesting a ceiling effect or diminished response to pregnancy. As the largest contributor of circulating EVs during pregnancy and a sex-specific determinant of offspring growth, we examined the mid-gestation placenta. Although male placentas had a larger transcriptional response to MPS than females, both increased cellular metabolic gene expression, and decreased trophoblast differentiation genes, suggesting disruption in metabolic signaling and placental development. To better understand interactions with the maternal compartment, we examined the transcriptome of the uterus apposing these placentas. In contrast to the placentas, there were robust sex differences in the uterine transcriptome where tissue apposing male placentas showed changes in genes involved in cellular metabolism, but tissue apposing female placentas showed changes in genes involved in innate immunity. Finally, we characterized the mid-gestation fetal brain transcriptome and determined MPS offspring reduced expression of genes involved in neuronal development relative to controls. Together, these data suggest a lasting effect of preconception stress on maternal physiology, detected in circulating EVs that may play an important role as both a peripheral biomarker of maternal state and mode of transmission of MPS effects via changes in their cargo.

Disclosures: Y. Cisse: None. **W. Huang:** None. **M.A. Kane:** None. **T. Bale:** None.

Poster

P602. Early-Life Stress: Molecular Mechanisms and Cellular Effects

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Program #/Poster #: P602.05

Topic: F.04. Stress and the Brain

Support: NIEHS ES028202
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NIMH MH104184
NIMH MH108286

Title: Stress-induced dysregulation of the hypothalamic extracellular matrix shapes HPA axis development

Authors: *K. R. MONTGOMERY, M. S. BRIDI, L. M. FOLTS, T. L. BALE;
Pharmacol., Univ. of Maryland Sch. of Med., Baltimore, MD

Abstract: Lifetime maternal exposure to chronic stress and adversity represent significant risk factors for neuropsychiatric disorder development in their offspring. However, the exact mechanisms by which preconception experiences are transmitted to the next generation and influence neurodevelopment are not well understood. We established a novel mouse model of maternal preconception stress (MPS) where female, but not male, offspring of MPS dams show heightened stress reactivity in adulthood, a hallmark of many neuropsychiatric disorders. To examine the hypothesis that the sex-specific effects of MPS on stress responsivity involve lasting changes to the paraventricular nucleus of the hypothalamus (PVN), the master regulator of the hypothalamic-pituitary-adrenal (HPA) axis, we performed PVN Sim1 neuron-specific RNA sequencing in control and MPS postnatal day 21 (PN21) offspring. Remarkably, differential gene expression analysis showed more than 2200 genes differed in PVN neurons between MPS and control female offspring at PN21. Subsequent analysis revealed only 63 differentially expressed genes between MPS and control male offspring, suggesting that the observed female-specific adult stress phenotype may result from altered PVN neuron development. Functional analysis of the PN21 MPS female differentially expressed genes revealed significant enrichment of genes involved in the production and remodeling of the extracellular matrix (ECM). We then compared gene expression patterns using RNA sequencing in PVN neurons from MPS and control offspring in adulthood and found that female MPS offspring again had the highest enrichment of differentially expressed genes related to the extracellular matrix. Taken together, these data suggest that MPS imparts enduring sex-specific changes to the formation and maintenance of neuronal extracellular matrix in the PVN. The ECM undergoes considerable maturation during development, transitioning from a juvenile state to the adult state beginning in the second postnatal week. This transition coincides with the closure of plasticity windows in specific brain regions, though the majority of this work has been conducted solely in males and in non-hypothalamic regions. Our results suggest that dysregulation of either the developmental rate or the composition of the ECM could lead to lasting effects on PVN circuitry formation that persist into adulthood. Ongoing pharmacological and chemogenetic manipulations will examine the role of hypothalamic ECM in shaping the adult HPA response and how perturbations to neuronal activity during critical developmental windows alter ECM formation and HPA axis organization.

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Poster

P602. Early-Life Stress: Molecular Mechanisms and Cellular Effects

Location: Virtual Only

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Program #/Poster #: P602.06

Topic: F.04. Stress and the Brain

Support: NIEHS ES028202
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NIMH MH104184
NIMH MH108286

Title: Extracellular vesicles as novel stress signals altering placental and fetal development

Authors: *H. ZIERDEN[1], K. E. MORRISON[2], B. M. NUGENT[1], T. L. BALE[3];
[1]Univ. of Maryland Sch. of Med., Baltimore, MD; [2]West Virginia Univ., Morgantown, WV;
[3]Pharmacol., Univ. of Maryland Baltimore, Baltimore, MD

Abstract: Maternal stress is linked to a variety of adverse fetal and pregnancy outcomes. In our well-established mouse model of early prenatal stress (EPS), we observe impaired cognitive function, alterations in metabolic programming, and heightened stress sensitivity, specifically in male offspring. A novel contribution to this stress signaling during pregnancy is extracellular vesicles (EV) largely secreted by the placenta. EVs are cell-derived nanoparticles that participate in intercellular communication by transporting signaling factors, including proteins and noncoding RNA, between cells and tissues. We first examined the role of EVs in stress signaling by isolating circulating EVs from pregnant mice, comparing control and EPS-exposed groups and performing small RNA-sequencing and proteomic analyses to analyze the content of these EVs. We injected labeled EVs into naïve dams, and utilized *in vivo* imaging (IVIS) to identify changes in EV trafficking to maternal and fetal compartments, and to detect transcriptomic changes in placental and fetal tissues. We observed that EVs from control and EPS-exposed dams were similarly trafficked to the placenta and did not appear to enter the fetal compartment. In determination of changes in EV characteristics, we found that the size of circulating EVs remains consistent over the course of gestation (~120 nm), but the concentration of EVs decreases as gestation progresses. Additionally, we observed that the surface charge (zeta potential) of EVs isolated over the course of pregnancy trended towards becoming more neutral. In addition, we found a reduction in overall EV concentration from stress dams compared to controls, and observed differences in the cargo signatures of EVs isolated from the two groups. We hypothesize that vesicle size, concentration, and surface charge of circulating EVs could influence signaling and EV interactions with fetal tissues. Together, these studies provide insight into the role EVs play in promoting stress signals in both maternal and fetal circulation, their interaction at the level of the placenta, and the impact of prenatal stress on important signaling dynamics between maternal and fetal compartments during gestation.

Disclosures: H. Zierden: None. **K.E. Morrison:** None. **B.M. Nugent:** None. **T.L. Bale:** None.

Poster

P602. Early-Life Stress: Molecular Mechanisms and Cellular Effects

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Program #/Poster #: P602.07

Topic: F.04. Stress and the Brain

Support: RO1 Grant 361711

Title: Exposure to a low resource environment alters rat maternal care

Authors: *J. FLOWERS[1], C. ARDEKANI[3], A. JEAN-LOUIS[1], N. SEKHAWAT[1], D. A. BANGASSER[2];
[2]Dept. of Psychology and Neurosci. Program, [1]Temple Univ., Philadelphia, PA; [3]Univ. Of California Davis, Davis, CA

Abstract: Growing up in a low resource environment can induce effects on offspring that endure through adulthood. However, few studies investigate how raising offspring in a low resource environment affects parental brains and behavior. Therefore, our lab utilized the limited bedding/nesting (LBN) manipulation in rodents to mimic this environment. In LBN, dams and pups are put in a cage with a barrier to prevent access to bedding, no enrichment, and only one paper towel for nesting materials from post-natal (PND) days 2-9. This contrasts with control dams who have ample bedding, nesting, and enrichment. The LBN manipulation shifts maternal care such that dams spend more time engaged in pup-directed behavior (e.g., nursing, grooming pups) and less time engaged in self-care (e.g., self-grooming, resting outside the nest). This increase in pup-directed behavior does not prevent ill effects in offspring. This study extends the characterization of LBN dam behavior by assessing pup retrieval. We observed the dams' retrieval latency for each pup and the latency to crouch over the pups once they were all retrieved. Our preliminary results suggest that both LBN (n=5) and control dams (n=4) retrieve their first pup at similar times, but the LBN dams were quicker to retrieve all of their pups to the nest [$t(2)=.30$, $p=.11$]. Once retrieved, both LBN and control dams crouched over the nest at a similar rate. Ongoing studies are also assessing whether LBN dams are more defensive. When combined with our prior findings that LBN dams engage in more pup-directed behaviors, these data suggest that a low resource environment drives dams to be more focused on their pups, suggestive of a hyperarousal phenotype.

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Poster

P602. Early-Life Stress: Molecular Mechanisms and Cellular Effects

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Program #/Poster #: P602.08

Topic:

Support: NIDA R01 DA049837
NIDA Diversity Supplement DA049837

Title: Early life adversity causes sex-specific transcriptional changes in the basolateral amygdala of rats

Authors: *A. CUARENTA[1], C. C. BAVLEY[4], R. KARBALAEI[4], S. FAMULARO[4], M. E. WIMMER[2], D. A. BANGASSER[3];
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Abstract: Early life trauma can increase risk for the development of a variety of psychiatric disorders later in life including depression, anxiety, and substance use disorder—all disorders where dysregulation of the basolateral amygdala (BLA) is reported. However, early life stress that is not overwhelming can have an "inoculating" effect that promotes the development of resilience in adulthood. To understand how early life adversity impacts the basolateral amygdala, we used a rodent model of early life stress called the limited bedding and nesting model (LBN), which mimics a low resource environment by restricting a dam's access to nesting materials during the pups first week of life. Previously we found this manipulation produces a resilient-like phenotype against addiction-related behaviors. For example, LBN reduces impulsive choice, a BLA-mediated behavior, particularly in male rats. We therefore sought to delineate possible molecular underpinnings that promote stress-induced resilience. RNA sequencing was conducted to delineate the effect LBN had on the transcriptional profile of the BLA in adult rats (male control, $n = 4$; female control, $n = 5$; male LBN, $n = 5$; female LBN, $n = 4$). We used rank-rank hypergeometric overlap (RRHO) analysis to compare overall gene expression pattern in males and females induced by LBN. We found that LBN induced sex-specific changes in transcription. Specifically, we see significant distinction between genes upregulated in males and downregulated in females due to LBN. We narrowed our analysis to genes showing a significant difference between control and LBN and found 209 differentially expressed genes (DEGs) in females and 149 DEGs in males. These gene expression changes were predominantly sex specific as only 11 genes were altered by LBN in males and females. Heatmaps organized by fold change of LBN DEGs displayed different patterns of upregulated and downregulated genes in males and females. Of interest *Grin1*, the gene for the glutamate ionotropic receptor NMDA type subunit 1, was downregulated by LBN in females and pathway analysis found LBN - induced changes in glutamate pathways in females. In males, pathway analysis found LBN-induced changes in the MAPK pathways and several MAPKs, such as *MAPK10*, were

upregulated in LBN compared to control males. These analyses highlight unique patterns of gene expression LBN induces within the BLA in a sex-specific manner. Future work will focus on validating top targets with RNAscope and manipulating them in a cell-type specific manner to link changes to addiction-related behavior. Altogether, this work furthers our understanding of the neurobiological underpinnings of stress inoculation.

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Poster

P602. Early-Life Stress: Molecular Mechanisms and Cellular Effects

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Program #/Poster #: P602.09

Topic: F.04. Stress and the Brain

Support: NIH Grant ES028202
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NIH Grant MH108286

Title: Dynamic changes in stress-induced PVN-CRF neuron activity during juvenile development

Authors: *M. S. BRIDI, T. L. BALE;
Pharmacol., Univ. of Maryland Baltimore, Baltimore, MD

Abstract: Stress dysfunction and HPA axis dysregulation are associated with vulnerability to neuropsychiatric disorders. Adverse events or trauma experienced during vulnerable developmental periods such as during pubertal maturation increase disease risk across the lifespan. The paraventricular nucleus of the hypothalamus (PVN) integrates critical information regarding stressful stimuli, and the output of PVN corticotropin-releasing factor (CRF) neurons regulates hormonal and autonomic stress responses required for survival. While adult PVN CRF neurons are highly plastic and quickly return to baseline activity during stress recovery, determination of when this homeostatic setpoint is established during postnatal development is not understood. Further, despite the vulnerability of adolescents to stress-related neuropsychiatric disease, juvenile PVN anatomy and function has received limited attention. To examine how CRF neurons mature relative to HPA axis function over a juvenile period of development, and how adversity experience, modeled as social isolation stress exposure in mice, during this vulnerable period alters this trajectory, we utilized *in vivo* fiber photometry to measure PVN-CRF neuron activity while mice were exposed to acute or chronic stressors, with concurrent collection of tail blood to assay corticosterone levels. Stress-induced CRF calcium activity is

examined in younger animals and compared with their corticosterone response that decreased with age. The impact on this trajectory will be examined in juvenile mice while exposed to chronic social isolation stress following weaning. Our studies are focusing on a developmentally-programmed trajectory of PVN activity and HPA axis activation that is vulnerable to disruption by stress challenge during a specific maturational period. Further investigation must determine the relative contributions of afferent input and intrinsic CRF neuronal properties.

Disclosures: M.S. Bridi: None. **T.L. Bale:** None.

Poster

P602. Early-Life Stress: Molecular Mechanisms and Cellular Effects

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Program #/Poster #: P602.10

Topic: F.04. Stress and the Brain

Support: IA/E/18/1/504310
SreePVF/G/BS/19/1

Title: Accelerated aging associated with a history of early stress: role of mitostasis and neuroinflammation

Authors: *P. R. CHAUDHARI[1], A. SINGLA[1], S. E. FANIBUNDA[2], P. TIWARI[1], S. SURYAVANSHI[1], U. KOLTHUR-SEETHARAM[1], A. D. B. VAIDYA[3], V. A. VAIDYA[1]; [1]Dept. of Biol. Sci., Tata Inst. of Fundamental Res., Mumbai, India; [2]Kasturba Hlth. Society-MRC and Vaidya Lab-TIFR Alliance, Mumbai, India; [3]Med. Res. Centre, Kasturba Hlth. Society, Mumbai, India

Abstract: Early life stress can evoke life-long alterations in anxiety and depression-like behaviour, and accelerate neuronal aging and cognitive decline. However, the molecular and cellular changes evoked by early trauma that contribute to these persistent phenotypes remain only partially understood. Maintaining a fine balance of mitostasis (mitochondrial biogenesis and mitophagy) is essential to the health of neurons. Mitochondrial dysfunction and neuroinflammation are implicated as a key precursor events that can drive neurodegenerative changes in the aging brain, and are linked with increased risk for stress-related psychopathology. Here, we sought to address the influence of a history of early stress on mitochondrial biogenesis, function, and mitophagy, as well as the consequences on both peripheral and central inflammation, focussing in the brain on the hippocampus which is particularly susceptible to neuroinflammation. A history of maternal separation (MS), an early stress model in rodents, results in enhanced adult anxiety, depression-like behaviour, accelerated aging, and perturbed neuroendocrine stress responses that endure across the life span. We carried out the MS paradigm in Sprague Dawley rats, which involved separation of the entire litter from the dam,

once daily for 3 hours from postnatal day 2 (P2) to P14. The influence of MS was then assessed at multiple time points across the life-span in key limbic neurocircuits, in particular in the hippocampus, prefrontal cortex and the hypothalamus of both male and female rats using cellular, molecular, neuropharmacological, imaging, and behavioural approaches. We provide evidence that a history of MS is associated with a dysregulation of multiple mitochondrial measures in key limbic brain regions of middle-aged MS rats. We noted a robust dysregulation in mitostasis, with a reduction in mitochondrial biogenesis and an increase in mitophagy, associated with a deterioration of mitochondrial function, accompanied by an inflammatory state evoked in the hippocampus and prefrontal cortex as MS animals age. We also analysed serum for circulating proinflammatory signatures and observed a robust induction in proinflammatory molecules, commencing as early as 6 months and noted to worsen as animals hit middle-age. These studies motivate future investigation to address whether strategies that intervene and ameliorate the inflammation associated changes can also reverse the mitochondrial and inflammatory changes that we observe following early stress.

Disclosures: P.R. Chaudhari: None. **A. Singla:** None. **S.E. Fanibunda:** None. **P. Tiwari:** None. **S. Suryavanshi:** None. **U. Kolthur-Seetharam:** None. **A.D.B. Vaidya:** None. **V.A. Vaidya:** None.

Poster

P754. Opioids: Reinforcement, Seeking, and Reinstatement I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P754.01

Topic: G.09. Drugs of Abuse and Addiction

Support: DA050116
DA050116-02S1

Title: Goal-directed decision making is disrupted during morphine withdrawal

Authors: *B. HALBOUT, Z. A. SPRINGS, C. R. HUTSON, K. K. BODINAYAKE, S. B. OSTLUND;
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Abstract: Drug-related cues are known to play several distinct roles in regulating drug-seeking and drug-taking behaviors. Drug-related cues can directly motivate drug use and relapse. Moreover, growing evidence shows that these cues can be distracting and may therefore disrupt ongoing cognitive control processes that support adaptive, goal-directed decisions, which might pave the way for maladaptive drug-seeking behavior. While clinical research on this subject has gained traction, there have been few such preclinical studies. We developed a novel rodent behavioral model to study the disruptive influence of morphine-related cues on goal-directed

action selection. Adult male Long Evans rats (n = 7-8 / group) were first trained with two different action-outcome contingencies (e.g., left press --> grain & right press --> sucrose). This was followed by a cue learning phase, during which separate multimodal contextual cues were paired with repeated morphine ($CS_{MORPHINE}$) or saline (CS_{SALINE}) administration. We then tested the rats' ability to choose between the two actions based on expected reward value using a well-established outcome devaluation task. This test took place in the presence of $CS_{MORPHINE}$ or CS_{SALINE}, immediately after morphine administration (Early Withdrawal) or following 21 days of withdrawal (Late Withdrawal) allowing us to determine the tendency of the morphine-paired cue to transiently interfere with goal-directed behavioral control (i.e., sensitivity to outcome devaluation). We found that morphine-exposed rats were insensitive to devaluation when tested in Early but not Late Withdrawal and that this effect was not dependent on $CS_{MORPHINE}$ or CS_{SALINE}. Brief re-exposure to morphine in Late Withdrawal also re-instated insensitivity to outcome devaluation. On the other hand, control groups that received saline administration in the presence of both contextual cues remained sensitive to outcome devaluation. Together, these data suggest that acute morphine withdrawal is associated with a deficit in goal-directed decision making. Ongoing experiments using sucrose licking tasks are assessing the influence of withdrawal and morphine-paired context on the motivational and hedonic processes that govern reward consumption.

Disclosures: B. Halbout: None. **Z.A. Springs:** None. **C.R. Hutson:** None. **K.K. Bodinayake:** None. **S.B. Ostlund:** None.

Poster

P754. Opioids: Reinforcement, Seeking, and Reinstatement I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P754.02

Topic: G.09. Drugs of Abuse and Addiction

Support: NIDA IRP

Title: Evaluation of Oliceridine as a high selectivity, low potency μOR radioligand for molecular imaging

Authors: *E. VENTRIGLIA[1], S. LAM[1], Y. SHAHAM[1], J. BONAVENTURA[2,3], M. MICHAELIDES[1];
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Abstract: A μOR radioligand with a low potency— compared to that of currently available fentanyl derivate radioligands— would reduce the pharmacological activation of the radioligand during molecular imaging studies, ideally without compromising affinity and selectivity of the

μOR. In this research, we investigate the biased μOR agonist Oliceridine (TRV-130) as a potential radioligand candidate via molecular imaging and behavioral paradigms. Using *in vitro* competition and *ex vivo* GTPγS binding assays in rodents, we have demonstrated Oliceridine's high affinity and low efficacy for brain μOR across both sexes. Similarly, using *in vivo* ARG, we found that pharmacological doses specifically bind to μOR rich areas of the rat brain. On the other hand, [^{3}H]Oliceridine appears not to enter the brain at trace doses and is instead sequestered within ventricles. This suggests that trace doses but not larger, pharmacological doses of Oliceridine are actively excreted from the brain by efflux pumps expressed throughout endothelial cells of the blood brain barrier. To overcome this, we found that co-injection of 1mg/kg Tariquidar, a P-glycoprotein efflux pump blocker, increased [^{3}H]Oliceridine brain uptake in μOR rich areas of the brain in *in vivo* ARG. To characterize the addiction-relevant behavioral response towards Oliceridine in an enhanced uptake condition, we are employing transgenic P-glycoprotein 1 knock-out mice in a conditioned place preference paradigm. Our results serve to facilitate the utilization of Oliceridine as a radioligand when co-administered with a P-gp blocker, enabling trace doses to bind to brain μOR with minimal G-protein recruitment.

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Poster

P754. Opioids: Reinforcement, Seeking, and Reinstatement I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P754.03

Topic: G.09. Drugs of Abuse and Addiction

Title: Calcium permeable AMPA receptors in the nucleus accumbens core and shell mediate the incubation of oxycodone craving

Authors: J. R. NOWLIN[1], B. W. C. WONG[1], A. R. ZIMBELMAN[2], K. CONNESS[1], B. O'BRIEN[1], J. GREENBERG[1], M. VOGEL[1], ***M. T. STEFANIK**[1];
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Abstract: The enduring craving for drugs of abuse is a major contributing factor to the relapse to drug seeking. Cue-induced craving progressively intensifies ('incubates') over weeks of forced abstinence or withdrawal. This 'incubation of craving' has been observed during withdrawal from many different classes of abuse in rodents and humans, yet relatively little is known about this phenomenon or its mechanisms regarding the widely-prescribed prescription opioid, oxycodone (Oxy). Incubation for psychostimulants depends on time-dependent strengthening of AMPA receptor (AMPAR) transmission onto medium spiny neurons in the nucleus accumbens

(NA) through the accumulation of high-conductance, GluA2-lacking, Ca^{2+}-permeable AMPARs (CP-AMPARs). Recent studies have shown that oxycodone craving incubates, and our lab has identified CP-AMPAR emergence in the NA during this timeframe. However, it is not known if these receptors are functional contributors to the incubation of oxycodone craving. In the current study, we microinjected NASPM, a selective CP-AMPAR antagonist, directly into the nucleus accumbens core (NAcore) or shell (NAshell) subregions immediately before a seeking test. We hypothesized that Oxy craving would incubate and that if Oxy incubation was neurobiologically similar to incubation for other drugs of abuse, drug-seeking behavior would be significantly reduced in rats that received NASPM microinjections directly into the NA. Male Sprague Dawley rats underwent 10 days of extended-access Oxy or saline self-administration (6hr/day, 0.15mg/kg/infusion). In a counterbalanced design, immediately before 30-minute cue-induced seeking tests on withdrawal day 1 (WD1; when craving is low) and withdrawal day 15 (WD15; when craving has incubated), animals received bilateral microinjections of either NASPM (40 µg/side) or aCSF. Results show that NASPM microinjections into either subregion significantly reduced the incubation of drug craving in oxycodone self-administering animals on WD15 but not on WD1, and had no impact on saline animals. Together, these studies provide critical insight on the anatomical locus for CP-AMPAR action and its functional role in driving incubated oxycodone craving.

Disclosures: J.R. Nowlin: None. **B.W.C. Wong:** None. **A.R. Zimbelman:** None. **K. Conness:** None. **B. O'Brien:** None. **J. Greenberg:** None. **M. Vogel:** None. **M.T. Stefanik:** None.

Poster

P754. Opioids: Reinforcement, Seeking, and Reinstatement I

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Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant DA048336

Title: Enhancement of compulsive-like opioid self-administration, and seeking, that persists despite two different forms of punishment-induced abstinence.

Authors: *S. HONEYCUTT, M. PALADINO, A. MUKHERJEE, G. LONEY;
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Abstract: Nicotine and opioid use disorders (OUDs) are highly comorbid. Critical to the formation and maintenance of OUDs are drug-associated memories (e.g., contextual cues). Contextual cues can become associated with the reinforcing and aversive properties of drugs, which then coordinate later motivated behaviors. Our lab previously demonstrated that systemic and intra-insular cortex treatment with nicotine modulates contextual opioid conditioning and

enhances opioid intravenous self-administration (IVSA), supporting the role of nicotine in modulating opioid-associated contextual learning in a manner that facilitates OUDs. Here, we employed a model of punishment-induced abstinence wherein remifentanil (RMF) IVSA was explicitly unpunished in one context while infusions in a different context resulted in contingent punishment. These models are often used for their translational value because they model the self-imposed abstinence observed in humans which can follow experience with the deleterious consequences of drug use. We examined the effect of nicotine (0.4 mg/kg) or saline treatment, in rats, prior to RMF (3.2 µg/kg/infusion) IVSA sessions. RMF IVSA occurred in two alternating contexts. One context was unpunished, and, in another context, infusions were punished by one of two forms of punishment. In one group, we used an interoceptive punisher (escalating histamine adulteration). In the second group, we used an exteroceptive punisher (escalating foot-shock intensity). Subsequently, we examined the motivation to obtain RMF in progressive-ratio and extinction sessions in both contexts. We found that, regardless of punisher, rats treated with nicotine, relative to saline, took more RMF regardless of punishment and had higher breakpoints in both the unpunished and punished contexts. Additionally, nicotine treated rats displayed decreased latencies to first RMF infusion and first earned foot-shock. Similarly, nicotine treatment slowed extinction of RMF seeking and increased the propensity for cue exposure to induce relapse to RMF seeking within the punished context. Comparisons of the two forms of punishment revealed differential effects such that histamine adulteration was less effective than footshock and did not support measures of contextual discrimination between unpunished and punished contexts. In summary, nicotine enhances compulsive-like opioid seeking, and bolsters against negative consequences of drug taking in punishment associated contexts. However, the form of punishment differentially effects the continued abstinence from opioid seeking.

Disclosures: S. Honeycutt: None. **M. Paladino:** None. **A. Mukherjee:** None. **G. Loney:** None.

Poster

P754. Opioids: Reinforcement, Seeking, and Reinstatement I

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Program #/Poster #: P754.05

Topic: G.09. Drugs of Abuse and Addiction

Title: Effects of clonidine on the expression of morphine withdrawal symptoms precipitated by naloxone in the mouse and in the rat

Authors: G. FROGET, C. FROGER-COLLEAUX, ***D. PUSHETT**, B. PEAN, C. PERRAUDEAU, V. CASTAGNE;
Porsolt SAS, Le Genest-Saint-Isle, France

Abstract: Naloxone is widely used to treat opioid overdose, although it induces serious side effects. Clonidine, an alpha-2-adrenergic agonist, is frequently prescribed to attenuate symptoms

of opioid detoxification. In this study, we aimed to evaluate the effects of clonidine in two rodent models of morphine withdrawal, precipitated by naloxone administration. Male and female Swiss mice (ten per group) were made morphine-dependent following seven administrations over two days. Two hours after the last morphine administration, withdrawal (assessed by jumping behavior) was precipitated by the injection of naloxone. The effect of clonidine on the number of jumps was observed over a 10-minute period. Male Sprague Dawley rats (four per group) were made morphine-dependent with twice daily administrations of escalating doses over ten days. Two hours after the last morphine administration, withdrawal was precipitated by naloxone injection and the effects of clonidine on body weight, body temperature, behavioral symptoms and stool consistency, were assessed. Clonidine significantly reduced the number of jumps induced by naloxone injections in morphine-dependent mice. The effects were more marked in female (-73%, $p < 0.01$) than in male mice (-55%, $p < 0.05$). In morphine-dependent rats, clonidine reduced the fecal output (-100%), did not affect the loss of body weight and potentiated hypothermia (-1.5°C max). Conversely to its effects in the mouse, clonidine increased naloxone-induced jumping behavior (+ 5262%). The naloxone-precipitated withdrawal model in the mouse using jumping as the main endpoint is a good screening tool to evaluate pharmacological treatments, potentially alleviating the side effects of opiate detoxification. The rat model using fecal output as the main endpoint, while less amenable to screening studies, allows for the evaluation of a broader range of potential symptoms associated with the opiate detoxification.

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Poster

P754. Opioids: Reinforcement, Seeking, and Reinstatement I

Location: Virtual Only

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Program #/Poster #: P754.06

Topic: G.09. Drugs of Abuse and Addiction

Support: Engdahl Family Foundation

Title: Sex differences in distress-related behavior and reinstatement of opioid seeking in mice that witness a conspecific under predatory threat

Authors: T. MATVEEVA[1,2], Z. L. MATTI[2], C. W. COLLINGE[2], E. H. TRESSLER[4], ***J. C. GEWIRTZ**[2,3];
[1]Psychology, Cornell Univ., Itheca, NY; [2]Psychology, [3]Neurosci., Univ. of Minnesota, Minneapolis, MN; [4]Biol., U. Minnesota, Minneapolis, MN

Abstract: Witnessing other people in distress ("observational distress") is a significant source of pathogenic stress in human populations. We have shown in male mice that witnessing a conspecific mouse under predatory threat induces a variety of distress-related behaviors in the observer, including observational distress-induced reinstatement (ODIR) of opioid seeking. Here, we investigated whether male and female mice showed divergent responses following the observation of a conspecific under predatory threat.Male and female C57/Bl6 mice were trained in a conditioned place preference (CPP) paradigm, in which one chamber was paired with morphine (15mg/kg), and the opposite chamber with saline. Animals underwent 4 days of conditioning in each chamber, followed by extinction. Mice then observed the distress of a same-sex conspecific exposed to a male Sprague-Dawley rat. Distress-related behaviors (freezing, escape, burrowing, etc.) were recorded. In separate groups, we also investigated the role of oxytocin, which we and others have found to be critically important in the expression of observational distress. The exogenous neuropeptide was administered intranasally (20 μg/kg) either acutely or chronically, and the oxytocin antagonist L-368,899 hydrochloride was administered systemically (IP; 10 mg/kg). In male mice, observational distress induced reinstatement of CPP, as well as aggression towards a conspecific, primarily when the observer and demonstrator were familiar with each other. Moreover, the degree of reinstatement in the observers was predicted by the number of defensive and aggressive behaviors shown by their respective demonstrators. Female showed distress and reinstatement irrespective of their familiarity with their dyadic partner. In further contrast to males, they exhibited huddling behavior, not aggression, when returned to their home cage. Administration of oxytocin increased observational distress in male unfamiliar observers, thereby eliminating the familiarity bias. In both sexes, administration of the oxytocin antagonist rescued observational distress-related behavior.These data reveal sex-related differences in responding to the distress of another. Our paradigm may offer a means of further investigating sex differences in the induction of psychosocial stress and in stress-induced reinstatement in mice.

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Poster

P754. Opioids: Reinforcement, Seeking, and Reinstatement I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P754.07

Topic: G.09. Drugs of Abuse and Addiction

Support: R01DA035943

Title: Fentanyl self-administration by rats engages the central nucleus of the amygdala

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Abstract: Opioid use disorders (OUD) can be conceptualized as maladaptive 'wanting' or craving of opioids, regardless of potential consequences or alternative reinforcers. The central nucleus of the amygdala (CeA) has been demonstrated to modulate motivation for natural and drug reinforcers, but there has been limited exploration of its role in self-administration models of OUD. To assess the role of the CeA in opioid self-administration, we trained male (n=20) and female (n=10) Sprague-Dawley rats to self-administer the synthetic opioid, fentanyl (0.5μg/i.v. infusion), on an FR1 schedule in 2-hr daily sessions. Rats achieved robust and stable intake after training for 28 sessions. Using GABA agonists, baclofen and muscimol, we found that CeA inactivation attenuated the mean number of fentanyl infusions, supporting a role for this region in fentanyl reinforcement. To probe the role of specific CeA transmitters previously implicated in appetitive processes, we microinfused either the opioid receptor antagonist, naltrexone, the 5-HT_2 receptor agonist, 2,5-dimethoxy-4-iodoamphetamine (DOI), or the dopamine receptor antagonist, flupenthixol, prior to self-administration sessions. Blockade of CeA opioid receptors by naltrexone increased intake and decreased inter-response intervals, consistent with CeA opioid receptors being key mediators of fentanyl reward. DOI reduced fentanyl intake, particularly during the first 15 minutes of the session when motivation to take drug was likely highest, indicating that 5-HT_2 receptors in the CeA modulate drug consumption when motivation to consume drug is high. While flupenthixol administered to the CeA failed to alter mean intake, it did shift the distribution of inter-response intervals, increasing the frequency of intervals under 120 seconds. None of these effects interacted with sex. Taken together, these results demonstrate that the CeA is engaged by fentanyl self-administration when subjects have been trained under conditions that do not typically induce strong dependence.

Disclosures: E. Chaloux-Pinette: None. **M. Castro:** None. **P.H. Janak:** None.

Poster

P754. Opioids: Reinforcement, Seeking, and Reinstatement I

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Topic: G.09. Drugs of Abuse and Addiction

Support: OVPR 2019-OSU CHS
TSET/OCASCR

Title: Differential rearing effects on the gut-brain axis after oxycodone precipitated withdrawal in adolescent rats

Authors: A. G. TORRES[1], Z. SIMSEK[1], S. RIVERA[2], T. RICHARDSON[2], S. ASSEFA[3], G. KOEHLER[3], *D. VAZQUEZ SANROMAN[1];
[1]Anat. and Cell Biol., [2]Col. of Osteo. Med., [3]Microbiology and Biochem., Oklahoma State Univ. Ctr. for Hlth. and Sci., Tulsa, OK

Abstract: Chronic use of opioids leads to the development of dependence. Environmental enrichment (EE) prevents oxycodone-induced withdrawal in adult rats. After chronic oxycodone use, adolescents are more susceptible to gastrointestinal disorders (nausea, constipation) compared to adults, with effects on intestinal homeostasis. However, opioid effects on the gut-brain axis activity during oxycodone withdrawal remain unknown. Brain-derived neurotrophic factor (BDNF) plays important roles in controlling cell survival in the nervous system and enhancing gastrointestinal motility. Hence, 1) we will investigate correlations of differential rearing with changes in the adolescent gut microbiome and 2) determine the effects of oxycodone-induced withdrawal in brain/gut BDNF levels. **Methods:** Adolescent male Sprague-Dawley rats were raised in isolated condition (IC), standard social condition (SC), or enriched condition (EC). On postnatal day 51 oxycodone dependence was initiated using a passive injection model (1, 2, 3, and 5 mg/kg) every 12 h for 5 days. Chronic oxycodone administration facilitated tolerance and physical dependence, demonstrated by withdrawal symptoms. On the 6th day of administration, rats received a naloxone injection (1 mg/kg) precipitating withdrawal. Brains were collected for proBDNF and mBDNF immunoassay analyses. Gut microbiota compositions were analyzed using 16S rRNA gene amplicon sequencing in baseline and time-course fecal samples as well as in intestinal samples contents. **Results:** IC rats showed an increase in sniffing, grooming, and rearing, while all withdrawal features were not observed in the EE rats. Social isolation followed by oxycodone withdrawal enhanced proBDNF and mBDNF expression in the Prefrontal Cortex. Differential rearing conditions correlated with compositional changes in the gut microbiota.

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Poster

P754. Opioids: Reinforcement, Seeking, and Reinstatement I

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Topic: G.09. Drugs of Abuse and Addiction

Support: MRC Grant MR/N02530X/1

Title: Neural and behavioural signature of the compulsive relapsing nature of heroin addiction in the rat

Authors: *M. FOUYSSAC[1], A. BELIN-RAUSCENT[1], M. SIDDLE[2], M. PUAUD[1], B. J. EVERITT[1], D. J. BELIN[1];
[1]Psychology, Univ. of Cambridge, Cambridge, United Kingdom; [2]Developmental Neurosciences, Univ. Col. London, London, United Kingdom

Abstract: Addiction has been hypothesised to result from a transition from volitional drug use to the compulsive seeking and taking of drugs that occurs only in a subset of recreational users. The neural and behavioural factors underlying the vulnerability to switch to compulsive drug seeking are increasingly being understood for stimulant drugs, but less so for opiates. Here we present a novel procedure in the rat that has enabled the identification of behavioural and molecular markers of the vulnerability to develop compulsive heroin seeking behaviour. In this novel procedure, the individual tendency to persist in seeking heroin in a drug-free state despite contingent punishment (mild footshock) was revealed in 25 individuals from a large cohort of 89 Sprague Dawley rats with an extended history of heroin seeking maintained by heroin-associated cue presentations. In a series of longitudinal studies, drug naive rats were tested for several addictive behaviour-relevant traits including anxiety, decision making, sensation and novelty seeking as well as reward (sweetness) preference, prior to being trained to seek heroin under the influence of the conditioned reinforcing properties of heroin-paired cues for several weeks (a second-order schedule of reinforcement). The compulsive nature of heroin seeking behaviour was then measured over several sessions during which mild footshock punishment was delivered contingent on seeking responses prior to, but not during, the administration of heroin. The individual tendency to persist in seeking heroin in the face of punishment was shown to be predicted by a sign-tracking phenotype while that to relapse after punishment-induced self-abstinence was associated with a high anxiety trait. This individual vulnerability was associated with an alteration in the adenosinergic system in the nucleus accumbens in that rats identified as highly compulsive had higher mRNA and protein levels of the adenosine 2A receptor in the ventral striatum as compared to low compulsive rats. These adaptations were further shown to predict compulsive heroin seeking at the population level. Together, these data indicate that high anxiety and a propensity to ascribe incentive value to conditioned stimuli predict the vulnerability to express the compulsive relapsing nature of heroin addiction that may be mediated by the adenosinergic system in the ventral striatum. These data offer new avenues to developing therapeutic strategies to target the compulsive relapsing nature of heroin seeking that represents a hallmark feature of addiction.

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Poster

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Topic: G.09. Drugs of Abuse and Addiction

Support: NIH P20GM113109

Title: Taste Reactivity to Sucrose in Differentially Reared Rats Altered by Mu Opioid Receptor Functioning

Authors: *D. LAUX[1], T. J. MOSER[2], M. C. AZUMA[1], J. K. GOMENDOZA[1], M. CAIN[3]; [2]Dept. of Psychological Sci., [1]Kansas State Univ., Manhattan, KS; [3]Psychological Sci., Kansas Sate Univ., Manhattan, KS

Abstract: It is well established that rearing in isolated environments during adolescence increases incentive salience for reinforcers, compared to animals reared in enriched environments. Our current research suggests that isolated rats emit less hedonic responses ('liking') to sucrose in a taste reactivity paradigm, despite their increased operant responding ('wanting') for sucrose. Interestingly, Mu (μ) opioid receptors (MORs) are critical for determining hedonic value. While it is established that MOR function is critical for hedonic responding, MOR function during hedonic responding has not been investigated in differentially reared rats. In the current experiment, we examine changes in hedonic responding for sucrose in differentially reared rats, while enhancing MOR function via morphine injections. Male and female Long-Evans rats arrived on postnatal day 21 and were randomly assigned to rear in isolated (IC), standard (SC) or enriched (EC) conditions for 30 days. Post-weaning, rats were implanted with indwelling intraoral fistulas. Following recovery, rats underwent taste reactivity trials for sucrose, utilizing a within-subjects design with a counterbalanced order for morphine injections (0, 2 and 4 mg/kg, sc) and sucrose concentrations (0, 0.1M and 0.5M). Taste reactivity data collected via video recording was analyzed frame by frame to quantify orofacial hedonic and aversive responses. Our data suggests that high doses of morphine reduces hedonic responding for sucrose only in IC rats. Comparatively, EC rats with high doses of morphine display increased hedonic responding for sucrose. These data indicate that activation of MORs may decrease 'liking' in isolated rats, and increase 'liking' in enriched rats. These results suggest that isolation rearing may alter MOR function to change incentive salience for reinforcers.

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Poster

P819. Cortical Networks I

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Title: Dopamine facilitates associative memory coding in the lateral entorhinal cortex

Authors: *J. LEE[1], H. JUN[1], S. SOMA[2], T. NAKAZONO[3], K. IGARASHI[1];
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[3]Fukushima Med. Univ., Fukushima, Japan

Abstract: Dopamine plays a central role in motivation and reward. Accumulating evidence shows that dopaminergic inputs in the ventral tegmental area (VTA)-striatum circuit are critically involved in reward-based learning. However, it remains unclear how dopamine reward signals control the entorhinal-hippocampal circuit, another brain network critical for learning and memory. Here we show that dopaminergic inputs from the VTA/substantia nigra to the lateral entorhinal cortex (LEC) are critical for cue-reward associative memory. Optogenetic inhibition of LEC dopamine inputs, or inhibition of LEC layer 2a fan cells, impaired the learning of olfactory cue-reward association, although the retrieval of pre-learned memory was spared. Photometry recordings revealed that LEC dopamine transmits novelty-induced reward expectation signals. Using optogenetic-assisted electrophysiological recording during associative learning, we found a cue-reward association coding of LEC fan cells, where representations of newly learned rewarded cues gradually overlapped with that of a pre-learned rewarded cue while separating them from the representation of a pre-learned unrewarded cue. This coding of LEC fan cells was disrupted by the inhibition of LEC dopamine inputs. These results indicate that dopamine functions as a powerful memory gatekeeper for the entorhinal cortex to encode cue-reward associative memory, pointing to the involvement of the entorhinal cortex in dopamine-mediated reinforcement learning.

Disclosures: J. Lee: None. **H. Jun:** None. **S. Soma:** None. **T. Nakazono:** None. **K. Igarashi:** None.

Poster

P819. Cortical Networks I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P819.02

Topic: H.08. Learning and Memory

Support: NSERC PGSD2 - 535097 - 2019

Title: Medial prefrontal cortex routes relevancy of ongoing events to the nucleus reuniens

Authors: *X. YU[1], K. TAKEHARA-NISHIUCHI[2];
[1]Cell and Systems Biol., [2]Dept. of Psychology, Univ. of Toronto, Toronto, ON, Canada

Abstract: Established literature suggests the medial prefrontal cortex (mPFC) works concertedly with the hippocampus (HPC) during the consolidation and retrieval of event memories; however, their interaction during encoding remains unclear. By using a HPC-dependent differential associative learning task, we have shown that the mPFC changes its network activity patterns depending on whether a neutral conditioned stimulus (CS) is predictive of an upcoming aversive stimulus. Moreover, augmenting this relevancy-selective activity facilitated the formation of stimulus associations. This neural signal could thus be a mechanism through which the mPFC enhances the contrast between relevant and incidental events and facilitates the encoding of the former in the hippocampus. Since the mPFC lacks direct projections to the HPC, the exact pathways through which this relevancy signal reaches the HPC remains unknown. To address this point we used fiber photometry to monitor mPFC axon terminals in the nucleus reuniens (RE) and mediodorsal thalamus (MD), the former of which possess direct HPC projections. Terminal calcium activity was recorded as subjects were trained in a differential trace eyeblink conditioning task wherein one CS was paired with a mild eyelid shock (CS+) and the other was presented by itself (CS-). Before conditioning, presentations of the CS by themselves elicited comparably small increases of calcium activity in terminals within the RE and MD. Once stimulus pairings started, mPFC terminals in the RE began exhibiting significantly larger calcium responses to the CS+ compared to the CS-. Responses to the CS+ also increased in magnitude across conditioning sessions. Conversely, in terminals within the MD, both the CS+ and CS- elicited more comparable calcium responses. Furthermore, the magnitude of the responses to either CS type did not significantly increase across sessions. These findings suggest that mPFC output to the RE differs based on the predictive qualities of the CS, and that this differentiation was much less apparent with output to the MD. The relevancy signal is thus likely to be routed to the RE, whilst information more related to the physical sensory aspects of a stimuli are routed to the MD. We therefore propose that the mPFC exerts executive control over hippocampal encoding by signaling the relevance of ongoing events via RE.

Disclosures: X. Yu: None. **K. Takehara-Nishiuchi:** None.

Poster

P819. Cortical Networks I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P819.03

Topic: H.08. Learning and Memory

Support: NIH Grant R01MH114870
NIH Grant P41EB019936

Title: Accuracy on recognition task preceding resting state scan predicts differential patterns of correlation between default mode and dorsal attention networks in individuals with depressive disorders compared to healthy controls.

Authors: S. SATZ[1], Y. O. HALCHENKO[2], R. RAGOZZINO[1], M. LUCERO[1], M. L. PHILLIPS[3], H. A. SWARTZ[3], ***A. MANELIS**[3];
[1]Psychiatry, Univ. of Pittsburgh Med. Center, Univ. of Pittsburgh, Pittsburgh, PA; [2]Psychology, Dartmouth Col., Hanover, NH; [3]Psychiatry, Univ. of Pittsburgh, Pittsburgh, PA

Abstract: *Introduction:* Previous studies suggested that resting state functional connectivity is affected by experiences preceding resting state. Little is known about the impact of memory task performance on subsequent functional connectivity within and between resting state networks in individuals with and without depressive disorders (DD). Characterizing this relationship is important for understanding aberrant memory consolidation in depression. *Methods:* 52 individuals with DD (age=28.2±0.9, 42 female*)* and 45 healthy controls (HC, age=29.0±1.0, 33 female*)* completed clinical interviews, and a food and object encoding task followed by a forced-choice recognition test. A 5-min resting state fMRI scan was administered immediately after the forced-choice task. Resting-state data were preprocessed using *fmriprep* and band-pass filtered (0.01Hz-0.1Hz). Resting state networks were determined using ICA across all 97 participants and were cross-correlated with the 7-network solution from Yeo (2011) using *fslcc*. The resulting networks served as inputs to *dual-regression* for within-network analyses and *FSLNets* for between-network analyses. We tested how diagnostic status moderated the relationship between memory accuracy and connectivity measures within and between resting state networks. *Results*: There was no significant effect of diagnosis-by-memory interaction on within-network connectivity. *FSLNets* revealed that the correlation between functional connectivity in the default mode network (DMN) comprised of the medial prefrontal cortex, posterior cingulate cortex and hippocampal formation, and functional connectivity in the dorsal attention network (DAN) comprised of the inferior temporal, parietal, and prefrontal cortices, was significantly associated with the interaction between recognition accuracy and diagnostic status (p=0.02). Specifically, a stronger positive correlation between these two networks was observed in individuals with DD who showed higher memory accuracy prior to resting state. No such effect was observed for HC. *Conclusion:* The DMN and DAN typically anticorrelate. Our findings that individuals with DD, but not HC, showed stronger positive correlation between these two networks in the context of accurate memory performance preceding resting state, suggests aberrant post-task memory consolidation processes. Such aberrant memory consolidation may lead to continuous “play back” of previously learned information and underlie rumination on negative thoughts and experiences in depression. Previous findings that DMN activity is related to the level of detail in thoughts partially supports this account.

Disclosures: S. Satz: None. **Y.O. Halchenko:** None. **R. Ragozzino:** None. **M. Lucero:** None. **M.L. Phillips:** None. **H.A. Swartz:** None. **A. Manelis:** None.

Poster

P819. Cortical Networks I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P819.04

Topic: H.08. Learning and Memory

Support: NIMH (1R01MH125557)

Title: The role of sleep in the formation of indirect memory associations

Authors: *T. TADROS[1], M. V. BAZHENOV[2];
[1]UCSD, La Jolla, CA; [2]Dept. of Med., Univ. of California San Diego, La Jolla, CA

Abstract: Relational memory, the ability to make and remember associations between objects remains an essential component of mammalian reasoning. In relational memory tasks, it has been shown that periods of offline processing, such as sleep, are critical to improving one's ability to make indirect associations or transitive inferences. For example, one may learn to associate two items (A and B) and later learn to associate B with a new element (item C) during the wake state. After briefly learning these associations, a subject can recall item B (also called "linking" item) when presented with item A or C, but is less adept at recalling item C. Behavioral research has shown that the duration of slow-wave sleep (SWS) following such is significantly correlated with the subject's ability to recall item C when presented with item A. Based on the empirical evidence that sleep improves relational memory, in this study, we built a Hodgkin-Huxley-based model of the thalamo-cortical network to understand how SWS can lead to improvements in an unordered relational memory task. The cortical network was composed of two layers, both including and excitatory and inhibitory neurons; the first layer represented the perception of an individual object (e.g. visual cortex) and the second layer represented higher-order processing (e.g. associative cortex). Feedforward connections from the first layer to the second layer and recurrent connections within the second layer were plastic and modified through spike-timing dependent plasticity (STDP) rules during training and sleep. The model was first trained on a paired associate inference task, where four pairs of items (e.g., A+B, B+C, X+Y, Y+Z) were presented to the network. After which, the model was able to recall direct associations (e.g., A->B, B->C) learned during the waking state. However, the indirect relational association (e.g., A->C) was not learned or was unreliable. After a period of SWS, the model's ability to recall these indirect associations was significantly improved, highlighting the importance of SWS in relational memory. In agreement with empirical data, we found that the duration of SWS significant correlates with the improvement in relational memory after sleep. Importantly, we found that replay during sleep increases synaptic connections between neurons representing the linking (common) item (B) and neurons representing the unlinked associations (A, or C). This change in synaptic connectivity led to a greater ability to recall the unlinked items when its

indirect pair was presented. Our study predicts that sleep can reactivate pathways to and from the linking item to the unlinked objects to form indirect associations.

Disclosures: T. Tadros: None. **M.V. Bazhenov:** None.

Poster

P819. Cortical Networks I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P819.05

Topic: H.08. Learning and Memory

Title: Predicting real-world mathematical proficiency and learning from brain activity

Authors: *Q. LIN[1], N. SHAIKH[1], N. B. TURK-BROWNE[1,2];
[1]Psychology, [2]Wu Tsai Inst., Yale Univ., New Haven, CT

Abstract: Mathematics are an integral part of all levels of education and useful in many aspects of everyday life, yet mathematical abilities vary dramatically across individuals. Mathematicians can reason through a sea of complex equations while some struggle with simple arithmetic like calculating the tip at a restaurant. What neural mechanisms explain such individual differences in math proficiency and learning? Here we test the hypothesis that how the brain responds to math instructional videos will reveal an individual's existing understanding of advanced concepts and their readiness to learn new concepts. We recruited 15 participants with a range of expertise, from novices with little college-level math exposure (e.g., first-year students not intending to major in STEM) to experts with extensive quantitative training (e.g., PhDs in a quantitative STEM field). After completing a written baseline exam on various topics in college-level math (linear algebra, calculus, probability) on Day 1, participants watched Khan Academy videos about these topics while undergoing fMRI on Day 2. On Day 3, participants completed another exam on these topics with a new set of questions. By projecting fMRI activity evoked during the videos into a common space using shared response modeling, an algorithm for functional alignment, we were able to predict baseline test performance on Day 1 in a cross-validated manner. Shared responses in default mode network regions such as the angular gyrus and precuneus carried the most predictive power. In addition to predicting baseline math proficiency, fMRI activity during the videos enabled us to predict who would improve the most after watching the videos in terms of test performance on Day 3. After identifying template voxel activity patterns linked to baseline math proficiency, we showed that the emergence of these patterns during learning predicted the magnitude of behavioral improvement. Taken together, our study demonstrates how combining advanced fMRI analysis methods with naturalistic stimuli can uncover the neural architecture supporting the acquisition of abstract concepts. This approach holds promise for identifying neural templates that could be targeted for cognitive

training of mathematical expertise through real-time neurofeedback. More generally, this study adds to a growing literature that grounds education and pedagogy in the brain.

Disclosures: Q. Lin: None. **N. Shaikh:** None. **N.B. Turk-Browne:** None.

Poster

P819. Cortical Networks I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P819.06

Topic: H.08. Learning and Memory

Support: Kavli pilot grant from the Rockefeller University
National Institutes of Health R00MH109652
Searle Foundation
Mathers Foundation

Title: Targeted Prefrontal Projections Drive Feature Recognition and Memory Recall

Authors: *N. YADAV[1,2], C. NOBLE[2], J. NIEMEYER[2], A. TERCEROS[2], J. D. VICTOR[1], C. M. LISTON[1], P. RAJASETHUPATHY[2];
[1]Feil Family Brain and Mind Res. Inst., Weill Cornell Med. Col., New York, NY; [2]Lab. of Neural Dynamics & Cognition, The Rockefeller Univ., New York, NY

Abstract: The neural basis underlying the relationship between a contextual memory and its constituent features is not well understood; in particular, where features are represented in the brain and how they drive memory recall. To gain insight into this question, we developed a virtual reality based behavioral task where mice learned to associate multisensory contexts with rewarding or aversive outcomes. During retrieval sessions, they were presented with partial features of the original multi-modal context and challenged to recall the associated contextual memory. We performed longitudinal imaging in hippocampus as mice performed this task and identified robust representations of global context but not of individual features (9% conjunctive vs 1.1% feature-selective neurons, n=3 mice, 9 sessions). By studying cortical inputs to hippocampus, we found that inhibition of anterior cingulate cortex (AC) led to silencing of a substantial proportion of context-selective neurons in CA1 and to behavioral deficit in feature-based memory recall. Single neurons in AC displayed feature-selectivity as well as mixed-selectivity to combinations of features, in contrast to the strong conjunctive tuning that was observed with CA1 neurons. These multiplexed single neuron activity gave rise to robust population level representations of features. Interestingly, we observed that AC feature representations lag hippocampus context representations during training, but dynamically reorganize to lead and target recruitment of context ensembles in hippocampus during recall (CDF as a function of latency, KS test, $p<0.001$). This AC-CA1 interaction is further enhanced

by the saliency of the memory, as suggested by tight coupling of feature-context ensembles in reinforced compared to non-reinforced contexts (ACfeature-CA1context correlations, $c_{Aversive}$ =0.43, $c_{Neutral}$=0.23, $p<0.05$). Together, we provide neurophysiological insights into how feature representations access long-range episodic representations to drive memory recall. Ongoing work is aimed at understanding how feature representations are selected and stabilized during learning.

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Poster

P819. Cortical Networks I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P819.07

Topic: H.08. Learning and Memory

Support: NINDS Grant R21NS095094

Title: Dissociation between left inferior frontal cortical thickness and encoding-related fMRI activity in temporal lobe epilepsy

Authors: *S. S. KAFAFI[1], P. F. HILL[1], B. C. LEGA[2], M. L. RACENSTEIN[1], D. R. KING[1], M. D. RUGG[1];
[1]Ctr. for Vital Longevity and Dept. of Brain and Behavioral Sci., Univ. of Texas at Dallas, Dallas, TX; [2]Neurosurg., UT Southwestern Med. Ctr., Dallas, TX

Abstract: Cross-sectional and longitudinal studies have established that patients with temporal lobe epilepsy (TLE) show accelerated cortical thinning in comparison to healthy adults. However, the impact of these structural differences on memory function in TLE patients is under-explored. The purpose of the present study was to examine whether global or regional cortical thickness moderates the neural correlates of successful memory encoding in TLE. Male and female TLE patients (N=16) and age-matched healthy controls (N=20) were scanned with fMRI while they studied a series of word lists. Following the presentation of each list and a brief distractor task, participants attempted to recall as many of the study items as possible. The scanning session was followed by a post-scan test of recognition memory that employed a mixture of studied and novel words. Thus, we were able to identify fMRI 'subsequent memory effects' (greater encoding-related activity for later remembered than later forgotten items) based on two different memory tests. There were no significant differences in recall performance between the patients and controls, although recognition memory performance was significantly lower in the patients. Highly robust subsequent memory effects predictive of later free recall and recognition were evident in the left inferior frontal gyrus, among other regions. These effects did

not significantly differ in magnitude between the two groups. For each participant, we extracted estimates of global and left IFG cortical thickness, and left and right hippocampal volume, from their T1-weighted MRIs. Both of the thickness metrics were significantly lower in the TLE patients, while there were no group differences in hippocampal volume. Across participants, neither estimate was reliably associated with recall or recognition memory performance. The findings suggest that cortical thinning in TLE has little or no impact on memory performance or the neural correlates of successful memory encoding, at least as these correlates are manifest in the LIFG.

Disclosures: S.S. Kafafi: None. **P.F. Hill:** None. **B.C. Lega:** None. **M.L. Racenstein:** None. **D.R. King:** None. **M.D. Rugg:** None.

Poster

P819. Cortical Networks I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P819.08

Topic: H.08. Learning and Memory

Support: SFARI (Simons Foundation Autism Research Initiative)
EASTBIO DTP Studentship, BBSRC, UKRI

Title: Increased prefrontal cortex gamma power in a rat model of Fragile X Syndrome

Authors: *J. W. MOORE[1], L. KOSTRZEWA[1], A. ASIMINAS[1,2], D. E. ARKELL[1,2], P. A. DUDCHENKO[3], P. C. KIND[1,2], E. R. WOOD[1,2];
[1]Ctr. for Discovery Brain Sci., Univ. of Edinburgh, Edinburgh, United Kingdom; [2]Simons Initiative for the Developing Brain, Edinburgh, United Kingdom; [3]Div. of Psychology, Univ. of Stirling, Stirling, United Kingdom

Abstract: Fragile X Syndrome (FXS) is one of the most common inherited forms of intellectual disability and autism spectrum disorder. Mouse and rat models of FXS have been produced through knockout (KO) of the *Fmr1* gene, and numerous learning and memory deficits have been observed in these models. Previous data from our lab suggest that the *Fmr1*$^{-/y}$ rats fail to show memory for object-place-context associations in a spontaneous object exploration recognition memory task, which is reliant on intact connections between medial prefrontal cortex (mPFC) and hippocampus (Till et al. 2015, Hum Mol Genet, Asiminas et al. 2019, Sci Trans Med). More generally, interactions between these areas are important for adaptation to novelty and cognitive flexibility, which have been shown to be affected in FXS patients (Schmitt et al. 2019, Brain Sci). The current study aimed to investigate whether differences in network activity within, or functional connectivity between, mPFC and hippocampus could underlie such deficits. Tetrodes were implanted into the mPFC and hippocampus of adult male *Fmr1*$^{-/y}$ and littermate

wild-type (WT) rats, to record single units and local field potentials (LFP) from both areas simultaneously, while the animals freely explore familiar and novel environments and object configurations, including an object-in-place task that is dependent on mPFC-hippocampus connections. Our analysis of LFPs indicates that power of gamma (30-80 Hz) oscillations in the mPFC was significantly increased in KOs compared to WTs, in both familiar and novel environments, while mPFC theta power, and power of oscillations in the hippocampus, were no different between genotypes. Further analyses of interaction mechanisms, such as coherence, phase-amplitude coupling, and spike-phase locking are currently ongoing. Elevated cortical gamma power has been observed previously using surface EEG in the mouse *Fmr1*$^{-/y}$ model (Lovelace et al. 2018, Neurobiol Dis), and in FXS patients (Wang et al. 2017, J Neurodev Disord). Our translation of this finding to *Fmr1*$^{-/y}$ rats indicates increased cortical gamma power as a robust preclinical biomarker, and suggests mPFC network dysregulation which could underlie cognitive inflexibility.

Disclosures: J.W. Moore: None. **L. Kostrzewa:** None. **A. Asiminas:** None. **D.E. Arkell:** None. **P.A. Dudchenko:** None. **P.C. Kind:** None. **E.R. Wood:** None.

Poster

P819. Cortical Networks I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P819.09

Topic: H.08. Learning and Memory

Support: National Institute of Mental Health, NIH, Division of Intramural Research (ZIAMH002920)
National Institute of Neurological Disorders and Stroke, NIH, Division of Intramural Research

Title: Distinct deficits of short-term versus extremely long-term repetition priming following lateral versus anteromedial frontal cortex damage

Authors: S. C. MILLEVILLE[1], ***S. J. GOTTS**[1], J. H. WITTIG, JR[2], S. K. INATI[3], K. A. ZAGHLOUL[2], A. MARTIN[1];
[1]Lab. of Brain and Cognition, NIMH/NIH, Bethesda, MD; [2]Surgical Neurol. Br., [3]Office of the Clin. Director, Natl. Inst. of Neurolog. Disorders and Stroke, NIH, Bethesda, MD

Abstract: Object repetition commonly leads to long-lasting improvements in identification speed and accuracy, a behavioral facilitation referred to as “repetition priming”. Neuroimaging and transcranial magnetic stimulation (TMS) studies have most often implicated the involvement of left lateral frontal cortex in repetition priming, although convergent evidence from neuropsychological studies is lacking. In the current study, we examine the impact of surgical

resection for the treatment of epilepsy on the magnitude of repetition priming at relatively short-term (30-60 minute delay) and extremely long-term (3 months) delays in 41 patients with varying seizure foci and resection locations (17 left anterior temporal, 10 right anterior temporal, 5 frontal, and 9 other). Overall, patients exhibited significant repetition priming at both short-term and long-term delays. However, patients with frontal resections (largely anterior and medial frontal) differed significantly from those with right anterior temporal resections in showing fully intact short-term priming but absent long-term priming. Supplementing this sample with a set of 10 recovered aphasic patients (having previously suffered a left hemisphere stroke), we further compared priming magnitudes in patients with damage that included versus spared left lateral frontal cortex. Priming following left lateral frontal damage was indeed significantly reduced at the short-term delay (30-60 minutes) for these patients, consistent with the results of previous neuroimaging and TMS studies. Taken together, the results suggest the differential involvement of lateral and anteromedial frontal regions in mediating repetition priming at short-lag and long-lag timescales, respectively.

Disclosures: S.C. Milleville: None. **S.J. Gotts:** None. **J.H. Wittig:** None. **S.K. Inati:** None. **K.A. Zaghloul:** None. **A. Martin:** None.

Poster

P967. Computational and Analysis Tools VI

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P967.01

Topic: I.06. Computation, Modeling, and Simulation

Support: NSF IOS-155925
NSF DBI 2014217

Title: Topological inference of stimulus space reveals low-dimensional geometry in fly and mouse olfaction.

Authors: M.-C. WU[1], ***Y. BERCHENKO-KOGAN**[1], M. WACHOWIAK[2], V. ITSKOV[1]; [1]Mathematics, Pennsylvania State Univ., University Park, PA; [2]Neurobio., Univ. of Utah, Salt Lake City, UT

Abstract: Sensory coding in olfaction remains poorly understood. Unlike vision, where the stimulus and receptor code can be simply defined, the complexity of the “odor space” as well as the complexity of the odorant receptor (OR) repertoire has hindered our understanding of odor coding and processing. For example, there is no consensus on key defining features of odor sensation such as the dimensionality of odor perceptual or odor receptor space. It is commonly conjectured that the ORs transform a high-dimensional “chemical space” to a relatively low-dimensional space of neuronal representations.

We developed a novel mathematical method for the inference of basic geometric features of a stimulus space from neural activity. This method makes no substantial assumptions on the underlying stimulus space, instead it assumes that the receptive fields are *convex* in some natural coordinate system in the underlying olfactory space. The convexity of sensory receptive fields is well-documented in many other sensory modalities, such as vision and spatial navigation. The key feature of this method is that it is insensitive to the inherent (and usually unknown) monotone non-linearities of neuronal responses.
It turns out that it is possible to infer basic geometric features of the underlying stimulus space, such as the *non-linear* dimension, based on a noisy and sparse data that is available for the activity of olfactory receptors in flies and mice. Our method converts the activity of each OR into a combinatorial summary that records relative rankings of neuronal responses to odorants per each OR. This data preserves basic information about the geometry of the underlying stimulus space, such as the dimension. We then use topological methods to infer the *non-linear* dimension of the underlying space.
We used our methods to estimate basic geometric features of OR representations in two distant species: Drosophila and mice. We compared the geometric features of neural representations in Drosophila antennal odorant receptors with those in the mouse olfactory bulb, using previously published datasets as well as newly collected data from the mouse. We found that the non-linear dimension of the olfactory coding space is low in both species. Our results indicate that the dimensionality on per-receptor basis is significantly different between these species, as the fly has substantially smaller olfactory receptor repertoire, than the mouse. Our findings establish that the non-linear transformation performed by the olfactory receptors compresses the high-dimensional chemical space into a low-dimensional olfactory space, at the level of the olfactory receptor neurons.

Disclosures: M. Wu: None. **Y. Berchenko-Kogan:** None. **M. Wachowiak:** None. **V. Itskov:** None.

Poster

P967. Computational and Analysis Tools VI

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P967.02

Topic: I.06. Computation, Modeling, and Simulation

Support: NIH R01 NS120581
NSF DMS-1951165

Title: Topological data analysis of spontaneous activity in the zebrafish optic tectum

Authors: *J. PAIK[1], E. HANSEN[2], G. SUMBRE[2], C. P. CURTO[1];
[1]Mathematics, The Pennsylvania State Univ., University Park, PA; [2]Ecole Normale Superieure, Paris, France

Abstract: Techniques using topological data analysis can help us find structure in correlation matrices that traditional linear algebra techniques cannot. One cause of this problem is that notions like rank and spectra are not numerically robust to nonlinearities upon entries. An attempt to address this problem are through Betti curves, introduced in [1], which are matrix statistics that are invariant to monotone nonlinearities upon entries. In this study, we use Betti curves (β_k) to analyze spontaneous activity from the zebrafish optic tectum, data collected using two-photon light sheet microscopy. The calcium imaging signal from ~4,000 neurons are clustered into functional assemblies and a correlation matrix is computed. From there, we calculate Betti curves on submatrices induced by assemblies using Ripser. What we find are identically zero β_k for $k > 0$, which are clearly different to the Betti curves for a random assembly, which have nontrivial β_k. In [2], the authors showed that an all-positive symmetric matrix with an underlying rank of one has identically zero Betti curves β_k for $k > 0$. Hence the correlation matrices are consistent with that of a rank one matrix. We note that in contrast to Betti curves, the singular values of the correlation matrix induced by both the assembly and the random assembly are consistent with that of a full rank matrix. We propose that this low rank structure is a signature of the attractor-like dynamics observed in the zebrafish optic tectum.

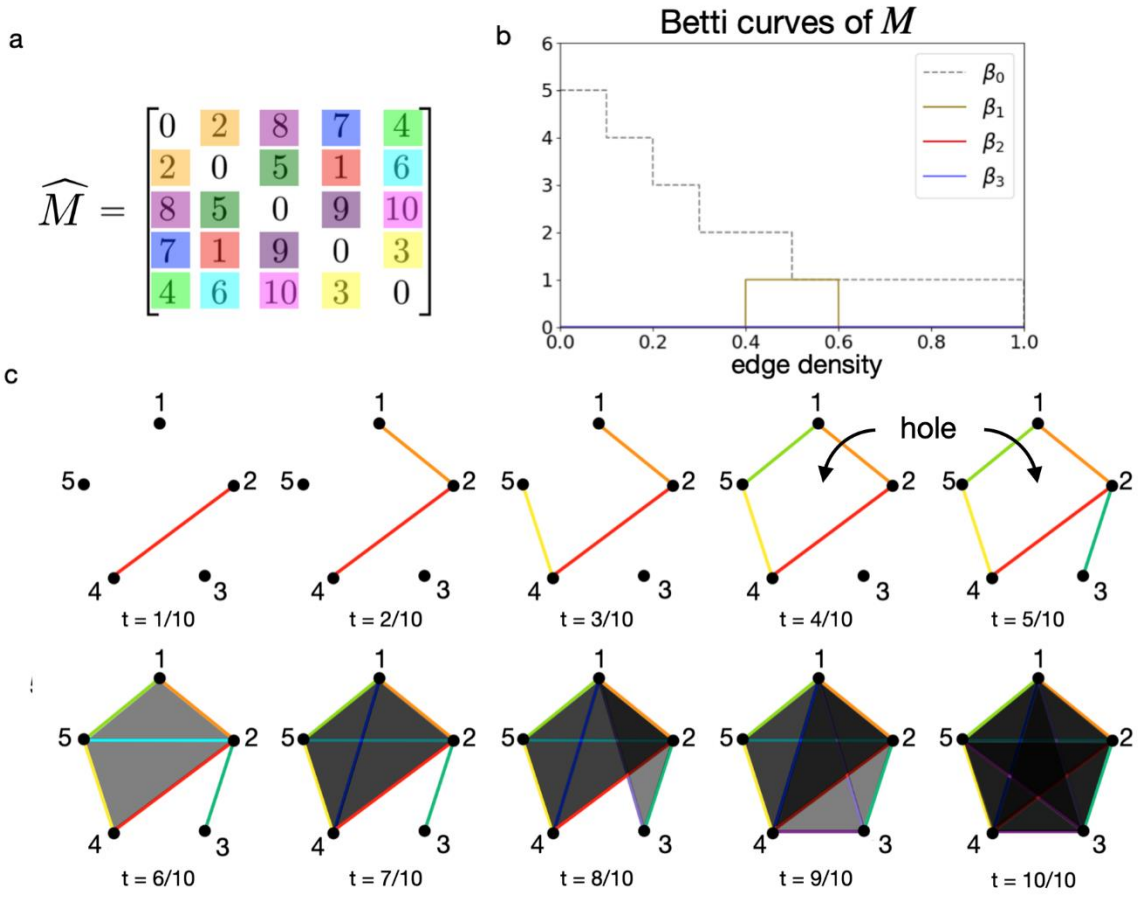


Fig. 1. In (a), we see the ordering matrix of a rank four matrix. (b) The Betti curves induced by which are the same for any matrix with the same relative ordering of entries as . (c) We depict the construction of Betti curves, by adding edges one by one relative to their weight in

References

1.Giusti et al. Clique topology reveals intrinsic geometric structure in neural correlations. PNAS 112(44), 13455-13460, 2015.
2.Curto et al. Betti curves of rank one matrices. Geometric Science of Information, 2021. (in press)

Disclosures: J. Paik: None. **E. Hansen:** None. **G. Sumbre:** None. **C.P. Curto:** None.

Poster

P967. Computational and Analysis Tools VI

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P967.03

Topic: I.06. Computation, Modeling, and Simulation

Support: NIH SPARC Program Grant 1OT3OD025349

Title: Sparc: enhanced microscopy image analysis tools for the production of fair neuromorphological data

Authors: M. L. HEAL, A. E. SULLIVAN, C. R. BATES, M. GULENKO, *S. TAPPAN, P. J. ANGSTMAN, J. R. GLASER;
MBF Biosci. - Microbrightfield LLC., Williston, VT

Abstract: Microscopy image analysis techniques have been constantly evolving with the onset of new microscope devices, advanced histological preparations, and cutting-edge computational technologies. Though the output of such image analysis software often allows for robust qualitative and quantitative analysis, the data files are frequently absent of file-level metadata about the subject and anatomical region from which the data was derived. Moreover, the use of cross-species anatomical ontologies (e.g., UBERON, FMA) have seldom been used in neuroanatomical reconstruction pipelines due to a lack of software tools that serve anatomical terms from these ontologies directly to the user at the time of neuron reconstruction. As public-access data portals (e.g., sparc.science), open science, and collaborative academic ecosystems become more common, the need for this rich machine-readable metadata at the file level becomes more crucial. To help prepare researchers for this shift in paradigm, we have developed tools for findable, accessible, interoperable, and reusable (FAIR) reconstruction of multi-species, multi-organ, and multi-scale neuroanatomical regions. Unlike any other software, this toolset supports the use of anatomical ontologies at the time of data creation through a direct API

connection to the SciCrunch knowledge infrastructure for accessibility of current, FAIR, and organ-specific terminology lists curated by anatomical experts. It is also equipped with verification methods to ensure all annotations have globally unique and persistent identifiers. The output file format produced by this toolset has a comprehensive and publicly available specification that richly characterizes all elements of the neuromorphological data. The amalgamation of these novel tools will facilitate scientific discovery by producing meaningful and sustainable data that can be searched for, shared, and repurposed.

Disclosures: **M.L. Heal:** A. Employment/Salary (full or part-time):; MBF Bioscience. **A.E. Sullivan:** A. Employment/Salary (full or part-time):; MBF Bioscience. **C.R. Bates:** A. Employment/Salary (full or part-time):; MBF Bioscience. **M. Gulenko:** A. Employment/Salary (full or part-time):; MBF Bioscience. **S. Tappan:** A. Employment/Salary (full or part-time):; MBF Bioscience. **P.J. Angstman:** A. Employment/Salary (full or part-time):; MBF Bioscience. **J.R. Glaser:** A. Employment/Salary (full or part-time):; MBF Bioscience.

Poster

P967. Computational and Analysis Tools VI

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P967.04

Topic: I.06. Computation, Modeling, and Simulation

Support: NIH Grant 1OT3OD025348-01
NIH Grant 1OT3OD025347-01
NIH Grant 1OT3OD025349-01
NIH Grant 1OT2OD030541-01
SPARC Program

Title: Collaborative, sustainable, and FAIR neurosciences in bioelectronic medicine: the SPARC Data Resource Center

Authors: *E. NEUFELD[1], J. B. WAGENAAR[2], P. HUNTER[3], M. E. MARTONE[4], B. DE BONO[5], S. TAPPAN[6], N. KUSTER[7];
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Abstract: Stimulating the autonomic nervous system (ANS) to modulate organ physiological function holds enormous therapeutic promise, but the underlying mechanisms are poorly understood. The NIH Stimulating Peripheral Activity to Relieve Conditions (SPARC) program

aims to improve targeted therapies by providing public access to high value datasets, maps, tools, and predictive simulations to support the development of new, safe, and effective neuromodulation devices. Over 100 SPARC teams world-wide work on ANS mapping, manipulation/observation tools, and therapeutic translation. To assure an integrative approach and facilitate collaborative open science, SPARC has established the Data Resource Center (DRC). It i) stores, organizes, and disseminates data generated by the SPARC program, ii) establishes standards (e.g., on minimal metadata and formats) and provides curation and knowledge management services, iii) generates interactive visualizations of nerve-organ anatomy and function, and iv) offers a modelling, simulation, and data analysis platform. The SPARC Portal (https://sparc.science/) hosts data (currently > 30Tb) and computational studies, which can be found through knowledge-base and map-driven search, viewed, downloaded, or executed. The SPARC Portal provides unified access to the SPARC platform comprising: 1) Pennsieve, a cloud-based data management platform to manage, curate, and publish large scientific datasets,offers dynamic, interactive, and modular visualizations of nerve, organ and body anatomy, and function across a range of species; 2) SciCrunch, a dynamic vocabulary management system used to develop community-driven ontologies and organize curation metadata into graphs to power smart queries; 3) technology from MBF Bioscience that supports image segmentation, co-registration, annotation, and visualization; 4) the ApiNATOMY framework to define relationships between anatomy components and the nervous system; and 5) Flatmap/Scaffold views that provide multi-scale visualization of relationships and data; 6) o^2S^2PARC, an online, cloud-based, open, extensible platform for computational (neuro-)sciences, serves to predict the effects of ANS neuromodulation on organ function. Computational models and data analysis pipelines can be hosted, shared, executed, documented, and coupled. These infrastructure and approaches ensure that datasets are scientifically interpretable, reproducible and FAIR (Findable, Accessible, Interoperable and Reusable). The integration of these resources in the SPARC platform will accelerate progress in the field of bioelectronics medicine, the wider life-sciences and beyond.

Disclosures: E. Neufeld: None. **J.B. Wagenaar:** None. **P. Hunter:** None. **M.E. Martone:** None. **B. de Bono:** None. **S. Tappan:** None. **N. Kuster:** None.

Poster

P967. Computational and Analysis Tools VI

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P967.05

Topic: I.06. Computation, Modeling, and Simulation

Title: The Allen Cell and Structure Segmenter: An open source toolkit and Napari plugin for segmenting 3D intracellular structures in microscopy images

Authors: *K. A. GERBIN, J. CHEN, .. ALLEN INSTITUTE FOR CELL SCIENCE TEAM, S. M. RAFELSKI;
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Abstract: The Allen Institute for Cell Science uses high-replicate live cell imaging to understand the principles by which cells reorganize in health and disease. We aim to predict cell behavior by building a state space of structural signatures in human induced pluripotent stem cells (hiPSCs) to differentiated cell types, while building open source image analysis tools and reproducible datasets.
Towards this goal, we generated a dynamic imaging pipeline using endogenous fluorescently-tagged hiPSC lines. Each line expresses a monoallelic EGFP-tagged protein that represents a major cellular structure or organelle, including neuron-relevant structures such as the nuclear envelope (Lamin B1), nucleolus (Fibrillarin and Nucleophosmin), mitochondria (Tom20), endoplasmic reticulum (Sec61 beta), peroxisomes (PMP34), cell membrane (CAAX), actin filaments (Beta-actin), microtubules (alpha-tubulin), and more. Thousands of high-resolution 3D images are acquired for each structure and used to develop quantitative image-based assays and computational models. However, accessible cell and structure segmentation tools remain a challenge in the field, limiting the extraction of meaningful information from imaging data.
With this in mind, we developed the Allen Cell and Structure Segmenter, a Python-based open source toolkit and Napari plugin for 3D segmentation of intracellular structures. This toolkit combines classic and iterative deep learning workflows to streamline the identification of successful 3D segmentation algorithms for a wide range of intracellular structures. To make this tool broadly applicable across diverse cell types and structures, we 1) formulated a classic image segmentation workflow based on a minimal number of selectable algorithms and tunable parameters, 2) successfully applied this workflow to over 30 intracellular structures; whereby the workflows can be selected via a lookup table, 3) developed this workflow and lookup table into a user-friendly plugin for the open source image viewer Napari, and 4) implemented an iterative deep learning workflow using the results of these classic segmentation algorithms with minor manual inspection to generate a ground truth for training deep learning models.
This open source segmentation toolkit facilitates quantitative single-cell cell biology in cell types from pluripotent stem cells to neurons, leveraging state-of-the-art algorithms in computer vision for specific image segmentation needs and challenges. The Allen Cell and Structure Segmenter, as well as our suite of open source tools, methods, data, and other resources are available free to the public on our website allencell.org.

Disclosures: K.A. Gerbin: None. **J. Chen:** None. **.. Allen Institute for Cell Science Team:** None. **S.M. Rafelski:** None.

Poster

P967. Computational and Analysis Tools VI

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P967.06

Topic: I.06. Computation, Modeling, and Simulation

Support: Conacyt A1-S-36984
Conacyt 704048
Conacyt 287002 CB-2016-01
SIP20211217

Title: In silico analysis on the stability of calpain-10

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Abstract: Calpain-10 is a non-lysosomal calcium-sensitive cysteine protease that has been proposed to play an important role in the development of diabetes mellitus and neuropathic pain as some of its potential targets are voltage-gated sodium channels (Nav). Calpains hydrolyze their substrate by separating domains from target proteins and, consequently, activate or inhibit their targets. Although multiple proteins have been identified as targets for calpains, how they recognize and interact with their targets and how they recognize the hydrolysis site remains a mystery. Here we use a bioinformatic approach to obtain a full-length molecular model of Calpain-10. The 3D structure of mouse Calpain-10 (666 aa) was constructed to visualize different conformations adopted over to 250 ns time lapse, by molecular dynamics (MD), in a simulated environment at 310 and 400 K. Calpain-10 is gradually denatured, especially the α-helix in domain I that loses its conformation throughout DM. The unfolding of the α-helix in domain I may be relevant to regulate the activation of Calpain-10 since, in contrast to calpains 1 and 2, domain I remains bound to Calpain-10 after activation. Domain III also exhibit conformational changes that could be involved in calcium binding for subsequent protease activation. This description helps us understand the functionality and stability of Calpain-10 and better understand its activation mechanisms and will help us develop more models of interactions with target proteins.

Disclosures: L.M. Arratia-Cortés: None. **A.V. Vega:** None. **A. Zamorano-Carrillo:** None.

Poster

P967. Computational and Analysis Tools VI

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Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P967.07

Topic: I.06. Computation, Modeling, and Simulation

Title: Generative adversarial networks for parameter inference in populations of neuroscience models

Authors: *T. RUMBELL[1], S. SAGHAFI[2], C. O. DIEKMAN[2], J. KOZLOSKI[1], V. GUREV[1]; [1]IBM - TJ Watson Res. Ctr., Yorktown Heights, NY; [2]Dept. of Mathematical Sci., New Jersey Inst. of Technol., Newark, NJ

Abstract: In computational neuroscience, parametric families of mechanistic models are often employed to simulate neural activity from a set of recordings. In population of models (PoM) scenarios (a.k.a. stochastic inverse problems (SIP)), the parametric family is fit to a population of observed data from the model output domain, transferring a dataset of real-world observations collected across a population of individuals into the latent space of mechanistic parameters. Neural systems exhibit striking heterogeneity within populations at all scales, while at the same time multiple model parameter sets can result in equivalent activity, rendering the models noninvertible. Furthermore, these models typically have an intractable likelihood, making parameter inference challenging and inaccessible to traditional techniques. Recent advances in deep learning have enabled development of new approaches to related problems. Deep learning networks for amortized inference in likelihood-free settings have been applied in several neuroscience modeling problems [1]. However, deep learning methods have not been applied in SIP to construct parametric families of models. We present a generative adversarial network (GAN) approach to solve SIP formulated as a constrained optimization problem. We developed these methods to be generically applicable to mechanistic model parameter inference across domains, and designed them with flexible configuration that can incorporate prior information directly into the network architecture [2]. Here, we apply our GAN methods for parameter inference in neuroscience modeling problems, using models at several scales for demonstration. First, we used the Morris-Lecar model of neuronal spiking and inferred diverse populations of parameters that exhibit either Hopf or saddle-node on invariant cycle bifurcations. Second, using a Hodgkin-Huxley (HH) model [3], we inferred a population of ionic conductance parameters that are coherent with a distribution of measured rebound spike delays after hyperpolarization. Third, using a large network of HH models, we inferred distributions for global excitation and inhibition parameters that result in activity across the network matching network spiking statistics. Across these examples, we demonstrate several advanced parameter inference methods facilitated by our GAN framework, such as amortized SIP using a conditional GAN, and our constrained optimization formulation of SIP using either a neural network surrogate model to recreate the mechanistic model function, or a differentiable implementation of the mechanistic model ODEs directly incorporated into the inference architecture.

Disclosures: **T. Rumbell:** A. Employment/Salary (full or part-time):; IBM Research. **S. Saghafi:** None. **C.O. Diekman:** None. **J. Kozloski:** A. Employment/Salary (full or part-time):; IBM Research. **V. Gurev:** A. Employment/Salary (full or part-time):; IBM Research.

Poster

P977. Electrical Methods to Modulate Neural Activity I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P977.01

Topic: I.08. Methods to Modulate Neural Activity

Support: Marie Slodowska-Curie Grant 713673
"la Caixa" Foundation (ID 100010434), LCF/BQ/IN18/11660012
Ministerio de Economía, Innovación y Competitividad (Spain) Grant #RTI2018-099282-B-I00B

Title: Auricular transcutaneous vagus nerve stimulation acutely modulates memory persistence and brain connectivity in mice

Authors: C. BRAMBILLA PISONI[1], E. MUÑOZ MORENO[3], I. GALLEGO AMARO[1], R. MALDONADO[1,4], A. IVORRA[2], G. SORIA[3,5], *A. OZAITA[1,4];
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Abstract: Electrical brain stimulation techniques take advantage of the intrinsic plasticity of the nervous system and may have a wide range of therapeutic applications. In this context, vagus nerve stimulation (VNS) is an approved treatment for refractory epilepsy and drug resistant depression. Among the different modalities of VNS, a non-invasive form of VNS, auricular transcutaneous VNS (atVNS), is under investigation for different applications, including cognitive improvement. Brain regions relevant for memory performance are sensitive to vagal nerve afferents, although the impact of atVNS in brain activity has not been fully elucidated. We aimed to study the efficacy of atVNS in modulating memory persistence and brain connectivity in CD-1 male mice. Using a previously described atVNS device (Brain Stimulation 13:494, 2020), we targeted the left *cymba con*chae. We performed acute atVNS with a standard stimulation protocol under light isoflurane anesthesia. We found that novel object-recognition memory persistence was enhanced when atVNS was delivered just after, but not 3 h after, the familiarization phase. We obtained brain samples for bilateral c-Fos analysis, performing the electrostimulation protocol immediately after the familiarization phase, in various brain areas of the brainstem, hippocampus, amygdala, frontal cortex, thalamus and cortical regions of the default mode network (DMN). We first observed that atVNS did not modify overall c-Fos density in any of the brain regions analyzed. Then, correlation analysis was performed between c-Fos densities in each pair of regions to obtain matrices of significant relations in activity under non-stimulated and electrostimulated conditions. Such analysis revealed that atVNS coupled the activity of left and right *locus coeruleus*, increasing also its connection with the dentate gyrus (DG). Furthermore, the electrostimulation reinforced the relation in activity of all subregions of the hippocampus (DG, CA1 and CA3), and revealed a marked enhancement in the correlated activity between hemispheres both in prelimbic and infralimbic cortex, although it did not have marked effects on the prefrontal-retrospenial axis of the DMN. Moreover, evaluation of global brain network metrics showed a marked increase in integrity, segregation and strength properties

in the stimulated brain. Indeed, overall connectivity was found significantly increased in atVNS-exposed brains. In conclusion, our results provide new insights in the effects of atVNS on memory performance and brain connectivity, and open the possibility to extend our knowledge of the biological mechanisms of bioelectronics in medicine.

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Poster

P977. Electrical Methods to Modulate Neural Activity I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P977.02

Topic: I.08. Methods to Modulate Neural Activity

Support: UK Medical Research Council - MRC grant No: MR/R01213X/1
NIH SPARC Program (1OT2OD026545-01)

Title: Fast neural EIT for imaging of the spontaneous neural traffic of the cervical vagus nerve and its validation by spatially selective neuromodulation and MicroCT (SPARC).

Authors: *E. RAVAGLI[1], S. MASTITSKAYA[1], N. THOMPSON[1], J. PERKINS[2], A. GOURINE[1], K. ARISTOVICH[1], D. HOLDER[1];
[1]Univ. Col. London, London, United Kingdom; [2]Royal Vet. Col., London, United Kingdom

Abstract: Introduction: Fast neural EIT is a novel imaging tool which allows imaging of impedance changes caused by neuronal depolarisation. It was validated in rat sciatic nerve [1] but never used for autonomic nerves. The purpose of this work is to perform EIT imaging of spontaneous and evoked neuronal activity in cervical vagus nerve to assess the presence of fascicles with organ-specific functions. We validate the images with spatially selective neuromodulation (SSN) [2] and demonstrate the possibility of future use of EIT and SSN for precise closed-loop neuromodulation.
Methods: In N=4 pigs, multi-electrode cuff arrays for SSN (2 rings, 14 electrodes each) and EIT (1 ring, 14 electrodes) were applied on the left cervical vagus nerve. End-Tidal CO2, heart rate and EMG were recorded to monitor SSN-evoked changes in pulmonary, cardiac, and laryngeal activity, respectively. SSN was performed as in [2]. Biphasic pulses were applied at 50µs, 20Hz for recurrent laryngeal and pulmonary fascicles, and 1ms, 20Hz for cardiac fascicles. Amplitude required to evoke or modulate laryngeal, pulmonary and cardiac activity was 238±102µA, 355±105µA and 1038±634µA, respectively. EIT measurements (200µA, 6KHz) were performed over 14 pairs, 10min each. Spontaneous EIT was performed for pulmonary and cardiac activity, whereas laryngeal activity was evoked by recurrent laryngeal nerve stimulation at 50µs, 20Hz, 1.5mA. EIT data were subject to coherent averaging and image reconstruction. Vagus nerves

were dissected post-mortem for MicroCT tracing of organ-specific fascicles.
Results: EIT allowed reconstruction of three spatially separated images for laryngeal, pulmonary and cardiac fascicles. In all four animals, cross-sectional positions of laryngeal, pulmonary and cardiac SSN peaks followed the same order. For EIT, angular distances of pulmonary and cardiac activation peaks from laryngeal were 118±32° and 126±77°, respectively. For SSN, they were 125±36° and 158±57°, respectively.
Conclusions: EIT and SSN results suggest organotopic or function-specific fibre organisation in the cervical vagus nerve. EIT enables imaging of spontaneous activity within the cervical vagus nerve and evoked activity of large myelinated fibres. Together with SSN, this enables the possibility of closed-loop neuromodulation. Work in progress is analysis of MicroCT data to fully validate EIT, as well as translation of EIT and SSN to human applications.
References:
[1] Ravagli et al., 2020, Nat Commun. 11:6241[2] Aristovich et al., 2021, J Neurosci Methods. 352:109079

Disclosures: E. Ravagli: None. **S. Mastitskaya:** None. **N. Thompson:** None. **J. Perkins:** None. **A. Gourine:** None. **K. Aristovich:** None. **D. Holder:** None.

Poster

P977. Electrical Methods to Modulate Neural Activity I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P977.03

Topic: I.08. Methods to Modulate Neural Activity

Support: DoD SBIR SB173-001

Title: Protective anti-inflammatory reflex caused by electrical stimulation of the splenic neurovascular bundle

Authors: *C. KAISER, E. HAAG, I. SONDH, D. ZACHS, M. NEWHOFF COLLINS, H. LIM; Univ. of Minnesota, Minneapolis, MN

Abstract: Systemic inflammation is decreased by activation of the neuro-immune mechanism termed the Cholinergic Anti-inflammatory Pathway (CAP), providing a promising treatment for chronic and acute inflammatory conditions including rheumatoid arthritis, Crohn's disease, inflammatory bowel disease, sepsis, and acute kidney injury. In CAP, efferent vagus and splenic nerve activity leads to release of norepinephrine in the spleen, triggering a signaling cascade involving CD4 T-cells and macrophages that leads to inhibition of pro-inflammatory cytokine release. CAP activation by electrical stimulation of the cervical vagus nerve (VNS) in animal models decreased pro-inflammatory cytokines such as TNF-alpha, IL-1beta, and IL-6. Nearly all published research on CAP targets the vagus nerve for stimulation. However there remains

disagreement in the field about the direct anatomical pathway from the vagus nerve to the splenic nerve. To characterize the effects of CAP activation directly associated with the splenic nerve near the end-organ, our group designed a study using electrical stimulation of the splenic nerve (SNS) at its central branch proximal to innervation into the spleen, where it is embedded in a neurovascular bundle (NVB). Results demonstrated the effect of direct SNS in the absence of VNS. We assessed efficacy of SNS for treatment of acute inflammation in anesthetized rats injected with lipopolysaccharide endotoxin (LPS), a model of sepsis. LPS stimulates macrophages to heighten secretion of pro-inflammatory cytokines. Inflammation state was monitored for up to two hours post-LPS injection. Cytokine levels were assessed within circulating blood every 30 minutes and within the spleen hourly. Immunohistochemistry was performed on splenic nerve and whole spleen tissue to verify location of the splenic nerve within the neurovascular bundle using neurofilament and tyrosine hydroxylase antibodies and to assess tissue damage. Additionally, electrophysiology was performed to characterize nerve activation in response to SNS therapy. Central branch SNS provided a protective effect against LPS-induced inflammation. Cytokine levels for TNF-alpha and IL-1beta were decreased in animals that were treated with SNS compared to LPS positive controls, indicating lower systemic inflammation. Compound action potentials were successfully recorded along the splenic neurovascular bundle, providing near-organ neural activity verification of SNS therapy. No significant tissue damage was observed on the stimulated NVB compared to controls in histology. These results confirm the safety and efficacy of SNS as an alternative to VNS for activation of CAP.

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Poster

P977. Electrical Methods to Modulate Neural Activity I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P977.04

Topic: I.08. Methods to Modulate Neural Activity

Support: Medtronic Inc.

Title: A Potential Novel Treatment for Hypoglycemia Utilizing Sub-Diaphragmatic Vagus Nerve Stimulation: Demonstration of a Rapid Increase in Plasma Glucose in Rat and Swine Models of Diabetes with Vagal Stimulation

Authors: *J. J. WAATAJA[1], D. GAL[1], S. M. GOETZ[2], R. K. NIHALANI[1];
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Abstract: Over thirty (34.2) million Americans are diabetic w/ 1.5 million new cases per year. Hypoglycemia negatively affects diabetics. The average type 1 diabetic experiences two episodes

of symptomatic hypoglycemia/week. Severe hypoglycemia (need for a 3rd party assistance) has an annual incidence of 1.0 - 1.7 episodes per patient per year. Every year 73% of insulin dependent type 2 diabetics experience hypoglycemic episodes and 15% have severe episodes. Hypoglycemia can cause loss of consciousness, stroke, coma or death. Repeated hypoglycemic episodes have been linked to cardiovascular disease. Treatments typically involve consuming 15 grams of a fast-acting carbohydrate. However, this treatment is not ideal for nocturnal hypoglycemia & contraindicated for severe hypoglycemic episodes. Use of insulin pumps for insulin dependent diabetics is limited to ~1% of insulin dependent diabetic population. Insulin pump therapy in conjunction w/ continuous glucose monitor (CGM) technology decreases the risk of hypoglycemia, but still remains a meaningful problem in insulin dependent diabetics. Thus, there is opportunity for bio-electronic treatment modalities for management of hypoglycemia. The vagus nerve innervates organ systems that regulate plasma glucose (PG). It has been demonstrated that vagus nerve stimulation increases pancreatic endocrine hormone release and in some circumstances will induce an increase in PG; likely due to glucagon secretion. Here we tested different bio-electronic stimulation parameters delivered to the sub-diaphragmatic posterior vagus nerve (containing celiac fibers innervating the pancreas) in rat and pig diabetic model systems. First, 1 Hz stimulation demonstrated a significant 43±14 mg/dL rise in PG within 15 min in type 2 diabetic Zucker rats compared to sham. Next, we tested stimulation in an alloxan treated type 1 diabetic swine model following insulin induced hypoglycemia. Fasting PG was 446±30 mg/dL. IV injection of insulin caused a dramatic decrease in glucose to levels below 60 mg/dL. One to 2 IV injections of dextrose were required to induce a sustained increase in PG. In experimental conditions stimulation was applied following a 33% drop in PG. Responses were demonstrated with frequencies of 5 and 10 Hz; yielding a significant 38±13 mg/dL rise in glucose by 10 min. Following experiments histopathology was performed of the liver, pancreas, nerve and brain. All tissues were healthy. This methodology has the potential to work in concert with CGM technology to trigger stimulation on demand as a treatment for hypoglycemic episodes.

Disclosures: J.J. Waataja: A. Employment/Salary (full or part-time):; ReShape Lifesciences Inc. **D. Gal:** A. Employment/Salary (full or part-time):; ReShape Lifesciences Inc. **S.M. Goetz:** A. Employment/Salary (full or part-time):; Medtronic Inc. **R.K. Nihalani:** A. Employment/Salary (full or part-time):; ReShape Lifesciences Inc..

Poster

P977. Electrical Methods to Modulate Neural Activity I

Location: Virtual Only

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Program #/Poster #: P977.05

Topic: I.08. Methods to Modulate Neural Activity

Support: DARPA grant N66001-17-2-4018

Title: Effects of non-invasive trigeminal nerve stimulation on visuomotor adaptation

Authors: *D. E. ARIAS, C. A. BUNEO;
Arizona State Univ., Tempe, AZ

Abstract: Trigeminal nerve stimulation (TNS) is a novel neuromodulation technique where low-intensity electrical stimulation is delivered to branches of the trigeminal nerve to modulate brain activity. The rationale behind TNS is the activation of ascending trigeminal pathways to brain stem nuclei known to influence activity in cortical structures. TNS has been shown to improve the symptoms of major depression, drug-resistant epilepsy, migraine, and other disorders. Stimulation of the vagus nerve, which has similar therapeutic profiles and projections as the trigeminal nerve, has shown the ability to enhance plasticity in motor areas. However, it is unknown whether TNS can exert similar effects. For this reason, this study explored the effects of TNS on visuomotor adaptation. Fifty-six (56) right-handed healthy subjects (26 women, 30 men; 18-36 y/a), were randomly assigned to 3 groups based on the stimulation frequency used: 120 Hz (n=19), 60 Hz (n=19) and sham (n=18). Participants performed a visuomotor rotation task that involved center-out reaching movements to eight targets arranged in a vertical plane. Four blocks of trials were performed as follows: two baseline blocks with veridical visual feedback, one adaptation block involving a 30° CCW rotation of hand visual feedback, and one washout block where the rotation was removed. TNS was applied for 20 min before the 2nd baseline block using two electrodes placed on either side of the forehead, thereby targeting the ophthalmic branches of the trigeminal nerve. Each participant set the intensity to a sub-maximum tolerable level (mean currents; 120 Hz: 3.01 ± 0.76 mA, 60 Hz: 3.43 ± 0.83 mA). No current was applied to the sham group. To minimize variations in emotional states across subjects, during the stimulation phase participants viewed images of neutral valence and arousal obtained from the International Affective Picture System. Visuomotor performance was quantified by the directional error (DE), defined as the angular difference between the hand position and the virtual feedback at peak velocity. DE across the adaptation block was fitted to a double exponential model to capture the fast and slow learning processes that drive motor adaptation. Preliminary analyses showed that the 60 Hz group had the slowest adaptation among groups. In addition, learning rates were initially similar between the 120 Hz and sham groups, but gradually diverged, with the 120 Hz group exhibiting slightly faster rates than sham later in the adaptation block. The results suggest that TNS is associated with frequency-dependent effects on visuomotor adaptation, which provides new information on the potential use of TNS in motor applications.

Disclosures: D.E. Arias: None. **C.A. Buneo:** None.

Poster

P977. Electrical Methods to Modulate Neural Activity I

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Program #/Poster #: P977.06

Topic: I.08. Methods to Modulate Neural Activity

Support: DARPA Targetted Neuroplasticity Training (TNT)

Title: Using vagus nerve stimulation to shape neuromodulation of the neocortex

Authors: Z. H. MRIDHA[1,3], J. W. DE GEE[1,3], ***S. LEE**[2,3], S. ALEEM[1,3], E. NEYHART[1], J. REIMER[1], M. THOMPSON[1,3], K. JASPE[1,3], M. J. MCGINLEY[1,3,4];
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Abstract: It is now known that neuromodulators are responsible for dynamically adjusting the functional brain state to changes in the environment. Dysfunction of neuromodulator signaling is central to many neurological and psychiatric disorders, including Parkinson's disease, depression, epilepsy and addiction. We have recently reported that electrical stimulation of the mouse vagus nerve activates cholinergic axons in neocortex (Mridha et al. 2021). Acetylcholine is heavily involved in modulating neuronal excitability, plasticity, cortical state and cognitive tasks such as attention and memory. Vagus nerve stimulation (VNS) thus provides a promising avenue to treat a broad range of disorders and perhaps shape brain state.
Here, we leverage a combination of two-photon (2P) advanced imaging techniques and genetically encoded fluorescent sensors to characterize the dynamics of VNS-evoked changes in acetylcholine concentration with high spatiotemporal resolution. The sensors were developed by coupling neurochemical-sensing G protein-coupled receptors (GPCRs) with a circularly-permuted fluorescent protein (Jing et al. 2018). We injected the specific $GRAB_{ACH}$ sensors to the auditory cortex (A1) and recorded the VNS-evoked acetylcholine concentration as well as the pupil dilatation. Corroborating our previous cholinergic axon results, we here find that VNS parametrically increases cholinergic concentration in neocortex.
We also show that pupil dilation is a reliable and convenient biosensor for successful VNS. Pupil size is widely used as a non-invasive metric of brain state. Previous studies, including ours, have shown links between pupil size and the activity in major neuromodulatory systems including the basal forebrain acetylcholine system (Reimer et al., 2016; Mridha et al. 2021). Exploiting this relationship, we are working on closed-loop applications based on real-time pupil detection in combination with VNS. Applications range from "clamping" the brain's arousal state at the optimal level to perform a certain cognitive task, to guide VNS parameter selection for more targeted neuromodulation in a range of disorders.
References Mridha, Z et al.: Graded recruitment of pupil-linked neuromodulation by parametric stimulation of the vagus nerve. *Nat. Commun. 12, 1539 (2021).* Jing, M. et al.: A genetically encoded fluorescent acetylcholine indicator for in vitro and in vivo studies. *Nat. Biotechnol.* **36**, 726–737 (2018). Reimer, J., McGinley, M. J., Liu, Y., Rodenkirch, C., Wang, Q., McCormick, D. A., & Tolias, A. S. (2016). Pupil fluctuations track rapid changes in adrenergic and cholinergic activity in cortex. *Nature communications*, *7*(1), 1-7

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Poster

P977. Electrical Methods to Modulate Neural Activity I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P977.07

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Grant R01-HL-141560
LivaNova PLC

Title: Differential activation of locus coeruleus neurons by vagus nerve stimulation bursting parameters

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Abstract: Vagus nerve stimulation (VNS) is used to treat epilepsy and depression, and additional uses such as heart failure and rheumatoid arthritis are under investigation. The noradrenergic center locus coeruleus (LC) is a key mediator for effects of VNS in the brain; however, the optimal stimulation parameters for ideal neuromodulatory effects remain unknown. Therefore, the current study examines neuronal response profiles during 11 VNS paradigms for ideal LC activation. Extracellular activity was recorded in left LC for chloralose-anesthetized rats (n=12) using stereotaxically-placed, high-impedance tungsten electrodes. VNS paradigms were delivered in a pseudorandom fashion for 5 consecutive cycles via a bipolar cuff electrode around the left cervical vagus nerve. These 11 predetermined VNS paradigms utilized variable standard frequencies and bursting characteristics to alter LC activity. Recorded neurons (n=83) were divided into categories based on firing rate changes from baseline during VNS: non-responders, negative, positive, and super responders. Additionally, timing response profiles of neurons were assessed: correlation of firing to VNS on periods, changes in synchrony between LC neurons during VNS, and latency/spike density during bursting VNS paradigms. Responses to stimulation are reliable and have expected latency, which implies causation by VNS. Overall, standard VNS paradigms drive greater responders initially, while bursting paradigms yield more positive responders across the 5 VNS cycles ($p<0.05$). Increasing VNS frequency yields higher super responders ($p<0.05$). For bursting paradigms, shorter intervals and fewer pulses per burst show more responders ($p<0.05$). Furthermore, standard paradigms show more cells negatively correlate with VNS ($p<0.01$). Greater numbers of cells are positively correlated with VNS with

more pulses per burst (p<0.05), with shorter intervals between bursts, and as standard VNS frequencies increase (p<0.01). LC synchrony is increased during VNS for the majority of bursting paradigms (p<0.05), but not for standard paradigms. For standard paradigms, 20 Hz is ideal for driving positive LC activation, while 5 Hz best negatively activates LC. Increased synchrony during bursting VNS suggests common input/network recruitment that may be ideal for nuclei like LC with strong phasic response profiles, with median frequencies and pulse numbers/short intervals having the strongest positive response. Stronger negative LC responses are observed with longer bursting intervals and lower frequencies. These results may provide insight into ideal parameters that will help maximize clinical efficacy of VNS.

Disclosures: A. Farrand: None. **V. Jacquemet:** None. **R. Verner:** A. Employment/Salary (full or part-time):; LivaNova PLC. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); LivaNova PLC stock options. **E. Beaumont:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; LivaNova PLC funding to independently conduct study.

Poster

P977. Electrical Methods to Modulate Neural Activity I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 10:45 AM - 11:45 AM

Program #/Poster #: P977.08

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Grant 1R01HL152162-01A1

Title: Electrical stimulation of subdiaphragmatic vagal trunk slows development of hypertension in the SHR

Authors: *E. W. DIRR[1], K. J. OTTO[2], J. ZUBCEVIC[1];
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Abstract: Introduction: The vagus nerve facilitates bidirectional communication between the gut and brain. A low vagal tone has been described in many conditions characterized by inflammation and gut dysbiosis, including hypertension (HTN). However, the role of the sub-diaphragmatic gut-projecting vagal nerve branches in control of blood pressure (BP) remains unclear. This study examines the effects of augmenting subdiaphragmatic vagal signaling on development of HTN in spontaneously hypertensive rat (SHR), a rodent model of neurogenic age dependent HTN characterized by gut dysbiosis. **Methods**: Juvenile male SHR (N=10, 8 wo) were implanted with telemetric BP transducers and either a silicone cuff electrode (STIM, N=5) or sham cuff devoid of electrode sites (SHAM, N=5) on the ventral subdiaphragmatic vagal

trunk. Following recovery and baseline BP recordings, STIM rats were connected to a stimulus generator and subjected to biphasic (600 μA, 500 us/phase, 25 Hz) vagus nerve stimulation (10 minutes/ session, 3 sessions/ day, 5 days/ week). SHAM rats were subjected to similar handling but with no stimulation. 24 hr continuous BP recordings were obtained once per week for six weeks in all rats. Analysis of cecal bacterial abundance and composition was completed using the Illumina platform 16s bacterial sequencing at endpoint. **Results**: Starting at five weeks after stimulation onset, diastolic BP in SHAM rats was significantly higher compared to baseline (ΔDBP= +26.7 mmHg, $p < 0.001$). This was not observed in STIM rats (ΔDBP= +15.2 mmHg, $p = 0.15$). Four weeks after the onset of stimulation, we observed a significantly elevated systolic (ΔSBP= +22.5 mmHg, $p = 0.002$) and mean BP (ΔMBP= +16.7 mmHg, $p = 0.009$) in SHAM rats alone when compared to baseline. Both SHAM and STIM groups showed a significantly elevated SBP and MBP at weeks five compared to baseline, which remained elevated for the duration of the study. Gut bacterial sequencing showed no significant differences in diversity or abundance of big bacterial phyla between groups. However, the abundance of bacterial orders of *Anaeroplasma*, *Anaeroplasmataceae*, and *Anaeroplasmatales* was higher in the SHAM versus STIM group. **Conclusions**: Bioelectric restoration of sub-diaphragmatic vagal signaling may be beneficial in reparation of gut-brain neural axis and cardiovascular homeostasis. This is demonstrated through alleviated increase in DBP and delay in increase in SBP and MBP in the SHR following chronic vagal stimulation. These changes may be due to effects on afferent vagal signaling and/or gut microbiota induced by subdiaphragmatic vagal stimulation.

Disclosures: E.W. Dirr: None. **K.J. Otto:** None. **J. Zubcevic:** None.

Poster

P977. Electrical Methods to Modulate Neural Activity I

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Program #/Poster #: P977.09

Topic: I.08. Methods to Modulate Neural Activity

Support: NIH Grant U18EB029353
NIH Grant R01DE021798
NSF GRFP

Title: Wireless Endovascular Nerve Stimulation with a mm-sized MagnetoElectric Powered Bio-ImplanT (ME-BIT)

Authors: *J. CHEN[1], P. KAN[2], Z. YU[1], F. ALRASHDAN[1], R. GARCIA[2], A. SINGER[1], C. LAI[1], B. AVANTS[1], S. CROSBY[3], M. M. FELICELLA[2], A. ROBLEDO[2], J. D. HARTGERINK[1], S. A. SHETH[4], K. YANG[1], J. T. ROBINSON[1];
[1]Rice Univ., Houston, TX; [2]Univ. of Texas Med. Br., Galveston, TX; [3]Neuromonitoring Associates, Houston, TX; [4]UTHealth McGovern Med. Sch., Houston, TX

Abstract: Advancements in bioelectronic medicine have enabled the treatment of various disorders through neuromodulation. However, existing approaches often require invasive surgeries to reach therapeutic nerve targets and implantation of centimeter-sized devices. Here we demonstrate that it is possible to stimulate peripheral nerves from within blood vessels using a millimeter-sized wireless stimulator that can be delivered minimally invasively through a percutaneous catheter. By taking advantage of the vascular system that accompanies nerves as part of the neurovascular bundle, stimulation leads can be directed through the blood vessels to specific nerve targets that are difficult to reach with traditional surgeries. This would significantly lower the barrier to entry for neuromodulatory treatment approaches because of the reduced risk. These mm-sized implants are achieved through the use of magnetoelectric materials to deliver data and power to a digitally programmable 0.8 mm^2 CMOS system-on-a-chip. As a proof-of-principle, we demonstrate that these MagnetoElectric powered Bio-ImplanTs (ME-BITs) can stimulate peripheral nerves both directly and from within the blood vessels in rodent and porcine models. The wireless endovascular nerve stimulator (EVNS) shown here opens the door for the possibilities of a new class of low risk and highly precise endovascular bioelectronic medicine with direct access to difficult to reach nerve targets and analytes within the blood for fully closed-loop systems without the need for open surgery.

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Poster

P024. Development: Cytoskeletal Regulation of Dendrite Growth

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P024.01

Topic: A.05. Axon and Dendrite Development

Support: 5R01MH118444-03

Title: The role of KIF11 in modulating microtubule dynamics and dendritic transport

Authors: *J. WINGFIELD[1], S. V. PUTHANVEETTIL[2];
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Abstract: During mitosis the cell relies on a several different microtubule motors to dynamically manipulate the microtubule architecture for proper cell division. Interestingly, several of these motors are expressed in postmitotic neurons, including KIF11 (kinesin-5) which walks on antiparallel microtubules to separate the spindle poles. A recent study from our lab found that knockdown of KIF11 in mouse primary hippocampal cultures leads to increased dendritic

arborization, the number of synapses, and synaptophysin and piccolo expression (Swarnkar et al. 2018). We sought to investigate the mechanism by which KIF11 regulates dendritic arborization by first examining how KIF11 modulates microtubule dynamics in dendrites. We expressed Dendra-EB1 in primary hippocampal cultures and utilized two different KIF11 inhibitors: one that prevents KIF11 from binding to microtubules (Ispinesib) and one that locks KIF11 in the rigor state, bound to microtubules (BRD9876). Ispinesib treatment significantly increased the frequency of retrograde EB1 comets compared to both the vehicle (~330%) and BRD9876 (~480%). However, BRD9876 had no significant effect on the frequency of EB1 comets in either the anterograde or retrograde direction. Curious if a change in microtubule dynamics would broadly impact microtubule-based transport, we visualized trafficking of mitochondria in primary hippocampal cultures by employing MitoTracker© Green. Intriguingly, Ispinesib treatment led to a significant decrease in anterograde transport compared to the vehicle (~240%) and BRD9876 (~380%); as well as an increase in retrograde transport of mitochondria compared to the vehicle (~260%) and BRD9876 (~180%). Again, we found BRD9876 had no significant effect on the trafficking of mitochondria compared with the vehicle. Given that BRD9876 prevents KIF11 from walking on microtubules and addition of this drug had no impact on EB1 comets of mitochondrial transport, we suspect KIF11 is already in the rigor state and acts to stabilize microtubules in dendrites. We reason that the observed increase in dynamic minus-end-out microtubules upon KIF11 inhibition is what ultimately leads to increase in arborization, and synapse number seen with KIF11 knockdown. We propose a new model for dendritic arborization in where dynamic minus-end-out microtubules initiate the formation of a new dendritic branches.

Disclosures: J. Wingfield: None. **S.V. Puthanveettil:** None.

Poster

P024. Development: Cytoskeletal Regulation of Dendrite Growth

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P024.02

Topic: A.05. Axon and Dendrite Development

Support: NIH NS086082
GSU 2CI Neurogenomics

Title: The chaperonin CCT is required for dendritic elaboration through cytoskeletal regulation

Authors: *E. N. LOTTES, B. TETE, D. N. COX;
Neurosci. Inst., Georgia State Univ., Atlanta, GA

Abstract: Neuronal proteostasis is integral to the growth and maintenance of intricate and polarized cellular architectures. Chaperones are a key part of proteostatic maintenance

machinery, and ensure proper conformation of other proteins. While chaperones are particularly important in neurons, they have been under-studied in dendrites. Chaperonin-containing tailless complex polypeptide-1 (CCT) is an ATP-dependent chaperonin comprised of eight subunits which come together to form a barrel-shaped complex. CCT is thought to fold anywhere from 1-15% of the cellular proteome. Two of its most notable clients are actin and tubulin - major cytoskeletal components essential to the development and maintenance of dendritic arbors. Although CCT has been studied in axons, especially in conjunction with Hereditary Spastic Paraplegia, a genetic disease marked by advancing neuropathy, the potential function of CCT is not well understood in dendrites. Therefore, we investigated the role of CCT in the dendrites of *Drosophila melanogaster* multidendritic (md) sensory neurons of the peripheral nervous system using neurogenetics; *in vivo* imaging; and neuromorphometric quantitative analyses. Loss-of-function (LOF) genetic analyses support recently published work showing that disruption of each of the eight CCT subunits results in dendritic hypotrophy in dendritically complex Class IV (CIV) sensory neurons. Dendritic complexity plateaus in early third instar stage in CCT LOF conditions, revealing a failure of elaboration due to LOF of the chaperonin. Despite findings in CIV neurons, a LOF screen of simpler Class I, II and III neurons did not reveal any hypotrophy due to loss of CCT. Transcriptomic analyses of these neuron classes reveal highest levels of expression in CIV neurons and lowest in CI neurons. Together, these findings suggest that different neuronal types have differential reliance on CCT function. This is bolstered by our cytoskeletal investigations, which have revealed that homozygous mutation of CCT5 leads to decreased levels of stable microtubules in CIV but not CI neurons. Through live multi-channel imaging we have confirmed that loss of CCT results in both decreased stable MT and F-actin signal throughout the dendritic arbor. Polarity of MT EB1 comets is preserved in CCT LOF conditions, though the comets are fewer and slower. Early work on the CCT co-chaperone, Prefoldin (PFD), suggest that PFD LOF results in phenotypically similar but milder hypotrophy and reductions in cytoskeletal signal. Together, our findings indicate that CCT is necessary for the elaboration of dendritic arbors through regulation of the cytoskeleton in complex CIV md neurons.

Disclosures: E.N. Lottes: None. **B. Tete:** None. **D.N. Cox:** None.

Poster

P024. Development: Cytoskeletal Regulation of Dendrite Growth

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P024.03

Topic: A.05. Axon and Dendrite Development

Support: NIH Grant EY024884
NIH Grant EY021146
NIH Grant EY032392

Title: Fat3 acts through independent cytoskeletal effectors to coordinate asymmetric cell behaviors during polarized circuit assembly

Authors: *E. AVILES[1], A. KROL[1], S. J. HENLE[1], J. BURROUGHS-GARCIA[2], M. R. DEANS[2], L. V. GOODRICH[1];
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Abstract: The correct localization of neurons, their processes, and their synapses is crucial for the proper flow of information within a neural circuit. This polarity emerges sequentially in development, starting with directed migration of neuronal precursors, which subsequently elaborate neurites that form synapses in specific locations. Although several molecules required for synapse formation between specific partners have been identified, less is known about the molecular mechanisms that control where synapses form relative to other neurons in the circuit. The retina offers an excellent example of the link between the morphology of an individual neuron and its position in the circuit. In the retina, synapses occur exclusively in the outer and the inner plexiform layers (IPL). Amacrine cells (ACs), whose cell bodies reside either in the inner nuclear layer or retinal ganglion layer, make connections with other retinal cells in the IPL. Thus, developing ACs must extend processes that are oriented towards the IPL, regardless of where their cell bodies are positioned. We focused on AC development to study the molecular mechanisms that govern neural circuit polarity. In other organs, Fat cadherins sense position and polarize individual cells by inducing localized changes in the cytoskeleton that are coordinated across the tissue. Similarly, the polarized organization of the retina depends on Fat3, an atypical cadherin that controls where neurons migrate, which processes they retract, and where their synapses form. Here, we show that Fat3 acts through discrete modules in its intracellular domain (ICD) to coordinate these diverse features of circuit assembly. We found that Fat3-ICD interacts with cytoskeleton regulators, including Kif5B, Ena/VASP proteins, and the WAVE regulatory complex (WRC). By using CRISPR/Cas-9 to delete specific modules from the Fat3-ICD in mice, we found that neuronal migration, neurite retraction, and synapse localization are controlled by independent sets of effectors. Removal of binding motifs to WRC and Kif5B, but not Ena/VASP, affected AC migration. Consistently, the WRC is mislocalized in Fat3-deficient ACs, and deletion of Abi1 - a critical component of the WRC - resulted in AC migration defects. On the other hand, all three Fat3-ICD motifs are required for neurite retraction and, in their absence, synapses occur outside of the IPL. Unexpectedly, upon deletion of most of the Fat3-ICD, extra neurites were retained but they did not form ectopic synapses, suggesting that Fat3 acts in an independent pathway for synapse localization. Thus, Fat3 serves as a molecular node to coordinate different asymmetric cell behaviors across development.

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Poster

P024. Development: Cytoskeletal Regulation of Dendrite Growth

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P024.04

Topic: A.05. Axon and Dendrite Development

Support: March of Dimes Foundation
Whitehall Foundation Research Award
National Science Foundation
National Institute of General Medical Sciences
National Institutes of Health

Title: The kpc-1 3'UTR promotes dendritic transport and local translation of mRNAs to regulate dendrite branching and self-avoidance of a nociceptive neuron

Authors: *M. SHIH[1], Y. ZOU[2], H. CHIU[3], T. FERREIRA[4], N. SUZUKI[1], W. ZOU[5], C.-F. CHUANG[1], C. CHANG[1];
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Abstract: Mechanical stimuli on the skin of *C. elegans* are detected by the dendritic arbors of PVD nociceptive neurons, which provide uniform sensory coverage outside the head region across the entire animal. Through genetic screens, we isolate three mutants that display profound dendrite self-avoidance defects in PVD neurons. Studying dendrite self-avoidance in *C. elegans* is likely to provide new mechanistic insight into the process as the well-known self-avoidance molecule Dscam is absent from the *C. elegans* genome. Through whole genome sequencing, we identify the responsible mutations in the *kpc-1* gene. Compared to wild-type animals, a strong *kpc-1* mutant allele exhibits secondary dendrite branching defects whereas a weak *kpc-1* mutant allele displays tertiary dendrite self-avoidance defects. Here, we show that the *kpc-1 3' UTR* is required for *kpc-1*'s functions in both dendrite branching and dendrite self-avoidance. The *kpc-1* 3'UTR facilitates *kpc-1* RNA localization to branching points and contact points between sibling dendrites. Using fluorescence recovery after photoconversion, we show that the *kpc-1* 3'UTR promotes local protein synthesis in the distal segment of PVD dendrites. We identify an important secondary structural motif in the *kpc-1* 3'UTR required for tertiary dendrite self-avoidance. We demonstrate that over-expression of *kpc-1* leads to greater self-avoidance without limiting initial dendrite outgrowth, supporting a direct role of *kpc-1* in self-avoidance. Animals with *dma-1* receptor over-expression display similar secondary dendrite branching and tertiary dendrite self-avoidance defects that are suppressed with *kpc-1* over-expression, which suggests that DMA-1 is a potential KPC-1 target that is down-regulated by KPC-1. Our results support a model where KPC-1 proteins are synthesized locally at branching points and contact points between neighboring dendrites to locally down-regulate DMA-1 receptors to promote dendrite branching and self-avoidance.

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Poster

P024. Development: Cytoskeletal Regulation of Dendrite Growth

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Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P024.05

Topic: A.05. Axon and Dendrite Development

Support: NIH NS086082

Title: The PP2A serine/threonine phosphastase complex regulates cell-type specific cytoskeletal organization to promote dendritic diversification

Authors: *S. BHATTACHARJEE[1], E. N. LOTTES[1], S. NANDA[2], G. A. ASCOLI[2], D. N. COX[1];
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Abstract: Uncovering the molecular mechanisms that regulate dendritic diversification is essential to understanding the formation and modulation of functional neural circuitry. Transcription factors (TFs) have been widely implicated in regulating dendritic diversity but the molecular mechanisms by which this is achieved remain incompletely understood. Previous studies in our lab have demonstrated that the TF Cut regulate cell-type specific dendritogenesis through cellular pathways that converge on cytoskeletal architecture in *Drosophila* multi-dendritic (md) neurons. Neurogenomic and phenotypic analyses identified the highly conserved PP2A serine/threonine phosphatase complex as a downstream effector of Cut. The PP2A complex is composed of a catalytic subunit encoded by *microtubule star (mts),* a scaffolding subunit encoded by *PP2A-29B* and one of four alternate regulatory subunits encoded by *widerborst (wdb), twins, well-rounded (wrd)* and *CG4733.* Mutant analyses of *mts* and *PP2A-29B* reveal severe reductions in dendritic arborization with *wdb* functioning as the relevant regulatory subunit in Class IV (CIV) md sensory neurons. In contrast, mutations in *mts* and *PP2A-29B* leads to increased dendritic complexity via *de novo* filopodia formation in Class I (CI) md sensory neurons. Mechanistically, while Mts and PP2A-29B are also part of the STRIPAK complex, our data indicate that the PP2A and STRIPAK complexes function in parallel to regulate dendritic architecture. Cellularly, live imaging reveals that *mts* mutations leads to microtubule (MT) destabilization and increased MT turnover. In addition, dynamic EB1::GFP imaging reveals Mts is required to maintain both dendritic and axonal MT polarity. In contrast to its effects on MT, *mts* knockdown leads to F-actin reorganization with a proximal shift towards the soma and actin hyperstabilization. Loss of *mts* has distinct effects on organelle trafficking in these neurons. In CIV neurons, *mts* knockdown leads to reductions in dendritic

localization of organelles including mitochondria and satellite Golgi outposts while CI neurons show increased Golgi outpost trafficking along the dendritic arbor. Further, *mts* mutant neurons exhibit defects in neuronal compartmentalization with selective markers of dendrites and axons being detected in opposing compartments. At a regulatory level, phenotypic and immunohistochemical analysis reveal that β-tubulin subunit 85D is a common PP2A target in CI and CIV md neurons while the TF FoxO may be a target of the complex in CI neurons. Collectively, these studies provide novel insights into the functional roles of the PP2A phosphatase in promoting dendritic diversity.

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Poster

P024. Development: Cytoskeletal Regulation of Dendrite Growth

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Program #/Poster #: P024.06

Topic: A.05. Axon and Dendrite Development

Support: NIH KL2

Title: Initiation vs. elongation: determining the specific contribution of Ca_V1 channels to activity-dependent neurite outgrowth in cerebellar granule neurons

Authors: *A. L. PARKER, B. MEADE, H. WEN, A. J. WILLIAMS;
Dept. of Psychiatry, Univ. of Iowa, Iowa City, IA

Abstract: The neuropsychiatric disease-associated Ca_V1 channels are critically involved in neurodevelopment. It has been shown in multiple neuronal subtypes that KCl-induced activity induces dendritic growth through Ca_V1 channels. These measures of dendritic growth have largely used global measurements of dendritic growth, such as total dendritic length and dendritic branching. While these data are useful for identifying general phenotypic changes due to Ca_V1 channels, they combine the output of multiple biological processes, namely dendritic initiation and elongation. Dendritic initiation is the process by which actin-derived processes are extended from existing structures. Conversely, dendritic elongation is the microtubule-driven invasion and stabilization of dendrites. These two processes are mediated by different cell signaling pathways yet existing studies of Ca_V1 channels have not distinguished between them. Therefore, the underlying mechanism for how Ca_V1 channels specifically impact activity-dependent dendritic growth is not yet known.
In this study, I use sparsely labelled cerebellar granule neurons to specifically measure the number and length of the neurites for each individual cell to determine how activity-dependent neurite initiation and elongation are respectively impacted. I measured both primary neurites

(directly from the soma) and secondary neurites (branching from existing neurites) to determine how each of these components are impacted.
This study shows that KCl-induced activity is significantly associated with increased dendritic number and not dendritic length. Therefore, KCl-induced activity impacts dendritic initiation, not dendritic elongation. I next assessed the contribution of Ca_V1 channels to KCl-induced neurite initiation through the use of both a Ca_V1 channel antagonist and agonist. I showed that Ca_V1 channels impact the number of primary neurites induced by KCl without affecting the number of secondary neurites. This indicates that Ca_V1 channels are specifically involved in activity-dependent primary dendrite initiation, not secondary dendrite initiation.
These data clarify existing knowledge on the relationship between Ca_V1 signaling and dendritic growth and provide a framework for connecting the neurodevelopmental function of these channels to long-term changes in neuronal structures and function. The next steps of this research will be to identify the downstream signaling pathways and mechanisms for how Ca_V1 channels affect primary neurite initiation in cerebellar granule neurons.

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Poster

P024. Development: Cytoskeletal Regulation of Dendrite Growth

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Topic: A.05. Axon and Dendrite Development

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Sigma Xi Grant-in-Aid of Research award to Mr. Pemberton

Title: Activation of the G protein-coupled estrogen receptor (GPER) promotes neural development in rat embryonic (E18) hippocampal neurons

Authors: *F. XU[1], M. ROSATO[1], K. PEMBERTON[1], C. DELEON[2], C. ARNATT[2];
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Abstract: Accumulating evidence has proven that the steroid hormone estrogen plays a pivotal role in the nervous system, ranging from its development and function to neuroprotection after injury. The mechanistic actions of estrogens in the nervous system were originally restrained to classical, nuclear estrogen receptors (ERs), ERα and ERβ, which regulate gene transcription processes. Recently, the seven transmembrane domain of G protein- coupled receptor 30 (GPR30) has been identified to be a third estrogen selective receptor, namely the G protein-coupled estrogen receptor (GPER). Although mounting evidence has suggested GPER's

profound roles in neural survival and protection, a caveat is that most studies have primarily looked at the effects of estrogen and GPER in mature neurons derived from postnatal to adult animals or after neural network has been established. GPER's roles and working mechanisms in early neuronal development, especially the development of neurites (axons and dendrites), is severely lacking. In this study, we investigated the role of GPER activation in neural development of the primary rat embryonic (E18) hippocampal neurons. Our preliminary data revealed that activation of GPER by selective agonist G1 and non-selective agonist 17β-estradiol (E2) significantly increased neurite outgrowth. GPER activation also increased neuronal excitability and intracellular calcium (Ca^{2+}) in hippocampal neurons. All these effects were significantly inhibited by GPER selective antagonist G15, implicating GPER's specific effects. The rise in Ca^{2+} involved extracellular entry via Ca^{2+} channels and phospholipase C (PLC)-mediated intracellular Ca^{2+} store release. RNA sequencing data revealed the significant upregulation of transcripts for protein kinases such as Ca^{2+}/calmodulin-dependent protein kinase II (CaMKII) and Akt (known as Protein Kinase B), as well as axonal/dendritic proteins such as the collapsing response mediator protein (CRMP) and growth associated protein 43 (GAP 43). Future studies
of these signalling pathways and neuron cytoskeletal proteins in GPER action will advance our mechanistic understanding of estrogen' effects in the nervous system and provide new knowledge for promoting nervous system development.

Disclosures: F. Xu: None. **M. Rosato:** None. **K. Pemberton:** None. **C. DeLeon:** None. **C. Arnatt:** None.

Poster

P078. Comparative Anatomy: Evolution

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P078.01

Topic: A.10. Development and Evolution

Support: MacBrainResource NIH RO1 MH113257

Title: Neurochemical dissection of the complex micro-structure of the stria terminalis in infant and adult macaque monkey

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Abstract: The stria terminalis (ST) is a bidirectional fiber tract anchored in the amygdala and bed nucleus. While the general C-shaped trajectory is well-established, there is much less known about the micro-organization, especially in macaque monkeys, the subject of the present study.

In parallel histological series reacted for antibodies against fiber or cellular elements, we addressed this question in five brains from one adult (8 years) and 4 infants (2.5 months old). Brains were frozen sectioned at 50μm in the coronal plane, with 1 mm within series spacing. Both the dorsal and ventral ST (dST, vST) were investigated, standardized to the anterior-posterior (AP) extent of the lateral geniculate nucleus (~7 mm). This gave several specific results: 1) In the dST and vST, myelin-poor bands or patches were evident and coincided with dense fiber bundles positive for calbindin (CB), calretinin (CR), or tyrosine hydroxylase (TH). These were medially biased, dorsal-ventrally oriented in the vST, but more medio-lateral in dST. In vST, fibers positive for neuropeptide Y (NPY) or somatostatin (SOM) were laterally biased, avoiding the ventralmost edge of the ST. Fibers positive for choline acetyltransferase were preferentially located dorsolateral (vST). 2) Large thalamostriatal blood vessels bordered and partly invaded the dST (medially) and vST (laterally and ventral) and were often associated with TH and NPY fibers. 3) Small clusters and scattered neurons, visualized by neuronal nuclei protein (NeuN), occurred across the AP extent. Neuron number was larger for dST than vST; and dST and vST counts, averaged over 5 sections per brain, were less numerous in the adult brain (dST: 70.4 ± 25.6 (SD), vST: 34.4 ± 17.7) than in the 2.5M infant brains (dST: 105.8 ± 3.9 (SEM), vST: 67.9 ± 3.4). 4) Subgroups of neurons were verified as positive for CR (the largest number), NPY, CB, or SOM. For the infants and adult respectively, the average number of neurons per section (vST) was CR (14.6 ± 1.9; 7.0 ± 1.0), NPY (5.1 ± 1.0; 2.5 ± 1.3), CB (3.9 ± 0.9; 4.0 ± 1.9), and SOM (n=2 infants: 3.8 ± 1.6; 2.8 ± 1.5). For dST, equivalent counts were CR (19.8 ± 2.6; 6.2 ± 1.6), CB (9.9 ± 0.5; 5.6 ± 6.0), NPY (8.6 ± 0.8; 3.0 ± 1.2), and SOM (for n=2 infants: 6.9 ± 0.7; 3.0 ± 1.6). 5) Cell size and shape are heterogeneous within each subpopulation. Neurons have previously been reported in the ST (rodent: Alheid et al., 1998; primate: Martin et al., 1991), and considered part of the extended amygdala. However, the conspicuously lower numbers in the adult brain recall the developmental cell loss typical of telencephalic white matter neurons (Kostovic and Rakic, 1980), raising the possibility that these neurons are intrinsic to the ST.

Disclosures: M. Sakharkar: None. **K.S. Rockland:** None. **A. Duque:** None.

Poster

P078. Comparative Anatomy: Evolution

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Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P078.02

Topic: A.10. Development and Evolution

Support: NIH Grant NS020926
Brains and Behavior Seed Grant (Georgia State University)

Title: White matter connectivity patterns in tool-using primates

Authors: *A. MURPHY[1], K. A. PHILLIPS[3,4], S. F. BROSNAN[5,6,7,8], E. E. HECHT[2];
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Abstract: Extensive tool-use is one of the hallmark features of human behavior. While human tool-use exceeds that of other species in complexity, other primates have shown the ability to utilize tools. Chimpanzees and bonobos (last common ancestor (LCA) with humans of 6-7 million years ago (Ma)) show tool-use behaviors in the wild. Other species, such as Rhesus macaques (LCA with humans of 25Ma), can use tools in captive settings but do not do so extensively in the wild. A group of even more distantly related primates, capuchins (LCA with humans of 40Ma), show convergent evolution of percussive stone tool use with humans and chimpanzees. Like humans, capuchins show high relative brain to body mass as well as sophisticated social behaviors including cooperation. Previous work comparing humans, chimpanzees and rhesus macaques has suggested that greater tool-using ability corresponds to more distributed patterns of temporal-parietal and fronto-parietal white matter connectivity. However, it is currently unclear whether similar neurological changes underly the ability to use tools in Old World and New World primate lineages. To address this question, 20 capuchins (*Sapajus apella*) were scanned at 3T. T1 images were acquired at a voxel size of 0.5mm isotropic, and diffusion-weighted imaging was acquired at 1.0mm isotropic at 128 directions with 11 B0s. Probabilistic tractography was performed using TRACULA. Seed regions-of-interest were areas of the brain associated with the social learning of tool use, including the ventral premotor area, ventrolateral prefrontal cortex, intraparietal lobule, superior temporal sulcus, and inferior temporal cortex. The thresholded extent of projections from these regions were compared both to humans and to other tool-using non-human primates. By comparing connectivity across distantly related primates who have displayed tool-using behavior, it may be possible to pinpoint specific patterns of connectivity that are associated with tool-use. This can allow for a better understanding of what changes in the brain may underly the evolution of this ability.

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Poster

P078. Comparative Anatomy: Evolution

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Program #/Poster #: P078.03

Topic: A.10. Development and Evolution

Support: NSF IOS 1457291

Title: Neuroanatomical asymmetry in the canine brain

Authors: *S. A. BARTON[1], D. A. GUTMAN[3], T. M. PREUSS[4], M. KENT[5], E. E. HECHT[2]; [1]Human Evolutionary Biol., [2]Dept. of Human Evolutionary Biol., Harvard Univ., Cambridge, MA; [3]Dept Psychiatry & Behavioral Sci., Emory Univ. Dept. of Psychiatry and Behavioral Sci., Atlanta, GA; [4]Yerkes Natl. Primate Res. Ctr., Emory Univ., Atlanta, GA; [5]Univ. of Georgia, Athens, GA

Abstract: In the brains of both humans and our ape relatives, research has documented left/right asymmetries in gray matter morphology, white matter connections, and functional activations. These asymmetries have been linked to specialized behavioral adaptations including language, gestural communication, handedness, and tool use. Meanwhile, left/right asymmetries in behavioral tendencies have been recognized across the animal kingdom, suggesting a deep evolutionary origin for the lateralization of physiological mechanisms underlying behavior. However, it is unknown to what extent brain asymmetries underlying lateralized behaviors are a general feature of large, complex brains or a unique adaptation of the primate lineage. Canids and other carnivores evolved large, complex brains independently and convergently with primates, and therefore offer an opportunity to address this question. We examined T1-weighted MRI images of 62 domestic dogs from 33 breeds, opportunistically collected from a veterinary MRI scanner from dogs who were referred for neurological examination but were not found to show any neuropathology. Images were segmented into gray matter, white matter, and CSF. Gray matter segmentations were registered to a bilaterally symmetric, study-specific template using ANTS and then modulated by the Jacobian determinant of the warpfield, an index of where and how much each subject's scan had to deform to come into alignment with the template. We then left/right flipped these images and computed original-flipped and flipped-original maps. Finally, we applied one-sample t-tests for the two subtraction images using Monte Carlo permutation testing. Volumetrically asymmetric regions of gray matter include portions of temporal, frontal, and piriform cortex. We additionally assessed the relationship between these gray matter asymmetries and historical functional specializations across breeds such as herding, guarding, and companionship. These results help inform our understanding of the neural basis of behavior in dogs, and more generally, suggest that asymmetry may be a common feature underlying the evolution of large brains and complex behavior across clades.

Disclosures: S.A. Barton: None. **D.A. Gutman:** None. **T.M. Preuss:** None. **M. Kent:** None. **E.E. Hecht:** None.

Poster

P078. Comparative Anatomy: Evolution

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Program #/Poster #: P078.04

Topic: A.10. Development and Evolution

Support: Biotechnology and Biological Sciences Research Council (BBSRC) UK [BB/N019814/1]

Title: Insight into hominoid brain evolution: A white matter atlas of the lar gibbon

Authors: *K. L. BRYANT[1], A. A. KHRAPITCHEV[2], M. BERTELSEN[4], P. R. MANGER[5], J. SALLET[6,3], R. B. MARS[3,7];
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Abstract: Determining the unique specializations of the human brain requires an understanding of how the long distance connectivity has been modified and reorganized since our evolutionary divergence from other primates. Recent work by our group (Bryant et al., 2020) elucidated putative human and chimpanzee white matter specializations that arose since our divergence from our recent common ancestor with chimpanzees, by comparing the major white matter bundles of those two species using macaques as an outgroup. However, to identify which specializations are unique to hominids (great apes) and which are unique to hominoids (great and lesser apes) requires the characterization of hylobatid (lesser ape) white matter organization. We produced a white matter atlas using probabilistic diffusion tractography on an ex vivo diffusion-weighted scan of a lar gibbon (Hylobates lar) brain from Copenhagen Zoo. The brain was formalin-fixed within 24 hours, stored in phosphate buffered saline, and scanned in fluorinert. Diffusion-weighted resolution was 0.6 mm * 0.6 mm, b-value = 4000 s/mm2. Protocols seeding in the bodies of 24 major white matter tracts, originally derived from protocols designed for humans and macaques (De Groot et al., 2013; Mars et al., 2018) were adapted based on our previous comparative investigations (Bryant et al., 2020). Tractography was carried out in native space using FSL's XTRACT (Warrington et al., 2020) and thresholded to produce maps of 24 major white matter tracts (fig. 1). Tracts were projected to the surface by multiplying the tracts with a matrix describing the tractography of the grey/white matter boundary to the whole brain (O'Muircheartaigh & Jbabdi, 2017), creating a cortical termination maps. Ongoing work will use exploit this map to create a formal comparison of great ape, lesser ape, and Old World monkey connectional anatomy to offer insight into the evolutionary trajectory of white matter organization in hominoid evolution (cf. Mars et al., 2018).

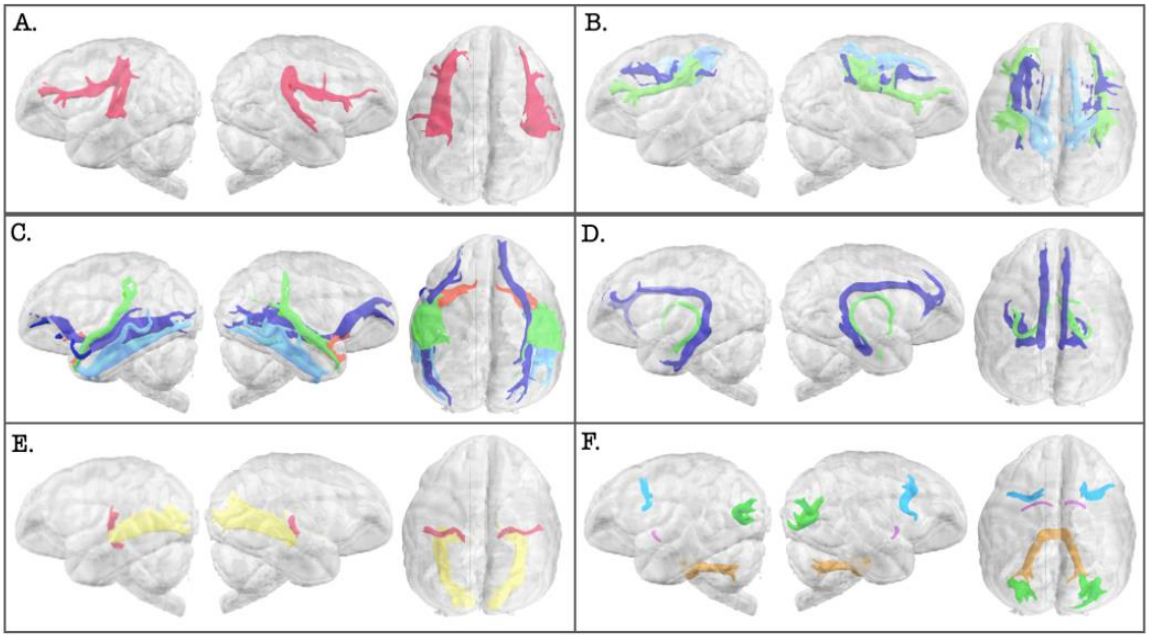


Figure 1: Thresholded probabilistic tracts in the lar gibbon. A: arcuate fasciculus; B: superior longitudinal fasciculi; C: temporal tracts (inferior longitudinal fasciculus, inferior fronto-occipital fasciculus, uncinate fasciculus, middle longitudinal fasciculus; D: cingulum and fornix; E: acoustic and optic radiations; F: short tracts - middle cerebellar peduncles, vertical occipital fasciculus, frontal aslant, anterior commissure.

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Poster

P078. Comparative Anatomy: Evolution

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Program #/Poster #: P078.05

Topic: A.10. Development and Evolution

Support: NSF DRL 1631563
NIH Office of the Director, P51 OD011132

Title: Individual variation in chimpanzee white matter connectivity, gray matter morphology, and behavior.

Authors: *S. VIJAYAKUMAR[1], W. D. HOPKINS[2], T. M. PREUSS[3], E. E. HECHT[1]; [1]Dept. of Human Evolutionary Biol., Harvard Univ., Cambridge, MA; [2]Dept. of Comparative Med., Univ. of Texas MD Anderson Cancer Ctr., Bastrop, TX; [3]Yerkes Natl. Primate Res. Ctr., Emory Univ., Atlanta, GA

Abstract: In order to explain how evolved differences in brain anatomy give rise to behavioral adaptations, comparative neuroscience researchers have primarily focused on inter-species comparisons of the large-scale organization of the brain. However, within-species analyses of variation in behavior and brain anatomy can also provide valuable information toward this topic. Interestingly, comparative studies suggest that the degree of variability in the gray matter morphology and sulcal anatomy of association regions is not only higher in humans as compared to chimpanzees, but also less heritable, putatively indicating that these regions evolved to support experience-dependent, culturally-transmitted behaviors. Given that the white matter pathways connecting the association regions have expanded in humans as compared to other primates, the underlying white matter connectivity seems likely to be driving these individual differences. To address this possibility, we studied 65 adult chimpanzees, ages ranging from 9 to 51 years (mean = 22, s.d. = 8.75, 41 female) that were housed at the Yerkes National Primate Research Center. Ethical approval was obtained from The Institutional Animal Care and Use Committee. T1 scans ($0.7 \times 0.7 \times 1.0$ mm^3) and diffusion scans ($1.8 \times 1.8 \times 1.8$ mm^3) in 60 directions were acquired using a Siemens 3T Trio scanner. To delineate the individual differences driven by white matter connectivity, we performed probabilistic tractography between all gray matter voxels and constructed a voxel-wise connectivity matrix. Coefficient of variation mapping of voxel-based connectivity showed the most variation in the orbitofrontal cortex, cingulate cortex, precentral gyrus, postcentral gyrus, anterior temporal lobe, lateral parietal lobe, and lateral occipital cortex. In order to identify individual variation in cortical grey matter organization, we used voxel-based morphometry. Grey matter images were registered to a symmetric study-specific template and modulated by Jacobians to account for local differences due to non-linear spatial transformations. Regions that showed the most individual variation included the orbitofrontal cortex, motor cortex, posterior parietal cortex, and parts of the visual cortex. We additionally analyzed associations between individual variation in connectivity and gray matter morphology and behavioral metrics of tool use, handedness, and communicative gesture. Because individual variation is the foundation on which natural selection acts, these findings have relevance for our understanding of how human-specialized behavioral skills might have evolved via adaptations to brain organization.

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Poster

P078. Comparative Anatomy: Evolution

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Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P078.06

Topic: A.10. Development and Evolution

Title: Description and characterization of the adult gray short-tailed opossum (Monodelphis domestica) brain: Focus on tyrosine hydroxylase-expressing neurons in the midbrain

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[3]Dept. of Psychological Sci., The Univ. of Texas Rio Grande Valley, Brownsville, TX

Abstract: Description and characterization of the adult gray short-tailed opossum (*Monodelphis domestica)* brain: focus on tyrosine hydroxylase-expressing neurons in the midbrain
Cristian M. Botello, John L. VandeBerg, and Mario Gil.
Mus musculus is one of the most widely used animal models to study central dopamine systems (Phifer-Riley & Nachmann, 2015). However, the research community needs alternatives to rodents to study the mammalian brain. The *Monodelphis domestica* is a marsupial, their pups are born underdeveloped and easily accessible, and are thus excellent models for tracking neurodevelopment (Baggott & Moore, 1990). The present study includes three objectives. The first was to consider the similarities and differences between the *Mus Musculus* and the *Monodelphis* brains. This was done by viewing Nissl-stained brain sections from each animal and comparing neural structures. We observed major species differences in the forebrain. The corpus callosum in the mouse brain is prominent, whereas it is absent in the Opossum. There are also differences in the general structure of the hippocampus and ventricles. As expected, posterior brain structures are similar between the two species. Further research is needed to determine what these differences could mean in behavior and cognition.The second objective was to characterize tyrosine hydroxylase (TH), a marker for dopamine neurons, expression in the *Monodelphis* brain. However, research is needed to see if commercially available TH antibodies, which are well researched in mice, can also be used to study the *Monodelphis* brain. Based on our preliminary results, we observed a stronger signal from the Pelfreeze TH antibody when compared to the EMD Millipore antibody. The final objective was to look at the differences in TH expression in the Ventral Tegmental Area (VTA) and the Substantia Nigra (SN) of Opossums that were exposed to a social stimulus compared to animals that were not exposed to a social stimulus. Animals in the experimental group were exposed to another animal for 10 minutes, then were sacrificed, and perfused 60 minutes after the social stimulus. Those in the control group were not exposed to social stimulus. We are currently processing brain tissue from these different groups to test the hypothesis that a social stimulus increases TH protein in the midbrain of the *Monodelphis*. Given that *Monodelphis* is a model for neurodevelopmental research and that there is not a lot of literature on their use in neuroscience, our work can contribute to the characterization of central dopamine systems in this innovative animal model.

Disclosures: C.M. Botello: None. **J.L. VandeBerg:** None. **M. Gil:** None.

Poster

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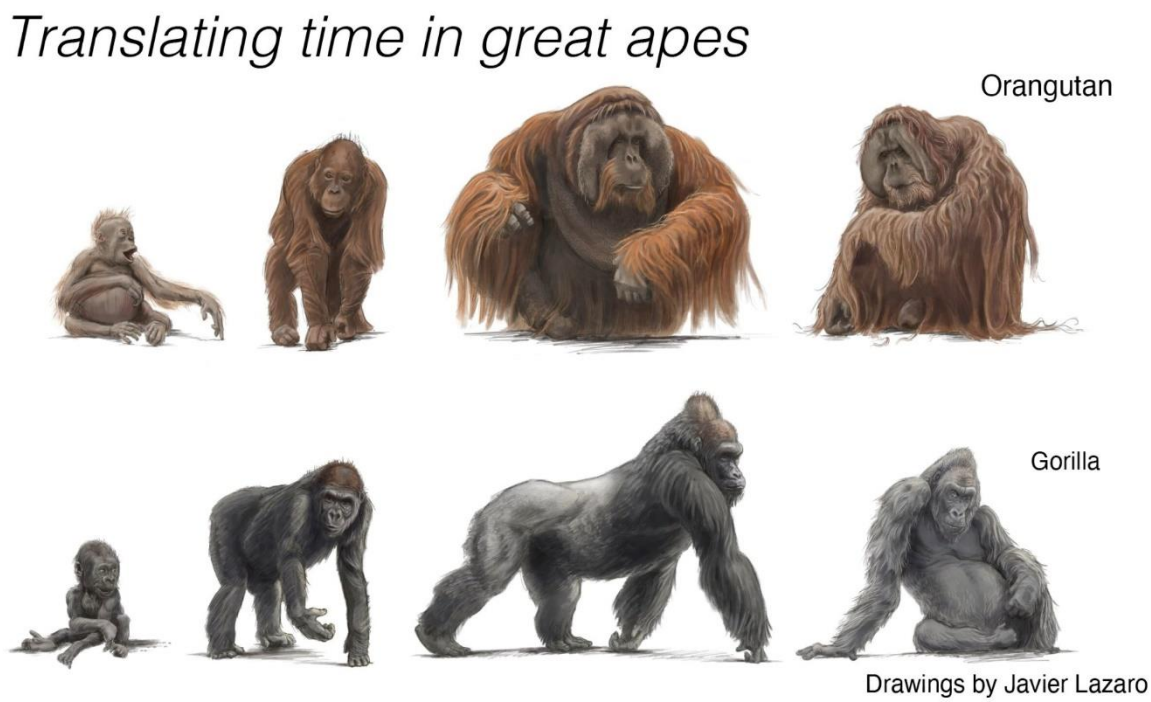
Topic: A.10. Development and Evolution

Support: NICHD R21HD101964 to CJC
COBRE 5P20GM103653
NIGMS P20GM103446 to CJC

Title: Translating time from transcriptomics and structural variation in great apes reveals old age as a distinctive human trait.

Authors: *K. OFORI[1], J. SCHWARTZ[1], B. RIGBY DAMES[2], C. BAUCUM[3], A. VAN DER KOUWE[4], E. TAKAHASHI[4], C. J. CHARVET[1];
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Abstract: How the unique capacities of human cognition arose in evolution is an enduring question of interest. Yet, we still lack appropriate procedures to align ages across primates, and this lacuna has hindered progress in understanding the evolution of biological programs in humans. We found corresponding ages across humans and five non-human primates (e.g., orangutan, gorillas) by collecting 379 time points from abrupt and gradual changes in behavior, anatomy, and transcription. In addition, we collected high-resolution diffusion MR scans of mammals close to birth (e.g., mouse, marmoset), which we used to track the relationship between brain growth and pathway maturation. We used these data to identify how schedules of development and aging vary across species. We found that the lifespan is extended in humans relative to apes after ages are aligned in different species. The extended lifespan of humans is concomitant with biological processes (e.g., brain atrophy) observed in aged humans that are difficult to detect in studied great apes. This dataset identifies corresponding ages across primates species, and accounts for the observation that biological pathways in aged humans are difficult to detect in great apes. Our study prompts a reevaluation of the use of model systems to understand aging, given that very old age is a period of life that has no counterpart in great apes.



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Poster

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Program #/Poster #: P078.08

Topic: A.10. Development and Evolution

Support: NSF IOS 1457291

Title: White matter connectivity of the cerebellum in the Russian farm-fox experiment

Authors: *C. N. ROGERS FLATTERY[1], S. VIJAYAKUMAR[1], J. LEVINE[2], J. MCENEANEY[2], L. N. TRUT[3], A. V. KUKEKOVA[4], J. B. SMAERS[5], E. E. HECHT[1]; [1]Dept. of Human Evolutionary Biol., [2]Harvard Univ., Cambridge, MA; [3]Inst. of Cytology and Genet. of the Siberian Br. of Russian Acad. of Sci., Novosibirsk, Russian Federation; [4]Dept. of Animal Sciences, Col. of ACES, Univ. of Illinois at Urbana-Champaign, Urbana, IL; [5]Dept. of Anthrop., Stony Brook Univ., Stony Brook, NY

Abstract: Above and beyond its classical association with motor coordination and motor learning, the cerebellum is thought to play a role in cognition. In particular, the lateral hemispheres of the cerebellum have been implicated in attention, executive function, language,

and social cognition. Most of this research has been done in humans and non-human primates, and little is known about cerebellum-cortical connectivity in clades that independently evolved large brains, such as canids. Here, we examined cortico-cerebellar connectivity in silver foxes (Vulpes vulpes) from the farm-fox domestication experiment. These foxes have been bred to create strains displaying divergent social behavior, thus they provide an excellent model for understanding how strong selection pressure for social behavior shapes neural circuits. We looked across strains of silver foxes for variation in cerebellum structure and connectivity. Previous research revealed volumetric differences in the cerebellum across these strains. In our study, we used diffusion tensor imaging (DTI) tractography in 10 tame, 10 unselected, and 9 aggressive/avoidant foxes to examine cortico-cerebellar connectivity across strains. Images were collected with 60 diffusion directions at a resolution of 300 cubic microns using a Bruker 9.4T MRI. We defined five cerebellar regions of interest: the anterior lobe; lobule HVI; lobule HVII; lobules HVIII, HIX, and HX; and the vermis. Among these ROIs, the anterior lobe was the best predictor of total brain size. In general, aggressive and conventional foxes had greater overlap in their patterns of connectivity compared to tame foxes. In all strains, the vermis showed the most extensive connectivity with the frontal cortex, in contrast to previous results from primates showing frontal connectivity with the lateral cerebellar hemispheres. This may be related to the frontal cortex in canids being more limbic-associated than in primates. Additionally, we also used a data-driven approach to divide the cerebellum into five clusters based on connectivity patterns with the cerebrum. These clusters corresponded moderately well to our a priori anatomical ROIs. In the tame foxes, clusters putatively corresponding to HVI and HVII were shifted more laterally compared to the conventional and aggressive foxes. Overall, our results suggest a congruence between anatomically defined lobules in the cerebellum and connectivity patterns. Our results also suggest that strong selection pressure on social behavior can affect cortico-cerebellar connectivity and that adaptive change in cerebellar morphology may be linked at least in part to connectivity changes.

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Poster

P078. Comparative Anatomy: Evolution

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P078.09

Topic: H.06. Social Cognition

Support: NSF 1846201

Title: Japanese and rhesus macaques differ in amygdala cannabinoid 1 receptor-immunoreactive axon density

Authors: *D. N. JONES[1], H. N. SMITH[1], K. N. HIRTER[1], E. L. MUNGER[1], P. R. HOF[2], C. C. SHERWOOD[3], M. RAGHANTI[1];
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Abstract: Japanese and rhesus macaques differ in amygdala cannabinoid 1 receptor-immunoreactive axon density
Endogenous and exogenous cannabinoids signal through the cannabinoid 1 receptor (CB1R) to modulate various aspects of social behavior, including aggression and anxiety. In rodents and primates, CB1R expression in the basolateral amygdala is dense and cannabinoid signaling in this region has been reported to influence social behavior. Little is known about how endocannabinoid signaling in the amygdala contributes to primate social diversity. The behaviorally diverse and species-rich cercopithecoid monkey genus *Macaca* is an ideal model for addressing this topic. Japanese (*Macaca fuscata*) and rhesus macaques (*M. mulatta*) display similar social styles in some respects; however, there is evidence to suggest they differ in their stress response, amygdala structure, and monoaminergic signaling. To further assess the molecular basis of social style in Japanese and rhesus macaques, we used immunohistochemistry and stereological methods to compare CB1R-immunoreactive (CB1R-ir) axon density in the basolateral amygdala, which is comprised of the lateral, basal, and accessory basal nuclei. Our study sample included 6 Japanese and 5 rhesus macaques. Repeated-measures ANOVAs were used to evaluate species differences, with amygdala region as the within-subjects measure and species as the between-subjects factor. This revealed significant main effects for species and area (p values < 0.05) with no interaction. Post hoc tests revealed higher CB1R-ir axon density in the basal and accessory basal nuclei of rhesus macaques compared to Japanese macaques. Our results suggest that CB1R-mediated signaling in the lateral nucleus of the amygdala is comparable between the two species, while the differences we observed in the basal and accessory basal nuclei may contribute to the nuanced behavioral differences observed between them.

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Poster

P078. Comparative Anatomy: Evolution

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P078.10

Topic: A.10. Development and Evolution

Support: NSF-IOS 1457291

Title: Neurodevelopmental scaling is a major driver of brain-behavior differences in temperament across domestic dog breeds

Authors: *E. E. HECHT[1], I. ZAPATA[2], *C. E. ALVAREZ[3], D. A. GUTMAN[4], T. M. PREUSS[5], M. KENT[6], J. SERPELL[7];
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Abstract: Dog breeds vary in temperament, including traits like aggression, anxiety, and trainability. These breed differences are both significant and highly heritable. However, the underlying neural bases are unknown. In this study, we analyzed breed-average scores for the 14 major dimensions in the Canine Behavioral Assessment and Research Questionnaire, a well-validated measure of canine temperament, in relation to structural MRI scans of 62 dogs of 33 breeds. We observed significant relationships between several behavior categories and brain morphology. Source-based morphometry analyses revealed that morphologically covarying gray matter networks involved in the flight-or-fight response and social processing were significantly linked to stranger-directed fear and aggression - putatively, the main behavioral responses under selection during wolf-to-dog domestication. Additionally, voxel-based morphometry analyses revealed that expansion in broad regions of cortex was significantly associated with higher scores for trainability. Conversely, expansion in distributed subcortical regions were significantly linked to C-BARQ categories for fear, aggression, and other “problem” behaviors. In striking correspondence with models of developmental constraint on brain evolution, these results overlapped closely with regional volume changes associated with changes in total brain size. These results suggest that disproportionate enlargement of later-developing regions in larger-brained dogs is at least partly responsible for the well-established link between dog body size and behavior.

Disclosures: E.E. Hecht: None. **I. Zapata:** None. **C.E. Alvarez:** None. **D.A. Gutman:** None. **T.M. Preuss:** None. **M. Kent:** None. **J. Serpell:** None.

Poster

P078. Comparative Anatomy: Evolution

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Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P078.11

Topic: H.06. Social Cognition

Support: NSF BCS-1846201

Title: Serotonin innervation of the ventral pallidum and nucleus accumbens is conserved among primates.

Authors: *H. N. SMITH[1], D. N. JONES[1], E. MUNGER[1], P. R. HOF[2], C. C. SHERWOOD[3], M. RAGHANTI[1];
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Abstract: Previously identified differences in serotonin innervation have been proposed to underlie differences in behavior, such as personality style and sociability. Contrasting serotonergic fiber densities have been found in the amygdala of chimpanzees versus bonobos, and humans and apes are known to have more serotonin than monkeys in the dorsal and medial caudate nucleus and dorsal putamen. Our present work builds on earlier results by examining serotonergic axon innervation density in the nucleus accumbens and ventral pallidum, two important nodes in the reward system. The present sample included humans (n = 6; NIH NeuroBioBank), pigtailed macaque monkeys (n = 5; National Primate Research Center, University of Washington), and capuchin monkeys (n = 6; Alpha Genesis). All individuals were adult and free of neuropathological alterations. Brain sections were immunohistochemically processed for serotonin transporter (SERT) (Millipore, MAB 5618), and stereological methods (SpaceBalls probe, MBF Bioscience) were used to quantify the length density of SERT-immunoreactive axons and neuron densities from adjacent Nissl-stained sections. Repeated measures ANOVA was used to evaluate differences of SERT-immunoreactive axon densities and neuron densities among species. The main effect of brain region was significant ($F_{1,2}$ = 12.25, p = 0.004) with greater SERT innervation in ventral pallidum compared to the nucleus accumbens in all species. The main effect of species and the interaction of species x brain region were not significant. Based on these results, the serotonergic system in the nucleus accumbens and ventral pallidum appears to be evolutionarily conserved in the amount of innervation supplied to neurons among human and other anthropoid primates.

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Poster

P087. Ionotropic Glutamate Receptors: AMPA, Kainate, and Delta Receptors

Location: Virtual Only

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Program #/Poster #: P087.01

Topic: B.02. Ligand-Gated Ion Channels

Support: UNAM-PAPIIT IA203421
R21-AG048506
P01-AG000538

RF1AG057558

Title: Learning differentially increases the synaptic levels of AMPA receptor subunits (GluA1-GluA4) in the rodent hippocampus: a single-synapse approach

Authors: *G. A. PRIETO[1], C. J. RODRIGUEZ-ORTIZ[2], M. KITAZAWA[3], C. W. COTMAN[2]; [1]UNAM, Inst. de Neurobiología, Juriquilla, Mexico; [2]UC Irvine, Irvine, CA; [3]Med., Univ. California, Irvine, Irvine, CA

Abstract: Decades of experimental work support the idea that synapses are the anatomical substrate for experience-dependent plasticity in the brain. Changes in the synaptic strength underlie learning and memory via the accumulation of glutamate AMPA receptors (AMPAR) at the surface of excitatory synapses. AMPAR are tetrameric receptors constituted by multiple combinations of four subunits: GluA1 GluA2, GluA3, and GluA4. However, the relative contribution of each AMPAR subunit on learning and memory is relatively unexplored. Here, we introduce Fluorescence Analysis of Single-Synapse Potentiation induced by Learning (FASS-PiL), a flow cytometry-based method to quantify surface levels of all four GluA-AMPAR subunits, in parallel, in isolated synaptosomes after a learning episode in rodents. We evaluated surface levels of all four AMPAR subunits in synaptosomes of mice trained in a learning protocol commonly used for studying episodic memory in the hippocampus (e.g., Object Location Memory (OLM) and Object Recognition Memory (ORM) tasks). Briefly, after exploring novel objects in a novel environment for 10 min, mice were returned to their home cage for 60 min. After hippocampus dissection, synaptosome isolation and immunostaining, samples were analyzed via flow cytometry. We first demonstrated that FASS-PiL is a simple and sensitive method to track all four GluAs at the synaptosome surface (each GluA subunit was paired with the presynaptic marker Neurexin-1beta to focus on synaptosome particles containing both pre- and postsynaptic compartments). Notably, by profiling hundreds of events, our data showed that training increases the number of hippocampal synaptosomes expressing high levels of GluA1 and GluA2, but not GluA3 and GluA4 (vs control animals). This result indicates that plasticity-related mechanisms underlying learning are AMPA-subunit-specific in hippocampal synapses. Our approach could provide the basis for protocols to study behavioral-relevant mechanisms of plasticity directly at the synapse, with single-event resolution.

Disclosures: G.A. Prieto: None. **C.J. Rodriguez-Ortiz:** None. **M. Kitazawa:** None. **C.W. Cotman:** None.

Poster

P087. Ionotropic Glutamate Receptors: AMPA, Kainate, and Delta Receptors

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P087.02

Topic: B.02. Ligand-Gated Ion Channels

Support: NIH Grant HL154512
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Title: α2δ-1 Promotes Synaptic Expression of Calcium Permeable-AMPA Receptors by Disrupting Heteromeric GluA1/GluA2 Assembly in the Hypothalamus in Hypertension

Authors: *J.-J. ZHOU, S.-R. CHEN, J.-Y. SHAO, H. CHEN, H.-L. PAN;
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Abstract: The native AMPA receptors (AMPARs) in the brain are calcium impermeable and mainly composed of GluA1/A2 heteromers. AMPARs lacking GluA2 are highly calcium permeable (CP-AMPARs) and their currents show inward rectification at positive membrane potentials. Increased prevalence of CP-AMPARs in the paraventricular nucleus (PVN) of hypothalamus augments sympathetic outflow in hypertension. However, the molecular mechanisms underlying this AMPAR synaptic plasticity in hypertension remain unknown. In this study, we determined the role of α2δ-1 in regulating AMPAR phenotype in the hypothalamus in spontaneously hypertensive rats (SHR). Co-immunoprecipitation analysis showed a significant reduction in GluA1/GluA2 heteromers in the synaptosome and endoplasmic reticulum (ER)-enriched fractions of the hypothalamus in SHR compared with normotensive Wistar-Kyoto rats (WKY). Whole-cell recordings in brain slices showed an inward rectification of current-voltage (I-V) relationship of AMPAR-mediated excitatory postsynaptic currents (AMPAR-EPSCs) in retrogradely labeled, spinally projecting PVN neurons in SHR. Inhibition of α2δ-1 with gabapentin increased GluA1/GluA2 heteromers in ER-enriched fractions of the hypothalamus and abolished the inward rectification of AMPAR-EPSCs in labeled PVN neurons in SHR. Co-immunoprecipitation analysis revealed that α2δ-1 physically interacted with both GluA1 and GluA2 in the hypothalamus in rats and humans. The protein levels of α2δ-1-GluA1 and α2δ-1-GluA2 complexes in the hypothalamus were significantly greater in SHR than in WKY. Furthermore, disruption of the α2δ-1-AMPAR interaction with an α2δ-1 C terminus peptide restored heteromeric GluA1/GluA2 in the ER and normalized the inward rectification of AMPAR-EPSCs in labeled PVN neurons in SHR. These findings indicate that α2δ-1 is an important AMPAR-interacting protein that promotes synaptic predominance of CP-AMPARs in the PVN via disrupting heteromeric GluA1/GluA2 assembly in hypertension.

Disclosures: J. Zhou: None. **S. Chen:** None. **J. Shao:** None. **H. Chen:** None. **H. Pan:** None.

Poster

P087. Ionotropic Glutamate Receptors: AMPA, Kainate, and Delta Receptors

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

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Topic: B.02. Ligand-Gated Ion Channels

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Title: The differential role of auxiliary subunits in the assembly of AMPA receptors

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Abstract: AMPA receptors (AMPARs) are ionotropic glutamate receptors that transmit the majority of fast excitatory synaptic transmission. Regulation of the number and composition of AMPARs at the postsynaptic membrane is essential for the process of learning and memory, and AMPAR dysregulation is implicated in various brain disorders. In the brain, AMPARs associate with auxiliary subunits that can affect their functional activity, however auxiliary subunit regulation of AMPAR assembly is unclear. Here, we addressed the role of auxiliary subunits in the assembly of AMPARs using a combination of blue native gel-electrophoresis, immunocytochemistry, and electrophysiology. We find that AMPARs co-expressed with cornichon-2 (CNIH-2) and cornichon-3 (CNIH-3) enhance the tetramerization process, whereas co-expression with other auxiliary subunits including stargazin (TARP-2), TARP-8, and GSG1-L does not. To study the basis for this, we mutated key residues in the AMPAR transmembrane domain which attenuates tetramerization and co-expressed these AMPARs with auxiliary subunits. Destabilized tetramerization leads to increased receptor dimers and significantly reduced tetramer formation. We found that when AMPAR mutants are co-expressed with CNIH-2 or CNIH-3, the tetramer fraction was efficiently rescued compared to co-expression with other auxiliary subunits. Our results suggest that auxiliary subunits influence receptor tetramerization through the transmembrane domain with CNIHs capable of stabilizing the transmembrane region and rescuing disrupted receptor assembly. In contrast, other auxiliary subunits demonstrate a much weaker influence on tetramerization compared to CNIHs. We conclude that CNIHs differ from other auxiliary subunits in their ability to influence the stages of AMPAR biogenesis.

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Poster

P087. Ionotropic Glutamate Receptors: AMPA, Kainate, and Delta Receptors

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P087.04

Topic: B.02. Ligand-Gated Ion Channels

Support: ZIA HD008914
ZIA HD008869

Title: Neto proteins differentially modulate the gating properties of Drosophila glutamate receptors

Authors: *T. HAN, M. SERPE;
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Abstract: *Drosophila* neuromuscular junction (NMJ) is a glutamatergic synapse similar in composition and physiology to excitatory synapses in the mammalian CNS. In flies as in humans, synapse strength and plasticity are determined by the interplay between expression of different ionotropic glutamate receptor (iGluR) subtypes, *Drosophila* NMJ utilizes two distinct postsynaptic complexes that co-exist within individual postsynaptic densities; they are heterotetrameric complexes composed of three shared subunits, GluRIIC, -IID and -IIE, and either -IIA (type-A) or -IIB (type-B). We have previously discovered Neto (Neuropillin and Tolloid-like) as an obligatory subunit of the fly NMJ iGluR complexes, essential for receptors recruitment and function at the *Drosophila* NMJ. Neto belongs to a family of highly conserved auxiliary proteins that have an ancestral role in the formation and modulation of glutamatergic synapses. *Drosophila* neto encodes two isoforms, Neto-α and -β, that share highly conserved extracellular motifs and have different cytoplasmic domains, generated by alternative splicing. Either isoform could confer functionality to NMJ iGluR complexes reconstituted in heterologous systems, indicating that *Drosophila* Neto is required for iGluR function. To examine the role of Neto isoforms on the modulation of NMJ iGluRs gating properties, we expressed both recombinant receptors in HEK293T cells and measured their properties using outside-out patch-clamp technique. Here we report for the first time the biophysical properties of fly NMJ iGluRs of defined subunit composition. We found that Neto is absolutely required for the function of both type-A and -B receptors; in the absence of Neto, these receptor complexes are expressed on the cell surface, but do not elicit any detectable currents. Both of Neto-α and -β have similar effects on single channel conductance and deactivation rates for both receptor subtypes. However, the two Neto isoforms differentially modulate the desensitization rates of type-A and -B receptors. Pre-treatment with Concanavalin A increases the single channel open times, whereas extracellular PhTx blocks the channels to various extents. In outside-out patches the IV relationship is linear but shows biphasic rectification in the presence of intracellular spermine. Finally, we compared the deactivation kinetics of receptor channels in outside-out patches and larval muscle membrane and found that native, extrajunctional channels have a slower decay kinetics than the reconstituted channels. Our study reveals that Neto is not only required for functional channels but also increases the diversity of the receptor properties.

Disclosures: T. Han: None. **M. Serpe:** None.

Poster

P087. Ionotropic Glutamate Receptors: AMPA, Kainate, and Delta Receptors

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Program #/Poster #: P087.05

Topic: B.02. Ligand-Gated Ion Channels

Support: NIH Grant 5T32GM007464-42

Title: Kainate and AMPA receptors control error correction during navigation along chemosensory gradients

Authors: *P. MALDONADO-CATALA[1], P. J. BROCKIE[2], J. E. MELLEM, JR[3], D. MADSEN[1], A. V. MARICQ[4];
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Abstract: Understanding how the nervous system controls behavior across vastly different timescales has been a challenge in neuroscience. During navigation, animals must perform moment to moment corrections as well as long-term modulation of their behavior to navigate efficiently. The nematode *C. elegans* suppresses backwards locomotion and performs corrections in steering during navigation towards chemical attractants. However, how this simple nervous system can control behavior at short timescales (steering) and longer timescales (reversals) is unknown. In this study, we show how a single pair of key interneurons called RIA, in the nematode control error-correction during navigation by adjusting steering and suppressing reversals. Interestingly, this is achieved by expressing two classes of ionotropic glutamate receptors, the AMPAR and Kainate receptor are expressed in RIA. These two classes of receptors differentially regulate behavior during navigation by 1) controlling moment to moment corrections in steering and 2) long-term suppression of reversals. Electrophysiological analysis reveals a mechanism for this neural control of error-correction. The AMPAR provides large-rapid activation of the interneuron that acts as a negative-feedback to control steering during navigation in the timescale of hundreds of milliseconds. The Kainate receptor provides small-amplitude but long lasting activation that allows for the suppression of reversals which occur in the timescale of tens of seconds. In this study, we tie the physiology of different classes of glutamate receptors to information processing and behavior. Taken together, our results provide a fundamental mechanism for how a simple neural circuit can control behavior across different timescales by expressing different classes of glutamate receptors which confer different properties to the activation of the neuron.

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Poster

P087. Ionotropic Glutamate Receptors: AMPA, Kainate, and Delta Receptors

Location: Virtual Only

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Topic: B.02. Ligand-Gated Ion Channels

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Title: Activation of extrasynaptic kainate receptors drives hilar mossy cell activity

Authors: *C. RAMOS[1], S. LUTZU[2], M. YAMASAKI[3], Y. YANAGAWA[4], K. SAKIMURA[5], S. TOMITA[6], M. WATANABE[7], P. E. CASTILLO[8];
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Abstract: Mossy cells (MCs) of the dentate gyrus (DG) are key components of an excitatory associative circuit established by reciprocal connections with dentate granule cells (GCs). MCs are implicated in place field encoding, pattern separation and novelty detection, as well as in brain disorders such as temporal lobe epilepsy and depression. Despite their functional relevance, little is known about the determinants that control MC activity. Here, we examined whether MCs express functional kainate receptors (KARs), a subtype of glutamate receptors involved in neuronal development, synaptic transmission and epilepsy. Using mouse hippocampal slices, we found that bath application of submicromolar and micromolar concentrations of the KAR agonist kainic acid induced inward currents and robust MC firing. These effects were abolished in GluK2 KO mice, indicating the presence of functional GluK2-containing KARs in MCs. In contrast to CA3 pyramidal cells, which are structurally and functionally similar to MCs, and express synaptic KARs at mossy fiber (MF) inputs (i.e., GC axons), we found no evidence for KAR-mediated transmission at MF-MC synapses, indicating that most KARs at MCs are extrasynaptic. Immunofluorescence and immunoelectron microscopy analyses confirmed the extrasynaptic localization of GluK2-containing KARs in MCs. Finally, blocking glutamate transporters, a manipulation that increases extracellular levels of endogenous glutamate, was sufficient to induceKAR-mediated inward currents in MCs, suggesting that MC-KARs can be activated by increases in ambient glutamate. Our findings provide the first direct evidence of functional extrasynaptic KARs at a critical excitatory neuron of the hippocampus.

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Poster

P087. Ionotropic Glutamate Receptors: AMPA, Kainate, and Delta Receptors

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P087.07

Topic: B.02. Ligand-Gated Ion Channels

Support: Turkish Ministry of National Education YLSY Scholarship
British Neuroscience Association

Title: Mechanisms of kainate receptor regulation of AMPA receptor surface expression and function

Authors: *B. P. YUCEL, J. D. NAIR, K. A. WILKINSON, R. SEAGER, A. FLETCHER-JONES, J. M. HENLEY;
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Abstract: Kainate receptors (KARs) and AMPA receptors (AMPARs) are tetrameric glutamate-gated ion channels that play critical roles in neuronal excitability, signal transduction and synaptic plasticity. Their dysfunction has been strongly implicated in neurological and neurodegenerative disorders including Alzheimer's disease, autism spectral disorders (ASD), and epilepsy. However, the precise inter-relationship, relative synaptic localisations of KAR - AMPAR complexes, and crosstalk between their signalling pathways, are not well understood.Our lab recently demonstrated that different patterns of KAR activation can bidirectionally regulate AMPAR surface expression and synaptic strength. Depending on the KAR stimulation protocol used, we observe either KAR-mediated LTP (long-term potentiation) or LTD (long-term depression) of AMPARs. To explore the molecular pathways that underpin KAR regulation of AMPARs we are now investigating whether the receptor complexes and/or individual subunits interact using co-immunoprecipitation techniques both in HEK293T cells by exogenously expressing KAR and AMPAR subunits, and in rat cortical neurons overexpressing KARs. Our data show that in both HEK293T cells and neurons the AMPAR subunit GluA1 co-immunoprecipitates with the KAR subunit GluK2. Furthermore, contrary to some previous reports, using high resolution confocal microscopy and immunolabelling we show that KARs and AMPARs can localise to the same dendritic spines at the same time. Building on these initial data we are testing the hypothesis that, at least at some synapses, KARs regulate AMPARs either by binding of individual AMPAR and KAR subunits or, perhaps more likely, through common interacting proteins.

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Poster

P087. Ionotropic Glutamate Receptors: AMPA, Kainate, and Delta Receptors

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P087.08

Topic: B.02. Ligand-Gated Ion Channels

Title: Noradrenaline excites serotonin dorsal raphe neurons via glutamate delta 1 receptor-dependent ionic current

Authors: *S. C. GANTZ[1], K. MOUSSAWI[2], H. S. HAKE[3];
[1]Mol. Physiol. and Biophysics, Univ. of Iowa, Iowa City, IA; [2]Psychiatry, Univ. of Pittsburgh, Pittsburgh, PA; [3]Psychology, Univ. of Washington, Seattle, WA

Abstract: *In vivo* studies using rodent models have shown that the dorsal raphe - the largest serotonergic nucleus and the predominant source of serotonin in the brain - is controlled by noradrenaline afferents. In this brain region, noradrenaline activates $G\alpha_q$-coupled α1-adrenergic receptors (α1-A_R), causing action potential firing and serotonin release. In the absence of noradrenaline, dorsal raphe neurons are either silent or fire slowly and erratically demonstrating necessity of α1-A_R signaling. Despite this crucial role, the ion channels responsible for α1-A_R-mediated depolarization were unknown. Here, we show in adult C57BL/6J male and female mouse brain slices that α1-A_R-mediated excitatory synaptic transmission was mediated by the ionotropic glutamate receptor homolog cation channel, glutamate delta 1 receptor ($GluD1_R$). $GluD1_R$-channels were constitutively active under basal conditions carrying tonic inward current and synaptic activation of α1-A_Rs augments tonic $GluD1_R$-channel current. The mechanism underlying this coupling has not been resolved but was GTP-dependent. We found that α1-A_R-mediated excitatory synaptic transmission was inhibited by the amino acids D-serine and glycine, the glutamate receptor antagonist, kynurenic acid, and completely blocked by the calcium-permeable glutamate receptor-channel blocker, NASPM. Provided the limitations of pharmacological strategies, we then used a CRISPR/Cas9 viral strategy to introduce a deleterious mutation to the gene encoding $GluD1_R$ (*Grid1*) and functionally deleted $GluD1_R$ channels from dorsal raphe neurons. Following functional deletion of $GluD1_R$, the α1-A_R-dependent inward current was eliminated. Further, loss of dorsal raphe $GluD1_R$-channels produced an anxiogenic phenotype. Thus, $GluD1_R$-channels are responsible for α1-A_R-dependent induction of persistent pacemaker-type firing of dorsal raphe neurons and regulate dorsal raphe-related behavior. Given the widespread distribution of these channels, ion channel function of $GluD1_R$ as a regulator of neuronal excitability is proposed to be widespread in the nervous system.

Disclosures: S.C. Gantz: None. **K. Moussawi:** None. **H.S. Hake:** None.

Poster

P087. Ionotropic Glutamate Receptors: AMPA, Kainate, and Delta Receptors

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P087.09

Topic: B.02. Ligand-Gated Ion Channels

Title: Standing current by the delta-1 glutamate receptor is independent of g protein activity in central neurons

Authors: *D. S. COPELAND, S. C. GANTZ;
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Abstract: G protein-coupled receptors (GPCRs) have grown to be large targets for therapeutics due to their versatility. As the pool of pharmaceuticals targeting these receptors expands, a deeper understanding of the actions of GPCRs has become more relevant. One of the many functions of GPCRs is action on ion channels to influence membrane potential and, ultimately, neurotransmitter release. The glutamate delta-1 receptor ($GluD1_R$) showcases this mode of action, where activation of $G\alpha_q$PCRs produces current through the $GluD1_R$ ion channel pore in a G protein-dependent manner. The $GluD1_R$ channel also carries a standing inward current, henceforth referred to as a tonic current (Gantz et al. 2020). The aim of this study was to investigate constitutive $G\alpha_q$PCR activity by manipulating G protein activity and monitoring changes in this tonic current. Brain slices were collected from male and female C57bl/6j mice and whole-cell patch-clamp recordings were made from dorsal raphe serotonin neurons at 35° C. After blocking NMDA, AMPA and kainate receptors, inhibitors or activators of G protein activity were delivered internally via the patch pipette. Tonic $GluD1_R$ currents were then measured by bath application of the pore-blocker NASPM. This produced apparent outward currents by blocking inward sodium flow and allowed us to quantify the amplitude of tonic current. Neurons treated with standard GTP-based internal solutions, as a control, had a -19.3±2.7 pA tonic current (n=8), consistent with our previous study. Neurons dialyzed with a non-hydrolyzable GTP analog (Gpp-NHp) produced a sizable potassium leak current through GIRK channels (49.7±8.2 pA, n=16). But Gpp-NHp had no effect on the tonic $GluD1_R$ current (p=0.72, Mann-Whitney test). We next targeted the alpha1-adrenergic receptor (alpha1-A_R), as our previous work demonstrated activation of alpha1-A_R augments $GluD1_R$ channel currents. Bath application of the alpha1-A_R inverse agonist prazosin had no effect on the tonic $GluD1_R$ current (n=7, p=0.50, Mann-Whitney test), suggesting a negligible role of on-going alpha1-A_R activity in the tonic $GluD1_R$ current. Lastly, following dialysis with a non-hydrolyzable GDP analog (GDPβS) which arrests G protein activity, there was no change in the tonic $GluD1_R$ current (-28.7±4.9 pA, n=13, p=0.27, Mann-Whitney test). This result held true after depleting $G\alpha_q$ responses via bath application of noradrenaline, and after multiple applications of NASPM. These data suggest that if present, constitutive $G\alpha_q$PCR activity has minimal impact on the tonic $GluD1_R$ current. Further, our results call into question the primary form of gating this mysterious, clinically-relevant receptor-channel.

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Poster

P087. Ionotropic Glutamate Receptors: AMPA, Kainate, and Delta Receptors

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Program #/Poster #: P087.10

Topic: B.02. Ligand-Gated Ion Channels

Support: NIH 2R01MH080047
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Title: Fluorescent lifetime imaging of CAMKII during behavioral time scale plasticity at CA1 synapses

Authors: *A. JAIN[1], R. YASUDA[2];
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Abstract: Hippocampal place cells, once formed, are stable for a given environment and can last for many days suggesting that an activity dependent synaptic plasticity underlies the induction/modulation of place cells. A recent study by Bittner et al showed that a novel non-hebbian form of plasticity induces CA1 place cell formation, where seconds long calcium plateau integrates with CA1 inputs to induce place cells. In our study, we characterized this behavioral time scale plasticity (BTSP) at single synapses in CA1 neurons. Using glutamate uncaging in hippocampal slices, we paired 5 uncaging pulses at 1 Hz frequency to somatic depolarization (induced by 600 pA current injection for 300 ms) with a 750 ms delay between them and found that potentiation can be induced in a synapse specific manner whether uncaging pulses were given before or after depolarization. In these experiments, when uncaging pulses precedes the depolarization, excitatory postsynaptic potential (EPSP) potentiation occurred by 76 ± 23% in stimulated spines and when depolarization was induced before the uncaging pulses, EPSP amplitude increased by 93 ± 26% in stimulated spines. As CA1 synapses receive inputs from different brain regions, we induced BTSP at different types of CA1 synapses using the same protocol and found that BTSP occurred specifically in the proximal apical dendrites and not in basal dendrites or distal dendrites. Generation of seconds long calcium plateau likely occurs via inputs from entorhinal cortex (EC) and provides instruction to the CA3-CA1 synapses during place cell formation. It is unknown what biochemical pathway transduces the signal from EC-CA1 synapses to CA3-CA1 synapses. Previous two-photon fluorescent life time imaging (2pFLIM) of CAMKII sensor showed stimulus locked CAMKII activity during long-term potentiation that lasts for 1-3 seconds, making it an ideal candidate to integrate EC and CA3 inputs during BTSP. We first confirmed the requirement of CAMKII in BTSP by doing experiments in slices made from T286A transgenic mice, where we failed to induce BTSP. However, surprisingly, preliminary 2pFLIM of CAMKII sensor during BTSP at single synapses showed no activation of CAMKII at single synapses and found a delayed CAMKII activity almost 30-40 second following BTSP induction. This suggests that maybe CAMKII is not the biochemical signal that integrates EC inputs to CA1 neurons. Further experiments will confirm

the CAMKII activity and identify the protein that integrates inputs during BTSP. Understanding activation profiles of kinases in behaviorally relevant plasticity mechanisms will provide insights into their specific functions during behavior

Disclosures: A. Jain: None. **R. Yasuda:** None.

Poster

P087. Ionotropic Glutamate Receptors: AMPA, Kainate, and Delta Receptors

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P087.11

Topic: B.02. Ligand-Gated Ion Channels

Support: The Leverhulme Trust

Title: Exploring the role of SENP3 in kainate receptor trafficking

Authors: *K. TALANDYTE, J. HENLEY, K. WILKINSON, R. SEAGER;
Univ. of Bristol, BRISTOL, United Kingdom

Abstract: SUMOylation is a protein based post-translational modification involving the conjugation of a small protein, SUMO, to substrate proteins to alter their stability, function or activity. The most common SUMO paralogues, SUMO-1-3, are attached to the substrate proteins through a three-step enzymatic cascade and can be removed by SUMO proteases (SPs). The biggest family of SPs are sentrin specific proteases (SENPs), which include SENP1-3 and SENP5-SENP7. There are multiple SUMO substrates identified in neurons, both in the nucleus, where SUMO is mostly localised, as well as in the cytosol, mitochondria and plasma membrane. Excitatory neurotransmission in the brain is mainly mediated by the neurotransmitter glutamate, which binds to three types of ionotropic receptors: AMPA, NMDA and kainate receptors (KARs). KARs are present pre-synaptically, where they regulate neurotransmitter release, post-synaptically, where they mediate synaptic transmission and plasticity and extra-synaptically, where they regulate neuron excitability. KARs seem to function both in a similar manner to AMPARs, by mediating synaptic plasticity and neurotransmission, and analogous to NMDARs, by inducing changes in surface expression of AMPARs to drive synaptic plasticity.
A widely expressed KAR subunit in the brain, GluK2, has been identified as a SUMO substrate in neurons. GluK2 C-terminus SUMOylation plays a role in GluK2-containing KAR trafficking in neurons. Moreover, phosphorylation of S868 on the GluK2 C-terminus by PKC in response to agonist stimulation leads to increased SUMOylation at K886 and GluK2 endocytosis. The aim of my work was to investigate the potential role of SENP3 in KAR stability as well as different functions of SUMO-1 and SUMO-2/3 in KAR trafficking.
I performed co-immunoprecipitation experiments to show that SENP3 interacts with the GluK2 subunit of KARs. Furthermore, I showed that phosphomimetic mutant S868D and WT GluK2

are SUMOylated with both SUMO-1 and SUMO-2/3 and that SENP3 is able to deSUMOylate WT GluK2 but has no effect on the phosphomimetic SUMO-1ylated GluK2. I am currently investing how SENP3 affects GluK2 stability, by using cycloheximide and MG132. My work has identified a novel role of SENP3 in GluK2 trafficking in neurons under basal conditions and suggests that different SUMO paralogs may have varying effects on GluK2.

Disclosures: K. Talandyte: None. **J. Henley:** None. **K. Wilkinson:** None. **R. Seager:** None.

Poster

P188. Systemic Factors in Brain Aging and Cognitive Decline

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P188.01

Topic: C.01. Brain Wellness and Aging

Title: Rates of amyloid deposition indicate widespread simultaneous accumulation throughout the brain

Authors: *M. R. LAPOINT[1], S. L. BAKER[2], S. M. LANDAU[3], T. M. HARRISON[1], W. J. JAGUST[3];
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Abstract: β-Amyloid (Aβ) plaques are a hallmark pathology of Alzheimer's disease (AD). Aβ begins to aggregate long before cognitive deficits. However, the progression of amyloid throughout the brain is not well understood. Using Pittsburgh Compound B (PIB)-PET imaging, we investigated the progression of regional Aβ accumulation in cognitively normal older adults (N = 195, mean age = 75.5 years). In cross-sectional analyses, elevated amyloid was found most often in frontal and parietal regions, and least often in occipital, medial temporal, and sensorimotor regions. Overall, however, individuals typically showed deposition in all or most regions or low levels throughout the brain. In a subset analysis of participants with longitudinal PIB-PET scans (N = 106, mean follow-up time = 4.71 years), we leveraged the well-known "inverted U" characteristic of amyloid-PET accumulation rates, where accumulation is fastest at intermediate levels, and slower at high and low levels of amyloid. By fitting longitudinal PET data to quadratic functions, we found that all examined regions reached their peak accumulation rates 24-28 years after an estimated initiation corresponding to the mean baseline PIB-PET signal in amyloid-negative older adults. We also found that individuals with the apolipoprotein-E (APOE) ε4 allele had faster amyloid accumulation in all brain regions, adjusting for baseline levels of amyloid. Finally, we focused on the association between amyloid accumulation and tau pathology, another hallmark of AD, in a further subset of participants who had a Flortaucipir-PET scan to measure tau (N =88). In all regions excluding the temporal lobe, faster Aβ

accumulation was associated with greater tau burden. The pattern of regions showing higher tau related to greater Aβ accumulation was similar regardless of the amyloid region being investigated. Overall, these results indicate that amyloid accumulates near-simultaneously in most brain regions, and that accumulation is associated with increased genetic risk of AD and higher tau pathology.

Disclosures: M.R. LaPoint: None. **S.L. Baker:** None. **S.M. Landau:** None. **T.M. Harrison:** None. **W.J. Jagust:** None.

Poster

P188. Systemic Factors in Brain Aging and Cognitive Decline

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P188.02

Topic: C.01. Brain Wellness and Aging

Title: Effect of congruency and ambiguity on memory performance across the adult lifespan

Authors: *L. VARGHESE[1], X. CHEN[1,2], W. JAGUST[1,2];
[1]Univ. of California, Berkeley, Berkeley, CA; [2]Lawrence Berkeley Natl. Lab., Berkeley, CA

Abstract: This study examines how stimulus properties, specifically the congruency and ambiguity between an object and a scene in a picture, affect memory of the picture in aging. A total of 273 participants from MTurk (aged 22-70 yrs old) completed a memory task online. They were first presented pictures depicting an object in a scene and judged if the object is likely or unlikely to be in the scene. Following a delay, participants were asked to identify pictures as old or new, with new items comprised of similar lures. Each studied picture was associated with two indices based on participants' likely/unlikely judgment during the study phase: congruency was based on whether participant judged the object-scene pair to be likely (congruent) or unlikely (incongruent); ambiguity was determined by the proportion of participants with a congruency judgment different from the majority, where higher values indicate the pair was perceived more ambiguous. Mixed-effects logistic regressions examined the effect of age, congruency/ambiguity, and their interaction on memory. We found that higher congruency predicted higher hit rates ($p < 0.001$): people were more likely to correctly identify an old image as old if they considered it a congruent pair. But congruency also led to more false memories ($p < 0.001$): people more often incorrectly identified a lure as an old image if they considered the initial stimulus congruent. A significant interaction between congruency and age was found for memory accuracy where older adults were more negatively affected by congruency ($p = 0.02$, Fig 1a). Ambiguity predicted both higher hit and false alarm rates ($p < 0.001$), and overall negatively affected memory accuracy ($p = 0.01$), with similar effects across ages (Fig 1b). In summary, congruency and ambiguity proved to be both harmful and beneficial for memory (high hit rates and high false alarm rates), with an overall negative influence on memory quality.

Interestingly, older people's memory is more influenced by congruency than young adults, likely suggesting that older adults rely more on schema for memory retrieval.

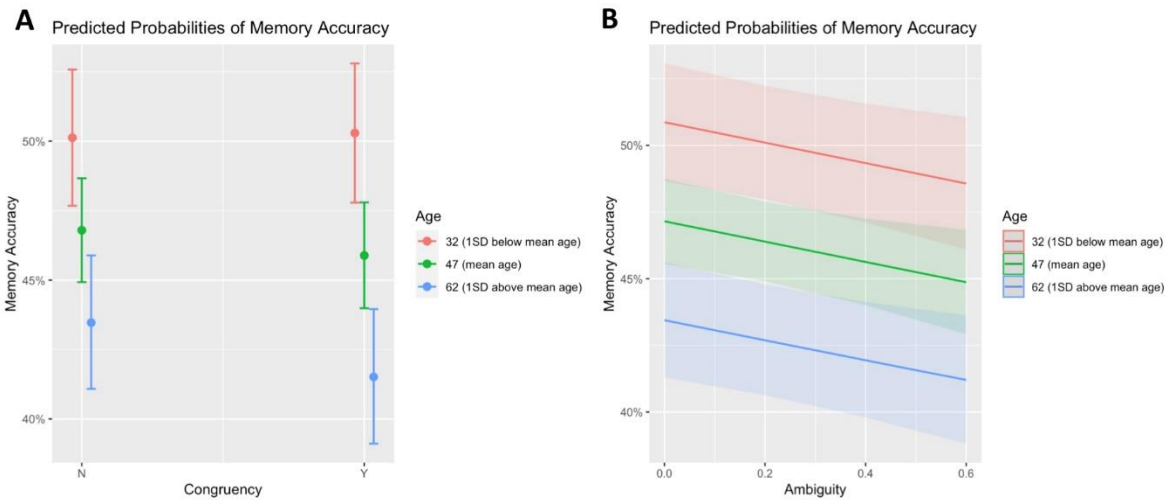


Figure 1: Logistic regression model of the probability of accurately remembering an image according to A) congruency (N=No (incongruent), Y=Yes (congruent)) and age ; B) ambiguity and age

Disclosures: L. Varghese: None. **X. Chen:** None. **W. Jagust:** None.

Poster

P188. Systemic Factors in Brain Aging and Cognitive Decline

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P188.03

Topic: C.01. Brain Wellness and Aging

Support: NIH Grant 2P30AG012839-26

Title: Mechanistic contributions of memory to decision-making impairments in Alzheimer's disease

Authors: *Z. ZHANG[1], S. MABOUDIAN[1], A. J. JACKSON[2], S. NGO[2], M. MANIVANNAN[2], A. S. KAYSER[2,1], M. HSU[1], W. CHIONG[2];
[1]Univ. of California, Berkeley, Berkeley, CA; [2]Univ. of California, San Francisco, San Francisco, CA

Abstract: Life is not a multiple-choice test. Because real-world decisions are often open-ended, with choice options conceived by decision-makers themselves, the quality of decisions may heavily rely on the retrieval of options, as failure to generate promising options limits, and often eliminates, the opportunity for choosing them (Zhang et al., 2021, PNAS). Because of the

demand on memory retrieval, such decisions may pose substantial challenges to patients with Alzheimer's disease (AlzD), the hallmark of which is substantial and worsening memory deficits. Our sample included Alzheimer's disease patients (N = 13), along with healthy controls (N = 16) and behavioral-variant frontotemporal dementia (bvFTD) patients (N = 13) as a dementia control group. The experiment consisted of (1) a semantic fluency condition, where subjects recalled as many items as possible from a category, (2) an internal-menu choice (IMC) condition, where subjects chose their two most preferred items from a category without a menu, and (3) an external-menu choice (EMC) condition, where subjects made the same choice with a predefined menu. The order of fluency and IMC was counterbalanced and EMC was always the last. Of primary interest is choice switches between IMC and EMC in the same category, with the rationale being that memory constraints in IMC would be lifted in EMC due to the provision of a menu. By examining the occurrence and nature of such switches and linking them with performances in fluency, we could characterize the extent and causes of decision impairments in different groups. As expected, Alzheimer's disease patients recalled fewer items in fluency than both control ($p < 0.005$) and bvFTD groups ($p < 0.01$) across categories. Importantly, this difference in memory retrieval capabilities were related to IMC to EMC choice switches: the Alzheimer's disease group made more switches than both bvFTD ($p < 0.05$) and controls ($p < 0.01$), and a significant correlation was observed across all groups between the average number of items recalled in SF and the average choice switches. In parallel, at the item level, we also found that subjects tended to switch to more difficult to recall items. Furthermore, Alzheimer's disease patients made higher memory-driven switches than controls, while bvFTD patients showed more value-driven switches.
These findings provide novel evidence for an overlooked source of decision impairments in Alzheimer's disease and connect models of open-ended decisions with longstanding findings of memory impairments in Alzheimer's disease. They bear implications for new interventions that help improve decision quality in Alzheimer's disease and related dementia.

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Poster

P188. Systemic Factors in Brain Aging and Cognitive Decline

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P188.04

Topic: C.01. Brain Wellness and Aging

Support: NIH Grants: K08AG058749 (KGR), F32AG050434-01A1 (KGR), K23 AG038357 (KAV), P50 AG023501, P01 AG19724 (BLM), P50-AG023501 (BLM & GDR), R01 AG045611 (GDR); AG034570, AG062542 (WJ); NS100440 (SSN), DC176960 (SSN), DC017091 (SSN), AG062196 (SSN)

Larry L. Hillblom Foundation: 2015-A-034-FEL and (KGR); 2019-A-013-SUP (KGR)
Alzheimer's Association: (PCTRB-13-288476)

Title: Abnormal neural oscillations reveal excitatory-inhibitory imbalance are distinctly associated with amyloid and tau depositions in Alzheimer's disease

Authors: *K. RANASINGHE[1], P. VERMA[1], C. CAI[1], X. XIE[2], K. KUDO[3,1], X. GAO[1], H. LERNER[1], D. MIZUIRI[1], A. STORM[1], L. IACCARINO[1], R. LA JOIE[1], B. MILLER[1], M. L. GORNO TEMPINI[1], K. RANKIN[1], W. J. JAGUST[4], K. A. VOSSEL[5], G. RABINOVICI[1], A. RAJ[1], S. S. NAGARAJAN[1];
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Abstract: Excitation-to-inhibition (E/I) imbalance is believed to be a key contributor of synaptic and network degeneration in Alzheimer's disease (AD). Extensive preclinical research on transgenic animal models of AD have demonstrated neuronal and circuit level E/I imbalance mediated by amyloid-beta (Aβ) and abnormally phosphorylated tau proteins. However, the mechanisms of E/I imbalance leading to disrupted networks and their relationships to Aβ and tau in humans remain poorly understood. In this multimodal imaging study in patients with AD (n=20 AD patients; n=35 age-matched controls), we first examined the spectral changes in oscillatory brain rhythms using magnetoencephalography and their relationships to tau and amyloid-beta (Aβ) accumulation in PET imaging (flortaucipir and ^{11}C-PiB, for tau and Aβ, respectively). Next, we estimated parameters of a linear neural mass model that best fit the observed spectra, and examined the relationships between model parameters for excitatory and inhibitory neural sub-populations, and tau and Aβ accumulations. We found that neuronal hypoactivity was associated with tau while hyperactivity was associated with Aβ. We also demonstrated E/I imbalance in patients with AD, depicted as abnormal excitatory and inhibitory neuronal parameters in a linear neural mass model that reproduced the empirical macroscopic power spectra (Figure 1). The abnormal excitatory and inhibitory neuronal parameters showed distinct associations with tau and Aβ—higher tau correlated with a longer excitatory time-constant, whereas higher Aβ correlated with a longer inhibitory time-constant. The current study demonstrates how abnormalities in the oscillatory power spectrum in AD is mechanistically linked to impaired function of excitatory and inhibitory neuronal populations that are in turn associated with tau and Aβ accumulations. The multimodal neuroimaging in AD patients in the current study demonstrate how different frequency oscillations are distinctly affected by Aβ and tau, and offer new perspectives for network stabilizing therapies.

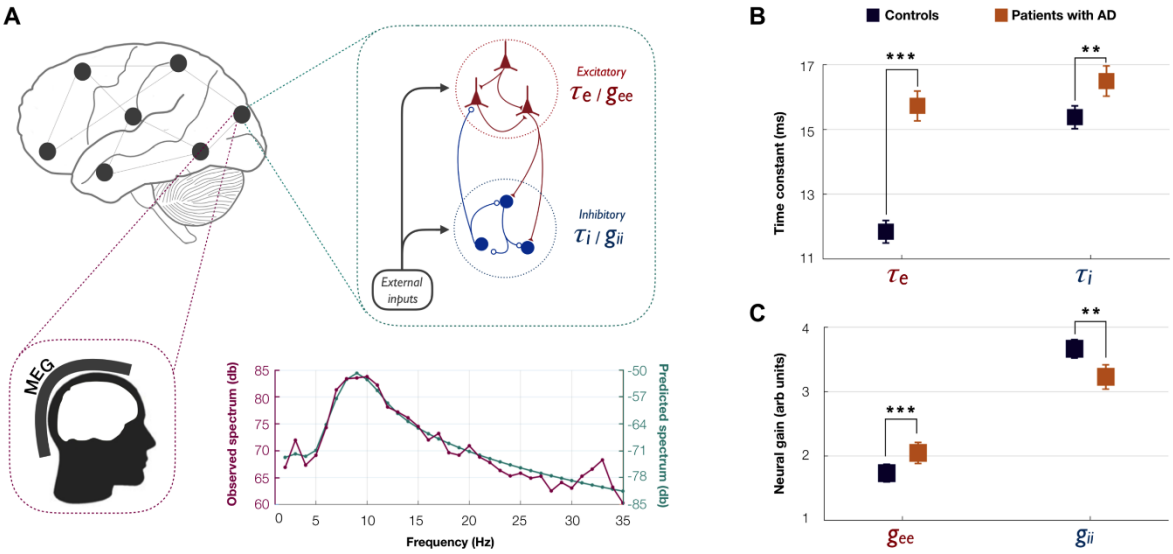


FIGURE 1
Linear neural mass model and its predictions of excitatory and inhibitory neuronal deficits in patients with AD. Schematic representation of the linear neural mass model and an example model prediction (A). Spectral graph model represents the local assemblies of excitatory and inhibitory neurons into lumped linear systems, at each region-of-interest (ROI). External inputs and outputs are gated through both excitatory and inhibitory neurons. The recurrent architecture of the two pools within a local area is captured by the neuronal time constants, τ_e and τ_i , and neural gain terms, g_{ee} and g_{ii}, indicating the loops created by recurrents within excitatory, inhibitory and cross-populations. At each ROI, the model delivers these parameters as it predicts the broad-band spectrum (1-35 Hz) optimized to the empirical spectrum derived from MEG. Patients with AD showed significantly increased neuronal time constants, τ_e and τ_i compared to age-matched controls (B). Patients with AD also showed increased excitatory neural gain (g_{ee}) and reduced inhibitory neural gain (g_{ee}) than controls (C). The markers and error-bars depict the least-square means and 95% confidence intervals. Abbreviations: AD, Alzheimer's disease; MEG, magnetoencephalography.

Disclosures: K. Ranasinghe: None. **P. Verma:** None. **C. Cai:** None. **X. Xie:** None. **K. Kudo:** None. **X. Gao:** None. **H. Lerner:** None. **D. Mizuiri:** None. **A. Storm:** None. **L. Iaccarino:** None. **R. La Joie:** None. **B. Miller:** None. **M.L. Gorno Tempini:** None. **K. Rankin:** None. **W.J. Jagust:** None. **K.A. Vossel:** None. **G. Rabinovici:** None. **A. Raj:** None. **S.S. Nagarajan:** None.

Poster

P188. Systemic Factors in Brain Aging and Cognitive Decline

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P188.05

Topic: C.01. Brain Wellness and Aging

Support: NIH Grant 1RF1AG071566
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NIH Grant U01 AG024904
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Cleveland Clinic Keep Memory Alive Young Investigator Award
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a private grant from Peter and Angela Dal Pezzo

Title: Sex Modulates the Pathological Age Effect on Caudate Functional Connectivity in Mild Cognitive Impairment

Authors: *Z. YANG[1,2], J. Z. K. CALDWELL[1], J. L. CUMMINGS[2], J. KINNEY[2], D. CORDES[1,2,3];
[1]Cleveland Clin., Las Vegas, NV; [2]Univ. of Nevada Las Vegas, Las Vegas, NV; [3]Univ. of Colorado, Boulder, CO

Abstract: Introduction: Brain aging is characterized by considerable heterogeneity, including the variability induced by demographic factors and symptomatic or presymptomatic pathology, such as mild cognitive impairment (MCI). While episodic memories are established and maintained by an interplay between medial temporal lobe and other cortical regions, the degradation of the frontostriatal system is suggested to be a driving factor of episodic memory decline in old adults, which dismisses a uni-factorial view of neurocognitive aging. In this study, we focused on evaluating whether and how sex mediates the pathological age effect on caudate functional topology in MCI. **Materials and Methods**: 277 functional magnetic resonance imaging (fMRI) sessions from 163 cognitive normal (CN) older adults and 309 fMRI sessions from 139 subjects with MCI were included in our analysis. Pearson's correlation was used to characterize the functional connectivity (FC) between caudate and each brain region, then caudate nodal strength was computed to quantify the overall caudate FC strength. Association analysis between caudate nodal strength and age was carried out in MCI and CN separately using linear mixed effect (LME) model with covariates (education, handedness, sex, Apolipoprotein E4 and intra-subject effect). Then LME model was applied to women and men separately within MCI group to evaluate age effect on caudate nodal strength and each region's FC with caudate. Finally, Pearson's correlation was used to assess the association of neuropsychological scores with age and caudate nodal strength. **Results**: MCI group had significantly stronger age-related increase of caudate nodal strength compared to the CN group. Analyzing women and men separately revealed that the age effect on caudate nodal strength among MCI participants is significant only for women (left: P=6.23x10^{-7}, right: P=3.37x10^{-8}), but not for men (P>0.3 for bilateral caudate). Caudate connectivity with ventral prefrontal cortex substantially contributed to the positive age effect on caudate nodal strength in women with MCI. Both positive and negative age effects on individual region's connectivity with caudate were observed in men with MCI. While both women and men with MCI demonstrated worse cognitive performance with older age, higher caudate nodal strength is significantly related to worse cognitive performance only in women but not in men with MCI. **Conclusion**: Stronger age-related increase of caudate nodal strength in MCI over CN is sex-dependent. Caudate nodal strength may be a sensitive biomarker of pathological aging in women with MCI.

Disclosures: Z. Yang: None. **J.Z.K. Caldwell:** None. **J.L. Cummings:** None. **J. Kinney:** None. **D. Cordes:** None.

Poster

P188. Systemic Factors in Brain Aging and Cognitive Decline

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

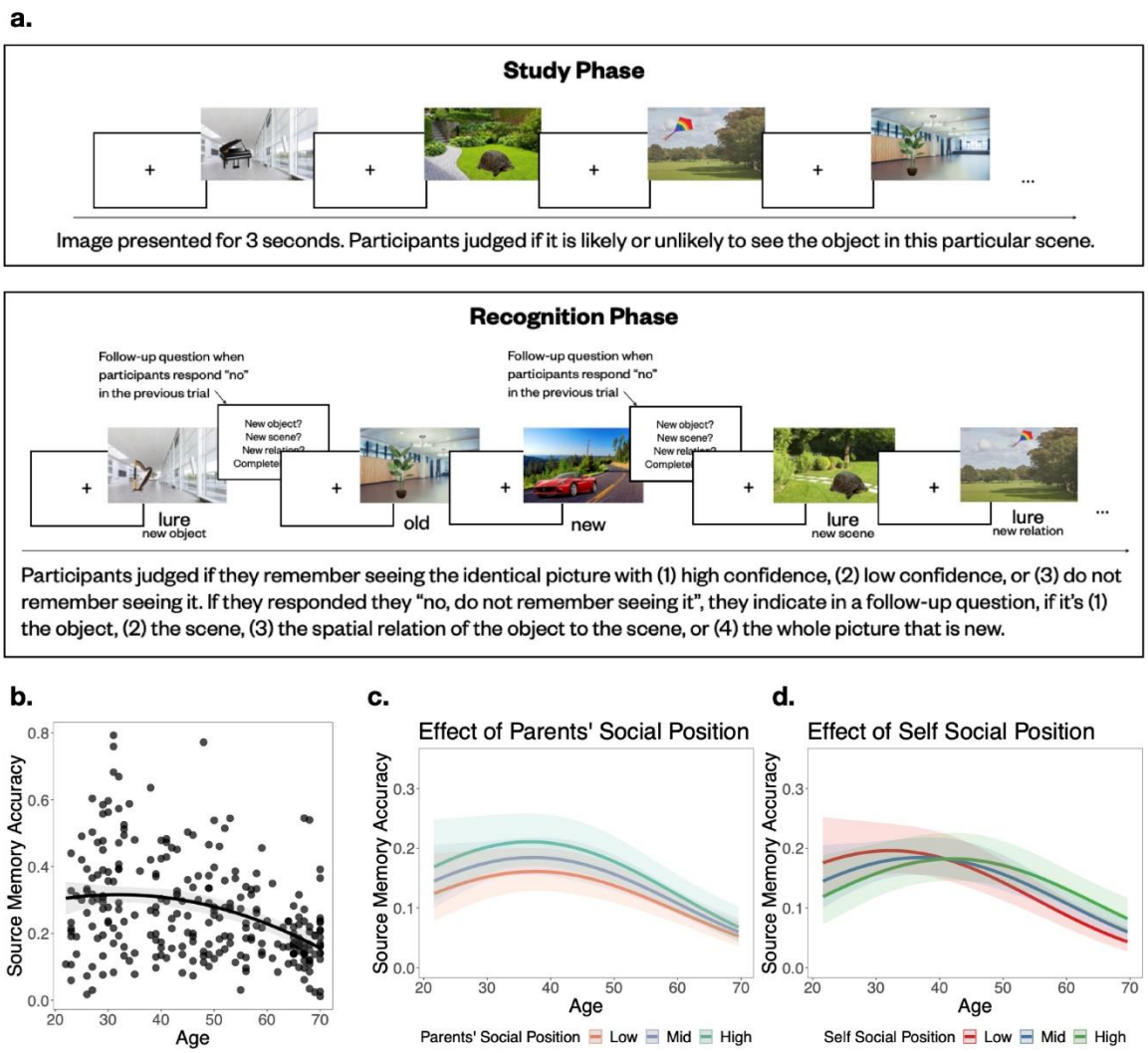
Program #/Poster #: P188.06

Topic: C.01. Brain Wellness and Aging

Title: Advantaged family and higher social position associated with better memory and delayed memory aging separately

Authors: *X. CHEN[1,2], L. VARGHESE[1], W. J. JAGUST[1,2];
[1]Univ. of California, Berkeley, Berkeley, CA; [2]Lawrence Berkeley Natl. Lab., Berkeley, CA

Abstract: Lower socioeconomic status has been shown to have life-long effects on cognition. In the present study, we investigated how social position of one's original family, as well as current social position, may influence an individual's memory ability across the adult lifespan. A total of 271 participants recruited from MTurk (aged 22-70 years old) completed a memory experiment online. During the experiment, they were presented pictures depicting an object in a scene, and later tested for their memory of the pictures. During the recognition phase, if a participant responded that the exact picture was not previously seen, they were asked whether the picture differed by object, scene, spatial relation, or entire picture (Fig 1a). The response accuracy for this question was used to index memory (i.e., source memory accuracy). Participants also reported their highest education and the occupation class of their most recent job. The same information for their original family was also collected. Using the education and occupation information, Hollingshead index of their current social position (self social position) and their original family (parents' social position) were calculated. We used mixed-effects logistic regressions to examine the effect of age, age^2, parents' social position, self social position, and their interactions on source memory accuracy. We found that memory performance decreases with age quadratically: older people exhibited significantly faster memory ability drop with increasing age (Fig 1b). Moreover, higher parents' social position was constantly associated with better memory performance (Fig 1c), whereas higher self social position was associated with a delayed performance drop in older age, which occurred later than those with lower social position (Fig 1d). The findings suggest that an advantaged family social position appears to be associated with an age-invariant benefit for memory across the lifespan; having a higher social position as an adult, on the other hand, may help "slow down" memory aging in later life.



Disclosures: X. Chen: None. **L. Varghese:** None. **W.J. Jagust:** None.

Poster

P188. Systemic Factors in Brain Aging and Cognitive Decline

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P188.07

Topic: C.01. Brain Wellness and Aging

Title: Subjective cognitive self-appraisal correlates with cortical thickness and tauopathy in aging

Authors: *K. ZHUANG[1], X. CHEN[1,2], K. E. CASSADY[1,2], S. L. BAKER[2], W. J. JAGUST[1,2]; [1]Helen Wills Neurosci. Inst., Univ. of California, Berkeley, Berkeley, CA; [2]Mol. Biophysics and Integrated Bioimaging, Lawrence Berkeley Natl. Lab., Berkeley, CA

Abstract: Self-awareness of cognitive decline is often impaired in patients with Alzheimer's Disease (AD). In normal aging, it is also common to observe inaccurate self-appraisal of cognitive ability. Understanding this phenomenon in cognitively healthy older adults (OA), especially those with early AD pathologies, is imperative to early detection of behavioral alterations related to AD. A total of 151 cognitively healthy OA (male = 62, mean age = 76.09) from the Berkeley Aging Cohort Study underwent neuropsychological testing and 1.5T structural MRI. A subset of 66 OA (male = 29, mean age = 78.36) further received amyloid-PET with [11C] PiB and tau-PET with [18F] AV-1451. Additionally, we included longitudinal cognitive data (mean (sd) follow up = 6.95 (3.48) years, 6.68 (3.09) sessions) of all subjects with PET. Self-appraisal rating was recorded after each task to estimate performance relative to peers of the same age, sex, and education. Average appraisal discrepancy was calculated by subtracting objective performance from self-appraisal rating (positive discrepancy = overestimation). We found that worse performers had lower self-appraisal ratings than better performers ($r = .323$, $p < .001$), but they still overestimated their abilities by having higher appraisal discrepancy ($r = -.621$, $p < .001$). Whole-brain vertex wise analyses revealed that greater overestimation was associated with thinner inferior-lateral temporal, fusiform, and rostral anterior cingulate cortices. Multiple linear regression models examined the relationship between amyloid, tau, and self-appraisal measures. Higher entorhinal tau was related to lower self-appraisal ratings regardless of amyloid status ($p = .019$). Higher entorhinal tau was also associated with higher appraisal discrepancy, or overestimation, in amyloid negative OA ($p = .018$). In addition, linear mixed effect models were used to explore longitudinal effects of biomarkers and objective performance on changes in self-appraisal measures and showed no significant longitudinal effects. In summary, the behavioral results were consistent with the previously described Dunning-Kruger effect. OA with AD pathology had lower self-appraisal ratings, consistent with the suggestion of subjective cognitive decline prior to objective deficit in AD. Additionally, performance overestimation was related to greater atrophy and higher entorhinal tau in OA with low amyloid. These results suggest that overestimation could implicate structural differences in medial and temporal regions in OA and that reduced performance appraisal could be an early indicator of AD pathology.

Disclosures: K. Zhuang: None. **X. Chen:** None. **K.E. Cassady:** None. **S.L. Baker:** None. **W.J. Jagust:** None.

Poster

P188. Systemic Factors in Brain Aging and Cognitive Decline

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P188.08

Topic: C.01. Brain Wellness and Aging

Support: NIH Grant RF1AG060754

Title: Deep brain stimulation of the cholinergic basal forebrain improves cognition in senescent male monkeys

Authors: K. R. PENNINGTON[1], C. BANNERJEE[3], F. VALE DIAZ[3], Y. ZHENG[5], A. V. TERRY, JR[4], J. CHIN[6], C. CONSTANTINIDIS[7], *D. BLAKE[2];
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Abstract: Prior work has documented short and long term improvements in higher cognitive function caused by deep brain stimulation of the cholinergic basal forebrain. In this work, we explored whether these findings extended to two senescent male Rhesus monkeys. Animals were pre-trained in on-cage touchscreen behaviors that tested working memory and sustained attention. Then, animals were implanted with electrodes in the nucleus basalis of Meynert targeting the region 2 mm posterior to the anterior commissure. Electrodes were coupled to wire leads and tunneled to an implantable pulse generator. The working memory behavior was a two AFC color discrimination, and delay length was set each day using adaptive tracking. Over 9-12 weeks of unilateral stimulation, working memory delays increased two to four-fold. Over the following 12 week period with no stimulation, working memory delays continued to be similarly elevated. Sustained attention was tested before and after, but not during, the first stimulation period. The behavior followed a continuous performance task design in which animals were rewarded for touching each white square in a stream of squares of difference colors. Hit rates and errors of commission both increased, with the balance favoring improved detection. Brains were processed post-mortem for histological reconstruction of stimulation site, and protein analysis from the stimulated vs the unstimulated cortical hemispheres. The results demonstrate that deep brain stimulation of the basal forebrain can cause improvements in working memory and sustained attention in aged male macaques, and that these changes are sustained for at least months after cessation. Senescent female monkeys are entered in a parallel study.

Disclosures: K.R. Pennington: None. **C. Bannerjee:** None. **F. Vale Diaz:** None. **Y. Zheng:** None. **A.V. Terry:** None. **J. Chin:** None. **C. Constantinidis:** None. **D. Blake:** C. Other Research Support (receipt of drugs, supplies, equipment or other in-kind support); Boston Scientific provides DBS devices via a PPP agreement.

Poster

P188. Systemic Factors in Brain Aging and Cognitive Decline

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P188.09

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Title: Alterations in resting-state functional connectivity in individuals with HIV: Parallels with aging

Authors: *M. C. MCCUSKER[1,2], B. J. LEW[3], J. O'NEILL[3], B. MORSEY[3], S. SWINDELLS[3], H. S. FOX[3], T. W. WILSON[2,3], G. E. DOUCET[2];
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Abstract: The increasing incidence of age-related comorbidities in people with HIV (PWH) has led to accelerated aging theories. Functional neuroimaging research, including functional connectivity (FC) using resting-state functional magnetic resonance imaging (rs-fMRI), has identified neural aberrations related to HIV infection. Yet little is known about the relationship between aging and resting-state FC in PWH. This study enrolled 121 PWH and 133 demographically matched controls spanning 22-72 years old who underwent rs-fMRI. We computed FC within and between the major resting-state networks in each sample. ANCOVA models were used to identify independent and interactive effects of HIV and aging. We also performed network-based statistical (NBS) analyses to characterize group differences in FC between multiple regions (n = 512). We found independent effects of age and HIV in between-network FC. The age-related increases were widespread, while the PWH displayed further increases above and beyond aging, specifically between the default-mode (DMN), executive control (ECN), and limbic networks. The NBS analyses showed similar results: PWH showed a significant reduction in FC primarily within the DMN, especially in the frontal and parietal regions, but also between the DMN and ECN irrespective of aging compared to uninfected controls. Meanwhile, PWH showed significant increases in FC between the DMN and ECN, especially in the frontal lobes, irrespective of aging relative to uninfected controls. Since both HIV infection and aging are associated with independent increases in between-network FC, HIV infection may be associated with a reorganization of the major brain networks and their functional interactions in a manner similar to aging.

Disclosures: M.C. McCusker: None. **B.J. Lew:** None. **J. O'Neill:** None. **B. Morsey:** None. **S. Swindells:** None. **H.S. Fox:** None. **T.W. Wilson:** None. **G.E. Doucet:** None.

Poster

P250. Parkinson's Disease: Cellular Mechanisms II

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Program #/Poster #: P250.01

Topic: C.03. Parkinson's Disease

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Title: Transcriptomic changes linked to age-dependent neuromelanin accumulation in a new Parkinson's disease mouse model

Authors: *N. PEÑUELAS[1], A. LAGUNA[1], H. XICOY[1], E. MARKIDI[1], M. GONZALEZ-SEPULVEDA[1], A. NICOLAU[1], M. LORENTE-PICÓN[1], J. COMPTE[1], A. PARENT[1], T. CUADROS[1], J. ROMERO-GIMÉNEZ[1], L. MIQUEL-RIO[2], G. PUJOL[1], B. RODRÍGUEZ-GALVÁN[1], L. GIMÉNEZ-LLORT[3], A. BORTOLOZZI[2], I. CARBALLO-CARBAJAL[1], M. VILA[1];
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Abstract: In Parkinson's disease (PD) there is a preferential degeneration of neuromelanin (NM)-containing neurons, especially neurons from the Substantia Nigra (SN) but also from the Ventral Tegmental Area (VTA) and Locus Coeruleus (LC). We generated a new NM-producing mouse model, based on the tissue-specific constitutive expression of human tyrosinase (hTyr) under the tyrosine hydroxylase (TH) promoter (tgNM), that mimics the distribution and age-dependent accumulation of NM in the human brain (i.e. catecholaminergic groups A1-A14). TgNM mice exhibited major PD features, including both motor and non-motor behavioral alterations, inclusion body formation, neuronal degeneration in lower brainstem areas (LC) together with neuronal dysfunction in higher brainstem areas (SN and VTA). In order to understand the mechanisms by which NM accumulation in specific brain areas ultimately interferes with the normal functioning of cells, we characterized genome-wide transcriptomic changes linked to the intracellular presence and progressive accumulation of NM in two NM-accumulating neuronal subpopulations (SN and VTA) that are known to be differentially susceptible to PD pathology. We selectively isolated single dopaminergic NM-containing neurons by laser capture microdissection from male and female wild-type and tgNM animals at 3 months, 12 months and 20 months of age (n=4-6 mice per group). We performed differential expression analysis, resulting in statistically significant differentially expressed genes at all ages (p-value<0.01 and $\log_2$FC><0.5). Gene-set enrichment analysis (GSEA) with Reactome Pathway Database led to the identification of altered biological pathways in tgNM related to

neuroinflammation, vesicle-mediated transport and lipid metabolism, transcription and translation, mitochondrial function and cell cycle (senescence) (False Discovery Rate<0.05). Targeted-based validation of candidate RNA species was performed in microdissected samples by quantitative real-time PCR and candidate biological pathways were validated at the protein level by western blot in dissected ventral midbrain tissues from biological replicates. The transcriptomic profiles identified in this project contribute to our understanding of selective vulnerability in PD and brain aging, and points to key biological pathways and molecular targets in prodromal and early PD.

Disclosures: N. Peñuelas: None. **A. Laguna:** None. **H. Xicoy:** None. **E. Markidi:** None. **M. Gonzalez-Sepulveda:** None. **A. Nicolau:** None. **M. Lorente-Picón:** None. **J. Compte:** None. **A. Parent:** None. **T. Cuadros:** None. **J. Romero-Giménez:** None. **L. Miquel-Rio:** None. **G. Pujol:** None. **B. Rodríguez-Galván:** None. **L. Giménez-Llort:** None. **A. Bortolozzi:** None. **I. Carballo-Carbajal:** None. **M. Vila:** None.

Poster

P250. Parkinson's Disease: Cellular Mechanisms II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P250.02

Topic: C.03. Parkinson’s Disease

Title: Pak6 regulates tfeb to enhance autophagy in neurons

Authors: *F. AGOSTINI[1], N. PLOTEGHER[1], R. AGOSTINIS[2], F. DE LAZZARI[1], S. COGO[3], M. BISAGLIA[1], E. GREGGIO[1];
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Abstract: Autophagy is a degradation and recycling mechanism which is essential for maintaining cellhomeostasis. This process is mainly controlled by the activity of the transcription factor EB (TFEB),that modulates the expression of several genes involved in the autophagic-lysosomal machinery.TFEB regulation and subcellular localization are mainly controlled by the binding of 14-3-3 proteins,which prevent its nuclear translocation.Accumulating evidence links autophagy dysregulation to the onset and progression of severalneurodegenerative diseases. Therefore, activation of TFEB and induction of autophagy areconsidered promising therapeutic approaches for the treatment of these pathologies. For thisreason, the major aim of this project is to characterize and validate a novel mechanism to enhanceautophagy and lysosomal function in neurons.Considering that the neuronal-enriched kinase PAK6 has been recently demonstrated to regulatethe interaction of 14-3-3 proteins with their binding partners, we hypothesized that PAK6 mayinduce TFEB nuclear translocation by phosphorylating 14-3-3s and promoting their dissociation fromTFEB.Taking advantage of both *in vitro* and *in vivo* models we assessed the

effect of PAK6 kinase activityon TFEB. First, we revealed that cells transfected with a hyperactive mutant form of PAK6 displayan increased TFEB nuclear translocation. We then tested whether PAK6 affects autophagy in SH-SY5Y neuroblastoma cells overexpressing wild type PAK6. We observed that PAK6 stimulatesautophagy, as assessed by the analysis of several autophagic markers and the level of TFEB.To explore whether PAK6 can activate autophagy also *in vivo*, we used *Drosophila melanogaster*downregulating *mbt*, the fly orthologue of mammalian PAK6. By performing western blots,lysosomal activity assays and immunofluorescence analysis, we found that neuronal-specificsilencing of *mbt* results in a reduction of autophagic activity and lysosomal function.Taken together, these results show a novel and conserved mechanism by which the kinase activityof PAK6 enhances autophagy/lysosome pathways through the activation of TFEB.Moreover, our data point to PAK6 as a potential key target to regulate autophagy specifically inneurons.

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Poster

P250. Parkinson's Disease: Cellular Mechanisms II

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Topic: C.03. Parkinson’s Disease

Support: PICT-2018-02738
PICT 2015 3687
PICT 2019-02416

Title: Increased D1/D5 receptors to Kv1.3 channel signaling causes striatal cholinergic interneurons hyperexcitability in parkinsonism and dyskinesia

Authors: *C. TUBERT[1], R. M. PAZ[2], A. M. STAHL[3], L. RELA[4], M. G. MURER[5];
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Abstract: Striatal cholinergic interneurons (SCIN) are the main source of striatal ACh. In Parkinson´s disease (PD), dopaminergic neurons that innervate the striatum degenerate, leading to a relative increase of cholinergic function that contributes to the generation of PD symptoms (Barbeau A, 1962, *C Med Ass J*). The gold standard therapy for PD is levodopa (L-dopa) administration, but prolonged treatment may result in adverse effects like dyskinesia (Kalia LV & Lang AE, 2015, *Lancet*). This highlights the need for the development of alternative therapies

for patients with advanced PD. Previously, we found that SCIN are hyperexcitable in a mouse model of PD as a result of a reduction in a current mediated by Kv1.3 channels, which is not explained by reduced channel expression (Tubert C *et al.*, 2016, *Cell rep*). Contrary to expectations, in a recent study we report that L-dopa administration at doses that induce dyskinesia not only does not revert the hyperexcitability observed in SCIN of parkinsonian mice but instead exacerbates the already increased excitability of these cells through undetermined mechanisms likely involving stimulation of the D1/D5 receptors in SCIN (Paz RM *et al*., 2021, *Mov dis*). Our aim is to identify these mechanisms. Because this enhanced hyperexcitability is observed in the off L-dopa medication condition, 24 hs after the end of a chronic L-dopa treatment, we hypothesize that physiologic activation of D1/D5 receptors results in a reduction of Kv1.3 current and a consequent increase in excitability, and that an alteration in D1/D5 signaling contributes to the enhancement of hyperexcitability observed in the off L-dopa medication condition. With *ex-vivo* electrophysiological recordings and classic pharmacology, we found that, in physiological conditions, activation of D1/D5 receptors increases SCIN excitability by reducing Kv1.3 current through a signaling pathway involving cAMP and Erk1/2 kinase. Moreover, in parkinsonian and dyskinetic mice, antagonizing cAMP signaling in SCIN restores normal excitability. Finally, preliminary results suggest that the cAMP pathway overactivity is due to an increased ligand-independent activity of D1/D5 receptors that entails a cAMP-dependent reduction in Kv1.3 current. Our results suggest that the design of new strategies for restoring or preventing the changes that occur in the D1/D5 receptors to Kv1.3 signaling pathway in SCIN may prove beneficial both in PD and in dyskinesia.

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Poster

P250. Parkinson's Disease: Cellular Mechanisms II

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Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P250.04

Topic: C.03. Parkinson’s Disease

Title: Loss of Bmal1 drives cell-autonomous dopaminergic neuron degeneration

Authors: *M. KANAN, P. SHEEHAN, J. PATTERSON, A. G. DAVIS, E. MUSIEK; Washington Univ. Sch. of Med., St. Louis, MO

Abstract: Circadian disturbances are among the most common and disabling non-motor symptoms of Parkinson’s disease (PD), affecting as many as 90% of patients. While disruptions in circadian rhythms are a known symptom of PD, the relationship between molecular timing mechanisms and the dopaminergic neurodegeneration of PD is poorly understood. Previous data has indicated a connection between circadian clock machinery and increased neuronal oxidative

stress, which is a known risk factor for PD-related neurodegeneration. To examine the relationship between circadian clock machinery and health of dopamine containing neurons in the substantia nigra pars compacta (SNpc), we examined the effect of tissue-specific deletion of the critical circadian clock transcription factor BMAL1 (BKO) on dopaminergic neuron degeneration. We performed stereological counts of tyrosine hydroxylase expressing (TH+) cells in the SNpc and found that global inducible BKO mice exhibited a significant reduction in dopaminergic neurons, suggesting Bmal1 may regulate dopaminergic neuron longevity. Global Bmal1 deletion also exacerbated dopaminergic neuronal loss in response to striatal injection of alpha-synuclein fibrils. Neuron-specific BKO mice showed a significant reduction of TH+ cells in the SNpc, while the astrocyte specific BKO mice did not, suggesting a cell-autonomous effect. We are currently examining transcriptional regulation of glutathione transferases and other oxidative stress pathways by BMAL1 as a possible mechanism. These findings suggest that neuronal BMAL1 may directly regulate the homeostasis of dopaminergic neurons, potential through upregulation of oxidative stress gens controlled by Bmal1.

Disclosures: M. Kanan: None. **P. Sheehan:** None. **J. Patterson:** None. **A.G. Davis:** None. **E. Musiek:** None.

Poster

P250. Parkinson's Disease: Cellular Mechanisms II

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Program #/Poster #: P250.05

Topic: C.03. Parkinson’s Disease

Title: Neurotoxic astrocyte secreted-exoDrp1 causes excessive mitochondrial fragmentation and neurodegeneration in Parkinson's disease.

Authors: *B. SEO, H. KO;
Inst. for Cell Engineering, Johns Hopkins Univ. Sch. of Med., Baltimore, MD

Abstract: Emerging evidence suggests that neurotoxic astrocytes play a role in the onset of neurodegenerative disorders including Parkinson’s disease (PD). However, it is still unclear how neurotoxic astrocytes contribute to neurodegeneration in PD. It speculates that toxic factors produced by the neurotoxic astrocytes may play a role in neurotoxicity, yet the toxic factors that mediate neurotoxicity are unknown. In order to identify neurotoxic factors secreted by neurotoxic astrocytes, we conducted mass spectrometry (MS) analysis combined with SILAC (Stable Isotope Labeling with Amino acids in Cell culture) using conditional medium collected from neurotoxic astrocytes (ACM) induced by conditional medium (MCM) collected from α-synuclein preformed fibrils (α-syn PFF)-activated microglia. Intriguingly, dynamin related protein 1 (Drp1) was identified from the MS analysis. Our preliminary studies indicate that Drp1 is secreted from neurotoxic astrocytes via exosomes (**exoDrp1**) and exoDrp1 is uptaken by

neurons. We also found transported exoDrp1 leads to mitochondrial fragmentation and neurotoxicity in neurons. More importantly, exoDrp1 aggravates human α-syn PFF-induced Lewy bodies (LBs) pathology. Based on our preliminary results, we have investigated the contribution of exoDrp1 in human dopaminergic neurodegeneration using both H9-human embryonic stem cell (hESC)-derived midbrain dopaminergic neurons and PD patient's induced pluripotent stem cells (hiPSCs)-derived midbrain dopaminergic neurons. The investigation will provide a new insight into the pathogenesis of PD and new strategy for the treatment of PD.

Disclosures: B. Seo: None. **H. Ko:** None.

Poster

P250. Parkinson's Disease: Cellular Mechanisms II

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Program #/Poster #: P250.06

Topic: C.03. Parkinson's Disease

Support: Canadian Institute of Health Research
Heart and Stroke Foundation of Canada
Ontario Graduate Scholarship
Parkinson's Foundation Summer Student Fellowship

Title: Activating transcription factor-4 mediated inhibition of mTOR signalling results in PUMA/Bax-dependent neuronal apoptosis in models of parkinson's disease

Authors: *M. D. DEMMINGS, K. C. HURLEY, E. TENNYSON, S. P. CREGAN;
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Abstract: Parkinson's Disease (PD) is the second most common neurodegenerative disorder and results from the progressive loss of dopaminergic (DA) neurons in the substantia nigra. The underlying disease pathology is characterized by alpha-synuclein aggregates, which form Lewy body inclusions and increase oxidative and ER stress. Unfortunately, the mechanisms by which DA neurons die remain to be fully understood and current treatments for PD only mitigate some of the clinical symptoms of the disease. Previously we have demonstrated that ATF4, a key mediator of the Integrated Stress Response (ISR), is upregulated in dopaminergic neurons following exposure to MPP+, 6-OHDA, or humanized alpha-synuclein pre-formed fibrils (PFFs). Specifically, we determined that ATF4 induction promotes neuronal death through transcriptional activation of known death genes including pro-apoptotic BH3-only protein PUMA. Interestingly, despite ATF4 being required for PUMA activation, chromatin immunoprecipitation experiments have revealed that ATF4 does not interact directly with the PUMA promoter. Given this, in the current study, we aim to characterize the indirect mechanism by which ATF4 activation signals PUMA-dependent neuron loss. Using mouse primary neurons,

we demonstrate that ectopic ATF4 expression results in activation of Trib3, SESN2, and DDIT4, which have previously been implicated to have inhibitory activity on mTOR. Indeed, ectopic expression of ATF4 in neurons also results in downregulation of mTOR signalling and increases in neuronal apoptosis. Importantly, our current investigations, show that ATF4-deficient neurons are resistant to mTOR inhibition induced by exposure to a-syn PFFs or PD toxins as compared to wild-type littermates. In addition, we describe that pharmacological inhibition of mTOR results in significant increases in PUMA activation and promotes PUMA-dependent dopaminergic neuron loss. Lastly, we demonstrate that both PD toxins and a-syn PFFs activate the PUMA/Bax intrinsic mitochondrial apoptotic pathway as marked by cleaved-caspase activation and subsequent neuronal loss. Ultimately, these results provide a novel signalling mechanism by which ATF4 indirectly promotes PUMA/Bax-mediated cell death through suppression of mTOR signalling and provides further evidence for targeting the ISR for the development of therapeutics for PD.

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Poster

P250. Parkinson's Disease: Cellular Mechanisms II

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Program #/Poster #: P250.07

Topic: C.03. Parkinson’s Disease

Support: 1F30AG066333-01

Title: Defective mitochondria-lysosome contact site dynamics in Parkin PD iPSC-derived dopaminergic neuronal models

Authors: *W. PENG, L. SCHRÖDER, P. SONG, Y. WONG, D. KRAINC;
Northwestern Univ., Chicago, IL

Abstract: Autosomal recessive mutations in in the E3 ubiquitin ligase PARK2 (Parkin) are associated with early-onset Parkinson's disease (PD). Although previous studies have implicated Parkin in the regulation of mitochondrial homeostasis via mitophagy, whether Parkin alters additional forms of mitochondria-lysosome interaction, and how loss-of-function mutations in Parkin lead to neurodegeneration remain incompletely understood. Here, we show that loss-of-function mutations in Parkin alter mitochondria-lysosome contact site dynamics in multiple cell types independent of mitophagy. Parkin regulation of mitochondria-lysosome contacts is dependent on its ubiquitination activity and occurs via stabilization of active, GTP-bound Rab7. Importantly, these defective mitochondria-lysosome contact site dynamics are recapitulated in Parkin PD patient iPSC-derived dopaminergic neurons, and can be rescued by modulating levels

of mitochondria-lysosome contact site tethering proteins. These findings highlight a novel role of Parkin in mitochondrial and lysosomal homeostasis and suggests Parkin regulation of mitochondria-lysosome contact site dynamics as an important pathway in both cellular homeostasis and disease.

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Poster

P250. Parkinson's Disease: Cellular Mechanisms II

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Program #/Poster #: P250.08

Topic: C.03. Parkinson’s Disease

Title: Deletion of Lag3 in the Microglia Prevents Neuropathology in Mouse Model of Parkinson Disease

Authors: *X. YANG[1], S. HA[1], H. LIU[2], N. WANG[2], S.-U. KANG[2], T. DAWSON[3], V. DAWSON[3], X. MAO[3];
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Abstract: Parkinson's disease (PD), the accumulation of aggregated α-synuclein (α-syn) is the typicalpathological hallmark disease. Substantial evidence has indicated that pathological α-syn canspread from cell to cell. The neuroinflammation induced by the spreading of pathological α-synsubsequently leads to progressive neurodegeneration and behavioral deficits. Previously, wehave identified a major receptor of the neuron that mediates the spreading of pathogenic α-synperformed fibrils (PFF), named lymphocyte activation gene 3 (Lag3) (Mao et al., Science. 2016).In the neuron, Lag3 mediates the endocytosis of pathogenic α-syn and contributes to theneuron-to-neuron transmission. Based on the previous study, we generated the Lag3 neuronalconditional knock out (LL-Nestin-Cre) mice (Zhang et al., Sci. Immunol. 2017), and found thatthe pathogenic α-syn transmission was reduced. The role of Lag3 in the neuron is wellestablished, however, the underlying mechanism of how α-syn pathology in the microgliainduces neuroinflammation is not clear. In our current study, we have explored the role of Lag3in microglia mediated neuroinflammation. To study the role of Lag3 in microglia, we havegenerated the microglial Lag3 conditioned knockout mice (LL-CX3CR1-ERT-Cre). In these mice,we evaluated how the absence of the Lag3 receptor in the microglia influence PD pathology.We performed intra-striatal injection of α-syn PFF in these mice and evaluated diseasepathology at varying time intervals. Our results indicate that deletion of Lag3 in the microgliaprevents α-syn mediated neuropathology abnormalities. Moreover, deletion of Lag3 in microgliaresulted in significant rescue of motor deficit compared to wild type (WT) mice. To

understandthe disease mechanisms, we performed our in-vitro experiments in the primary microglia culturefrom WT vs Lag3 KO mice. In-vitro study results show that deletion of Lag3 in the microgliainhibit release of pro-inflammatory cytokines and thereby reduce the reactive astrocytes.Furthermore, when the neurons are exposed with the conditional media from the stimulated glia
cells, it leads to significant attenuation of neurotoxicity. Overall, this study demonstrates theimportant role of Lag3 in the microglia. Targeting of microglia Lag3 could have beneficial effectsand open a new therapeutic avenue for neurodegenerative diseases.

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Poster

P250. Parkinson's Disease: Cellular Mechanisms II

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Program #/Poster #: P250.09

Topic: C.03. Parkinson’s Disease

Title: The binding of L-DOPA to the siderophore-binding protein Lipocalin-2

Authors: *S. ALHASSEN[1], A. ALACHKAR[3], M. KHOUDARI[5], M. SENEL[5], O. CIVELLI[4], W. S. ALHASSEN[2];
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Abstract: Parkinson’s disease (PD) is a neurodegenerative disease that is characterized by the degradation of dopaminergic neurons in the substantia nigra pars compacta causing tremors, rigidity, bradykinesia, and akinesia. Most strategies for treating PD largely depend on the replacement of lost dopamine. The gold standard for treating PD symptomatically is through levodopa (L-DOPA), which is the precursor to dopamine, however, this treatment has many issues, mainly the effectiveness of the treatment over time and the dyskinesia that may develop over chronic L-DOPA treatment. Lipocalin 2 (Lcn2) is a protein that’s function is to bind to siderophores, which are small iron-chelating compounds. By sequestering iron, Lcn2 prevents its use by bacteria and thus limits its growth. L-DOPA has also been shown to bind to iron through its catechol structure. We hypothesize that Lcn2 may play a role in PD and chronic L-DOPA treatment by transporting any excess L-DOPA that has bind to iron into the cell, which may have damaging effects. Using surface plasmon resonance imaging (SPRi), we can monitor interactions in real time, as well as their specificity, kinetics, and affinity. Through SPRi we can monitor if there is an interaction between Lcn2 and the L-DOPA-iron complex. We were able to show that this interaction does occur and is specific to iron and the catechol structure. Compounds with a similar catechol structure to L-DOPA, such as dopamine, also showed interaction, but to a lesser

degree. Compounds similar in structure to L-DOPA with slight modifications to the catechol structure showed very little to no interaction, suggesting that the catechol structure is necessary for the interaction to occur. Through immunohistochemistry, we stained human embryonic kidney (HEK293) cells that have been treated with Lcn2 and the L-DOPA-iron complex. Using select antibodies for both Lcn2 and L-DOPA, we would be able to see if Lcn2 is able to bring the complex into the cell. Chronic L-DOPA treatment causes tolerance and less effectiveness of the drug, leading to higher dosing of the treatment. As the dosing increases, there is an abundance of free L-DOPA that then binds to iron, which is then brought into the cell via Lcn2. Once in the cell, L-DOPA has a number of toxic and deteriorating effects, which may be the cause of some of the negative side effects of chronic L-DOPA treatment.

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Poster

P306. Blood Flow II

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Title: The effects of locomotion in the sensory cortex of awake APP/PS1 mice

Authors: *B. EYRE, O. SHABIR, L. LEE, J. BERWICK, C. HOWARTH;
Univ. of Sheffield, Sheffield, United Kingdom

Abstract: The brain is an energy-demanding organ, consuming approximately 20% of the body's energy supply[1]. In order to regulate its own energy demands the brain has developed a mechanism, known as neurovascular coupling (NVC). NVC ensures that when neurons are active, there is a rapid increase in cerebral blood flow to that same brain region[2]. Evidence suggests this mechanism is impaired in many neurological disorders, including Alzheimer's disease (AD)[3, 4]. Many studies have relied on anaesthesia to investigate neural activity and the subsequent hemodynamic responses. However, anaesthesia can have profound impacts on NVC - including dampening neural activity and reducing cerebral blood volume (CBV)[5]. Due to this, groups have started to investigate NVC using awake rodents[6, 7]. However, behaviour such as locomotion has the potential to confound sensory-evoked NVC responses. This study aims to examine NVC in an AD mouse model, in addition to examining the effects of short bursts of locomotion on sensory-evoked responses. Adult 9-12m APP/PS1 (B6;C3-

Tg(APPswe,PSEN1dE9)85Dbo/Mmjax) (AD n=8 (7 male, 1 female), WT n= 5 (3 m, 2 f)) and old 16-19m (AD n=4 (2 m, 2 f), WT n=4 (1 m, 3 f)) mice received a thinned window surgery. One-week after surgery animals were habituated to a spherical treadmill where 2-dimensional optical imaging spectroscopy (2D-OIS) was used to measure changes in cerebral hemodynamics, in response to a whisker stimulation and spontaneous trials with no stimulation. The experimenter was blinded from genotype during experiments and analysis. Statistical tests will assess whether there is an effect of the amount and timing (relative to 2s whisker stimulation) of short bursts of locomotion on evoked hemodynamic responses and whether there are any differences in evoked hemodynamic responses when comparing AD and WT mice; both adult and old age mice. Preliminary results suggest that short bursts of locomotion alter the amplitude of sensory-evoked responses, with the timing of locomotion (relative to the whisker stimulation) also altering evoked responses. Additionally, evoked hemodynamic responses appear preserved when comparing AD and WT mice. We suggest that during awake imaging, locomotion should be monitored to ensure results are not confounded by the effects of this behaviour. 1. Zlokovic, BV. (2008) Neuron 57, 178-201. 2. Attwell, D. et al. (2010) Nature 468, 232-243. 3. Kisler, K. et al. (2017) Nat Rev Neurosci 18, 419-434. 4. Tarantini, S. et al. (2017). Exp. Gerontol. 94, 52-58. 5. Gao, YR. et al. (2017). Neuroimage, 153, 382-398. 6. Huo, BX. et al. (2014) Journal of Neuroscience, 34, 10975-10981. 7. Tran, CHT. et al. (2018). Neuron, 100, 1133-1148.

Disclosures: B. Eyre: None. **O. Shabir:** None. **L. Lee:** None. **J. Berwick:** None. **C. Howarth:** None.

Poster

P306. Blood Flow II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P306.02

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Title: A Circuit Model of the Circle of Willis and its Clinical Applications

Authors: D. ANANDAYUVARAJ[1], *R. BAIG[2], E. PONTE[2], ***B. R. SHETH**[1];
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Abstract: Our current goal is to model blood flow in the Circle of Willis (CoW) in humans and leverage the model to pinpoint structural weaknesses in individual arteries of the CoW. The CoW is a circulatory anastomosis of several arteries at the bottom (inferior) side of the brain of reptiles, birds, mammals including humans that supplies oxygenated blood to over 80% of the brain. Computational fluid dynamics models of blood flow have investigated structure of arterial segments and junctions, turbulent flow and resulting stagnation. However, these models are not scalable to arterial networks, in part because of their computational complexity, and are thus limited in their clinical applicability. Sidestepping the above critique, we designed a zeroth-order

electrical circuit model of the CoW. Individual arteries of the CoW were modeled as resistors, with the internal carotid artery (ICA) and basilar artery modeled as inlets to the CoW, and the remaining arteries serving as outlets leaving to the rest of the brain. Difference in blood pressure along individual arteries was modeled using Poiseuille's Law, with experimental measurements of diameter and length of human arteries obtained from sources (Iqbal et al. 2013, Hillen et al. 1986, and five others) informing the circuit model. The range of anatomical dimensions reported in human studies was used to constrain randomly generated 100 million+ models (this further allowed us to model individual variation across the population). The flow (current) solution to each individual model was then computed using theorems from circuit analysis based on Kirchhoff's Current Law (KCL) and Kirchhoff's Voltage Law (KVL), and compared with empirical data on blood flow from the literature (e.g. Zarinkoob et al., 2015). We chose to analyze further only those models whose currents (flow rates) fell within the range of human variation for all CoW arteries. In particular, we modeled three of the most common variations in the CoW for which experimental flow data are available – a complete CoW, which is observed in one-half of the population, and two other variations, which together account for nearly 90% of the total variation. A preliminary regression analysis showed that the ICA, the Middle Cerebral Artery, Posterior Cerebral Arteries 1 & 2 had flow rates higher than expected across the distribution of ~1950 filtered models of the complete CoW, implying higher levels of shear stress in these arteries and likely sites for future aneurysms and stroke. Our work thus far provides a glimpse into clinical insights that can be gleaned from simpler models of blood flow. The modeling is being extended to more complicated networks and non-stationary blood flows.

Disclosures: D. Anandayuvaraj: None. **R. Baig:** None. **E. Ponte:** None. **B.R. Sheth:** None.

Poster

P306. Blood Flow II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P306.03

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIH Grant RF1 NS110054
NIH Grant R00 AG059893
Alzheimer's Association 2019-AARG-641299

Title: Spontaneous vasomotion propagates along cortical arterioles in the mouse brain with similar properties as sensory-evoked vascular reactivity

Authors: *L. P. MUNTING[1], M. G. KOZBERG[1], C. A. AUGER[1], O. BONNAR[1], L. HIRSCHLER[1,2], S. S. HOU[1], S. M. GREENBERG[1], B. J. BACSKAI[1], S. J. VAN VELUW[1]; [1]Massachusetts Gen. Hosp., BOSTON, MA; [2]Leiden Univ. Med. Ctr., Leiden, Netherlands

Abstract: Sensory stimulation, such as visual stimulation, evokes a local, vasodilation-mediated blood flow increase to the activated brain region, a process referred to as neurovascular coupling (NVC). Spontaneous vasomotion is a change in arteriolar diameter that occurs in the absence of sensory stimulation, at low frequency (~0.1 Hz) and has been suggested to be neuronally entrained (Mateo *et al.*, Neuron 2017). These slow fluctuations in vessel diameter are a driving force for paravascular soluble waste clearance (van Veluw *et al.*, Neuron 2020), the failure of which has been implicated in neurodegenerative disease. NVC-mediated vascular reactivity has been shown to propagate from capillaries along penetrating arterioles to surface arterioles, but it is unclear whether spontaneous vasomotion shows a similar propagation pattern. We therefore imaged arterioles in the visual cortex in 7 awake, head-fixed, male, C57Bl/6J mice (4-5 months old) with 2-photon microscopy over 3 imaging sessions per mouse. In each session, 5-minute time-lapses were acquired at rest and during visual stimulation (at frequencies between 0.025 and 0.1 Hz) to capture both spontaneous and visually evoked vessel diameter changes respectively. 15 µm-spaced regions of interest (ROIs) were drawn along the length of arterioles - on average 18 ROIs per vessel - in every dataset to extract local vessel diameter changes over time. After demeaning, detrending and low-pass filtering (<0.2 Hz), time profiles in each ROI were cross-correlated to the time profile in a reference ROI to determine if the local diameter change was leading or lagging that of the reference ROI. Linear regression was performed through the retrieved leads/lags to determine if there was a significant diameter change propagation (i.e., a wave) along the arteriole. Both at rest and during sensory stimulation, approximately half of the arterioles showed significant propagation of vessel diameter changes (46 % of the vessels during resting state, 59 % during visual stimulation). Evoked changes in vessel diameter were 29 % higher in amplitude than spontaneous changes ($p < 0.001$, Mann Whitney U), but wave detection did not correlate with the magnitude of the diameter change. The mean (± standard deviation) wave speed was 221 (± 102) µm/sec at rest and 253 (± 131) µm/sec during visual stimulation, and in both conditions, ~75 % of the waves travelled upstream. In summary, these results indicate that spontaneous vasomotion, like sensory-evoked vascular reactivity, propagates along cortical arterioles. This likely plays a role in paravascular brain clearance and supports the hypothesis that vasomotion is neuronally entrained.

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Poster

P306. Blood Flow II

Location: Virtual Only

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Program #/Poster #: P306.04

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIH Grant NS110069
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NSF Grant 1539034

Title: Optogenetic control of single-arteriole diameter in three dimensions in vivo

Authors: *P. O'HERRON[1], D. A. HARTMANN[2], K. XIE[1], P. KARA[3], A. Y. SHIH[4]; [1]Physiol., Augusta Univ., Augusta, GA; [2]Neurol. and Neurolog. Sci., Stanford Univ., Stanford, CA; [3]Univ. of Minnesota, Minneapolis, MN; [4]Ctr. for Developmental Biol. and Regenerative Med., Seattle Children's Res. Inst., Seattle, WA

Abstract: Arteries in the brain dynamically modulate their diameter as part of several crucial vascular functions. Arteries dilate and constrict in response to changes in blood pressure to maintain constant flow levels and to protect the microvasculature from pressure-induced damage. Also, arteries rapidly dilate in a spatially localized fashion when there is an increase in local neural activity - a process called functional hyperemia - which is the basis of hemodynamic imaging techniques like fMRI. Altered vascular function, including impaired functional hyperemia and reduced blood flow, is now seen as a contributing factor in many neurological disorders. Thus, there is a critical need for an accurate understanding of vascular dynamics and of how neural activity interacts with and shapes hemodynamic responses. Typically, studies of vascular function either indirectly activate vessels by driving neural activity (e.g. with sensory stimulation), or use pharmacological agents which can have wide-ranging effects and have poor spatial and temporal resolution. This has limited our ability to separate vascular properties from the actions of the rest of the neurovascular unit in hemodynamic studies. Here we present an experimental paradigm to optically probe the contractile function of arterioles in vivo with high spatiotemporal precision. This was done by expressing the excitatory opsin ReaChR in vascular smooth muscle cells and pericytes. Using a 594 nm light-emitting diode we were able to evoke widespread vasoconstriction across the cranial window. With a 1040 nm focused, pulsed laser for two-photon stimulation, we were able to evoke highly localized constrictions targeted to individual pial artery branches or penetrating arterioles. Our dual light-path imaging system allowed the optogenetic stimulation to be performed with simultaneous two-photon imaging to monitor vessel activity. Using a spatial light modulator, we were also able to constrict vessels both above and below the imaging plane. This is a powerful tool to assay vasoconstrictive function of single arterioles across 3-dimensional vascular networks in vivo. We demonstrate its usefulness for studying the conductance of vascular signals and modulating the dynamics of functional hyperemia.

Disclosures: P. O'Herron: None. **D.A. Hartmann:** None. **K. Xie:** None. **P. Kara:** None. **A.Y. Shih:** None.

Poster

P306. Blood Flow II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P306.05

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIH Grant NS097805
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NIH Grant NS100447

Title: Intranasal treatment with tissue plasminogen activators rescues neuromuscular and cognitive dysfunction triggered by amyloid-ß

Authors: K. UEKAWA, A. ANFRAY, J. SEO, N. CASEY, P. ZHOU, C. IADECOLA, *L. PARK;
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Abstract: Amyloid-β (Aβ) is a key pathogenic factor in Alzheimer's disease and suppress the increase in cerebral blood flow (CBF) evoked by neural activity (functional hyperemia). The protease tissue plasminogen activator (tPA) contributes to the NMDA receptor-dependent component of functional hyperemia (PNAS., 105:1073, 2008), and tPA deficiency is involved in the neurovascular and cognitive dysfunction triggered by Aβ (J Neurosci., 40:8160, 2020). Therefore, we investigated whether intranasal administration of recombinant tPA (rtPA) prevents the detrimental neurovascular and cognitive effects of Aβ in mice overexpressing the amyloid precursor protein with the Swedish mutation (tg2576). Male tg2576 mice and wild-type (WT) littermates (10 mice/group) were instilled intranasally with rtPA [20 µg/day (10µl/nostril at 1 mg/ml concentration); 5 days/week] or vehicle (veh; 10µl dH_2O/nostril) starting at 12 months of age and were examined after 3 months of treatment. In veh-treated tg2576 mice, resting neocortical CBF, measured by ASL-MRI, was attenuated (-25%) compared to veh-treated WT littermates (130±3 vs. 98±6 ml/100g/min; $p<0.05$). To test the effect of rtPA on neurovascular coupling, CBF was recorded by laser-Doppler flowmetry in the somatosensory cortex of urethane-chloralose anesthetized mice. In veh-treated tg2576 mice, the increase in somatosensory cortex CBF produced by neural activity evoked by whisker stimulation was attenuated (-40%) compared to veh-treated WT littermates (19±2% vs. 11±1; $p<0.05$). Similarly, the CBF increase elicited by neocortical application of NMDA (40 µM) was reduced in tg2576 mice (-23%) compared with WT littermates ($p<0.05$). Intranasal rtPA in tg2576 mice rescued the attenuation in resting CBF and functional hyperemia ($p<0.05$ from veh-treated tg2576). Next, we assessed nesting behavior, a sensitive test of executive function in mice. In veh-treated tg2576 mice, the nesting score was reduced compared to veh-treated WT mice (-40%; $p<0.05$). Intranasal rtPA rescued nesting behavior in tg2576 mice ($p>0.05$ from veh-treated WT), effects associated with improved resting CBF and functional hyperemia. The findings indicate that tPA supplementation prevents neurovascular and cognitive dysfunction in aged tg2576 mice and, as such, may be a therapeutic strategy to counteract the detrimental effects of Aβ on neurovascular and cognitive function.

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Poster

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Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Support: NIMH GRANT MH096625
NINDS GRANT NS105584
National NeuroAIDS Tissue Consortium (NNTC) and CHARTER NeuroBioBank

Title: A novel approach to detect and eradicate HIV brain viral reservoirs

Authors: *S. VALDEBENITO, E. EUGENIN;
Neuroscience, Cell Biol. and Anat., Univ. of Texas Med. Br., Galveston, TX

Abstract: The human immunodeficiency virus (HIV-1) compromises the central nervous system (CNS) in the infected population (37 million worldwide), resulting in neurological dysfunction that ranges from minor cognitive impartment to dementia in at least half of the individuals. The major obstacle to eradicate HIV infection is viral reservoirs (VR). The VR is defined as anatomical compartments where the virus is quiescent, and upon the disruption of the antiretroviral therapy (ART), HIV replication is reactivated. It has been demonstrated that a small population of human primary microglia/macrophages and astrocytes in vivo and in vitro survives the infection, silent the virus, and generates bystander damage/inflammation. We identified a unique metabolic profile of these cells to produce ATP from glutamate/glutamine. This is significant because glutamate is already dysregulated in the HIV CNS and the more abundant neurotransmitter in the brain. Thus, VR has an unlimited source of energy to survive for an extended time. First, we used a novel multi-component method (HIV-integrated DNA, mRNA, and viral proteins in the same assay) to identify, quantify and characterize VR in brain tissues, from patients with ART, and primary astrocytes and microglia. Later, we isolate the VR from primary astrocytes and microglia using laser capture microdissection and perform a metabolomics analysis, Seahorse, western blot, and mitochondrial sequence. We correlate our results using imaging mass spectrometry for human HIV-infected brain tissues. Our results show that a small population of astrocytes and microglia are VR in the brain, and they use glutamine as a source of energy. Furthermore, our results from imaging mass spectrometry showed a lipid dysfunction in the HIV-infected brain compare with the healthy controls. In conclusion, we propose that these metabolic changes help the VR survive infection and reveal a novel metabolic target to kill and eradicate HIV brain viral reservoirs.

Disclosures: S. Valdebenito: None. **E. Eugenin:** None.

Poster

P306. Blood Flow II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P306.07

Topic: F.06. Brain Blood Flow, Metabolism, and Homeostasis

Title: Cortical Spreading Depression (CSD) in Mouse Models of Alzheimer's Disease and Atherosclerosis

Authors: *O. SHABIR[1], L. M. T. LEE[2], B. EYRE[2], C. HOWARTH[2], S. B. WHARTON[3], S. E. FRANCIS[1], J. BERWICK[2];
[1]Dept. of Infection, Immunity & Cardiovasc. Dis. (Faculty of Medicine), [2]Dept. of Psychology, [3]Sheffield Inst. for Translational Neurosci., Univ. of Sheffield, Sheffield, United Kingdom

Abstract: Background: Cortical spreading depression (CSD) is physiological process that causes a wave of depolarisation and subsequent vasoconstriction of arterioles in the brain and is associated with migraine with aura, epilepsy and TBI. The aim of this study was to investigate CSD and neurovascular coupling in mouse models of Alzheimer's disease and atherosclerosis.
Methods: 9-12m old male C57BL/6J mice (WT; n=8), J20-hAPP(Sw,Ind) (AD; n=8), rAAV8-mPCSK9-D377Y + Western Diet (ATH; n=6) & J20-hAPP(Sw,Ind) + rAAV8-mPCSK9-D377Y + Western Diet (MIX; n=5) were used. Cerebral haemodynamics were imaged using optical imaging spectroscopy 3-weeks after a chronically recovered thinned window surgery. A terminal acute imaging session was performed with a multichannel electrode was inserted into the brain causing a mild 'pin-prick' of the brain resulting in CSD. Mice were perfused at the end and brains dissected for immunohistochemistry and qPCR studies.
Results: Electrode insertion into the brain resulted in a wave of haemodynamic changes that occurred in all mice. In WT mice, electrode insertion led to a small decrease in HbT followed by a robust HbT bounce back, immediately followed by a small, sustained vasoconstriction (reduced HbT) that persisted for a short duration. In J20-AD mice, electrode insertion caused a large vasoconstriction to occur which spread across the cortex in a strong wave of vasoconstriction that was followed by a very small, attempted recovery. This was masked by a large sustained and prolonged vasoconstriction of contiguous vessels that persisted for some time. The largest vasoconstriction post-CSD occurred in J20-AD mice, followed by PCSK9-ATH mice. A prolonged and sustained constriction below baseline persisted in all mice post-CSD, however, this effect was recovered to baseline in WT mice during the first stimulation experiment 35 minutes after the CSD occurred. In all disease mouse models, the constriction below baseline was more severe and persisted for a much longer time with a slower haemodynamic recovery.
Conclusions: CSD could be an important biomarker of baseline brain health (potentially relating

to levels of neuroinflammation), and insults such as TBI or systemic disease could worsen NVC causing metabolic deficits that could initiate or progress neurological disease. In addition, acute imaging preparations (i.e., electrode insertion or imaging immediately after surgery) have profound consequences on cerebral haemodynamics and this should be considered performing experiments during this phase.

Disclosures: O. Shabir: None. **L.M.T. Lee:** None. **B. Eyre:** None. **C. Howarth:** None. **S.B. Wharton:** None. **S.E. Francis:** None. **J. Berwick:** None.

Poster

P343. Brain Injury: Cellular and Molecular Mechanisms II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P343.01

Topic: C.10. Brain Injury and Trauma

Support: U.S. Department of Defense through the USU-UCSF Partnership: Brain Injury and Disease Prevention, Treatment and Research.
the Center for Neuroscience and Regenerative Medicine Biomarkers Core
the Brain Injury Program of the National Institute for Nursing Research

Title: Progression of Post-traumatic Neurodegeneration Relative to Genetic Modulation of Human Mutant Tau Expression in Mice

Authors: *F. YU[1], D. IACONO[2,3], D. P. PERL[3,4], C. LAI[5], J. GILL[5,4], T. Q. LE[1], G. SUKUMAR[1], P. LEE[3], C. DEVOTO[5], R. C. ARMSTRONG[1,4];
[1]Anatomy, Physiol. and Genet., [2]Neurol., [3]Pathology, [4]Ctr. for Neurosci. and Regenerative Med., Uniformed Services Univ., Bethesda, MD; [5]NINR, NIH, Bethesda, MD

Abstract: Improving diagnosis and treatment of mild-moderate traumatic brain injury (TBI) requires understanding how relatively subtle injury progresses to post-traumatic neurodegeneration with chronic stage atrophy. Tau is mainly localized in myelinated axons in healthy adult brain. The interaction of the tau phenotype with the progression of injury-induced pathology is poorly understood. We used concussive TBI models in male and female mice with 0 (*wt*), 1 (*hemi*), or 2 (*hom*) alleles of the P301S human tau mutation to modulate gene dose-dependent levels of tau expression, hyperphosphorylation, and aggregation. As tau pathology has been associated with repeated concussions, we first examined a repetitive mild TBI (r-mTBI) model of closed head injury, with impacts once per day on five consecutive days. Mice survived for 1 day, 6 weeks or 4 months after r-mTBI or sham procedures. Immunohistochemistry and Simple Western™ protein quantification of the injured brain region confirmed soluble total tau and phosphorylated tau levels were highest in *hom* mice. Simoa™ Neuro-4 multiplex assay analysis of blood samples showed increased serum neurofilament light chain (Nf-L), a marker of

axon damage, at 1 day after r-mTBI as compared to shams. By 4 months after either r-mTBI or sham procedure, *hom* mice had further elevated Nf-L along with increases of total tau. P301S *hemi* mice did not show elevated serum biomarkers in any conditions. Effects from both the injury and tau genotype increased astrogliosis and microglial activation in the corpus callosum. We next examined a single impact moderate TBI (s-TBI) model that produces more extensive axon damage and progressive atrophy of the corpus callosum, which is common in TBI patients with chronic symptoms. Neuropathology demonstrated similar corpus callosum atrophy induced by s-TBI in *wt*, *hemi*, and *hom* P301S genotypes. Neuro-4 multiplex assay analysis showed increased serum Tau and Nf-L at 1 day after s-TBI. By 4 months after either s-TBI or sham procedure, *hom* mice had further elevated serum Nf-L. Quantification of soluble proteins in the injured brain region confirmed increased total tau and phosphorylated tau in *hom* mice, which was not altered in sham vs s-TBI conditions. The combined results from r-mTBI and s-TBI models suggest that both the extent of mutant tau expression and the severity of injury can influence the extent of axon damage and neuroinflammation, but robust tau pathology does not necessarily accelerate TBI induced progression of white matter atrophy.

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Poster

P343. Brain Injury: Cellular and Molecular Mechanisms II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P343.02

Topic: C.10. Brain Injury and Trauma

Support: Mary Tucker Currie Professorship to JWG

Title: Morphine attenuates pain-induced hemorrhage following traumatic brain injury (TBI)

Authors: *G. A. GIDDINGS, P. A. BEAN, M. M. TARBET, D. T. JOHNSTON, J. A. DAVIS, E. T. LOUT, M. K. HENWOOD, J. W. GRAU;
Texas A&M Univ., College Station, TX

Abstract: Traumatic brain injury (TBI) from car accidents, combat violence, and falls, is often accompanied by additional injuries (polytrauma) that serve as a source of pain (nociceptive input). Previous work in our laboratory has shown that nociception is detrimental to recovery after spinal cord injury (SCI) or traumatic brain injury (TBI) and leads to increased lesion site hemorrhage (Turtle, 2019, Exp Neurol, 311, 115). The current study explored the circumstances under which pain input promotes hemorrhage after TBI. Male Sprague-Dawley rats received a moderate unilateral injury in the right frontal region using the Leica One device. In Exp. 1, rats

received an intradermal injection of the irritant capsaicin, or vehicle, to either the contralateral or ipsilateral hindpaw. Because sensory fibers in the spinal cord decussate, if afferent drive (rather than a systemic process) is critical, only contralateral pain should promote hemorrhage. Rats were sacrificed 3 hours after injection and perfused with saline. The extent of hemorrhage across the cortical surface was documented and hemoglobin content was assessed using the Drabkin's assay and immunoblotting. Only animals who received pain input on the contralateral hindpaw showed an increase in hemorrhage ($p < 0.01$). Prior work has shown that noxious electrical stimulation (shock) can foster hemorrhage after SCI and that this effect is not blocked by morphine. Exp. 2 examined whether the same is true after TBI. Rats received a systemic injection of morphine (20 mg/kg, i.p.) or its vehicle a day after the frontal region was injured. Thirty min later, half the animals in each condition received 6 min of shock applied to the tail. Blood pressure was measured. Tissue was collected 3 hr after shock treatment. Morphine produced an acute reduction in blood pressure. Shock did not have a lasting effect on blood pressure. Greater hemorrhage was evident on the cortical surface in vehicle treated shocked rats. Pretreatment with morphine attenuated this effect. Ongoing work is examining the circumstances under which pain input fuels hemorrhage after TBI, how it affects behavioral function, and the neurobiological processes involved.

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Poster

P343. Brain Injury: Cellular and Molecular Mechanisms II

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Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P343.03

Topic: C.10. Brain Injury and Trauma

Support: NIH Grant NS110898
PA Dept of Health SAP 410-007-9710

Title: Glucocorticoid receptor dysfunction in the ventral hippocampus mediates altered stress reactivity following pediatric traumatic brain injury

Authors: D. LENGEL[1], Z. L. ROMM[2], G. M. SMITH[3], N. W. SNYDER[4], J. W. HUH[5], ***R. RAGHUPATHI**[6];
[1]Neurobio. & Anat., Drexel Univ., Philadelphia, PA; [2]Neurobio. and Anat., Drexel Univ. Col. of Med., Philadelphia, PA; [3]Dept of Neurosci., [4]Cancer and Cell. Biol., Temple Univ., Philadelphia, PA; [5]Children's Hosp. of Philadelphia, Philadelphia, PA; [6]Neurobio. and Anat., Drexel Univ. Col. of Med. Neurosci. Program, Philadelphia, PA

Abstract: Children younger than 4 years old are at high risk for sustaining a traumatic brain injury (TBI) and are more likely to suffer psychosocial and behavioral deficits, which may become more prominent as they age into adolescence. We previously reported that TBI in 11-day-old rats resulted in an increase in open arm exploration in the elevated plus maze (EPM) at 4-weeks post-injury. The disruption of glucocorticoid receptor (GR) expression in the hippocampus similarly causes an increase in open arm activity, suggesting that these post-traumatic deficits may be a consequence of altered reactivity to acute stress. In this study, the role of glucocorticoid receptor (GR) function in the dorsal (DH) and ventral hippocampus (VH) in behavioral outcomes in the EPM was assessed at 4-weeks following injury to the 11-day-old rat. Brain injury resulted in a significant increase in open arm exploration in the elevated plus maze in male and female rats, in the absence of overt changes in locomotor activity. Although exposure to the EPM increased corticosterone levels in both sham and brain-injured animals, an increase in the hippocampal expression of the glucocorticoid-inducible gene, serum-and-glucocorticoid inducible kinase 1 (sgk1) mRNA following EPM exposure was only observed in sham-injured animals, suggesting that pediatric TBI may impair GR transcriptional activity. Viral transfection of the human GR (hGR) specifically within the VH reversed the increase in open arm exploration in the elevated plus maze following TBI. In addition, hGR transfection in the VH led to a significant increase in corticotropin releasing hormone (CRH) mRNA expression following exposure to the EPM in brain-injured animals. Together, these findings support an important role of GR function within the VH in the regulation of emotional responses during stress and suggest that impairments in GR function may contribute to the development of altered stress reactivity following pediatric brain trauma.

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Poster

P343. Brain Injury: Cellular and Molecular Mechanisms II

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Program #/Poster #: P343.04

Topic: C.10. Brain Injury and Trauma

Support: R56-NS-089523 (SGK)
R01-NS-095803 (SGK)

Title: Astrocyte-derived ApoE is required for the maturation of injury-induced hippocampal neurons and regulates cognitive recovery after traumatic brain injury.

Authors: *T.-S. YU[1], S. G. KERNIE[2];
[1]Columbia Univ. Med. Ctr., New York, NY; [2]Columbia Univ. Col. of Physicians and Surgeons, New York, NY

Abstract: Polymorphisms in the apolipoprotein E (ApoE) gene confer a major genetic risk for the development of late-onset Alzheimer's disease (AD) and are predictive of outcome following traumatic brain injury (TBI). Alterations in adult hippocampal neurogenesis have long been associated with both the development of AD and recovery following TBI and ApoE is known to play a role in this process. In order to determine how ApoE might influence hippocampal injury-induced neurogenesis, we developed a novel conditional system whereby functional ApoE from astrocytes was ablated just prior to injury. While successfully ablating 90% of astrocytic ApoE just prior to a closed cortical impact injury in mice, we observed an attenuation in the development of newly born neurons using a GFP-expressing retrovirus but not in existing hippocampal neurons visualized with a Golgi stain. Intriguingly, animals with a "double-hit", i.e. injury and ApoE conditionally inactivated in astrocytes, demonstrated the most pronounced impairments in the hippocampal-dependent Morris water maze test, failing to exhibit spatial memory after both acquisition and reversal training trials. In comparison, conditional knockout mice without injury displayed impairments but only in the reversal phase of the test, suggesting accumulative effects of astrocytic ApoE deficiency and traumatic brain injury on AD-like phenotypes. Together, these findings demonstrate that astrocytic ApoE is required for functional injury-induced neurogenesis following traumatic brain injury.

Disclosures: T. Yu: None. **S.G. Kernie:** None.

Poster

P343. Brain Injury: Cellular and Molecular Mechanisms II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P343.05

Topic: C.10. Brain Injury and Trauma

Support: Institutional Support from Northeast Ohio Medical University
Institutional Support from Akron Children's Hospital (Akron, OH)

Title: Characterization of melanopsin ganglion cell morphology in a mouse model of traumatic brain injury

Authors: *S. DEWAN, S. COOK, M. FUSILLO, M. A. SMITH;
Northeast Ohio Med. Univ., Rootstown, OH

Abstract: Traumatic brain injuries (TBIs) are a significant source of morbidity worldwide that can dramatically affect individuals at any point throughout their lifespan. TBIs can manifest in several different ways, but one of the most common is its impact on the visual system. Sleep-wake disturbances are increasingly recognized as a serious and often prolonged consequence in TBI and other chronic neurodegenerations, suggesting involvement of the non-image forming eye-brain pathways. A potential linkage between the occurrence of post-TBI image and non-

image forming visual impairments may reside within a unique subset of retinal neurons: melanopsin-expressing retinal ganglion cells (mRGCs). These retinal neurons link to brain sites responsible for the regulation of the circadian sleep-wake cycle, but also connect to a variety of other subcortical targets. While mRGC structure and function have been reported in other neurodegenerative conditions such as Parkinson's, Alzheimer's, and glaucoma, there is a continued lack of clear understanding regarding if and how mRGCs are affected following brain injury.
Previously, our lab has reported early neuronal signaling deficits accompanied by morphological changes in glaucoma and Alzheimer's mouse models. These deficits occur in RGCs that remain connected to their postsynaptic targets in the brain and may drive later axon and cell body loss. In this study we sought to determine if similar pre-degenerative morphological changes in mRGCs could be observed following traumatic brain injury. To accomplish this task, we used a combination of immunohistochemistry and high-resolution florescent microscopy to isolate mRGCs in the retina and the brain following a controlled cortical impact. Using Neurolucida 360 image analysis software, we measured dendritic field area, dendritic complexity (# of branch points and branches), soma area, and cell density to assess their general morphology and the integrity of mRGCs following injury. Our results indicated significant reduction in dendritic field complexity and area, despite an unaltered cell density in the retina of injured subjects 1-week after injury. These results suggest mRGC morphology is altered as a result of injury, which may impact their function. This could help explain non-image forming sequelae following brain trauma.

Disclosures: S. Dewan: None. **S. Cook:** None. **M. Fusillo:** None. **M.A. Smith:** None.

Poster

P343. Brain Injury: Cellular and Molecular Mechanisms II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P343.06

Topic: C.10. Brain Injury and Trauma

Support: NS110898
PA Dept of Health SAP 410-008-3087

Title: Role of glucocorticoid receptor dysfunction in the dorsal hippocampus in spatial learning deficits following pediatric traumatic brain injury

Authors: *D. LENGEL[1], Z. ROMM[1], N. SNYDER[2], J. W. HUH[4], G. M. SMITH[3], R. RAGHUPATHI[1];
[1]Drexel Univ., Philadelphia, PA; [3]Dept of Neurosci., [2]Temple Univ., Philadelphia, PA; [4]Children's Hosp. of Philadelphia, Philadelphia, PA

Abstract: Traumatic brain injury (TBI) in children younger than 4 years old leads to long-term deficits in cognitive function and learning ability that can persist or even worsen as children age into adolescence. In this study, the role of glucocorticoid receptor (GR) function in the dorsal hippocampus (DH) in chronic hippocampal-dependent cognitive function and synaptic plasticity were assessed following injury to the 11-day-old rat. Brain injury produced significant impairments in spatial learning and memory in the Morris water maze in male and female rats. These deficits were not associated with overt histopathology within the DHC. Brain injury also impaired Long Term Potentiation (LTP) in the CA1 region of the DH in both male and female rats. An assessment of the expression of GRs and glucocorticoid-associated genes in the DH immediately following the first day of water maze training revealed a significant decrease in the expression of the glucocorticoid-inducible gene, serum- and glucocorticoid-kinase 1 (sgk1) in brain-injured animals relative to sham-injured animals, suggestive of an impairment in GR transcriptional activity. Lentiviral transfection of the human GR (hGR) in the DH improved spatial learning and memory in the Morris water maze and also attenuated LTP deficits following TBI. Finally, GR overexpression in the DH resulted in a significant increase in the mRNA expression levels of sgk1, and the glutamate receptor subunits GluA1 and GluN1 following water maze training. Overall, these findings support an important role of dorsal hippocampal GR function in learning and memory deficits following pediatric TBI, and suggest that these effects may be related to the regulation of glutamate receptor subunit expression in the DH.

Disclosures: D. Lengel: None. **Z. Romm:** None. **N. Snyder:** None. **J.W. Huh:** None. **G.M. Smith:** None. **R. Raghupathi:** None.

Poster

P343. Brain Injury: Cellular and Molecular Mechanisms II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P343.07

Topic: C.10. Brain Injury and Trauma

Support: NS1108098

Title: The nicotinic acetylcholine receptor modulator AVL-3288 attenuates hippocampal-based cognitive deficits following repeated mild traumatic brain injury in adolescent rats

Authors: *T. MCCORKLE, R. RAGHUPATHI;
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Abstract: Sports-related concussions (SRC, a subset of mild Traumatic Brain Injury) in adolescents is a leading cause of long-term cognitive deficits. We have developed an age-appropriate and clinically relevant model of SRC in adolescent (35-day old) male and female Sprague Dawley rats by subjecting them to 3 separate episodes of an impact to the intact skull

(2.0mm depth at 5.5m/s velocity) over 7 days. Rats were tested for hippocampal-dependent spatial working memory at 1-and-4 weeks post-injury in the Morris water maze (MWM; female n=7/group 1 week and 4/group 4 weeks; male n=7/group 1 week and 8/group 4 weeks) and at 4 weeks in the novel object location (NOL) tasks (female n=8 uninjured (sham), 11 injured; male n=8/group). At 1 week, only male brain-injured animals exhibited significant deficits in locating the hidden platform in the MWM over the 5-day testing period compared to the male sham rats (sex*injury p=0.0007). However, both male and female brain-injured animals exhibited this deficit at 4 weeks (injury p=0.000002). Similarly at 4 weeks, both male and female brain-injured rats had significant difficulty recognizing the new location of an object they had been familiarized with in an earlier trial (injury p=0.000003). We hypothesized that impaired cholinergic transmission may be a mechanistic basis for these cognitive deficits. Positive allosteric modulation of nicotinic acetylcholine receptors (nAChR) via AVL-3288 was reported to diminish spatial learning deficits in moderately brain-injured adult male rats. Therefore, male and female rats underwent repeated mild injury or sham surgery in adolescence and then were injected with AVL-3288 at 30 minutes prior to testing the animals in either the MWM or NOL tasks at 4 weeks post-injury (female n=6/drug group, n=5/vehicle group; male n=6/group). Treatment with AVL-3288 attenuated injury-induced cognitive deficits in both tasks (treatment p=0.004 MWM, p=0.001 NOL) suggesting that reduced activity of the nAChR may underlie post-traumatic cognitive deficits.

Disclosures: T. McCorkle: None. **R. Raghupathi:** None.

Poster

P343. Brain Injury: Cellular and Molecular Mechanisms II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P343.08

Topic: C.10. Brain Injury and Trauma

Support: DOD Grant PR 130373
NIH Grant NS 085413

Title: Alterations in neuronal calcium activity after traumatic brain injury.

Authors: *P. SAWANT-POKAM[1], J. GAYNES[2], I. ANTHONY[1], K. BRENNAN[1];
[1]Dept. of Neurol., Univ. of Utah, Salt lake city, UT; [2]Univ. of Colorado, Denver, CO

Abstract: Traumatic brain injury (TBI) is increasingly recognized as a source of long-term neurological dysfunction that affects both military and civilian populations, with spreading depolarizations (SD) as known agents of clinical worsening after TBI. SD may also be involved in network plasticity after TBI. Using *in vivo* two-photon microscopy, we compared spontaneous and SD-associated calcium activity in layer 2/3 excitatory neurons after controlled cortical

impact (CCI) TBI in the hindpaw cortex (1-1.5 mm from the edge of a CCI lesion centered on barrel cortex) of Thy1-GCaMP6s mice. 48 hours after injury, neurons demonstrated a significant increase in cell volume, as expected given known edema at this time point, and prolonged but less frequent spontaneous intracellular calcium transients compared to sham animals. When we induced SD 48 hours after CCI, SD-associated calcium transients had a significantly greater area under the curve in TBI compared to sham animals. In contrast to changes in SD calcium transients, fluorescent imaging of extracellular glutamate signaling revealed no difference during SD between CCI and sham, suggesting that glutamate is not implicated in the larger SD calcium load. However, we also observed spontaneous 'plumes' of glutamate signaling in layer 2/3 somatosensory cortex of CCI mice. These events, which are suggestive of basal glutamate dysregulation, were absent in sham mice. Plume events also occurred in sham and CCI mice during SD propagation. The contrast between glutamate and calcium dynamics during SD after TBI suggests calcium-specific dysregulation that may be separable from glutamatergic activity. Yet glutamate plumes at baseline reveal that glutamatergic function is not normal either after TBI. Both phenotypes suggest a network under strain, with apparently separate mechanisms involved in two cardinal pathways of excitability and plasticity. Ongoing work seeks to understand these mechanisms.

Disclosures: P. Sawant-Pokam: None. **J. Gaynes:** None. **I. Anthony:** None. **K. Brennan:** None.

Poster

P343. Brain Injury: Cellular and Molecular Mechanisms II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P343.09

Topic: C.10. Brain Injury and Trauma

Support: RO1 NS117148
RO1 NS111378
RO1 NS 116838

Title: Thyroid hormone reverses cell-type specific pathways and circuits perturbed by traumatic brain injury

Authors: *G. ZHANG[1], N. PHI[1], I. AHN[1], Z. YING[1], G. DIAMANTE[1], N. WANG[1], I. CELY[1], R. LITTMAN[1], F. GOMEZ-PINILLA[1,2], X. YANG[1,3];
[1]Dept. of Integrative Biol. and Physiol., [2]Dept. of Neurosurg., [3]Inst. for Quantitative and Computat. Biosci., UCLA, Los Angeles, CA

Abstract: Traumatic brain injury (TBI) is a tremendous burden around the world affecting civilian, military, and athletes. The etiology of mild traumatic brain injury (mTBI) is complex

due to the tissue and cellular heterogeneity and the disrupted circuits within and between cell populations across the affected brain regions. that underlie cognitive impairments and subsequent neurological disorders. We previously employed single cell RNA sequencing (scRNAseq) to investigate the vulnerable cell types and cell-type specific changes in genes and pathways following mTBI in the hippocampus and frontal cortex, which revealed Ttr, a transporter of thyroid hormone thyroxine (T4), as a potential therapeutic target. Administration of T4 significantly reversed the cognitive impairment caused by mTBI. To understand the molecular targets of T4 treatment, we conducted scRNAseq of hippocampus and frontal cortex at the acute (24hr) and chronic (7days) phases of mTBI with or without T4 treatment at single cell resolution. We found that T4 treatment reversed the genes, pathways, and cell-cell interactions disrupted by mTBI at different timescales with distinct regional patterns. Our results provide a spatiotemporal dynamics of the molecular pathways and circuits underlying the therapeutic potential of T4 to mitigate mTBI induced cognitive and neurological disorders.

Disclosures: G. Zhang: None. **N. Phi:** None. **I. Ahn:** None. **Z. Ying:** None. **G. Diamante:** None. **N. Wang:** None. **I. Cely:** None. **R. Littman:** None. **F. Gomez-Pinilla:** None. **X. Yang:** None.

Poster

P343. Brain Injury: Cellular and Molecular Mechanisms II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P343.10

Topic: C.10. Brain Injury and Trauma

Support: NIH Grant R01-NS096012
NIH Grant F31-NS106806
NIH Grant R01-EY024890

Title: Traumatic brain injury to primary visual cortex produces long-lasting circuit dysfunction

Authors: *A. TIERNO[1], J. C. FRANKOWSKI[1], A. T. FOIK[1,2], J. R. MACHHOR[1], D. C. LYON[1], R. F. HUNT[1];
[1]Anat. and Neurobio., Univ. of California Irvine, Irvine, CA; [2]Ophthalamic Biol. Group, Intl. Ctr. for Translational Eye Res., Polish Acad. of Sci., Warsaw, Poland

Abstract: Primary sensory areas of the mammalian neocortex have a remarkable degree of plasticity, allowing neural circuits to adapt to dynamic environments. However, little is known about the effects of traumatic brain injury on visual circuit function. Here we used anatomy and *in vivo* electrophysiological recordings in adult mice to quantify neuron responses to visual stimuli two weeks and three months after mild controlled cortical impact injury to primary visual cortex (V1). We found that, although V1 was largely intact in brain-injured mice, there was

~35% reduction in the number of neurons that affected inhibitory cells more broadly than excitatory neurons. This reduction was layer-specific, with inhibitory neurons reduced in more superficial layers of the injured cortex and excitatory neuron loss throughout all cortical layers. V1 neurons showed dramatically impaired responses to visual stimuli and weaker orientation selectivity and tuning *in vivo*, consistent with loss of cortical inhibition. This deficit was present two weeks after injury and persisted long-term. As the first demonstration of cortical circuit dysfunction following central visual system neurotrauma, our results show a single, mild contusion injury produces profound and long-lasting impairments in the way V1 neurons encode visual input.

Disclosures: A. Tierno: None. **J.C. Frankowski:** None. **A.T. Foik:** None. **J.R. Machhor:** None. **D.C. Lyon:** None. **R.F. Hunt:** None.

Poster

P357. Neurodegeneration and Brain Injury: Clinical Biomarkers and Therapeutic Strategies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P357.01

Topic: C.10. Brain Injury and Trauma

Support: Healthy Brains for Healthy Lives

Title: Predicting recovery of consciousness in intensive care units

Authors: *S. BLAIN-MORAES, A. FRANTZ, C. MASCHKE, C. DUCLOS;
McGill Univ., Montreal, QC, Canada

Abstract: Background: Individuals who have suffered a severe brain injury typically require extensive hospitalization in intensive care units (ICUs), where critical treatment decisions are made to maximize their likelihood of recovering consciousness and cognitive function. These treatment decisions can be difficult when the neurological assessment of the patient is limited by unreliable behavioral responses. Reliable objective and quantifiable markers are lacking and there is both (1) a poor understanding of the mechanisms underlying the brain's ability to reconstitute consciousness and cognition after an injury and (2) the absence of a reliable and clinically feasible method of tracking cognitive recovery in ICU survivors. Our goal is to develop and validate a clinically relevant EEG paradigm that can inform the prognosis of unresponsive, brain-injured patients in the ICU.
Methods: We will recruit 200 continuously-sedated brain-injured patients across five ICUs. Between 24 h and 7 days post-ICU admission, high-density EEG will be recorded from behaviorally unresponsive patients before, during and after a brief cessation of pharmacological sedation. Once patients have reached the waking stage, they will be asked to complete an

abridged Cambridge Brain Sciences (CBS) battery, a web-based series of neurocognitive tests. The test series will be repeated every day during acute admission (ICU, ward), or as often as possible given the constraints of ICU and ward care. Following discharge, patients will continue to complete the same test series on weekly, and then monthly basis, for up to 12 months following injury.
Results: To date, we have recruited 20 patients to this study. We have recorded EEG from 15 patients with injuries ranging from stroke to severe traumatic brain injury. Further, we have begun cognitive testing in 10 patients and have CBS data for some patients up 6 months post-coma. Recruitment is ongoing at multiple sites across Canada. Preliminary results suggest that EEG may have predictive potential for both functional and cognitive outcomes following pharmacologically-induced coma.
Anticipated findings: We anticipate our findings will lead to an increased ability to identify patients, as soon as possible after their brain injury, who are most likely to survive, and to make accurate predictions about their long-term cognitive and functional outcome. In addition to providing critically needed support for clinical decision-making, this study has the potential to transform our understanding of key functional EEG networks associated with consciousness and cognition.

Disclosures: S. Blain-Moraes: None. **A. Frantz:** None. **C. Maschke:** None. **C. Duclos:** None.

Poster

P357. Neurodegeneration and Brain Injury: Clinical Biomarkers and Therapeutic Strategies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P357.02

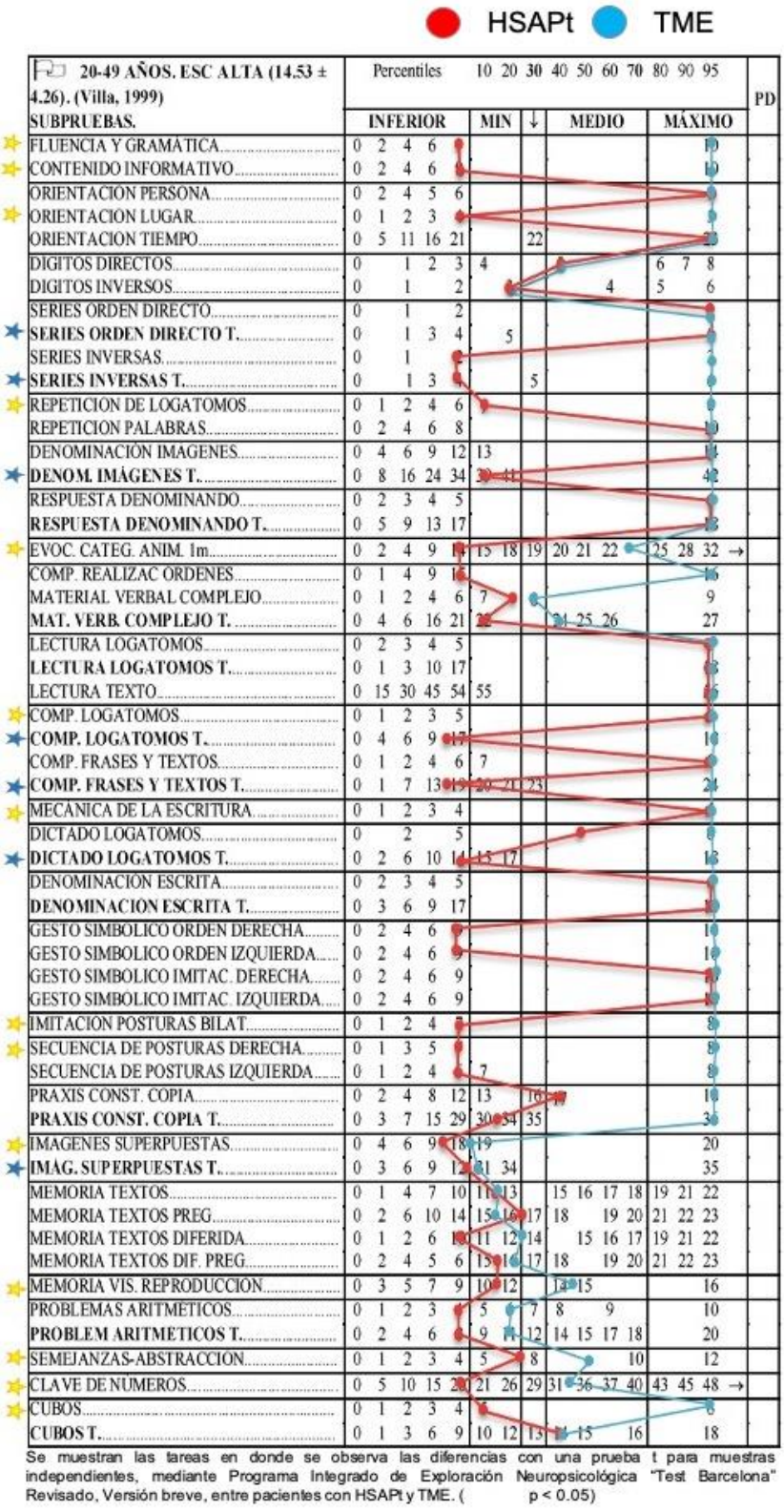
Topic: C.10. Brain Injury and Trauma

Title: Short-term cognitive profile of patients with Traumatic Subarachnoid Hemorrhage (tSAH) during the acute phase of the injury

Authors: *E. MORENO[1], M. RAMIREZ FLORES[2], J. CONTRERAS VACA[3];
[1]Psychology Fac., Univ. Nacional Autónoma De México, Ciudad De México, Mexico;
[2]Psychology Fac., Univ. Nacional Autonoma de México, Mexico City, Mexico; [3]Inst. Mexicano del Seguro Social, Mexico City, Mexico

Abstract: Traumatic Subarachnoid Hemorrhage (tSAH), a secondary event to Traumatic Brain Injury, with high morbidity and mortality, causing a high rate of disability, such as cognitive sequelae, hindering their social and occupational reintegration. Objective: This project sought to explore and identify the short term cognitive profile during the acute stage of injury in adult patients in order to identify the first cognitive domain deficits. Method: At the Regional General Hospital #2 "Doctor Guillermo Fajardo Ortiz" from "Instituto Mexicano del Seguro Social".

There were 15 patients with tSAH (age: 30; Schooling: 13.4 (3.83); M: 11; F: 4; Fisher scale: Grade II: 40%; Grade III: 40%; Grade IV: 20%), paired with a control group with Musculoskeletal Trauma (MT) by age and schooling; 15 patients (age: 30.8; Schooling: 13.2 (2.95); H: 10; M: 5). The Revised Integrated Neuropsychological Exploration Program "BARCELONA TEST" Mexican version was applied. Results: The tSAH group shows cognitive deficit in; Spatial Orientation, Fluency and Grammar, Language Processing Speed, Short and long-term verbal memory; Selective Attention, Perceptual Reasoning and Abstraction. Conclusion: The presence of the tSAH shows lower cognitive performance compared to the MT group, posing a higher response latency in areas associated with the evocation of information by semantic field and lexical comprehension; executive functions, main planning, supply and perceptual reasoning.



Se muestran las tareas en donde se observa las diferencias con una prueba t para muestras independientes, mediante Programa Integrado de Exploración Neuropsicológica "Test Barcelona" Revisado, Versión breve, entre pacientes con HSAPt y TME. ($p < 0.05$)

Disclosures: E. Moreno: None. **M. Ramirez Flores:** None. **J. Contreras Vaca:** None.

Poster

P357. Neurodegeneration and Brain Injury: Clinical Biomarkers and Therapeutic Strategies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P357.03

Topic: C.10. Brain Injury and Trauma

Title: Hormone levels impact concussion recovery assessment metrics in female varsity athletes - A preliminary study

Authors: *A. E. PIERIAS, D. J. GORBET, H. EDGELL, L. E. SERGIO;
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Abstract: Sports-related concussion is one of the most difficult injuries to diagnose and track. Research has provided evidence for the continued need of updating tools and metrics in hopes of increasing objectivity, specificity, and sensitivity. Research surrounding concussion has uncovered differences in the nature of injury, and recovery trajectory related to sex, previous history of concussion, age, and experience in sport. As such, it has become evident that research in the area of concussion should included males and females as separate groups, and account for as many other covariates as possible. Previous research from our lab noted sex-specific differences in performance on a complex visuomotor integration task and the reporting of emotionality symptoms in those with and without a history of concussion. This study seeks to expand on previous data by investigating the relationship between levels of estradiol, testosterone, and progesterone, and aspects of the sport concussion assessment tool (SCAT) along with a complex visuomotor transformation task in female varsity athletes. Participants included 30 female varsity athletes (16 with a history of concussion). All participants were asymptomatic by current standards at the time of testing. Participants were tested on two visuomotor skill tasks, one of which was a standard slide-finger-to-target on a touchscreen and the other was a complex transformation with a decoupled gaze/hand location and reversed visual feedback (requiring cognitive-motor integration). They also completed the SCAT3/5 assessment, and a saliva sample was collected. No significant group differences were found on any of the metrics - with and without the controlling for levels of estradiol, testosterone, or progesterone. When collapsed into a single group, progesterone was moderately positively correlated with overall performance on a visuomotor transformation task, and testosterone was moderately positively correlated with the cognitive-motor integration trials (two levels of dissociation) of the visuomotor transformation task. We suggest that these hormones may therefore have an impact on the sensitivity of current metrics and clinicians should take into account these impacts when making return to play decisions.

Disclosures: A.E. Pierias: None. **D.J. Gorbet:** None. **H. Edgell:** None. **L.E. Sergio:** None.

Poster

P357. Neurodegeneration and Brain Injury: Clinical Biomarkers and Therapeutic Strategies

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Program #/Poster #: P357.04

Topic: C.10. Brain Injury and Trauma

Support: VA RRD to AH K2-RX00949
VA RRD to AH I01 RX002916

Title: Customizing rTMS treatment targeting for co-occurring alcohol use disorder and mild traumatic brain injury using multi-modal neuroimaging data as neurobiological markers.

Authors: *A. HERROLD;
Edward Hines Junior VA Hosp., Hines, IL

Abstract: Alcohol use and mild traumatic brain injury co-occur, as well as, increase the risk, exacerbate symptoms, and worsen outcomes for each condition. While rTMS is an active area of investigation for the treatment of alcohol use disorder (AUD) and mild traumatic brain injury (mTBI) each in isolation, there is no consensus on the optimal rTMS site of stimulation for each of these conditions, let alone when they co-occur. Our objective was to customize rTMS targeting to improve alcohol craving among Veterans with co-occurring alcohol use disorder and mTBI (AUD+mTBI). We integrated data from a group-level fMRI analysis from a cross-sectional study to inform a subsequent pilot, ongoing, rTMS RCT recruiting from the same participants. To determine the target network, we first conducted a group-level analyses of alcohol cue reactivity fMRI task data collected from Veterans with AUD+mTBI (n=10), AUD only (n=6) and controls (n=5). Second, Neurosynth.org was used to determine areas of the brain most highly correlated with the left caudate locus of fMRI task activation. We used this aggregated functional connectivity database to select the cinguloopercular (CON) resting state network for rTMS targeting. Third, we customized the stimulation target by determining the site within the left DLPFC most correlated with the CON using individual-level rs-fMRI data utilizing published methods for a single AUD+mTBI Veteran. At the group level, Veterans with AUD+mTBI had significantly greater cue-induced activation ($p < 0.01$) in reward-related brain regions than healthy controls. Further, activity in the caudate hotspot was functionally connected to the CON. We identified the customized left DLPFC TMS target with strongest correlation to the CON at the individual level for a Veteran with AUD+mTBI. This target was anatomically distinct from that determined by more standard clinical methods. There were no AEs and the rTMS paradigm in the pilot RCT is safe and tolerable, thus far. We will examine how connectivity to the rTMS site of stimulation changed between pre- and post-rTMS and the relationship to symptom data to inform treatment responsiveness. Individualized rs-fMRI data can be used to customize TMS targets for AUD+mTBI with relevance to alcohol cue-induced

activation. Ongoing work will examine whether this customization improves alcohol-related outcomes after TMS.

Disclosures: A. Herrold: None.

Poster

P357. Neurodegeneration and Brain Injury: Clinical Biomarkers and Therapeutic Strategies

Location: Virtual Only

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Program #/Poster #: P357.05

Topic: C.10. Brain Injury and Trauma

Support: NIH Grant U54NS115266
NIH Grant 1R01AG057902-01

Title: Blood Vessel Proliferation in Chronic Traumatic Encephalopathy

Authors: G. A. ROSEN[1], D. KIRSCH[2], S. HOROWITZ[1], J. D. CHERRY[3], R. NICKS[4], H. KELLEY[4], R. MATHIAS[2], K. A. CORMIER[1], C. A. KUBILUS[2], J. MEZ[4], M. ALOSCO[4], Y. TRIPODIS[4], T. D. STEIN[5], V. E. ALVAREZ[1], A. C. MCKEE[3], *B. R. HUBER[1];
[1]VA Boston Healthcare, Boston, MA; [2]Boston Univ. Sch. of Med., Boston, MA; [3]Boston Univ., Boston, MA; [4]Boston Univ. Alzheimer's Dis. and CTE Ctr., Boston, MA; [5]Boston VA Med. Ctr., Boston, MA

Abstract: Chronic traumatic encephalopathy (CTE) is a neurodegenerative disease linked to exposure to repetitive head impacts (RHI). The diagnostic lesion for CTE is hyperphosphorylated tau protein (p-tau) in perivascular neurons with or without astrocytes at cortical sulcal depths. Simulations of impact forces experienced by athletes suggest that shear stress and deformation of the brain is greatest at sulcal depths. The shearing forces lead to inflammation within the depth of the sulcus. Neuroinflammation is associated with dysregulated neoangiognesis. We hypothesize that this type of injury stimulates neoangiogenesis through neuroinflammation and regional hypoxia. To investigate vascular proliferation in the context of RHI and CTE, we examined optically cleared sections of postmortem human dorsolateral frontal cortex from the VA-BU-CLF (UNITE) Brain Bank. Study participants included those with neuropathologically diagnosed CTE (n=10 severe, n=10 mild), RHI exposure without CTE (n=8), and RHI-naïve controls (n=7). The tissue was post-fixed using the SHIELD protocol and optically cleared using a modified version of passive CLARITY. Vasculature was stained with fluorescently labeled tomato lectin allowing high-resolution observation of three-dimensional vessel morphology. We quantitated variables including branch density and fractional volume to evaluate features associated with angiogenesis. In the grey matter at sulcal depths, branch density and fraction volume were significantly increased in CTE cases compared to RHI-exposed cases

without CTE. Furthermore, in the grey matter sulcus of non-CTE cases with a history of RHI exposure, blood vessel branch density was increased, and fraction volume was decreased compared to RHI-naïve controls. Blood vessel fraction volume in the grey matter sulcus correlated positively with the area of p-tau staining determined histologically. These findings demonstrate that vascular remodeling occurs at both the sulcus and gyral crest in those with a history of RHI compared to naive controls. In those with CTE, the magnitude of the vascular remodeling in the sulcal depth is greater than in RHI. Previous work has demonstrated that the depth of sulcus in CTE is also a focus of neuroinflammation. Together, these findings suggest RHI is associated with global gray matter neoangiogenesis, and in CTE lesions, chronic neuroinflammation further increases vascular changes.

Disclosures: G.A. Rosen: None. **D. Kirsch:** None. **S. Horowitz:** None. **J.D. Cherry:** None. **R. Nicks:** None. **H. Kelley:** None. **R. Mathias:** None. **K.A. Cormier:** None. **C.A. Kubilus:** None. **J. Mez:** None. **M. Alosco:** None. **Y. Tripodis:** None. **T.D. Stein:** None. **V.E. Alvarez:** None. **A.C. McKee:** None. **B.R. Huber:** None.

Poster

P357. Neurodegeneration and Brain Injury: Clinical Biomarkers and Therapeutic Strategies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P357.06

Topic: C.01. Brain Wellness and Aging

Support: APMRF
Together Strong

Title: A proteomics study of CSF protein biomarkers of Niemann-Pick Disease, Type C1

Authors: *W. LI[1], A. GRAYSON[2], Z. GOLDBERG[2], N. YANJANIN[3], E. BERRY-KRAVIS[2], F. PORTER[3], S. COLOGNA[1];
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Abstract: Nieman-Pick Disease, Type C1 (NPC1) is an autosomal recessive, fatal neurodegenerative disorder that is caused by mutations in *NPC1*, which encode a cholesterol transporter located on the membrane of the lysosome. As a result, unesterified cholesterol and sphingolipids accumulate in the lysosome, leading to many clinical phenotypes including hepatosplenomegaly, vertical gaze palsy, and progressive cerebellar and cortical degeneration. To date, there is no FDA-approved therapy for NPC1 patients, however, intrathecal treatment with VTS-270 (2-hydroxypropyl-beta-cyclodextrin, adrabetadex) and an oral drug, arimoclomol are being evaluated clinically. In order to identify biomarkers that can provide insight into disease pathophysiology, VTS-270 treatment status, and treatment response, our group is

investigating proteomic changes in cerebrospinal fluid (CSF) obtained from NPC1 patients who were treated with VTS-270 and healthy controls. In total, 17 NPC1 patient samples and 10 healthy controls have been analyzed. Briefly, CSF samples were depleted with a human serum albumin/IgG column to remove the most abundant proteins in the CSF. Proteins were then reduced, alkylated, and digested followed by isobaric labeling with a TMT10 multiplexing kit. Data were acquired on a Q-Exactive nanoLC-MS system and analyzed using the SEQUEST algorithm via the Thermo Proteome Discoverer software. Several differential proteins were found among the groups including Cytochrome c, Parvalbumin alpha, and Cathepsin D, implying that these proteins could be the potential biomarkers for VTS-270 treatment status and treatment response. Additional patient specimens are currently being evaluated as well as correlation analysis to establish the protein of responders and non-responders to VTS-270.

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Poster

P357. Neurodegeneration and Brain Injury: Clinical Biomarkers and Therapeutic Strategies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P357.07

Topic: C.01. Brain Wellness and Aging

Support: R01AG063881
R43AG061981

Title: Apoe4 polymorphism and reduced olfactory neural activation in aging

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Abstract: Objective: Olfactory dysfunction is associated with Alzheimer's Disease development. Our previous study revealed that odor identification tasks activated several olfactory brain regions more than odor memory tasks. In this study, we examined if apolipoprotein E ε4 (ApoE ε4) positivity contributed to reduced brain activations in the primary olfactory cortex regions were Alzheimer's neuropathology begins. **Method:** African American adults (N=11), who completed comprehensive battery of cognitive tests, odor psychophysical tasks, magnetic resonance imaging (MRI) under 3-Tesla scanner while performing episodic odor

memory tasks 55 - 75 years old, were genotyped for APOE polymorphisms. ApoE ε3 homozygote subjects were compared to carriers of ε4 allele on their performances in psychophysical tasks and cognitive score, and in the degree of neural activations in olfactory regions of interest. Areas of difference between the two groups were identified using an uncorrected t-statistic of 1.5, and FDR cluster-level correction of $p<0.05$ was used for multiplicity of tests (the cluster defining threshold was set at $p<0.01$). **Results:** There were 6 non-ApoE ε4 carriers (i.e., ε2/ε3 and ε3/ε3) and 5 ε4 carriers (i.e.ε3/ε4). Both groups were comparable educational, ethnic and other demographic characteristics. Though no significant differences in episodic odor memory tasks between groups, non- ε4 carriers had greater activation than ε4 carriers in several of the classical olfactory brain regions. No olfactory regions exhibited greater activation in ε4 carriers over the ε3 homozygotes. **Conclusion:** fMRI-based odor memory studies may be more potent than odor memory tasks alone in differentiating older non-ApoE ε4 carriers from ε4 carriers. Given the strong association between ε4 and AD development, if replicated this approach may contribute towards improved early detection of Alzheimer's disease.

Disclosures: M.S. Hipolito: None. **N. Rai:** None. **R. Seth:** None. **J.W. VanMeter:** None. **M. Shelby:** None. **M.M. Misiak-Christian:** None. **C. Nwaokobia:** None. **K.F. Manaye:** None. **T. Obisesan:** None. **E. Nwulia:** None.

Poster

P357. Neurodegeneration and Brain Injury: Clinical Biomarkers and Therapeutic Strategies

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Program #/Poster #: P357.08

Topic: C.01. Brain Wellness and Aging

Support: National Institute of Heath grant AG058748
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Alzheimer's Association Award AARF-17-530186

Title: Associations between catecholamine synthesis, locus coeruleus-entorhinal functional connectivity, and Alzheimer's pathology

Authors: *C. J. CIAMPA[1], J. H. PARENT[1], T. M. HARRISON[2], J. N. ADAMS[2], A. MAASS[3], W. J. JAGUST[2], A. S. BERRY[1];
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Abstract: The locus coeruleus (LC) is the brain's major source of the neuromodulator norepinephrine and is among the first brain regions to accumulate Alzheimer's disease (AD)-related tau pathology. Prominent models of AD pathophysiology posit that tau spreads trans-synaptically, first from LC to temporal regions, and is driven by neuronal activity. We probed relationships between LC catecholamine (norepinephrine/dopamine) synthesis capacity measured with [^{18}F]Fluoro-l-m-tyrosine (FMT) PET, temporal tau pathology measured with [^{18}F]Flortaucipir PET, and functional connectivity measured with 3T resting state fMRI. Participants were cognitively normal older adults (n = 50, mean age = 77) whose β-amyloid status was characterized with [^{11}C]Pittsburgh compound B (PiB) PET (16 [^{11}C]PiB+). LC functional connectivity analyses focused on correlating fMRI activity with entorhinal cortex (EC), which is structurally connected with LC and shows measurable tau pathology in normal aging. All regression models included age, sex, and years of education as covariates. Analyses testing relationships between LC catecholamine synthesis capacity and functional connectivity found lower LC [^{18}F]FMT net tracer influx (Ki) was related to higher functional connectivity between LC and anterolateral/transentorhinal cortex (t(44)=-2.09, p=.04), the first EC subregion to accumulate tau. We found no relationship between EC [^{18}F]Flortaucipir standardized uptake value ratio (SUVR) and LC-EC functional connectivity or LC [^{18}F]FMT Ki (both p > .44). However, higher LC-EC functional connectivity was associated with higher [^{18}F]Flortaucipir SUVR in a temporal tau meta-region of interest (EC, amygdala, hippocampus, fusiform, parahippocampal, middle temporal and inferior temporal gyri; t(39)=2.24, p=.03). A model including [^{11}C]PiB status revealed a significant [^{11}C]PiB by LC-EC functional connectivity interaction (t(37)=3.78, p=.001). Higher functional connectivity was associated with higher [^{18}F]Flortaucipir SUVR for Aβ+ but not Aβ- participants. Together, these findings link higher LC functional connectivity with greater temporal tau burden and lower LC catecholamine synthesis. These results are consistent with models proposing activity-mediated tau spread, and implicate lower LC catecholamine synthesis as a factor increasing one's susceptibility to tau spread. Future studies should investigate longitudinal relationships between catecholamine synthesis, functional connectivity, and AD pathology to better understand the influence of the LC-catecholamine system on aging trajectories.

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Poster

P357. Neurodegeneration and Brain Injury: Clinical Biomarkers and Therapeutic Strategies

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Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P357.09

Topic: C.01. Brain Wellness and Aging

Support: NIA R01 AG055449
NIA P30 AG028383
NIGMS S10 OD023573
UH3-NS100614

Title: Intracellular Rather Than Extracellular Diffusion MRI Measures Predict Working Memory Performance in Healthy Older Adults

Authors: *C. BAUER[1], V. ZACHARIOU[2], P. MAILLARD[4], A. CAPRIHAN[5], B. T. GOLD[3]; [2]Cognitive Neurosci. of Aging Lab., [3]Neurosci., [1]Univ. of Kentucky, Lexington, KY; [4]Univ. of California Davis, David, CA; [5]The Mind Res. Network, Albuquerque, NM

Abstract: Diffusion tensor imaging (DTI) is widely used in MRI research to estimate white matter health in the aging human brain, but little is known about which specific DTI indices are most predictive of cognitive function. While fractional anisotropy (FA) reflects diffusion associated with both intracellular (tissue) and extracellular (Free Water [FW]) compartments, recent multi-compartment models (FW-correction [includes FW-corrected FA and FW] and NODDI [includes intracellular volume fraction {ICVF} and orientation dispersion index {ODI}]) provide more specific intracellular or extracellular compartments. Here, we compared the strength of association between these different DTI metrics to both age and cognition (working memory [WM] performance). Ninety-nine healthy older adults (ages 60-85; 61 women) were recruited from the Lexington community and scanned at the University of Kentucky using a 3 Tesla MRI Siemens Magnetom Prisma with a 64-channel head coil. A 126-direction main DTI sequence using 4 b-values (0, 500, 1000, and 2000 s/mm^2), a reverse phase-encoding direction DTI sequence, and an fMRI sequence were acquired. Participants performed an in-scanner visual WM task (N-back; control, 1-back, and 2-back conditions) during fMRI acquisition with WM performance calculated using D-prime. A network of brain regions showing functional activation during the WM task was defined across participants using a group-level functional contrast (2-Back > [1-Back/2 + Compare/2]/2). White matter tracts connecting the functionally-activated working memory network was reconstructed using probabilistic tractography. Each participant's diffusion measures were extracted from skeletonized white matter tracts connecting WM regions using FSL's tract-based spatial statistics. Linear regression models with diffusion measures as the predictors and age or D-prime (with age as a covariate) as the dependent variables were conducted in SPSS controlling for gender. All diffusion measures except ODI significantly predicted age. However, only ICVF (p = 0.035) and FW-corrected FA (p = 0.061; marginal significance) specifically predicted D-prime. Our results suggest that intracellular-associated (axonal loss and demyelination) rather than extracellular-associated (neurodegeneration, vascular dysfunction, or neuroinflammation) diffusion may be more sensitive to WM performance.

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Poster

P357. Neurodegeneration and Brain Injury: Clinical Biomarkers and Therapeutic Strategies

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P357.10

Topic: F.04. Stress and the Brain

Support: CIHR Grant 173416

Title: Improvements in the classification of concussion cohorts using self-reported daily vs weekly stress reports

Authors: *J. L. LOEWEN[1,2], A. P. LAPOINTE[3,2,6], I. K. ONI[4,2,6], A. SOROUSH[4,2,6], J. N. RITCHIE[4], K. T. NEWEL[7], J. S. BURMA[5,6,2,8,9], J. F. DUNN[3,2,6];
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Abstract: Self-reported neuropsychological assessments in post-concussive cohorts have highlighted an increased risk of developing anxiety, depression, and/or stress disorders. The majority of these assessments are made using a single time point. However, repeated self-reported assessments of anxiety have demonstrated clinical potential. Minimal research has been conducted on the potential of repeated stress measures to increase the classification of individuals with a concussion history. This study consisted of 45 participants (67% female), 24 participants had concussion history (54% female, 23.91 ± 3.25 years old) and 21 participants had no concussion history (81% female, 23.31 ± 6.55 years old). Participants completed two separate questionnaires assessing self-reported stress. The Perceived Stress Scale (PSS) was administered once (day one of seven; Model 1) and the Daily Stress Questionnaire (DSQ; Model 2) was administered at the same time every day for seven consecutive days. We fitted two separate logistic models estimated using maximum likelihood to classify concussion history using stress, sex, handedness, and age as predictors. The models used stress measures obtained from the total PSS score and the daily total DSQ scores. Classification accuracy of concussion history was compared between both models. The first model correctly classified 62.5% of cases and no significant association between stress and concussion history was observed (beta = -0.04, 95% CI [-0.20, 0.11], p = 0.602; Std. beta = -0.20, 95% CI [-1.03, 0.55]). The second model correctly classified 82.2% of cases and identified statistically significant associations between stress and concussion history (beta = 1.95, 95% CI [1.22, 2.81], p < .001; Std. beta = -0.23, 95% CI [-0.76, 0.21). Daily measures of stress predicted concussed cohorts with 20% greater accuracy than singular measures. Our findings indicate that repeated self-reported stress measures better classify concussed cohorts and offer additional interaction between stress and concussion history

that weekly measures lack. Moreover, repeated measures of stress could prove useful in informing treatment and rehabilitation.

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Poster

P458. Subcortical Visual Pathways I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P458.01

Topic: D.07. Vision

Support: DFG Emmy-Noether KR 4062/4-1

Title: A developmental model for the emergence of orientation preference map in the mouse superior colliculus

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Abstract: The mouse superior colliculus (SC) has been shown to have an orientation preference map (OPM; Ahmadlou and Heimel, 2015; Feinberg and Meister, 2015; but see Chen et al., 2020). However, the OPM in the mouse SC does not represent all orientations equally as in the primary visual cortex. More specifically, different regions of the SC visual layers tend to overrepresent specific orientations, and a retinotopy-based concentric OPM pattern has been observed (Ahmadlou and Heimel, 2015). The underlying developmental mechanisms that give rise to the observed OPM in the mouse SC are still unclear. During development of the visual system, there are spontaneous activities spreading across the retina in wave-like patterns, where the stage III retinal (S3) waves are known to play an important role in circuit development, such as fine-scale retinotopic refinement. In the mouse SC, the S3 waves exhibit non-uniform propagating directions with net biases towards the caudal direction (Gribizis et al., 2019). In addition, the OFF retinal ganglion cells (RGCs) are recruited with a delay compared to ON RGCs during S3 waves (Kerschensteiner and Wong, 2008). Here, we hypothesize that these biases in the S3 wave propagating directions, together with the OFF-ON delay, could play a role in establishing the OPM in the mouse SC during development. To test this hypothesis, we studied a computational model of visual system development that incorporates the above-mentioned S3 wave features for examining the properties of the established OPM. By using the Hebbian learning mechanism to refine the RGC-SC connections, we show that the OFF delayed responses can segregate the ON- and OFF-RGC inputs to individual SC neurons, which locally

give rise to the orientation preference (Kremkow et al., 2016). Furthermore, we demonstrate that the biases in S3 wave propagating directions can mediate the formation of an OPM with a concentric pattern. In our model, the orientation preferences are represented based on their retinotopy, with each retinotopic location having biases in representing specific orientations, as observed in vivo (Ahmadlou and Heimel, 2015; Feinberg and Meister, 2015). In summary, our model suggests that OFF delayed responses of S3 waves can mediate local microscopic segregation of the ON-OFF receptive subfields of the postsynaptic SC neurons, whereas the biases in the wave propagating direction determine the overall macroscopic organization of the OPM. Taken together, these findings suggest that the S3 waves could instruct the formation of the OPM in the mouse SC by fine-tuning the receptive field of postsynaptic SC neurons.

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Poster

P458. Subcortical Visual Pathways I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P458.02

Topic: D.07. Vision

Support: James S. McDonnell Foundation Grant 220020516

Title: A simpler model can unify a broad range of phenomena in retinotectal map development

Authors: *S. S. JAMES, S. P. WILSON;
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Abstract: In the Chemoaffinity Theory, the retinotectal axon projection uses pairs of orthogonal signalling gradients in the retina to specify the eventual location of synapses formed on the tectum. Similar tectal gradients provide a coordinate system which allows each axon to approach a pre-specified destination. Although the ephrins have been shown to guide axons toward their destination, a complete account of the local interactions which recreate the topography of the retinal cells is lacking. In line with the assumptions of the Chemoaffinity Theory, a model developed by Simpson & Goodhill (2011) provides an account of the basic topographic arrangement of axons on the tectum, and reproduces the effects observed in a range of surgical and genetic manipulation experiments. However, this model does not represent chemotactic guidance as a local mechanism, and instead continuously provides each simulated axon with explicit information about the direction in which to grow to reach a pre-specified target. In addition to the inclusion of a global chemoaffinity term, this model includes a competitive interaction for space between growth cone agents and a receptor-ligand axon-axon interaction in order to account for the full set of experimental manipulations. Here, we replace the chemoaffinity term with a gradient-following model consisting of axonal growth cone agents that

carry receptor molecule expression at a level determined by the location of the soma on the retina. According to this entirely local model, growth cones moving on a simulated tectum are guided by two pairs of opposing orthogonal signalling molecules representing the ephrin ligands. We show that with *only* the chemoaffinity term and a receptor-ligand axon-axon interaction term (meaning that *all* growth cone interactions are by receptor-ligand signalling), a full range of experimental manipulations to the retinotectal system can be reproduced. Furthermore, we show that the observation that competition is *not* an essential requirement for axons to find their way (Gosse et al., 2008) can also be accounted for by this simpler model, due to the opposing influences of signalling gradient pairs. Finally, under the assumption of exponentially varying receptor expression in the retina, we demonstrate that ligand expression should either be exponential, if the receptor-ligand signal induces repulsion (i.e. gradient descent), or logarithmic, if the signal induces attraction (gradient ascent). Thus, we show that a model analogous to that which can explain thalamocortical patterning in the barrel cortex (James et al., 2020), can also account for the self-organization of topographic maps in the retinotectal system.

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Poster

P458. Subcortical Visual Pathways I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P458.03

Topic: D.07. Vision

Support: NIH Grant EY030507

Title: Brain-derived neurotrophic factor modulates retinal ganglion cell output synapses in the dorsolateral geniculate nucleus

Authors: *M. VAN HOOK;
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Abstract: Brain-derived neurotrophic factor (BDNF) is important in development and also plays roles in synaptic and neuronal plasticity. In the dorsal lateral geniculate nucleus (dLGN) of the thalamus - the primary projection target for retinal ganglion cells (RGCs) responsible for conscious vision, BDNF is present in RGC axons and the BDNF receptor TrkB is present on RGC axon terminals and post-synaptic thalamocortical (TC) relay neurons. The goal of this study was to determine how BDNF influences synaptic transmission and neuronal function within the adult dLGN. This was assessed with whole-cell patch clamp recordings from dLGN TC neurons in brain slices from adult mice (6-12 weeks old) of both sexes. RGC synapses (retinogeniculate/RG synapses) were activated in the presence of 60 μM picrotoxin either in parasagittal slices with an electrode positioned in the optic tract or in coronal slices via photic

activation of channelrhodopsin expressed in RGC axons (Chx10-Cre;Ai32 mice). Cortical feedback synapses were activated with an electrode positioned in the corticothalamic tract in parasagittal slices. Bath application of BDNF (20 ng/mL) led to an enhancement of RG synaptic transmission, as evidenced by an increase in excitatory post-synaptic current (EPSC) amplitude (27+/-6% increase, n = 18 TC neurons, p=0.016, paired t-test) and this was prevented by pre-treatment of the TrkB blocker cyclotraxin-B (p=0.74, n = 4 TC neurons). This was accompanied by a reduction in the frequency (p=0.006) and amplitude (p=0.008, n = 11 TC neurons) of miniature EPSCs and a small decrease in the AMPA/NMDA ratio (p=0.027, n = 8 TC neurons). In the presence of cyclothiazide (200 μM) and gamma-DGG (100 μM), BDNF had no effect on RG release probability, as assessed using paired pulse (50-1000 ms intervals) and 10 Hz train stimulation (3 s duration). However, BDNF did increase synaptic vesicle pool size, as indicated by an increase in Y-intercept of a line fit to the plot of cumulative EPSC amplitude. There was no statistically significant effect of BDNF on corticothalamic feedback synaptic strength; neither EPSC amplitude (p=0.3, n = 5 TC neurons) nor the paired pulse ratio (p=0.86) were altered following BDNF application. Finally, BDNF led to a slight TC neuron depolarization (3.6+/-0.9 mV, p = 0.0014, n = 14 TC neurons), but no statistically significant change in spiking (p=0.88, paired t-test, n = 7 TC neurons). These results indicate that acutely-applied BDNF enhances RG synaptic transmission by an increase in presynaptic vesicle pool size, although it also appears to influence post-synaptic receptors and membrane properties of TC neurons in the dLGN.

Disclosures:

Poster

P458. Subcortical Visual Pathways I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P458.04

Topic: D.07. Vision

Support: NIH/NEI R01 grant: EY030507
Nebraska Banker's Association Small Equipment Grant

Title: The role of retinal BDNF on the development of the dorsolateral geniculate nucleus

Authors: *E. BIERLEIN[1], A. BHANDARI[1], M. J. VAN HOOK[2];
[1]Ophthalmology and Visual Sciences, Pharmacol. and Exptl. Neurosci., [2]Ophthalmology & Visual Sci., Univ. of Nebraska Med. Ctr., Omaha, NE

Abstract: Brain-derived neurotrophic factor (BDNF) is a regulator of nervous system plasticity that plays important roles in the development and maintenance of synapses and neurons. BDNF is transported to the dorsolateral geniculate nucleus (dLGN), the visual relay center of the thalamus, and released by retinal ganglion cell axons, making this system an interesting model

for studying the role of BDNF in circuit development. However, the role of retinal BDNF in development has not been characterized in the dLGN. Thus, our goal was to determine how retinal BDNF influences the development of the dLGN by selectively deleting retinal BDNF using Cre-lox technology. We used a mouse line with Cre expression specific to the retina (Chx10-Cre) crossed with BDNFfl/fl mice to selectively delete BDNF from the retina to analyze the resulting changes in the structure and function of the dLGN in adult mice (9 to 14 weeks) of both sexes. To test for visual system function differences between Chx10-Cre;BDNFfl/fl (cKO) and control (C57Bl/6J or BDNFfl/fl mice), we analyzed eye tracking abilities in an optomotor (OMR) task using stimuli of varying spatial frequency and found that cKO mice spent less time tracking than controls (ctr) ($p=0.03$; cKO n = 16, ctr n = 10 mice, unpaired t-test). There were no differences in outer retinal light responses as seen in dark-adapted electroretinogram (ERG) ($p=0.7$ A wave, $p=0.9$ B wave; cKO n = 10 eyes, ctr n = 16 eyes, unpaired t-test), suggesting that OMR differences arose from changes in retinal ganglion cells or their projection targets in the brain. In whole-cell patch clamp recordings of miniature excitatory post-synaptic currents (mEPSCs) conducted on dLGN thalamocortical (TC) neurons, cKO mice had a lower area in cKO mEPSCs ($p=0.006$) likely due to differences in decay kinetics (cKO n = 17 cells, 8 mice; ctr n = 9 cells, 4 mice, nested t-test). To analyze TC neuron structural differences between cKO and ctr mice, we performed Sholl analyses on neurobiotin-filled TC neurons and found that cKO TC neurons were significantly less complex than ctr TC neurons ($p=0.03$, -7.183 ± 3.066 SEM; cKO n = 11 cells, 6 mice; ctr n = 10 cells, 4 mice). Analysis of binocular segregation using intravitreally injected cholera toxin b (CTb) tracers in prepubescent mice indicated no significant differences ($p=0.9$; cKO n = 7 mice, 13 sections; ctr n = 7 mice, 14 sections, unpaired t-test). Thus, retinal BDNF influences visual behavior as well as the structure and function of dLGN TC neurons. These results further our understanding of both developmental and neurodegenerative processes in the visual system.

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Poster

P458. Subcortical Visual Pathways I

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Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P458.05

Topic: D.07. Vision

Support: NIH Grant DP2EY027983

Title: Direct ipRGC input to the Substantia Innominata

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Abstract: The Substantia Innominata (SI) is a region of the basal forebrain which contains a population of cholinergic neurons that project diffusely throughout the cortex to modulate neural activity. This cholinergic neuromodulatory system is hypothesized to be involved in attention and arousal. The SI receives retinal input, but little is known about the retinal ganglion cell (RGC) types which project to this region or the circuits to which they contribute. To first characterize the extent of retinal projections to the SI, we intravitreally injected Cholera Toxin Subunit-B conjugated with a fluorescent protein, allowing us to visualize RGC terminals in the brain. We used an object-based colocalization analysis to count the number of RGC terminals in the SI within 1 μm of cholinergic neurons, identified based on the expression of choline acetyltransferase (ChAT). We observed RGC terminals closely apposed to cholinergic neurons, potentially making functional synapses. The number of RGC terminals near ChAT-positive neurons was significantly reduced when the one channel was rotated prior to the colocalization analysis. To identify the RGC types which project to the SI, we injected AAV-CAG-GFP into the SI to retrogradely label SI-projecting RGCs with GFP. Using cell-attached electrophysiology we recorded from retrogradely labeled RGCs in whole-mount retinas and classified the ganglion cells based on their light response. Many of these RGCs were the intrinsically photosensitive (ip)RGCs. In support of this, fluorescent labeling of ipRGC axons revealed substantial innervation of the SI by ipRGCs. Finally, we exposed mice to a 15-minute light pulse at 1000 lux and immunolabeled the SI for the immediate early gene c-fos to determine whether the SI receives light input from RGCs. Excitingly, we noted increased c-fos labeling in the SI of animals exposed to light in comparison to those exposed to dark only, suggesting that the SI receives functional light input from RGCs. Together, these data suggest that ipRGCs signal to neurons in the SI, suggesting a potential role for ipRGCs in regulating neuromodulatory networks of the brain based on environmental light conditions.

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Poster

P458. Subcortical Visual Pathways I

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Topic: D.07. Vision

Support: Kinghts Templar Eye Foundation Career Starter Grant to SGV
Gilbert Family Foundation Vision Restoration Initiative to ADH
NIH to ADH: EY027713

Title: Probing the role of retinorecipient target cells in visual circuit regeneration

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Abstract: Vision is the most important sense for humans to navigate the world and survive. Retinal ganglion cells (RGC) transmit light information in the environment to the brain and therefore represent a key bottleneck for vision. RGCs are vulnerable to damage in neurodegenerative diseases and injuries that cause blindness and impact millions of people, such as glaucoma and traumatic injury. *As RGCs and their axons are part of the central nervous system, they cannot repair themselves after injury*. One promising approach is to reapply fundamental mechanisms that drive the formation of visual circuits during development, to promote re-growth and connectivity of RGC axons with retinorecipient targets. During development, both RGCs and their targets play a key role in forming the retinofugal (eye-to-brain) pathway to allow functional vision. While many studies have focused on the cellular and molecular events occurring in the RGCs to promote regeneration, *far less is known about the role of target cells in RGC axon regeneration*. Here we are testing a novel idea that post-synaptic target cells in the retinofugal pathway can be leveraged to promote regeneration of visual circuits. During development, target-derived cues and neural activity are required in both, the RGCs and the target cells, to connect and refine RGC axons with their synaptic partners in the brain. Using a chemogenetic approach in a distal injury model, our data shows that *increasing electrical activity of neurons in the distal optic pathway can promote regeneration of RGC axons*. To investigate the specific contribution of retinorecipient target cells in this model, we identified a new mouse line to precisely activate cells that reside in the nucleus of the optic tract (NOT), a subcortical retinorecipient target in the accessory optic system that is responsible for image stabilization. Our preliminary data shows that specific activation of retinorecipient targets is sufficient to promote RGC axon regeneration, suggesting that target-derived cues are important to promote robust, accurate rewiring of injured visual circuits. These experiments will address the role of post-synaptic cells in CNS repair and will have widespread relevance for treating blinding diseases, stroke and traumatic brain injury.

Disclosures: S.G. Varadarajan: None. **O.S. Dhande:** None. **P. Le:** None. **A.D. Huberman:** None.

Poster

P458. Subcortical Visual Pathways I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P458.07

Topic: D.07. Vision

Title: Pathological and compensatory background mechanisms of hypoxia-induced long-term potentiation of NMDA neurotransmission in the visual retinocollicular pathway

Authors: *H. DUMANSKA, N. VESELOVSKY;
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Abstract: Hypoxia is the main accompanying factor of numerous diseases. Identification of the processes and mechanisms underlying the early stage of hypoxic injury of the visual retinocollicular pathway will benefit the future treatment of navigation, orientation, and visual attention impairments.We developed the unique in vitro model of the retinocollicular pathway – the coculture of dissociated retinal cells and superficial superior colliculus (SSC) neurons. In such a model, each synaptically connected pair of retinal ganglion cell (RGC) − SSC neuron reflects a single retinocollicular projection. Using paired patch-clamp and local superfusion technique, we recorded pharmacologically isolated NMDA−mediated postsynaptic currents in SSC neurons by generation action potentials in presynaptic RGCs under normal and hypoxic conditions. The decay time constant of postsynaptic currents was fit using a single exponential function.Hypoxia up to 5 min induces long-term potentiation of NMDA neurotransmission. Moreover, we observed a significant reduction of voltage-dependent magnesium blockade of evoked NMDA response during oxygen deprivation. Also, we analyzed the dynamics of the decay time constant of all NMDA−mediated currents under normal and hypoxic conditions. This analysis revealed that hypoxia initiates a rapid and irreversible decrease of the decay time (from 38.3 ± 5.6ms to 19.2 ± 6.8ms).Pathological hypoxia-induced potentiation of NMDA neurotransmission leads to an increase of calcium influx and, as result, to apoptotic cell death. We detected two bidirectional effects that accompanied such LTP with opposite effects. First, hypoxia reduces voltage-dependent magnesium blockade of NMDA receptor – one of the well-known protective mechanisms. This enhances the pathological effect of LTP of the NMDA transmission and causes an additional injury. Contrarily, the decrease of the decay time of NMDA currents during oxygen deprivation leads to the decrease of the calcium influx. Since kinetic and functional properties are determined by NMDA receptor subunit composition, such a decrease indicates an increase in NR2A-to-NR2B subunit ratio at the retinocollicular synapses. We assumed that rapid changes in subunit composition are associated with lateral receptor mobilization from adjacent locations, rather than with a new subunit expression.
The results obtained reflect that hypoxia-induced LTP of NMDA transmission is accompanied by bidirectional pathological and compensatory mechanisms that might be targeted to prevent hypoxia-involved lesions of the retinocollicular pathway.

Disclosures: H. Dumanska: None. **N. Veselovsky:** None.

Poster

P458. Subcortical Visual Pathways I

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Topic: D.07. Vision

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ERC 694401
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Title: Visual stimulus features are represented throughout the basal ganglia

Authors: *J. FABRE, A. J. PETERS, M. CARANDINI, K. D. HARRIS;
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Abstract: [Introduction] The basal ganglia are key structures in sensorimotor associations, receiving sensory input from the cortex which is thought to then influence downstream motor areas. For instance, neurons in the striatum have been shown to be selective for visual stimuli. However, it is not known whether selectivity is present in downstream structures and whether stimulus representations become increasingly more general. [Methods] To address this question, we used Neuropixels probes to record from the striatum, globus pallidus external (GPe), and substantia nigra reticulata (SNr) in head-fixed naive mice passively viewing a variety of visual stimuli: gratings of different orientation, spatial frequency, location, and an array of natural images. [Results] In the striatum, GPe and SNr, few neurons were visually-responsive. In all these structures, we found neurons responsive to visual images, and selective for orientation, spatial frequency, location and specific natural images. These neurons were generally selective for multiple features at once. We could also reliably decode natural images from striatal cell activity above chance level. [Conclusions] We conclude that neurons across the entire basal ganglia -- not just in striatum -- encode the properties of visual stimuli. We are currently comparing the visual responses between the striatum, GPe and SNr to understand the transformation that takes place between these nuclei. Given the role of basal ganglia in learning sensorimotor transformations, we are currently also recording activity in the striatum, GPe and SNr while mice perform a visual sensorimotor go/ no go task.

Disclosures: J. Fabre: None. **A.J. Peters:** None. **M. Carandini:** None. **K.D. Harris:** None.

Poster

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Title: The functional role of inhibitory neurons in the brainstem circuit mediating the optokinetic reflex

Authors: *A. LAVOIE, Y. HE, J. LIU, B. LIU;
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Abstract: Reflexes are critical to sustain life. For instance, the optokinetic reflex (OKR) is an involuntary eye movement that stabilizes retinal images. The OKR is mediated by neural circuits in the brainstem accessory optic system (AOS). Interestingly, there are various types of AOS neurons, defined by their anatomy, molecular markers, and functional properties, such as glutamatergic excitatory neurons and GABAergic inhibitory neurons. It is well known that AOS nuclei consist of rich population of inhibitory neurons. However, it remains unknown whether this genetically-defined population encodes different visual information compared to its counterpart, the excitatory population. Nor have the functional role of inhibitory neurons in the OKR been completely understood. To address these questions, I paired in vivo electrophysiology, optogenetics and chemogenetics. I found that inhibitory neurons differed in their functional properties and played an important role in modulating the functional properties of AOS neurons. These findings provide insights on how visual information is processed by brainstem circuits to insure appropriate reflexive behavior.

Disclosures: A. Lavoie: None. **Y. He:** None. **J. Liu:** None. **B. Liu:** None.

Poster

P458. Subcortical Visual Pathways I

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Program #/Poster #: P458.10

Topic: D.07. Vision

Title: Input from torus longitudinalis drives binocularity and spatial summation in the larval zebrafish tectum

Authors: A. L. TESMER, N. P. FIELDS, ***E. ROBLES**;
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Abstract: A common feature of vertebrate visual systems is that lower-order and higher-order visual areas are reciprocally connected. Feedforward projections confer visual responsiveness to higher-order visual neurons while feedback projections likely serve to modulate the responses of

lower-order visual neurons in a context-dependent manner. Optic tectum is the largest first-order visual brain area in zebrafish and is reciprocally connected with the torus longitudinalis (TL), a second-order visual brain area that does not receive retinal input. The functional role for feedback projections from TL to tectum has not been previously demonstrated. We demonstrate that TL feedback projections to tectum drive binocular integration and spatial summation in a specific tectal circuit. We performed genetically targeted, cell type-specific functional imaging in tectal pyramidal neurons (PyrNs) and their two input neuron populations: retinal ganglion cells (RGCs) and neurons in TL. PyrNs encode gradual changes in scene luminance using a complement of three distinct response classes that encode different light intensity ranges. Functional imaging of RGC inputs to tectum suggest that these response classes originate in the retina, supporting a model in which RGC input specifies PyrN functional class. In contrast, our data suggest that TL input serves to endow PyrNs with large, compound receptive fields that span both retinal hemifields. These findings reveal a novel role for the zebrafish TL in binocular integration and spatial summation.

Disclosures: A.L. Tesmer: None. **N.P. Fields:** None. **E. Robles:** None.

Poster

P511. Cerebellum: Cortex and Nuclei I

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Program #/Poster #: P511.01

Topic: E.02. Cerebellum

Support: European Union's Horizon 2020 Framework Programme for Research and Innovation under the Specific Grant Agreement No. 945539 (Human Brain Project SGA3)

Title: Non-linear frequency-dependence of neurovascular coupling in the cerebellar cortex: the vasodilation-vasoconstriction competition hypothesis

Authors: G. GAGLIANO[1], A. MONTEVERDI[1,3], S. CASALI[1], U. LAFORENZA[2], C. GANDINI WHEELER-KINGSHOTT[1,4], L. MAPELLI[1], ***E. D'ANGELO**[1,3];
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Abstract: Neurovascular coupling (NVC) is the process adjusting local cerebral blood flow according to neuronal activity (NA). Although NVC provides the basis for the blood-oxygen-level-dependent (BOLD) effect used in functional MRI (fMRI), the relationship between NVC and NA is still to be elucidated in detail. Recent studies in humans reported non-linear BOLD signals in the cerebellum during the execution of motor tasks with intensity-graded grip force application, with differences in vermis and hemisphere regions. This observation led to the

investigation of the NVC/NA relationship using a range of input frequencies in acute mouse cerebellar slices of vermis and hemisphere. Time-lapse imaging in bright-field conditions was used to detect capillary diameter changes following mossy fibers activation at different frequencies. Our results showed that capillary diameters increased in response to mossy fiber activation in the 6-300 Hz range, with a marked inflection around 50 Hz (vermis) and 100 Hz (hemisphere). The corresponding NA was recorded using high-density multi-electrode arrays and was correlated to capillary dynamics through a computational model, thus allowing us to dissect the main components of cerebellar granular layer responses recorded as local field potentials. In a previous work, we reported that in the cerebellar granular layer, NVC involves a balance between the NMDAR-NO pathway driving vasodilation and the mGluRs-20HETE pathway driving vasoconstriction. Simulations showed that the NMDAR-mediated component of the granular layer response was sufficient to explain the time-course of the capillary dilation but not its non-linear frequency-dependence, suggesting that the mGluRs-20HETE pathway could play a role at intermediate frequencies range. These parallel control pathways imply a *vasodilation-vasoconstriction competition hypothesis* that could adapt local hemodynamics at the microscale bearing implications for the interpretation of fMRI signals.

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Poster

P511. Cerebellum: Cortex and Nuclei I

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Topic: E.02. Cerebellum

Support: EU MSCA NEUSEQBOT (891774)
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R01MH093727

Title: Phyllum: a phy plugin for automatic cell classification of high-density neural recordings in the cerebellum

Authors: *F. NAVEROS[1,2], A. SÁNCHEZ LÓPEZ[1], J. F. MEDINA[1];
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Abstract: New high-density neural probes like Neuropixels offer unprecedented access to the brain because they allow us to record the activity of thousands of neurons simultaneously. To decipher the code embedded within the spike activity of the recorded neural populations, it is

critical to develop classification tools that can be used to assign these spikes to specific cell types. Here, we introduce *Phyllum*, a software package for automatic layer identification and cell classification of high-density electrophysiological recordings in the cerebellar cortex. *Phyllum* is designed to work with spike data that has been sorted with Kilosort and loaded into Phy - a widely-used graphical user interface optimized for curating data collected with high-density multielectrode arrays. We analyzed a series of Neuropixels recordings from different regions of the mouse cerebellar cortex in which the location of the probe was marked with a fluorescent dye and could be confirmed histologically. Neurons were classified with *Phyllum* using an automatic two-step algorithm that involved pre-assigning each individual spike cluster to one of the three layers within the cerebellar cortex (granular, Purkinje or molecular), followed by cell identification based on firing statistics and waveform properties. We found that this two-step process allowed *Phyllum* to outperform previous cell classification algorithms designed for analysis of single neuron recordings. To facilitate distribution and implementation of our software package, we are making *Phyllum* available as an open-source plugin for Phy.

Disclosures: F. Naveros: None. **A. Sánchez López:** None. **J.F. Medina:** None.

Poster

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Topic: E.02. Cerebellum

Support: R01NS112917
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EU MSCA NEUSEQBOT (891774)

Title: Neuropixels in the mouse cerebellar cortex

Authors: *A. SÁNCHEZ-LÓPEZ[1], S. OHMAE[1], F. NAVEROS[1,2], M. HEMELT[3], C. HULL[3], J. MEDINA[1];
[1]Neurosci., Baylor Col. of Med., Houston, TX; [2]Computer architecture and technology, Univ. of Granada, Granada, Spain; [3]Neurobio., Duke Univ. Sch. of Med., Durham, NC

Abstract: Much of what we know about the function of the cerebellar cortex comes from recordings of Purkinje cells in awake and behaving animals, which are typically made one cell at a time. In contrast, there is less information available about the activity of other cell types in the cerebellar cortex, or how these different cell types interact with each other. To fill this gap, we are currently using high-density Neuropixels probes to record the activity of neuron ensembles in two different regions of the mouse cerebellar cortex known to play a role in motor learning.

Here, we show that: (1) Neuropixels probes provide excellent stability and signal-to-noise during recordings in head-fixed mice, allowing us to measure the spontaneous, sensory and motor-related activity of 50-100 neurons simultaneously for long periods of time often lasting >3 hours, (2) Automatic spike sorting with Kilosort can achieve performance on par with that of manual sorting by expert users (<2% false positives and negatives), as long as the data is curated taking into account the known and idiosyncratic properties of cerebellar neurons, (3) Cell-type identity, which we are validating with opto-tagging experiments in some cases, can be ascertained by inspecting the waveform and the firing-pattern statistics (autocorrelogram, inter-spike-interval distribution, etc,) of each individual neuron, as well as its layer location within the cerebellar cortex, (4) Local interactions between neighboring neurons belonging to either the same or to different cell-specific groups, are common and easily detectable in crosscorrelograms. Altogether, our results provide the foundation necessary to analyze in vivo interactions between different cell types in the mouse cerebellum, providing a first glimpse at how the population activity is orchestrated during behavior.

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Poster

P511. Cerebellum: Cortex and Nuclei I

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Topic: E.02. Cerebellum

Support: Wellcome grant 536070, D-TRA, 173078

Title: Signal transformations along the cerebellar output pathway in behaving mice

Authors: *M. BEAU[1], R. RATTO[1], M. VARADI[1], D. COHEN[2], D. KOSTADINOV[1], M. HAUSSER[1];
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Abstract: To understand cerebellar function, it is crucial to understand how cerebellar output is generated. This remains an open question: the interplay between the Purkinje cells (PCs) -- the output neurons of the cerebellar cortex -- and the cerebellar nuclear cells (NCs) -- the ultimate output neurons of the cerebellum -- remains poorly understood. PCs are GABAergic, and they could influence NC output either by directly inhibiting them, or by entraining them via rebound spiking. Which of these two modes prevails in vivo in a physiological setting, and how this depends on PC synchrony, remains controversial.
We are addressing this problem by recording simultaneously from PCs and NCs using Neuropixels silicon probes inserted at an angle that allows us to reach monosynaptically

connected PCs and NCs. Using a combination of cell-type specific optogenetic tagging, prior knowledge about the firing properties of cerebellar cell types, and post-hoc 3D atlas registration of our recording sites, we identified PC (simple and complex spikes) and NC units in lobule simplex and interpositus nucleus respectively. We executed these recordings in mice performing a variety of behaviours: a motor task involving forelimbs coordination (wheel steering), locomotion, and reward prediction. We used cross-correlations to infer putative monosynaptic inhibition between PCs and NCs separated by up to a millimeter of tissue. We devised novel analysis strategies to account for sub-millisecond synchrony among local populations of PCs in order to disentangle direct inhibitory connections from confounding correlations caused by the synchrony of neighbouring PCs. Observing the relative firing of putatively connected PCs and NCs during behaviour, we found different signal transmission regimes in various behavioural contexts: anticorrelation during movement (wheel steering, locomotion), suggesting that PC inhibition predominates; and a positive correlation during reward prediction, suggesting that mossy fibres excitation predominates. We were also able to characterize the impact of complex spikes on nuclear firing: unexpectedly, nuclear cells were not inhibited but rather excited concomitantly with complex spikes, suggesting a domination of direct climbing fibre input over PC inhibition. These recordings constitute the first instance of direct readout of the impact of Purkinje simple and complex spikes on their target nuclear neurons, and shed light on the long debated signal transformations taking place along the cerebellar output pathway during behaviour.

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Poster

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Topic: E.02. Cerebellum

Support: NIH MH114047

Title: Structured connectivity of the cerebellar cortex enables noise-resilient preprocessing for pattern association

Authors: *T. NGUYEN[1], L. THOMAS[1], J. RHOADES[1], I. RICCHI[1], C. YUAN[1], A. SHERIDAN[2], D. HILDEBRAND[1], J. FUNKE[2], W. REGEHR[1], W.-C. A. LEE[3];
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Abstract: The cerebellum is critical for coordinated motor control and is implicated in complex behaviors, such as cognition and emotion. For motor coordination, the network is thought to detect and correct errors between expected and executed motor commands according to the forward model. How the network performs this task in the face of environmental and neural noise is unclear. The main feedforward circuit is relatively simple. Mossy fibers (MFs) carrying information from across the central nervous system provide input to large numbers of granule cells (GrCs) in the first layer of the network. The massive number of GrCs then converge by way of their parallel fibers in the second layer of the network to provide input to Purkinje Cells (PCs), the sole output of the cerebellar cortex. Influential theories on how information is processed that were established decades ago largely assumed random connectivity across the network; information theoretical models show that random connectivity can increase the encoding capacity of the first layer of the network. However, maximizing encoding capacity makes networks less resilient to noise. To understand how the circuit may address the tradeoff between encoding capacity and resilience, we used automated large-scale transmission electron microscopy and an automatic image segmentation pipeline using convolutional neural networks to map connectivity in the cerebellar cortex. In contrast to standard random connectivity models, we find that GrCs selectively sample MF boutons. Specifically, GrCs redundantly sample MF boutons, potentially increasing the resilience of the network to noise. GrCs also oversample specific MF boutons, potentially enhancing the transmission of specific information. Furthermore, adjacent PCs receive synapses from largely different populations of GrCs. However, PCs sample selectively from GrCs sharing MF bouton inputs. Taken together, we find redundant and selective connectivity across multiple network layers in the cerebellum that may enhance circuit resilience and help amplify and distinguish small differences in input patterns for a pattern association network.

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Poster

P511. Cerebellum: Cortex and Nuclei I

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Support: ERC Grant LOCOLEARN 866237
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Title: The role of distinct cerebellar circuit neurons in locomotion and locomotor learning

Authors: *J. R. JACOBS[1], M. R. CAREY[2];
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Abstract: The cerebellum is a brain structure involved in various sensorimotor and cognitive functions. For locomotor behaviors, the cerebellum is critical for keeping movements calibrated and coordinated across the body. Further, within the cerebellar circuit, various cell types have been mapped to distinct functions required for proper execution of movement. Here, we sought to ascertain how various cerebellar circuit neurons contribute to overground locomotion and locomotor learning on a split-belt treadmill in adult mice. We targeted Purkinje cells, granule cells, and a combination of granule cells and mossy fiber inputs for ablation via cell-type specific, cre-recombinase mediated expression of the diphtheria toxin receptor (DTR) and subsequent intraperitoneal administration of diphtheria toxin (DT). We found that ablations across all cell types caused an ataxic phenotype and impairments in locomotor learning, both of which scaled with the degree of cell death. Generally, the observed phenotypes in the two behaviours were similar to those previously reported for ataxic mutant mice, as forward motion of individual limbs was largely spared, while other 3D limb movements and interlimb coordination were impaired. Surprisingly, we also found that incomplete granule cell ablation led to outcomes commensurate with near/complete loss of Purkinje cells in both learning and overground locomotion. Overall, our results suggest that cell-specific manipulations within the cerebellar circuit yield overlapping phenotypes but gradations in outcome are observed in a cell-type dependent manner.

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Poster

P511. Cerebellum: Cortex and Nuclei I

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Topic: E.02. Cerebellum

Support: NIH Grant P30-DA048742
NIH Grant R01-NS112518

Title: Chronic, mesoscale Ca2+ imaging of cerebellar Purkinje cells in awake behaving mice using single photon microscopy

Authors: *M. L. STRENG[1], R. E. CARTER[2], K. TOGNERI[1], V. RAJENDRAN[1], S. B. KODANDARAMAIAH[1], E. I. KROOK-MAGNUSON[3], T. J. EBNER[4];
[2]Neurosci., [1]Univ. of Minnesota, Minneapolis, MN; [3]Univ. of Minnesota Syst., Minneapolis, MN; [4]Neurosci., Univ. of Minnesota Dept. of Neurosci., Minneapolis, MN

Abstract: A major challenge in cerebellar physiology is determining how the highly conserved cytoarchitecture of the cerebellar cortex underlies its contributions to ongoing behavior. Clusters of cerebellar Purkinje cells can be characterized by their parasagittal organization, expression of molecular markers, or by sources of common inputs: climbing fibers or parallel fibers. While a wealth of information is known about this cytoarchitectural organization, it is unclear how the functional information conveyed by Purkinje cells is organized throughout the cerebellar cortex, as most observations of Purkinje cell activity have been limited to individual cells or local populations. We recently developed a method for mesoscale recording of Purkinje cell Ca^{2+} activity in head fixed, behaving mice using transparent polymer windows of ~ $7mm^2$. This technique allows for the chronic recording of individual Purkinje cells across multiple folia, both vermis and simplex regions, bilaterally. Single photon Ca^{2+} imaging in PcP2-GCaMP6s mice, which exhibit sparse expression of GCaMP6s in Purkinje cells, enabled us to separately identify Ca^{2+} modulation that could be attributed to somatic activity (presumably partially reflective of parallel fiber modulation of simple spike activity) and dendritic activity (presumably reflective of climbing fiber driven complex spike activity) within the same recordings (consisting of ~100s of Purkinje cells). The chronicity of the approach allowed for recordings to be obtained in n = 6 mice over a period of ~3 months, with a minimum of ten recording sessions per mouse.To characterize Purkinje cell dendritic and somatic information, we performed blind source separation using the spatial Independent Component Analysis (sICA) algorithm JADER. At higher magnification, sICA identified independent components (ICs) corresponding to individual Purkinje cell dendrites or somata. The time courses of dendritic and somatic ICs appeared consistent with complex spike events and changes in simple spike activity, respectively. At the mesoscale level, sICA identified local populations of Purkinje cells, in which dendritic ICs were organized in the parasagittal domain, whereas somatic ICs were organized across a folium in the medial-lateral domain. Both dendritic and somatic ICs were highly modulated during behavior. Together, these results highlight an exciting new technique to characterize the functional properties of Purkinje cell populations across the cerebellar cortex. They also shed new light on the functional and spatial organization(s) of climbing fiber and parallel fibers systems and their ultimate influence on Purkinje cell output.

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Poster

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Topic: E.02. Cerebellum

Support: NCBS Graduate Student Fellowship, DAE, India

Title: Identifying Purkinje neuron state from calcium imaging data using machine learning

Authors: *A. VARMA[1], S. UDUPA[2], M. SENGUPTA[3], P. GHOSH[2], V. THIRUMALAI[1]; [1]Natl. Ctr. for Biol Sci., Bangalore, India; [2]Indian Inst. of Sci., Bangalore, India; [3]Washington Univ. Sch. of Med. in St Louis, St Louis, MO

Abstract: Purkinje neurons (PNs), the GABA-ergic principal neurons of the cerebellum, exhibit membrane potential bistability, firing either tonically or in bursts. Several studies have implicated the role of a population code in cerebellar function, with bistability adding an extra layer of complexity to the code. Calcium imaging is the method of choice to interrogate the activity of neuronal populations, however it does poorly at reporting the membrane potential state or the spiking patterns. Nevertheless, we hypothesised that the calcium signal might potentially contain information about the state of the PN.To bridge the gap between electrophysiology and imaging, we performed simultaneous patch-clamp electrophysiology and calcium imaging on larval zebrafish PNs. We used this ground truth dataset to develop a method of reconstructing the calcium signal given an electrophysiological trace. The calcium signal can be taken to be the convolution of spike timings with a GCaMP kernel. However, PNs have two sources of calcium - simple spikes (SS) and climbing fiber (CF) input. We treated each source as having its independent kernel and linearly summed their convolutions with corresponding event timings to produce a calcium signal reconstruction. The optimal combination of SS and CF kernels was found by minimizing the error between the reconstruction and the ground truth. We find that this method generalizes well across cells, and is independent of the state of the neuron.We then compiled electrophysiological recordings from PNs, grouped them by state, and applied this method on them to generate a large dataset of state-labelled calcium signal reconstructions (N=957 traces, from n=138 recordings). This data was used to train a 1-D convolutional neural network (1-D CNN) model to classify the state of a Purkinje neuron based on its calcium signal alone. A model with a single convolutional layer with 12 kernels, 5 samples long, and a stride length of 1 sample, performed best, with >85% accuracy on both training data as well as previously unseen data. Moreover, this network, which was trained only on reconstructions, identified PN state accurately even when presented with traces obtained from experiments. This tool opens up new avenues of research into understanding the role of Purkinje neuron bistability in cerebellar circuit function.

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Poster

P511. Cerebellum: Cortex and Nuclei I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P511.09

Topic: E.02. Cerebellum

Support: ANR-18-CE37-0006-01, WalkingCrossingNeurons
ANR-17-EURE-0017

Title: Acquisition of a complex locomotor task: activity of cerebellar molecular layer interneurons and gait dynamics

Authors: *A. ANDRY[1], H. STEIN[2], J. GABILLET[1], C. BATIFOL[1], N. CAYCO GAJIC[2], M. GRAUPNER[1];
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Abstract: The cerebellum is one of the key brain regions involved in motor coordination and learning. It is known that the cerebellum processes sensorimotor information to learn and refine the timing of movement patterns. However, the cellular underpinnings of such sensorimotor input integration and processing remain largely unknown. By combining calcium imaging with behavioral analysis, we study the role of a fundamental microcircuit for sensorimotor integration: the molecular layer interneuron (MLI) network, which provides a strong control over the output cells of the cerebellar cortex - the Purkinje cells. For this purpose, we implemented a complex locomotor task which allows us to perform calcium imaging while mice learn to walk on a runged treadmill over several sessions stretching over multiple days. To record longitudinally the activity in a large population of MLIs in lobulus simplex, GCaMP7f is expressed by stereotaxic injections of floxed AAV viral vectors of parvalbumin Cre mice. The runged treadmill is motorized which requires the animal to make steps of discrete lengths from rung to rung, involving a high level of sensorimotor integration. The high-speed behavioral video recordings are subsequently analyzed using DeepLabCut to extract paw locations. Individual paw trajectories are segregated into swing and stance phases based on relative speed and distance to the rungs.
Molecular layer interneurons in the cerebellar simplex exhibit pronounced locomotion-related activity: fluorescence increases globally in the MLI population upon locomotion onset and shows rich dynamics during walking. We first asked whether MLI calcium dynamics are related to the stepping pattern of the animal. To check for this, we aligned and averaged cell-specific transients to the beginning of swings. We found that a large proportion of recorded MLIs show swing-related transients consisting in fluorescence increases or decreases or combinations of both, depending on the cell. Swing-related transients of individual cells are paw-specific, as one would expect from the forelimb motor region. Our behavioral analysis of paw movements on the runged treadmill revealed that steps get longer and faster over the course of learning across days, consistent with results reported from the Erasmus ladder. While both novice and expert mice showed antiphasic paw movements of the forelimbs, fixed phase relations between hind- and forelimbs developed over days, resulting in a more regular, stability-centered gait. Our results shed light on the involvement of the cerebellar interneurons in generating coordinated movement in mammals.

Disclosures: A. Andry: None. **H. Stein:** None. **J. Gabillet:** None. **C. Batifol:** None. **N. Cayco Gajic:** None. **M. Graupner:** None.

Poster

P511. Cerebellum: Cortex and Nuclei I

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Program #/Poster #: P511.10

Topic: E.02. Cerebellum

Support: F31 NS113395-03
R01 NS114430

Title: Cerebellar associative learning underlies reach adaptation

Authors: *D. J. CALAME, A. L. PERSON;
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Abstract: A leading hypothesis of cerebellar function is that the cerebellum shapes a feedforward control policy to calibrate motor actions to meet a goal. Such a control policy is thought to be generated by Purkinje cells (PCs) which act as associative learners, shaping their simple-spike firing in response to cerebellar inputs based on complex spike-mediated plasticity. A critical component of this hypothesis is that PCs learn to use descending neocortical commands and sensory afferents to form an anticipatory control policy, and if these inputs unexpectedly change, PCs adapt to reestablish control. Here, we test this hypothesis by optogenetically stimulating cerebellar mossy fiber afferents originating in the basal pontine nucleus during a skilled reaching movement in mice (ChR2, 50-ms train). When stimulating in closed-loop during every trial, reach kinematics were perturbed time locked to the stimulus, but after several trials, reaches adapted to resemble baseline trials. During washout, when perturbation was removed, reaches displayed aftereffects in the direction opposing the initial deviation well timed to the position of stimulation, consistent with other paradigms of sensorimotor adaptation. Single-electrode and high-density cerebellar extracellular single-unit recordings targeted to the primary fissure, between Lob 4/5 and Simplex, showed that PCs encode reach kinematics, especially position of the paw, both at the population level and in individual cells. We hypothesized that time-locked adaptation could be explained by parallel fibers conferring continuous kinematic information to PCs throughout the reach, weighted by error-based plasticity. Using LASSO regression, we found that PC firing rates continuously represented upcoming kinematics across the reach epoch. To test whether motor context accounts for timed motor adaption, we modified the perturbation design by randomizing stimulus position during the stimulation block. Supporting the idea that sensorimotor context accounts for aftereffects, no aftereffects were observed following randomized-position perturbations. A cerebellar model recapitulated behavioral findings in both fixed and randomized-position

stimulation experiments and showed that reweighting inputs temporally associated with error occurred regardless of the position of a perturbation; however, aftereffects were accounted for by accumulated learning in temporally correlated inputs, absent in randomized-stimulation blocks. Together the data suggest that the cerebellum learns a control policy by ascribing cause and effect based on the reliability of temporal association of active inputs to errors.

Disclosures: D.J. Calame: None. **A.L. Person:** None.

Poster

P514. Cerebellum: Plasticity and Climbing Fibers

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P514.01

Topic: E.02. Cerebellum

Support: R01EY024712

Title: Cerebellar Abnormalities Associated with Loss of the Synaptic Scaffold CASK Follow a Degenerative Trajectory and Manifest Differentially Based on Sex

Authors: *P. PATEL[1], J. HEGERT[3], I. CRISTIAN[3], A. KERR[4], L. LACONTE[1], S. SRIVASTAVA[1], M. A. FOX[4], D. F. ENGLISH[2], K. MUKHERJEE[1];
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Abstract: A major open question in biomedical neuroscience is how singular genetic insults may affect different regions of the nervous system with varying specificity and severity. *CASK* is one such gene which associates with microcephaly and intellectual disability globally, but with a disproportionately severe hypoplasia of the cerebellum and hindbrain structures relative to forebrain structures. The CASK protein has been implicated in axon guidance, cell polarity, ion channel localization, active zone maintenance, and brain lamination. However, conflicting evidence exists on the nature of CASK's involvement in these functions and none explains the disproportionate effects on hindbrain structures. Here, we first demonstrate in human that complete absence of CASK produces no defect in brain lamination or neuronal migration but does manifest with severe cerebellar volume diminishment; this decrease in cerebellar volume is likely degenerative due to observed regression in motor skills and upregulation of glial fibrillary acid protein expression in the cerebellar cortex. Next, using a murine model where CASK is deleted from cerebellar cells post-developmentally, we mechanistically demonstrate that cerebellar hypoplasia is indeed due to late degeneration rather than developmental deficits. In the murine model, cerebellar development completes normally and gross motor skills are properly acquired by post-natal day 30 (p30), however by p100, cerebellar volume has decreased by approximately 60-70% with collapsing of the molecular layer and profound depletion of

cerebellar granule neurons (GCs) with no Purkinje cell (PC) death observed. Alterations in cerebellar morphology are accompanied by severe ataxia. In order to investigate the physiological mechanism of ataxia resultant from this degeneration, we will utilize single-unit recording from individual PCs to measure the simple spike to complex spike ratio observed in these projecting GABAergic cells – the only output of the cerebellar cortex. Recording from PCs in the context of GC loss, we will interrogate (1) the *in vivo* physiology of PCs in the absence of parallel fiber glutamatergic input and (2) the role of the complex/simple spike ratio on ataxia. Overall, the disproportionate cerebellar hypoplasia observed in human *CASK* mutations and murine models of CASK loss is degenerative in nature beginning at early ages. The X-linked nature of the *CASK* gene leads to expressivity patterns which result in plateau of degeneration in mosaic heterozygous mutations but devolving into full degeneration and epileptic encephalopathy in cases of hemizygous mutations in males having only one copy.

Disclosures: P. Patel: None. **J. Hegert:** None. **I. Cristian:** None. **A. Kerr:** None. **L. LaConte:** None. **S. Srivastava:** None. **M.A. Fox:** None. **D.F. English:** None. **K. Mukherjee:** None.

Poster

P514. Cerebellum: Plasticity and Climbing Fibers

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Program #/Poster #: P514.02

Topic: E.02. Cerebellum

Support: R01 NS072406
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R01 EY031972
Simons Collaboration on the global Brain 543031

Title: Pharmacological intervention reverses impaired learning in a mouse model of Fragile X syndrome, consistent with a saturation model

Authors: *A. SHAKHAWAT[1], J. BHATEJA[3], J. FOLTZ[2], J. L. RAYMOND[4];
[2]Neurobio., [1]Stanford Univ., Stanford, CA; [3]Dept. of Biol. Structure, Univ. of Washington, Seattle, WA; [4]Stanford Univ. Sch. of Med., Stanford, CA

Abstract: Experience-driven synaptic plasticity is thought to play a pivotal role in learning and memory. The enhancement of synaptic plasticity would seem to have the potential to enhance learning, however, it is often accompanied by the opposite—impaired learning. In the *Fmr1* knockout mouse model of Fragile X syndrome (FXS), there is enhanced associative long-term depression at the cerebellar parallel fiber-Purkinje cells synapses (PF-Purkinje cell LTD), but impaired cerebellum-dependent eye blink conditioning (Koekkoek et al. 2005). Here, we characterized oculomotor learning in Purkinje cell-specific *Fmr1* knockouts (L7- *Fmr1* mutant)

and used behavioral and pharmacological manipulations to test a saturation model of the detrimental effect of enhanced associative plasticity on learning. L7- *Fmr1* mutant mice exhibited the same, highly specific pattern of vestibular ocular reflex (VOR) learning deficits that we have found previously in other lines of mice with enhanced *or* impaired PF-Purkinje cell LTD. In particular, the L7- *Fmr1* mutant mice were impaired on learning to increase the VOR gain, whereas learning to decrease the VOR gain was spared. Moreover, the impairment of VOR-increase learning was more severe for high frequency (1 Hz) visual-vestibular training than low frequency (0.5Hz) training. Our previous experimental and theoretical work (Nguyen-Vu et al, 2016) suggested that the enhancement of PF-Purkinje cell LTD might impair learning by allowing spontaneous activity in the cerebellum to saturate LTD prior to training, making this mechanism unavailable to be recruited at the appropriate parallel-Purkinje cell synapses to support learning. This saturation model makes two predictions about interventions to reverse the learning deficits caused by enhanced synaptic plasticity, which were confirmed in our experiments. First, behavioral pre-training designed to reverse LTD at the PF-Purkinje cell synapses eliminated the oculomotor learning deficit in the L7- *Fmr1* mutant mice. Second, a pharmacological manipulation designed to suppress neural activity and thereby limit the induction of associative LTD during the pre-training period also reversed the learning deficit in the L7- *Fmr1* mutant mice. These results could have implications for the development of novel clinical approaches for patients with autism and other disorders caused by enhanced synaptic plasticity.

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Poster

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NIH R01 EY031972
Simons Collaboration on the Global Brain 543031

Title: Comparison of the calcium responses of Purkinje cells in the cerebellar flocculus to retinal slip and non-visual input

Authors: *A. S. FANNING, A. M. SHAKHAWAT, J. L. RAYMOND;
Neurobio., Stanford Univ., Stanford, CA

Abstract: Climbing fiber input to the cerebellum carries instructive signals that guide plasticity and learning by eliciting complex spikes accompanied by large calcium transients in Purkinje cells. In the flocculus of the cerebellum, climbing fibers have been viewed as encoding oculomotor errors in sensory coordinates, because the complex spikes in Purkinje cells convey retinal slip information. However, in the same neurons, complex spikes also can be driven by non-visual input. To assess their role in contributing to plasticity, we compared the calcium responses driven by the visual and non-visual input to these Purkinje cells. Although both drive complex spiking, multiple mechanisms can modify or shunt the calcium response that accompanies a complex spike. Therefore we recorded the calcium responses of a population of GCaMP6f-expressing Purkinje cells in awake behaving mice during optokinetic and/or vestibular stimulation to compare the calcium responses to these visual and non-visual inputs. During visual (optokinetic) stimuli and pairing of vestibular and visual stimuli, the calcium level increased during contraversive retinal image motion. During performance of the vestibulo-ocular reflex in the dark, calcium increased during contraversive head rotation and the associated ipsiverse eye movements. The amplitude of the non-visual calcium response was comparable to that during conditions with retinal image motion present that are known to induce oculomotor learning. Thus, the population calcium responses of Purkinje cells in the cerebellar flocculus to visual and non-visual input are similar to what has been reported previously for complex spikes, suggesting that multimodal instructive signals control the synaptic plasticity supporting oculomotor learning.

Disclosures: A.S. Fanning: None. **A.M. Shakhawat:** None. **J.L. Raymond:** None.

Poster

P514. Cerebellum: Plasticity and Climbing Fibers

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Topic: E.02. Cerebellum

Support: ANR SpinoCereLoco

Title: In vivo two-photon imaging of Purkinje cell activity during adaptation to sensorimotor mismatch

Authors: V. NGUYEN, *B. M. STELL;
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Abstract: To probe the internal models used by the cerebellar cortex during movement, we imaged Purkinje cells during adaptation to sensorimotor mismatches introduced into a simple reaching paradigm. A lick port was positioned in front of head-fixed mice and its lateral position was controlled in a 1:1 ratio via an electromechanically coupled joystick (gain = 1). Water

rewards were delivered from the lick port when it was positioned approximately 13 mm lateral to the center of the mouth, and could be retrieved when mice used the joystick to recenter the lick port in front of its mouth. This paradigm has several advantages: i) mice learn it very quickly (412 ± 110 trials on the 5th day on the setup; n = 8 mice), ii) sensorimotor mismatches can be repeatedly introduced within sessions by alternating different gains between the joystick and the lick port, iii) it engages cerebellar cortex on the surface of lobules IV/V of the vermis and simplex, which are easily accessible to *in vivo* electrophysiology and 2-photon imaging. When the gain between the joystick and the lick port was changed from 1 to 2 (i.e. the lick port moved twice as fast as the joystick) all mice overshot their mouths when attempting to retrieve water rewards but quickly adapted to the new gain within 5 - 10 trials by decreasing limb velocity and position, (n = 9 mice). Upon returning to control conditions mice increased limb velocity and position within 5 - 10 trials to adapt back to the control gain of 1. We used 2-photon imaging of GCaMP7 and GCaMP8 to monitor complex spikes in Purkinje cell dendrites during adaptation to changes in gain. Under control conditions we did not observe complex spikes linked to any component of the movement, but when the gain was increased, and sensorimotor mismatch occurred, we recorded movement related complex spikes from large regions of lobules IV/V in the vermis and simplex until the mouse adapted its limb kinematics to the new gain. These findings support classical models of supervised learning in which climbing fibers signal erroneous motor output to Purkinje cells via complex spikes.

Disclosures: V. Nguyen: None. **B.M. Stell:** None.

Poster

P514. Cerebellum: Plasticity and Climbing Fibers

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Fundação para a Ciência e a Tecnologia #PTDC/MED_NEU/30890/2017 (to MRC)

Title: Circuit dissection of neural instructive signals for associative cerebellar learning

Authors: *N. SILVA[1], J. RAMIREZ[1], D. PRITCHETT[1,2], M. CAREY[1];
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Abstract: Here we investigate the cellular basis of neural instructive signals for delay eyeblink conditioning, a cerebellum-dependent form of classical conditioning. In this paradigm, animals learn to blink to an initially neutral conditioned stimulus. Learning is driven by repeatedly pairing the conditioned stimulus with an air puff unconditioned stimulus (US) directed at the eye which drives an unconditioned response. Signals related to the air puff are conveyed to the cerebellum by climbing fibers, which provide strong synaptic inputs that elicit complex spikes in Purkinje cells. Complex spikes are thought to provide cellular instructive signals for plasticity that is hypothesized to underlie many forms of motor learning. However, the instructive stimuli that drive learning are often tightly associated with a reflexive movement - in this case, a blink in response to the air puff - that could be conveyed to the cerebellum via alternate pathways. It has therefore been difficult to dissociate climbing fiber activation from sensorimotor signals relating to these reflexive behavioral responses when identifying instructive signals for learning on the cellular level. Here we used cell-type specific perturbations of climbing fiber activity to test their necessity and sufficiency for learning. By combining behavioral, optogenetic and electrophysiological approaches, we further dissociate complex spike activity from reflexive movements. We find that optogenetically-evoked complex spikes can substitute for an air puff US to induce learning, even in the absence of an evoked blink. Further, evoked blinks that do not drive complex spikes do not drive learning. Finally, we found that in cases where expression of ChR2 in climbing fibers led to decreased probability of sensory-evoked complex spike responses, mice were unable to learn to an air puff US. However, under the same conditions, animals learned well to an optogenetically-evoked climbing fiber US. Together, these results support a necessary and sufficient role for climbing fibers as instructive signals for associative cerebellar learning.

Disclosures: N. Silva: None. **J. Ramirez:** None. **D. Pritchett:** None. **M. Carey:** None.

Poster

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Program #/Poster #: P514.06

Topic: E.02. Cerebellum

Support: 1DP2MH122398-01
T32CA154274

Title: Quantification of endogenous parvalbumin expression across Purkinje cell subtypes through CRISPR knockin of Cas9 fusions and the consequences for intrinsic firing properties

Authors: *C. BRANDENBURG[1], B. ALTAS[2], A. POULOPOULOS[3];
[1]Univ. of Maryland Sch. of Med., Baltimore, MD; [2]Pharmacol., Univ. of Maryland Sch. of Med., Baltimore, AL; [3]Pharmacol., Univ. of Maryland Sch. of Med., Baltimore, MD

Abstract: Although long used as a general PC marker, the calcium binding protein parvalbumin labels a subpopulation of PCs based on high and low/no expression, with a conserved distribution pattern across animals examined. The parvalbumin subtypes show differences in spontaneous firing that can be modified by altering calcium buffer content. These subtypes also show differential responses to potassium and calcium channel blockade, suggesting a mechanistic role for variability in PC intrinsic firing through differences in ion channel composition. Since antibody titrations for immunohistochemistry had varying results in quantifying parvalbumin expression, we use Cas9 fusions delivered to Purkinje cells via *in utero* electroporation to insert a fluorescent marker in the endogenous parvalbumin locus and quantify parvalbumin expression across cerebellar modules.

Disclosures: C. Brandenburg: None. **B. Altas:** None. **A. Poulopoulos:** None.

Poster

P514. Cerebellum: Plasticity and Climbing Fibers

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Program #/Poster #: P514.07

Topic: E.02. Cerebellum

Support: Wellcome Trust

Title: Fast and slow learning signals mediated by climbing fiber inputs to cerebellar Purkinje cells

Authors: *D. KOSTADINOV, M. BEAU, M. HAUSSER;
Univ. Col. London, London, United Kingdom

Abstract: Learning to execute goal-directed behaviors often consists of multiple phases - an early acquisition stage and later refinement stage. In the cerebellum, climbing fiber inputs to Purkinje cells are a key source of instructive signals for learning. These inputs are traditionally thought to drive learning by conveying sensorimotor errors, but there is increasing evidence that they can also implement other modes of learning, including reward-based reinforcement. It remains unclear, however, how these instructive signals emerge in climbing fibers, how they are used by the cerebellum during the acquisition and refinement phases of learning, and whether the same Purkinje cells mediate these different forms of learning. To address these questions, we have used two-photon calcium imaging to longitudinally record from the same populations of climbing fiber inputs to Purkinje cells over several weeks as head-fixed mice learned to execute

a virtual reality-based visuomotor integration task to obtain a delayed reward. Throughout training, we interleaved cued and unpredictable rewards between task trials. Once mice became proficient at the task, we introduced sensorimotor perturbations by altering the gain between object translation and wheel movement in single sessions. To quantify how sensory, motor, and reward-related activity were represented in climbing fiber inputs over learning, we built statistical models that use these task parameters to predict neural activity and compared how well models trained on sessions from expert mice were able to predict activity in naive mice and vice versa. We inferred several principles of how climbing fiber population dynamics change across learning on multiple time scales. In naïve mice, inputs already exhibited modulation by specific sensory and motor parameters, and these representations were generally maintained. In expert mice, responses to salient cues such as rewards became more reliable and could emerge in inputs that did not exhibit responses to these signals at the start of training. Moreover, responses to predictable stimuli were progressively suppressed. During gain change manipulations, the responses to operant rewards increased dynamically as animals adapted their behavior, supporting an active role in these signals for rapid learning. These data demonstrate that the same climbing fiber populations convey multimodal signals to Purkinje cells that are well-suited to drive learning of all variables that may assist animals in executing goal-directed behaviors, and that climbing fiber inputs, which are thought to be the main drivers of learning in the cerebellum, are themselves subject to instruction.

Disclosures: D. Kostadinov: None. **M. Beau:** None. **M. Hausser:** None.

Poster

P514. Cerebellum: Plasticity and Climbing Fibers

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Topic: E.02. Cerebellum

Support: European Union’s Horizon 2020 Framework Programme for Research and Innovation under the Specific Grant Agreement No. 945539 (Human Brain Project SGA3)

Title: Dendritic processing implements spike-timing dependent plasticity (STDP) in cerebellar Golgi cells

Authors: *T. SORBO, E. D'ANGELO;
Dept. of Brain and Behavioral Sciences, Unit of Neurophysiol., Univ. di Pavia, Pavia, Italy

Abstract: The existence of a precisely organized neuronal circuitry in the cerebellum is crucial for information processing and long-term synaptic plasticity induction. This latter, in various specific forms, determines persistent modifications in neuronal activity and synaptic

transmission providing the basis for learning and memory. In the cerebellum, it is through to be responsible for timing and adaption and, ultimately, for cerebellar control of movement and brain processing. Timing in the cerebellar circuit is tightly controlled by inhibitory interneurons. Among these, Golgi cells located in the granular layer play a primary role in modulating the input to Purkinje cells. Golgi cell receive inputs on apical and basal dendrites by parallel and mossy fibers, respectively. However, the mechanisms through which these neurons integrate input patterns is still unclear. In fact, little is known about how Golgi cells process the input signals, generate plasticity and modulate the gain and timing of signals conveyed to Purkinje cells. Recently, theoretical models predicted that spike-timing dependent plasticity (STDP) at mossy fiber-Golgi cells synapse would be a pivotal mechanism of Golgi cells inhibition at the cerebellar input stage. Furthermore, a computational model suggested the mechanisms through which Golgi cells dendrites might integrate and process the inputs transmitted by parallel and mossy fibers, implying that dendritic processing and plasticity in Golgi cells would depend on the spike time intervals between these two inputs. This project investigated, for the first time, Golgi cells STDP using whole-cell patch-clamp recordings in acute mice cerebellar slices. Mossy fibers spikes either anticipated or followed those on parallel fibers with a phase difference of ±75 ms. Indeed, STDP emerged, with peaks at ±20 ms, precisely matching computational predictions. Mechanistically, NMDA channel unblock at mossy fiber synapses on basal dendrites are the potential coincidence detectors of mossy and parallel fiber activity. This hypothesis is now under experimental testing. In summary, this work shows that dendritic processing is instrumental to STDP and opens new perspectives on the role of Golgi cells in determining learning and plasticity in the cerebellar circuit.

Disclosures: T. Sorbo: None. **E. D'Angelo:** None.

Poster

P514. Cerebellum: Plasticity and Climbing Fibers

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Topic: E.02. Cerebellum

Support: NIH Grant NS116854

Title: Excitatory and inhibitory synaptic currents in Olig2(+) eurydendroid neurons in larval zebrafish cerebellum during associative motor learning

Authors: *M. NAJAC, D. L. MCLEAN, I. M. RAMAN;
Neurobio., Northwestern Univ., Evanston, IL

Abstract: The larval zebrafish cerebellum is required for associative motor learning, such as learning to swim in response to a neutral visual stimulus that predicts a tactile stimulus to the tail.

During training, Purkinje cells alter their firing patterns, and the disruption of their activity prevents learning. Here, we studied excitatory and inhibitory synaptic currents (EPSCs and IPSCs) during learning in *Olig2(+)* eurydendroid neurons (ENs), a subset of Purkinje target cells, which are glutamatergic output neurons of the zebrafish cerebellum. We recorded EPSCs (at -60 mV) which result from direct input from parallel fibers and IPSCs (at 0 mV) from Purkinje cells, while monitoring fictive swimming from ventral root activity in immobilized larvae (dpf 6-8). The unconditioned stimulus was a mild electric tail shock triggering fictive swimming (unconditioned response, UR) and the conditioned stimulus was a low-intensity 2-s blue light. With repeated pairing of the shock to light offset, ~50% fish (n=22/41) acquired conditioned responses (CRs) after ~25 trials (range: 3-65). During fictive swimming (both CRs and URs), all *Olig2*(+) ENs (20/20) showed robust increases in EPSCs and IPSCs, with inhibition always far exceeding excitation, but only a few *Olig2*(+) ENs also showed light-associated synaptic activity without a motor response (6/20). Thus, synaptic activity was purely motor-associated in 70% of cells and mixed sensory-motor in 30%, with no obvious topographical distribution. The timing of swimming-associated PSCs differed for CRs and URs. For URs, PSCs in *Olig2*(+) ENs and the ventral root signal began simultaneously, but for CRs, PSCs preceded the ventral root signal by ~100 ms (n=21). The charge transfer during EPSCs as well as IPSCs was similar during CRs and URs that were not preceded by a CR. However, charge transfer was reduced during URs that followed CRs (IPSCs: n=18, p=0.01; EPSCs: n=20, p=0.002), consistent with previous observations in mice of reduced spiking to the US after learned responses in cerebellar output neurons. The US preceded by the CS evoked larger IPSCs than the US alone, even at the very first CS+US pairing. Over the course of learning, however, no gradual modification of synaptic currents was observed; the amount and timing of synaptic responses were simply toggled by the presence or absence of CRs. These results suggest that the synaptic plasticity leading to the emergence of CRs must occur upstream of parallel fiber synaptic signals, at the level of granule cells, consistent with imaging studies in mice, but that these modified synaptic inputs drive *Olig2*(+) ENs to contribute to the production of learned motor responses.

Disclosures: M. Najac: None. **D.L. McLean:** None. **I.M. Raman:** None.

Poster

P514. Cerebellum: Plasticity and Climbing Fibers

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Program #/Poster #: P514.10

Topic: E.02. Cerebellum

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Title: Chemoarchitecture of the human inferior olive

Authors: *G. SENGUL[1], U. TURE[2];
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Abstract: The inferior olive is the most prominent feature of the hindbrain, located in the ventrolateral part of the brainstem. It plays a major role in motor learning and motor coordination by providing the climbing fibers - one of the two major excitatory inputs to the cerebellar cortex. In this study, we have used numerous immunohistochemical markers to analyze the chemoarchitecture of the inferior olive of the human. A human brainstem was sectioned serially using a cryostat at a thickness of 60 µm, stained and observed under light microscopy. As in other animals, the inferior olive of the human consists of the principal, medial, and dorsal nuclei. We have analyzed transverse sections from Obex +7 and Obex +12, and observed substance P, tyrosine hydroxylase, neuropeptide Y, calbindin, calretinin, parvalbumin, calcitonin-gene-related peptide, choline acetyl transferase, cocaine-and amphetamine-related peptide (CART), GABA, glutamate, glycine, galanin, nitric oxide synthase, enkephalin, and vasoactive intestinal peptide immunoreactivities in all subnuclei - the principal, medial, and dorsal nuclei - of the inferior olive.

Disclosures: G. Sengul: None. **U. Ture:** None.

Poster

P520. Motor Circuits: Anatomy and Physiology

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P520.01

Topic: E.03. Basal Ganglia

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Programme "FIL-Quota Incentivante" of University of Parma and cosponsored by Fondazione Cariparma

Title: Laminar origin of corticostriate projections from prefrontal and rostral cingulate areas in the macaque

Authors: E. BORRA, M. RIZZO, M. LEO, *G. LUPPINO;
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Abstract: We have recently found that in the macaque frontal motor areas and frontal operculum corticostriatal (CSt) neurons projecting to the motor putamen are almost equally distributed in layers III, V, and VI, thus challenging the commonly assumed notion that CSt neurons in the macaque brain are primarily located in layer V (Borra et al. 2021, J Neurosci, 41:1455). In the

present study we examined whether this connectional feature applies also to prefrontal and rostral cingulate areas. To this aim, in 3 macaque monkeys we analyzed the laminar distribution of the CSt projections from the ipsilateral hemisphere after retrograde tracer injections in the caudate head (2 injections) and in the precommissural putamen (1 injection). After the tracer injections in the caudate head, cortical labeling was densest in prefrontal areas 9 and 8B and in cingulate areas rostral 24 and 32. Less dense labeling involved other lateral prefrontal areas (46, 45, and 8), area 14, and orbitofrontal areas 11, 13, and 12o. In areas rostral 24, 8B and in most of area 9, CSt labeled neurons involved in almost similar proportion mostly layers III and V. This laminar pattern was independent from variations in the local density of the labeled cells. A similar laminar pattern was observed also in area 45 and in orbitofrontal areas. However, in orbitofrontal areas there was a relatively high proportion of CSt labeled neurons also in layer VI (20% ca.). Furthermore, in a sector of area 8 there was labeling by far predominant in layer III. Finally, in areas 32, 14, and 46 and in the rostral part of area 9, labeled CSt cells were located predominantly in layer V. After the precommisural putamen tracer injection, in the prefrontal and rostral cingulate cortex the labeling was denser in areas 24 (mostly cingulate motor areas 24c and 24d), medial 9/8B, 46, 12r, and 12l. In all these areas labeled CSt cells mostly involved layer V. The present data suggest that, as for frontal motor areas, the laminar distribution pattern of CSt neurons largely varies independently from their density and from the projecting area/sector, but likely according to the target striatal zone. Thus, cortical areas appear to project in different ways to different zones of the striatum, so that different striatal zones are targets of characteristically weighted laminar projections from the various input areas.

Disclosures: E. Borra: None. **M. Rizzo:** None. **M. Leo:** None. **G. Luppino:** None.

Poster

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Program #/Poster #: P520.02

Topic: E.03. Basal Ganglia

Support: NIMH 5R01MH099114-08

Title: Altered spontaneous motor behavior and striatal circuit function in conditional mGluR5 knockout mice

Authors: *J. J. MARSHALL, J. XU, N.-H. YEH, T. NOMURA, J. N. ARMSTRONG, J. G. PARKER, A. CONTRACTOR;
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Abstract: Metabotropic glutamate receptor 5 (mGluR5) plays a crucial role in regulating plasticity at synaptic terminals formed by cortical pyramidal cells onto spiny projection neurons

(SPNs) in the striatum. Specifically, activation of mGluR5 is required for endocannabinoid dependent long-term depression (LTD) at these synapses. SPNs play key roles in regulating voluntary movement and reinforcement learning. Imbalanced activity between SPNs of the direct and indirect pathways (dSPNs and iSPNs) leads to abnormal motor function and is a feature of diseases affecting the basal ganglia, such as Parkinson's and Huntington's disease. However, the role of mGluR5 dependent plasticity in generating balanced spatiotemporal patterns of activity in these two pathways is unclear. To address this, we've generated mice that lack mGluR5 selectively in either direct or indirect pathway SPNs (dSPN and iSPN mGluR5 KO mice). Our characterization of these mice has revealed increased spontaneous locomotor activity in the open field in iSPN mGluR5 KO mice. Conversely, dSPN mGluR5 KO mice show decreased locomotor activity in the open field and when exposed to running wheels in the home cage. Additionally, dSPN knockout mice show reduced spontaneous expression of simple motor behaviors such as digging and nest building. While a variety of electrophysiological properties of dSPNs from these mice are normal when measured in an *in vitro* slice prep, *in vivo* microendoscopy shows that dSPN KO mice have changes in the spatiotemporal patterns of dSPN activation in the dorsal lateral striatum during spontaneous exploration of the open field. This demonstrates, therefore, that mGluR5 activation in dSPNs is required for the normal functioning of striatal circuits. Ongoing work is examining the role of mGluR5 dependent plasticity in mediating alterations in activity of the dorsal lateral striatum during motor learning.

Disclosures: J.J. Marshall: None. **J. Xu:** None. **N. Yeh:** None. **T. Nomura:** None. **J.N. Armstrong:** None. **J.G. Parker:** None. **A. Contractor:** None.

Poster

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Program #/Poster #: P520.03

Topic: E.03. Basal Ganglia

Support: European Research Council Consolidator Grant (646886–SynChI)
Israel Science Foundation grants (154/14 and 155/14)

Title: Collective dynamics of a sparse population of striatal neurons in freely moving mice

Authors: L. TIROSHI, Y. ATAMNA, *J. A. GOLDBERG;
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Abstract: Spiny projection neurons (SPNs) - the main population of striatal neurons - play a central role in movement initiation. Recent studies have shown that SPNs form spatially compact neural clusters of co-active neurons that code for the same types of movement. These clusters were extracted from groups of hundreds of SPNs that were imaged simultaneously with

microendoscopes. When endoscopically imaging densely packed neurons, an extensive background signal arising from neuropil activity is observed in addition to the somatic signals. Recent studies have characterized these neuropil signals and concluded that they are spatially highly-correlated and represent collective neural activity that is distinct from that of the somata. These results raise the question - could the spatially correlated neuropil signal be introducing spurious correlations among SPNs thereby creating an illusion of compact cell clusters operating together? To address this issue, we performed microendoscopic calcium imaging in a sparse population of putative SPNs (pSPNs, identified by immunoreactivity to either GAD or DARPP32). This approach allowed us to explore the collective pSPN activity while maintaining a good separation between the neuropil and somatic signals, as well as, between the signals from different somata. As a first step, we studied the relationship between pSPN activity and the self-initiated movements of freely moving mice. Next, mice underwent an operant conditioning paradigm, and we examined pSPN responses as they learned to associate an auditory cue with a reward. As previously reported, we found that pSPN activity is modulated around the onset of self-initiated movement, as well as following the presentation of task-related cues in the conditioning paradigm. Interestingly, we detected two types of responses around the various behavioral events - while the majority of responsive cells increased their activity following the relevant event, some pSPNs became significantly less active. By focusing on a sparse neuronal population we were able to track individual cells across imaging sessions. While the percentage of significantly responsive pSPNs remained stable, the identity of cells responding around the same type of behavioral event changed considerably from session to session. Finally, we found that the correlation between the activity of pSPN pairs does not depend on the distance between them, in contrast to previous reports of spatially compact clusters. Thus, by focusing on a sparse population of cells, we were able to uncover properties of the collective dynamics of striatal pSPNs that were not previously appreciated.

Disclosures: L. Tiroshi: None. **Y. Atamna:** None. **J.A. Goldberg:** None.

Poster

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Program #/Poster #: P520.04

Topic: E.03. Basal Ganglia

Support: NIH Center for Compulsive Behaviors Fellowship
NINDS Intramural NS003134

Title: Membrane potential dynamics during cholinergic transmission onto dopaminergic axons

Authors: *P. KRAMER, S. BRILL-WEIL, Z. KHALIQ;
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Abstract: Axons of dopamine neurons are locally modulated by striatal cholinergic interneurons through presynaptic nicotinic receptors (nAChRs). However, the nature of cholinergic transmission is unknown. The prevailing view based on electron microscopy is that cholinergic neurons signal largely through non-specific, slow volume transmission. Functional studies examining the regulation of striatal dopamine release show that synchronous activation of cholinergic interneurons can trigger dopamine release through activation of axonal nAChRs alone. Yet, the indirect nature of the methods used so far have been insufficient to allow precise examination of this axo-axonal transmission. To test this, therefore, we obtained patch-clamp recordings of the axonal membrane potential to precisely examine spontaneous and evoked cholinergic transmission onto striatal dopaminergic terminals. Our axonal recordings reveal the presence of clear phasic depolarizations that occur spontaneously at rates of 1-2 events per second, and which were abolished by nicotinic receptor antagonists. The cholinergic-evoked depolarizations had average rise times slightly less than 20 ms indicating the likely presence of a non-classical synapse. Tetrodotoxin-block of spike evoked transmission resulted in a 10-fold reduction in the frequency of spontaneous EPSPs but had no effect on amplitude, confirming that these events represent true miniature events resulting from quantal release. Paired-pulse stimulation of cholinergic interneurons with light-evoked opsins or electrical stimulation showed the marked synaptic depression. Importantly, we also show that the kinetics of nicotinic EPSPs are shaped by acetylcholinesterase, which limits the activation of nicotinic receptors distal to the site of ACh release. Thus, we provide the first comprehensive examination of membrane dynamics during axo-axonic synaptic transmission onto the dopaminergic axons. Although the large and extensively branching axonal arbors of dopamine neurons have led to the idea of global signaling, our data showing synaptic-like depolarizations suggests instead that local signaling within the arbor may also contribute to striatal subregion circuit dynamics.

Disclosures: P. Kramer: None. **S. Brill-Weil:** None. **Z. Khaliq:** None.

Poster

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Program #/Poster #: P520.05

Topic: E.03. Basal Ganglia

Support: R01 NS104089 NINDS

Title: Anatomical and functional characterization of a direct input from basolateral amygdala to dorsolateral striatum

Authors: *Z. B. HOBEL[1], J. L. PLOTKIN[1,2];
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Abstract: The dorsal striatum is crucial for motor learning and balance of action strategies, in part through dense inputs from sensory- and motor- related cortical areas. Limbic-associated brain regions, including the amygdala, also innervate the dorsal striatum, providing a mechanism for limbic-associated information to influence striatal action selection. Input to the striatum from the amygdala comes predominately from the basolateral nucleus (BLA) and densely innervates the dorsomedial (DMS) and ventral subdivisions of the striatum. If and how the BLA also innervates the functionally distinct neighboring dorsolateral striatum (DLS) is unclear. Here, we use a combination of anatomical circuit tracing, optogenetics, 2-photon imaging and slice electrophysiology to characterize the connectivity between the BLA and DLS in the mouse brain. We identified a population of DLS-projecting BLA neurons that is topographically distinct from the more numerous DMS-projecting population. These DLS-projecting BLA neurons form functional axo-spinous synapses on approximately 20-25% of DLS striatal spiny projection neurons. We further show that DLS- and DMS-projecting BLA neurons are integrated differently into prefrontal cortex circuits, with DMS-projecting BLA neurons sending axonal collaterals to the medial prefrontal cortex and DLS-projecting neurons sending axonal collaterals to more lateral prefrontal areas. Experiments aimed at understanding the role of this novel projection in modulating behavior are ongoing.

Disclosures: Z.B. Hobel: None. **J.L. Plotkin:** None.

Poster

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Program #/Poster #: P520.06

Topic: E.03. Basal Ganglia

Support: NIH Grant 1K99NS118053-01
Helen Hay Whitney Foundation
NIH Grant 5U19NS104649
Simons-Emory International Consortium on Motor Control

Title: Corticospinal populations broadcast complex motor signals to coordinated spinal and basal ganglia circuits.

Authors: *A. NELSON, B. ABDELMESIH, R. COSTA;
Columbia Univ., New York, NY

Abstract: How the brain and spinal cord transform neuronal activity into coordinated movement is a central question in neuroscience. Many models of motor control emphasize the role of sensorimotor cortex in directing movement, principally through the direct axonal projections corticospinal neurons (CSNs) make to the spinal cord. In addition to their intimate access to

motor output, CSNs also possess strikingly expansive supraspinal axon collaterals, the functional organization of which is largely unknown. Experimentally characterizing this architecture been limited by an inability to map, measure, and manipulate interconnected circuits defined by their synaptic targets. Using anatomical and electrophysiological circuit mapping techniques, we reveal dorsolateral striatum (DLS) as the preeminent target of CSN collateral innervation. Moreover, we show that CSNs are the only population of neurons that project directly to both spinal cord and DLS. CSNs evoke larger postsynaptic responses in D1- over D2-expressing striatal spiny projection neurons (SPNs), a bias that is mediated through more, rather than larger, synapses made on D1 SPNs. This supraspinal bias is reflected in the spinal cord: CSN populations that target D1 or D2 SPNs show preferential targeting of distinct spinal cord circuits, revealing coordinated spinal and forebrain connectivity. We combined multiphoton imaging of CSNs, transsynaptic rabies tracing from D1 or D2 SPNs, and a sequential lever press task to characterize the information encoded in the activity of CSNs and their collaterals in DLS. Contrary to more conventional perspectives, CSN populations encode not only individual movements and muscle output, but also higher-order information related to the onset and offset of motor sequences. Furthermore, similar patterns of complex activity are broadcast by CSN populations targeting different downstream striatal circuits. Our results reveal a logic of coordinated connectivity between forebrain and spinal cord circuits, where separate CSN modules broadcast similar muscle- and non-muscle-related information to downstream circuits, suggesting that differences in postsynaptic connectivity ultimately dictate motor specificity.

Disclosures: A. Nelson: None. **B. Abdelmesih:** None. **R. Costa:** None.

Poster

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Program #/Poster #: P520.07

Topic: E.03. Basal Ganglia

Support: European Research Council – FRONTHAL - 742595

Title: An alternative cortico-subcortical loop for motor coordination

Authors: *E. BOSZ[1,2], V. M. PLATTNER[3], M. A. DIANA[4], L. ACSADY[1];
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[3]Sainsbury Wellcome Ctr., London, United Kingdom; [4]Pierre et Marie Curie Univ., Paris, France

Abstract: Basal ganglia process frontal cortical signals which is then conveyed to thalamus via giant multisynaptic inhibitory terminals before they return to the cortex via the thalamocortical pathway, closing the loop. Unilateral interference with basal ganglia activity induces characteristic, unilateral rotation. The brainstem also contains several locomotor centers. Similar

to the basal ganglia the pontine reticular formation (PRF) also sends large multisynaptic terminals to the intralaminar thalamus (IL). Futhermore the PRF innervated by the basal ganglia. Activation of PRF fibers in the IL results in a strong motor phenotype, a complete behavioral arrest. In this study, we examined the cortical inputs of the PRF inhibitory cells (GlyT2+) and their function in order to assess whether frontal cortex is able to inhibit and/or pattern IL activity via a PRF inhibitory interface. We also tested the effect of optogenetic activation of inhibitory PRF cells on locomotor behavior. Injections of AAV-ChR2 into the frontal cortex (M2 and cingulate - Cg) of RBP4-Cre/Glyt2-eGFP double transgenic mice revealed that GlyT2+ neurons of the PRF receives L5 inputs. Cortical terminals contacted mostly mid caliber dendrites and spines of the GlyT2+ cells. In *in vitro* preparation photoactivation of frontal cortical fibers faithfully produced purely glutamatergic synaptic responses on PRF-GlyT2 cells. Juxtacellular recording and labelling in the PRF showed that photoactivation of frontal L5 cells evoked short latency APs with high probability in the PRF GlyT2+ cells at high stimulation frequency (10-20 Hz). Spontaneous rhythmic activity of Glyt2+ cells was tightly linked to the slow cortical oscillation. Pharmacological inactivation of the cortex led to decreased, irregular firing of the GlyT2+ neurons. Photoactivation of PRF Glyt2+ cells resulted in unilateral rotation. Photoactivation of PRF-IL GlyT2+ fibers led to significant decrease in the firing rate of IL cells *in vivo*, suggesting that frontal cortical activity may lead to decreased IL output in a selected IL cell population. Our results suggest that synchronous frontal cortical activity conveying a behavioral signal computed by frontal cortical regions can reliably activate inhibitory neurons of the PRF. PRF GlyT2 cells in turn transfer this to the IL as an inhibitory signal strongly affecting thalamocortical and thalamostriatal activity akin to the basal ganglia circuits.

Disclosures: E. Bosz: None. **V.M. Plattner:** None. **M.A. Diana:** None. **L. Acsady:** None.

Poster

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Topic: E.03. Basal Ganglia

Support: NIH Grant NS103037
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Title: Strengthening of cortico-striatal synapses from task-relevant motor cortex neurons during learning

Authors: F.-J. HWANG[1], R. H. ROTH[1], Y.-W. WU[2], Y. SUN[1], Y. LIU[1], J. B. DING[1]; [1]Stanford Univ., Stanford, CA; [2]Academia Sinica, Taipei, Taiwan

Abstract: The execution of skilled movements requires the coordinated activity of multiple brain regions and learning new motor skills involves adaptations of the connectivity between brain regions. The motor cortex and striatum are two key motor regions of the brain and play crucial roles in generating and organizing voluntary movement. Yet, how the synaptic connectivity between these regions is modulated during motor learning has not been defined. To assess this, we train mice on a single pellet reaching motor learning task and use the genetic fos-TRAP system to identify task-relevant neurons in the primary motor cortex (M1). We found that the degree of reactivation of this labeled neuronal population highly correlates with the level of mouse performance on the reaching task. By expressing channelrhodopsin (ChR2) in task-relevant neurons, we further probed the functional connectivity of cortico-striatal projection neurons onto spiny projection neurons (SPNs) in the dorsolateral striatum (DLS). Combining optogenetic stimulation and two-photon Ca2+ imaging in brain slices, we were able to identify the individual dendritic spines receiving inputs from task-relevant M1 neurons. We found that motor learning led to an increase in the number and strength of cortico-striatal inputs onto SPNs and that these inputs form local clusters along SPN dendrites. Together, our results indicate that a population of task-relevant neurons emerges in M1 during motor learning and that the projections of these neurons to the striatum are strengthened. This form of synaptic plasticity can functionally impact dendritic integration on dendrites of SPNs in the striatum and may play a role in the formation of precise motor sequences during learning.

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Poster

P579. Sensory Motor Systems I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P579.01

Topic: F.01. Neuroethology

Support: NIH R01NS110866
NSF CAREER 1943276
UCSB FRAP Awards

Title: An undergraduate team project to reconstruct neural circuits controlling Drosophila grooming behavior using an Electron Microscopy dataset for the fly's ventral nerve cord

Authors: N. HILL[1], P. LAWTON[1], K. ROSS[1], ***P. TANG**[1], M. TEITZ[1], D. WRIGHT[1], D. SYED[1], S. YOSHIKAWA[1], J. S. PHELPS[2], W.-C. A. LEE[3], J. H. SIMPSON[1];
[1]Univ. of California Santa Barbara, Santa Barbara, CA; [2]Harvard Univ., Cambridge, MA; [3]BCH / Harvard Med. Sch., Boston, MA

Abstract: Fruit flies perform sequential grooming behaviors, yet the neural pathways that coordinate these actions remain largely unknown. Recent, large-scale connectome efforts led to the collection of electron microscopy (EM) datasets for the fly nervous system, which allows for the investigation of neural circuits at synaptic resolution.
Since January 2021, a team of ~50 UCSB undergraduate students learned to trace neurons in the EM dataset of the adult female ventral nerve cord collected by the Lee and Tuthill labs (Phelps, et al 2021) using CATMAID, a collaborative tracing platform (Saalfeld et al 2009). This project provided remote research opportunities during COVID restrictions. Peer training is in development to incorporate new tracers and has expanded to include the Neuroglancer software/web interface (Dorkenwald et al 2020) as auto-segmentation data becomes available. Tracers were divided into three groups, led by student team leads and senior lab members. Each team reconstructed different neurons identified as critical for grooming in previous optogenetic activation screens:
I. wPN1 neurons are a pair of cholinergic interneurons that have dendrites in the wing neuropil and synaptic arbors in T3 on each side. These neurons were identified to affect coordinated wing grooming. GRASP data suggests that wPN1 receives synaptic input from wing campaniform sensilla, mechanosensory neurons embedded in the cuticle. A bundle of ~150 sensory neuron candidates has been partially reconstructed in the EM, and synaptic connections onto wPN1 have been identified by tracing.
II. 13A/13B inhibitory lineages: Neurons in the 13A/B lineage contain several distinct morphological types that project ipsilaterally in the T1 neuromere. Partial EM reconstructions have shown synaptic connections between some 13A neurons and subsets of leg motor neurons; some 13B neurons are indirectly connected to motor neurons through 13As. The catalog of projection types may illuminate how these inhibitory neurons contribute to leg coordination during grooming.
III. Mago-no-te (MGT): Activation of MGT promotes thoracic grooming. Light level and EM reconstruction of the pair of MGT neurons show two distinct branches. The ventral medial branch contains the post-synaptic sites between the T1 and T2 neuromeres, while the dorsal lateral branch contains a majority of the presynaptic sites in the T3 neuromere. Its pre-synaptic partners may include sensory neurons, a hypothesis that is being tested by EM neuron tracing. Complete tracing of these target neurons and mapping their synaptic connections will reveal new neural circuits that contribute to the regulation of fly grooming behavior.

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Poster

P579. Sensory Motor Systems I

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Program #/Poster #: P579.02

Topic: F.01. Neuroethology

Title: Roundup® And glyphosate exposure elicits proconvulsant behavior in caenorhabditis elegans

Authors: *A. S. NARAINE[1,2], I. SWEENEY[2], R. AKER[2], M. KALVEY[3], A. SURTEL[2], V. SHANBHAG[4], K. DAWSON-SCULLY[5,1];
[1]Max Planck Florida Inst. for Neurosci., Jupiter, FL; [2]Biol. Sci., Florida Atlantic Univ., Boca Raton, FL; [3]Lynn Univ., Boca Raton, FL; [4]Chem. and Physics, [5]Nova Southeastern Univ., Fort Lauderdale, FL

Abstract: As 3 billion pounds of herbicides are sprayed over farmlands every year, it is essential to advance our understanding of the influence that pesticides may have on neurological health and physiology of both humans and other animals. Herbicidal investigations, specifically those involving glyphosate-based herbicides, are often one-dimensional as the majority of studies examine glyphosate by itself, while farmers and the general public use commercial products which contain a myriad of chemicals in addition to glyphosate. Previous works have demonstrated neurotoxicity in *C. elegans*, but it has also been shown that humans suffer significant physiological effects at subtoxic thresholds. It was our contention that physiological effects could be exacerbated by dilutions far lower than those required to degrade neuronal tracts, and a rescue of those effects would demonstrate a significant dysregulation rather than outright neurodegeneration. We compared how glyphosate and Roundup® affect convulsant behavior in *C. elegans* and found that glyphosate (n=100) and Roundup® (n=104) both increase seizure-like behavior ($p<0.01$). Key to our initial hypothesis, we found that treatment with an antiepileptic drug (AED) rescued the prolonged convulsions ($p<0.001$). Notably, these effects were found at concentrations that are 1,000-fold dilutions of previous findings of neurotoxicity. In addition to prolonged convulsion, we discovered a significant percentage of nematodes exposed to Roundup® (33.5%) did not recover from their convulsions at all even compared to glyphosate (12.9%), however AED treatment completely alleviated the non-recovery status.

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Poster

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Program #/Poster #: P579.03

Topic: F.01. Neuroethology

Support: ANR-17-CE37-0019
CNRS-Uchicago collaboration grant

Title: Neural circuitry underlying sensorimotor decisions

Authors: *M. LEHMAN[1,2], S. AUTRAN[1], T. JOVANIC[1];
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Abstract: From finding nearby food to escaping a predator, animals must respond to sensory cues with appropriate motor actions in order to survive. The neural circuit mechanisms underlying the transformation of sensory information into appropriate motor output remain incompletely understood. It is for instance unclear how one action is selected among several possible responses to a single stimulus. In addition, how the selected action is specified by motor circuits depending on stimulus characteristics is not known either at the cellular and synaptic levels. To understand these mechanisms better, it would be important to map the entire neural circuitry from the sensory to the motor side in a model organism allowing functional testing of such circuits. Thanks to recent progress in connectomics, allowing the mapping of entire neural circuits in electron microscopy (EM) with synaptic resolution and to the genetic toolbox for cell-specific neuronal manipulations and monitoring, Drosophila larva presents itself as such a model. We will here present our progress on the investigation of the neural circuitry underlying larval responses to an aversive mechanosensory stimulus, the air puff. Drosophila larvae typically respond to the air-puff with either startle or escape behaviours (Jovanic et al, 2016). The selection of one action implies the inhibition of other, mutually exclusive, actions. Competitive interactions between the neural modules underlying startle and escape behaviors could thus exist in order to enable the selection between the two. Using inactivation and optogenetic activation of single neurons in behaving larvae combined with video tracking and automated behavioral detection (Masson et al., 2020), we determined the role of interneurons at different sensorimotor stages and found they are differentially involved in startle and escape behaviors. Some of these neurons promote one response and repress another suggesting they are involved in competitive interactions. Using EM-based connectomics, we are now mapping the startle and escape sensorimotor pathways and investigating where and how both pathways could interact. The connectivity motifs underlying these interactions should allow the execution of one behaviour at the expense of another. One possible connectivity pattern could be that of reciprocal inhibition of inhibition (Jovanic et al. 2016, Koyama and Pujala, 2016). Determining the neural circuit mechanisms underlying sensorimotor decisions in a model system where we can trace all connections and manipulate single neurons may bring insights into the neural circuit mechanisms of competitive interactions in general.

Disclosures: M. Lehman: None. **S. Autran:** None. **T. Jovanic:** None.

Poster

P579. Sensory Motor Systems I

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Program #/Poster #: P579.04

Topic: F.01. Neuroethology

Title: Differential Corpus Callosum Connections Across Mammalian Orders

Authors: A. GUZMAN, P. PATEL, N. A. SCHOTTLER, *W. GRISHAM;
Psychology, UCLA, Los Angeles, CA

Abstract: Prior work in this lab showed primates possessing proportionately larger neocortices than rodents & ungulates (but not carnivores). Here we analyzed long cortico-cortical connections, the corpus callosum (CC), in mammals with larger brains (primates, carnivores, and ungulates) as well as smaller (rodents). Images from the Comparative Mammalian Brain Collection and The Human Brain Atlas at Michigan State U. were used. Measures of CC thickness were taken from 3 ant-post levels (anterior-most section of CC, section where hippocampus appears, and posterior-most section of CC) in 42 species from the 4 orders. CC measures were made off the midline to avoid the hippocampal commissure. Measures of the neocortex (NC) thickness were made at the same level. For the raw data, the mean thickness of the corpus callosum scaled with the mean thickness of the cortex (slopes ~1.00) except in primates where the CC thickness outstripped the cortex and the slope exceeded 2.6. After correcting for phylogeny, the pattern of residuals was quite different--the slopes were all well below 1.0, and primates had a negative slope. ANOVAs--4 between (order) x 3 within (slice level) and covarying log10 brain size--were done on both raw and phylogeny corrected data. In both raw and corrected CC thickness, the CC was thinnest in ungulates. Similarly, raw and corrected neocortex analyses showed NC to be relatively thin in both ungulates and carnivores. Complex but similar interactions were revealed by analyses of both raw data ratios and corrected ratios of CC/NC: carnivore and rodent ratios did not change much across sections whereas primates' ratios were higher in the posterior section and ungulates' higher in the anterior. Our data are predicated on several assumptions: uniform size and density of CC axons, uniform size and density of cortical neurons, and thicker NC correlated with more CC projections. Nonetheless, our data showed that big-brained animals do not necessarily have thick NC & CC and that there is no straightforward relationship between CC and NC across orders.

Disclosures: A. Guzman: None. **P. Patel:** None. **N.A. Schottler:** None. **W. Grisham:** None.

Poster

P579. Sensory Motor Systems I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P579.05

Topic: F.01. Neuroethology

Support: NSERC Grant RGPIN-2016-04695

Title: Overactivation of the endocannabinoid system during early life alters sensorimotor development in zebrafish

Authors: *L. S. KHARA, D. W. ALI;
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Abstract: The medicinal and recreational use of cannabis has become more prevalent around the world. As cannabis compounds interact primarily with the endocannabinoid system (eCS), it is highly important to understand the fundamental roles played by the eCS during early development because perturbations of the eCS in the early stages of life are known to have detrimental impacts on neural development. In this study we aimed to explore how overstimulation of endocannabinoid (eCB) signalling during early embryogenesis affects the development of functional locomotor behaviors. In order to upregulate eCB signalling, zebrafish (*Danio rerio)* embryos were exposed to pharmacological inhibitors of the enzymes responsible for the degradation of the 2 endocannabinoids: anandamide (AEA) and 2-arachidonoylglycerol (2-AG), during the first 24 hours of development. We found that overactivation of eCB signalling resulted in hypolocomotion, as demonstrated through decreased contractile activity within the chorion of embryos at 1 day post fertilization (dpf), while 2 dpf embryos experienced a reduction in the distance and velocity of burst swimming. To ascertain whether sensorimotor development was altered, we examined the reflex response known as the short latency C-start escape at 2 dpf in response to a mechanical stimulus. Embryos treated with eCB degradation inhibitors displayed a significant increase in the response latency of initiating short latency C-start escape responses. Additionally, we sought to determine whether alterations in escape behaviors were also displayed through the sensation of alternative sensory modalities; therefore, we also examined C-start escapes in response to auditory/vibrational (AV) stimuli. Interestingly, we found that eCS overstimulation impairs the responsiveness to AV inputs, as animals treated with eCB degradation inhibitors demonstrated reduced rates of escape in response to sound at 5 dpf. These impairments were still present at 6 dpf as well, suggesting that there is a delay in the maturation of this response. Taken together, these findings indicate that tight regulation of the normal levels of eCB activity during early embryonic stages is critical for locomotor development and that the endocannabinoid system is essential for the maturation of sensorimotor circuits.

Disclosures: L.S. Khara: None. **D.W. Ali:** None.

Poster

P579. Sensory Motor Systems I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P579.06

Topic: F.01. Neuroethology

Support: ANR-17-CE37-0019

Title: State-dependent modulation of sensorimotor decisions

Authors: E. DE TREDERN, D. MANCEAU, V. SUS, ***T. JOVANIC**;
Paris-Saclay Inst. of Neurosci., Gif-sur-Yvette, France

Abstract: In face of aversive stimuli animals first need to decide whether to respond and then how to respond. In addition, they constantly need to balance their decisions to avoid dangers and threats with pursuing food search. These decisions are modulated by animal's internal state. Food-deprived animals were shown to be more likely to ignore aversive stimuli and to be more responsive to appetitive food-related stimuli. However, whether and how nutritional states affect non-feeding related decisions is not well understood. In order to address this question and investigate the neural circuit mechanisms underlying the state-dependent modulation of behavior, we combine automated behavioral detection, neuronal manipulation, functional imaging and electron microscopy in Drosophila larva. We have previously described a circuit that controls the decision between startle and escape-like behavior following a mechanical stimulus. We hypothesized that integration of other information, such as the feeding state, might occur through long-range projections that connect to this circuit. To investigate this, we first characterize how diet affect behavioral responses following a mechanical stimulus. Then, we describe how the activity of the already known neuronal substrates of decision making are differentially modulated by animal's nutritional status using 2-photon calcium imaging in intact larvae. Finally, we map additional substrates of decisions between startle and escape behaviors. Understanding the neural circuit mechanisms underlying simple decisions of whether and how to avoid aversive stimuli and how these decisions can be modulated by the internal state in the Drosophila larva where we can study neural circuit mechanisms with single cell and synaptic resolution across the nervous system, might shed light on the principles underlying theses processes across species.

Disclosures: E. de Tredern: None. **D. Manceau:** None. **V. Sus:** None. **T. Jovanic:** None.

Poster

P579. Sensory Motor Systems I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P579.07

Topic: F.01. Neuroethology

Support: Ph.D. Program Research Award, Robert C. Vackar College of Business and Entrepreneurship, The University of Texas Rio Grande Valley

Title: Effects of Environmental Enrichment on Neuroplasticity in Monodelphis domestica: Focus on Postsynaptic Density-95 and Brain-derived Neurotrophic Factor

Authors: *S. HEINGRAJ[1], E. I. ALANIZ[2], M. GIL[2,3,5], M. S. MINOR[1], I. PEREZ[2], K. RENTERIA[2], J. CANTU[2], A. HINOJOSA[4];
[1]Dept. of Marketing, The Univ. of Texas Rio Grande Valley, Edinburg, TX; [2]Dept. of Psychological Sci., The Univ. of Texas Rio Grande Valley, Brownsville, TX; [3]Sch. of Medicine, Dept. of Neurosci., The Univ. of Texas Rio Grande Valley, Harlingen, TX; [4]Dept. of Hlth. and Biomed. Sci., The Univ. of Texas Rio Grande Valley, Brownsville, TX; [5]Inst. for Neurosci., Harlingen, TX

Abstract: Environmental exposures and environmental influences play a vital role in brain development and may permanently impact the brain. In this study, we examined the effects of environmental enrichment (EE) in a non-traditional animal model, *Monodelphis domestica* (n = 40: $n_{NonSocialNonEnrichment(control)} = 6$, $n_{NonSocialEnrichment} = 14$, $n_{SocialNonEnrichment} = 19$, $n_{SocialEnrichment} = 7$), using the combination of behavioral indicators and immunohistochemical markers (i.e., postsynaptic density-95 (PSD-95) and brain-derived neurotrophic factor (BDNF)), particularly in the frontal cortex and hippocampus. *Monodelphis* has recently gained popularity as an alternative animal model for neurodevelopmental research, allowing researchers to examine the impact of environmental influences at early development stages.In Experiment 1 (i.e., the object-place recognition test), there was a statistically significant interaction effect of the number of times the animals interacted with old and new objects ($F(3,35) = 120.66$, $p < .001$), despite the sex and housing conditions. Females, in particular, interacted more with new objects compared to males, which may indicate sex differences in cognitive ability ($M_{FemaleOldObject} = 16.62$, $M_{FemaleNewObject} = 47.14$, $M_{MaleOldObject} = 19.06$, $M_{MaleNewObject} = 39.78$, $F(1,37) = 5.98$, $p < .01$). There were no differences between each enrichment condition. Based on the open field test (Experiment 2), latency to immobility of animals in the non-social enrichment condition was marginally significantly higher compared to the social enrichment condition ($U = 234.5$, $p = .09$). There were marginal significant differences in latency distribution to immobility according to the types of housing conditions ($\chi^2 = 7.143$, $p = .067$). The difference regarding the mobility duration between sex was also statistically significant ($F(1, 32) = 4.922$, $p < .05$). However, there were no significant differences between the conditions in the rotarod test (Experiment 3). The insignificant results may have been due to the fact that we exposed the animals to only one inanimate object in the EE condition.We also did a pilot test to examine PSD-95 and BDNF, particularly in the frontal cortex and hippocampus, and found the expression of the markers in these areas. We are currently conducting immunohistochemistry to examine the effects of social and EE on neuroplasticity. We expect that the PSD-95 and BDNF in the EE condition will be significantly increased compared to the control and social enrichment conditions. Together, this research may offer researchers an alternative animal model and opportunity to examine the impact of EE on neuroplasticity.

Disclosures: S. Heingraj: None. **E.I. Alaniz:** None. **M. Gil:** None. **M.S. Minor:** None. **I. Perez:** None. **K. Renteria:** None. **J. Cantu:** None. **A. Hinojosa:** None.

Poster

P591. Vocal/Social Communication: Non-Avian II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P591.01

Topic: F.01. Neuroethology

Title: Uncovering developmental changes in the neural circuits that regulate the production of mouse ultrasonic vocalizations

Authors: *N. PRANIC, C. KORNBREK, Z. HE, C. CHAE, K. A. TSCHIDA;
Psychology, Cornell Univ., Ithaca, NY

Abstract: Vocalizations are an essential means of communication in social animals, both human and non-human. Animals vocalize throughout their lifetimes, and vocalizations produced at different developmental stages serve distinct roles in behavior and are in turn elicited by distinct cues and contexts. Mouse (*Mus musculus*) pups produce ultrasonic vocalizations (USVs) in response to isolation and cold exposure, eliciting retrieval behavior from the mother, whereas adult mice produce USVs in response to social partners**.** However, the neural circuits that underlie the production of USVs in mouse pups remain poorly described. In addition, whether and how these circuits change over development as mice shift from producing isolation-induced USVs to producing social USVs remains unknown. To address these questions, we're applying a dual labeling strategy to examine developmental changes in the populations of neurons that are active during USV production at different ages. First, we will use an activity-dependent labeling strategy (TRAP2 transgenic mice crossed with Ai14 mice) to fluorescently label neurons throughout the brain that are active when mouse pups (PND 8) produce isolation-induced USVs. At a later point in development, USVs will be elicited from these same mice (either isolation-induced USVs at a later point in early development or social USVs in adolescence or adulthood), and Fos immunolabeling will be performed to compare neuronal populations that are active during USV production at different developmental stages. These experiments will reveal neuronal populations that regulate USV production across development, as well as shed light on the neural circuit changes that enable the emergence of social USV production in adolescent mice.

Disclosures: N. Pranic: None. **C. Kornbrek:** None. **Z. He:** None. **C. Chae:** None. **K.A. Tschida:** None.

Poster

P591. Vocal/Social Communication: Non-Avian II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P591.02

Topic: F.01. Neuroethology

Title: Midbrain circuits for context-dependent vocalization in the mouse

Authors: *P. ZIOBRO, Z. HE, A. SADANGI, K. A. TSCHIDA;
Cornell Univ., Ithaca, NY

Abstract: Vocalizations are an essential means of communication in humans and more broadly in mammals. To communicate effectively, animals must vocalize in the right way at the right time, and accordingly, animals produce different acoustic categories of vocalizations in different behavioral contexts that serve different communicative functions. Despite the importance of vocalization to mammalian social behavior, the neural circuits that allow animals to produce different acoustic types of vocalizations according to social context remain undescribed. To address this question, we study the neural circuits that underlie vocalization in mice, which produce ultrasonic vocalizations (USVs) during same-sex and opposite-sex social interactions and lower frequency squeaks in painful or aversive contexts. The midbrain periaqueductal gray (PAG) contains neurons essential for vocalization in mammals, and a recent study identified a specialized population of neurons in the PAG of the mouse whose activity is both necessary and sufficient for the production of USVs (referred to as PAG-USV neurons; Tschida et al., 2019). In the current study, we ask whether PAG-USV neurons are active during the production of squeaks, to begin to understand whether these neurons are uniquely important for USV production, or alternatively, contribute to the production of multiple acoustic types of vocalization.

To test whether PAG-USV neurons are active during the production of squeaks, we employ an activity-dependent labeling strategy (TRAP2 transgenic mice crossed with Ai14 mice) which permits the expression of fluorescent protein in neurons throughout the brain that were recently active during behavior. PAG-USV neurons are fluorescently labeled following social encounters marked by USV production in male and female mice. Ten days later, these same mice are either presented again with a social partner to elicit USV production a second time or are subjected to a mild footshock paradigm to elicit the production of squeaks. Fos immunostaining is then performed to label neurons that were active during the second behavior session, and the overlap of neurons within the PAG labeled during the first and second behavior sessions will be examined. These experiments will shed light on the functional organization of the midbrain circuits that allows animals to select and to produce the appropriate type of vocalization for a given behavioral context.

Disclosures: P. Ziobro: None. **Z. He:** None. **A. Sadangi:** None. **K.A. Tschida:** None.

Poster

P591. Vocal/Social Communication: Non-Avian II

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Program #/Poster #: P591.03

Topic: F.01. Neuroethology

Support: 5F31DC017879-03
5R01MH117778
2R01DC013826

Title: Adaptive control of vocal suppression in the mouse brain

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[1]Duke, Durham, NC; [2]Neurobio., Duke Univ., Durham, NC; [3]Duke Univ. Dept. of Neurobio., Durham, NC

Abstract: Vocal communication is a key social behavior by which humans and other mammals form and maintain social bonds, convey information about social status and mating fitness, and engage in cooperative behavior. The decision to vocalize is regulated by the ongoing environmental context, including the presence of external sensory and social cues, as well as the animal's own internal state and past experience. Though vocalizations are an important medium for sexual and social signaling between conspecifics, in many respects, the decision *not* to vocalize is equally important, as vocalizing in the wrong context may increase the risk of unnecessary energy expenditure, of retaliation by a dominant conspecific, or of attracting the attention of eavesdropping predators or parasites. Despite extensive research on circuits necessary for vocal production, little is known about forebrain networks that *suppress* vocalizations in response to appropriate environmental cues.
Mice produce complex ultrasonic vocalizations (USVs) in a variety of social contexts, allowing researchers to study the circuits underlying vocal control using advanced genetic tools. The midbrain periaqueductal gray (PAG) is a critical node in the vocal motor circuit whose activity is obligatory for vocalizations. Previously, we identified a subpopulation of PAG-projecting amygdala neurons on the border of the central and medial amygdala ($Amg_{C/M\text{-}PAG}$ neurons) whose activity is sufficient to suppress ongoing vocalizations via direct inhibition of a subpopulation of PAG neurons that are active during USV production. In the current study we sought to further characterize the identity and function of these neurons. We showed that $Amg_{C/M\text{-}PAG}$ neurons suppress vocalizations in both male and female mice across social contexts and that $Amg_{C/M\text{-}PAG}$ neurons express estrogen receptor alpha (Esr1). We then imaged the activity of $Amg_{C/M\text{-}PAG}$ neurons during social and non-social interactions. Finally, we traced the direct monosynaptic inputs to $Amg_{C/M\text{-}PAG}$ neurons and interrogated the function of inputs to this region from the medial preoptic area. Together these experiments improve our understanding of how $Amg_{C/M\text{-}PAG}$ neurons regulate adaptive vocal control.

Disclosures: V. Michael: None. **S. Xiao:** None. **R.D. Mooney:** None.

Poster

P591. Vocal/Social Communication: Non-Avian II

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Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P591.04

Topic: F.01. Neuroethology

Support: NIH Grant DP5-OD023116
UCSF PBBR

Title: A novel reticular oscillator in the brainstem synchronizes neonatal crying with breathing

Authors: *P. WEI[1], M. COLLIE[1], B. DEMPSEY[2], G. FORTIN[2], K. YACKLE[1];
[1]Univ. of California San Francisco, San Francisco, CA; [2]Inst. de Biologie de l'Ecole Normale Supérieure, Paris, France

Abstract: Human speech can be divided into short, rhythmically-timed elements, similar to syllables within words. Even our cries and laughs, as well as the vocalizations of other species, are periodic. However, the cellular and molecular mechanisms underlying the tempo of mammalian vocalizations remain unknown. Here we describe rhythmically-timed neonatal mouse vocalizations that occur within single breaths, and identify a brainstem node that structures these cries, which we name the intermediate reticular oscillator (iRO). We show that the iRO acts autonomously and sends direct inputs to key muscles in order to coordinate neonatal vocalizations with breathing, as well as paces and patterns these cries. Furthermore, we identify a set of molecular markers that labels the iRO neurons. These results reveal that a novel mammalian brainstem oscillator embedded within the conserved breathing circuitry plays a central role in the production of neonatal vocalizations.

Disclosures: P. Wei: None. **M. Collie:** None. **B. Dempsey:** None. **G. Fortin:** None. **K. Yackle:** None.

Poster

P591. Vocal/Social Communication: Non-Avian II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P591.05

Topic: F.01. Neuroethology

Title: Calling from the heartbeat: cardiovascular gating of vocal behavior in freely moving macaque monkeys

Authors: *C. RISUENO SEGOVIA[1], O. KOC[1], P. CHAMPEROUX[2], S. R. HAGE[3];
[1]Ctr. For Integrative Neurosci., Tubingen, Germany; [2]European Res. Biol. Ctr., Baugy, France; [3]CIN, Univ. of Tuebingen, Tuebingen, Germany

Abstract: Communication is a keystone of animal behavior. However, the physiological states underlying and gating natural vocal signaling are still largely unknown. In this study, we investigated the correlation of affective vocal utterances with concomitant cardiorespiratory mechanisms. We telemetrically recorded electrocardiography, blood pressure, and physical activity in six freely moving and interacting cynomolgus monkeys (Macaca fascicularis). Our results demonstrate that vocal onsets are strengthened during states of sympathetic activation, and are phase locked to a slower Mayer wave and a faster heart rate signal at ~2.5 Hz. Vocalizations are coupled with a distinct peri-vocal physiological signature based on which we were able to predict the onset of vocal output with high precision using three machine learning classification models. These findings emphasize the role of cardiorespiratory mechanisms in gating vocal onsets to optimize arousal levels and minimize energy expenditure during natural vocal production.

Disclosures: C. Risueno Segovia: None. **O. Koc:** None. **P. Champeroux:** None. **S.R. Hage:** None.

Poster

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Program #/Poster #: P591.06

Topic: F.01. Neuroethology

Support: NSF#1754332

Title: Two functions for one tract: the tradeoff of vocalizations and breathing in male rats

Authors: C. P. SCHAEFER, IV, ***T. RIEDE**;
Midwestern Univ., Glendale, AZ

Abstract: Breathing movements are governed by metabolic demands. They can be modulated by locomotion or emotional responses. Vocal production requires adaptations of breath control since the airflow through the respiratory tract is the source for vocal sounds like laughing, crying, human speech or calling in nonhuman mammals. Previous work from our lab suggests that vocal patterns in male rats are entrained to prevalent breathing movements. The use of a particular breathing pattern (e.g., eupneas or sighs) is reflected in vocal characteristics. In order to test this hypothesis further, the objective of this work was to determine whether increased breathing rates due to an increased metabolic demand, would affect vocal production. We investigated this in

awake and spontaneously behaving rats. Rats produce 22-kHz vocalizations during forced treadmill running. Those calls do not differ in fundamental frequency during a noxious context in which animals freeze. After 3 weeks of training, we recorded breathing movements and vocalizations in 8 male SD rats, ages 4 to 6 months, at rest and during running. If laryngeal vocal movements are entrained to prevailing breathing movements, we expected that vocal characteristics would change during increased breathing rates. As predicted, some vocal characteristics changed during running. Call duration shortened with decreased eupnea duration and increased breathing rates. Fundamental frequency, although overall not different to a non-running context, demonstrated a positive correlation with running speed. Unexpected was the observation of numerous spectral features not observed in a non-running context, such as frequency jumps, call interruptions, mid-call transitions to audible screams and absence of sound during a vocal exhalation. Those spectral features suggest that operation of the laryngeal valve is challenged during running since ultrasonic whistle production requires precise motor control of the laryngeal valve and subglottal pressure. Finally, we investigated how non-vocal eupneas and sighs were affected by the presence of vocalizations. Both breath types became longer and the inhale-exhale ratio of eupneas became smaller in the vicinity of vocal breaths. Sighs were produced less frequently during episodes of high vocal rates as if the two patterns were mutually exclusive. In sum, data support the hypothesis that vocal patterns are entrained to prevailing breathing patterns. Results suggest also that there is a reciprocal relationship. The non-vocal breathing movements are affected by the presence of vocal breathing. The study highlights the integrative complexity of breath control and vocal production.

Disclosures: C.P. Schaefer: None. **T. Riede:** None.

Poster

P591. Vocal/Social Communication: Non-Avian II

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Program #/Poster #: P591.07

Topic: F.01. Neuroethology

Support: ERC Starting Grant 851210
DFG Grant 329518246

Title: Social cues interact with intrinsic circuit dynamics to drive the choice between two communication signals in Drosophila

Authors: E. STEINFATH, A. KHALILI, *J. CLEMENS;
European Neurosci. Inst., Göttingen, Germany

Abstract: Communication is multi-modal - when we interact, we speak, gesticulate, and touch. However, how the different acoustic, visual, and tactile cues are selected and coordinated to

support communication is unclear. We address this issue in the vinegar fly which thanks to its complex social behavior and genetic toolbox is ideal for dissecting the neural basis underlying communication behaviors. During courtship, male flies produce two types of communication signals to woo the female: air-borne “courtship song” is produced by moving the wing, while substrate-borne “vibrations” are transmitted via the legs. The sensory cues and the neural circuits that drive the courtship song are well studied. However, little is known about the sensory and neural basis underlying the production of vibrations, and the cues and circuits that select between song and vibrations. Here we identify the context cues that drive the choice between the two communication signals and provide first insight into the circuit underlying that choice. To address this issue, we developed an experimental setup to record the interactions of flies during courtship alongside the male song and vibrations. Statistical modeling based on pose tracking suggests that the switch from song to vibration is induced by female stationarity, which we causally tested by optogenetically controlling female walking during courtship. We then identified two clusters of neurons - called pC2 and P1 - which control the switch between song and vibrations in an opposing fashion: Optogenetic activation of pC2 neurons in solitary males induced song during and persistent vibrations after activation. By contrast, activation of P1 neurons induced vibrations during and after activation, while song only occurs sparsely after activation. Sensory cues provided by the presence of a female overwrite these intrinsic dynamics resulting in more song production after the activation. Our results suggest that the choice between two communication signals in Drosophila is likely driven by inhibitory cross-talk between the pC2 and P1 neurons or their downstream targets. Social cues, which could differentially activate pC2 or P1, interact with the complex circuit dynamics arising from this cross-talk to choose the context-appropriate signal. Overall, this provides first insight into the logic underlying the choice between different communication signals via mutual inhibition modulated by social cues.

Disclosures: E. Steinfath: None. **A. Khalili:** None. **J. Clemens:** None.

Poster

P591. Vocal/Social Communication: Non-Avian II

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Program #/Poster #: P591.08

Topic: F.01. Neuroethology

Support: NIH R01 NS117469

Title: Nucleus ambiguus cell loss and inflammation pair with vocal changes in an adult rat model of Parkinson disease

Authors: E. H. DAO, S. A. LECHNER, J. M. WELSCH, ***C. A. KELM-NELSON**;
Dept. of Surgery | Div. of Otolaryngology, Univ. of Wisconsin-Madison, Madison, WI

Abstract: The pathology underlying cranial motor dysfunction, including vocalization, is poorly understood for progressive degenerative disorders including Parkinson disease (PD). Complete loss of function of the *Pink1* gene in a rat model causes an early and progressive form of the disease, analogous to the inherited familial type. In both humans and rats, the brainstem and larynx are mechanistically responsible for vocal motor functions. Male *Pink1*-/- rats show vocal communication differences compared to wildtype controls including changes in ultrasonic vocalization acoustics that affect functional communication, as well as alterations within vocal motor brainstem regions (*e.g.*, nucleus ambiguus, increased α-synuclein) and larynx muscle biology (*e.g.*, thyroarytenoid muscle, increased centralized nuclei and decreased intrinsic force). However, it is unknown whether (A) nucleus ambiguus motoneuron cell density is related to ultrasonic vocalization acoustics in this PD model or (B) if alterations in nucleus ambiguus gene expression contributes to behavioral changes in this model. In this study, male *Pink1*-/- rats were tested for vocalization acoustics during a recording session and then injected with Fluorogold, a retrograde tracer, unilaterally into the vocal fold. After 7 days, immunohistochemistry and unbiased stereology were used to count Fluorogold-positive motoneuron cell bodies. Relationships between cell numbers and vocal acoustics were assessed; there was a significant positive correlation between number of cells and duration of the calls; suggesting that reduced motoneuron density may compromise vocal duration. On a separate group of rats, RNA sequencing was used to examine nucleus ambiguus differential gene expression in male *Pink1*-/- rats compared to age-matched wildtype controls (n=513 up-regulated; n=327 down-regulated genes). Using the ENRICHR tool, the top up-regulated biological pathways (*genes*) included several inflammatory pathways: interleukin (IL) activation of NF-kappa β (*Sqstm1;Prkcz*), IL-1 (*Pik3r2;Irak4;Sqstm1;Prkcz*), IL-5 (*Gsk3a;Rps6kb2;Pik3r2*), IL-6 (*Gab1;Pik3r2;Agt*) signaling pathways as well as cell death (*Aph1b;Ubb;Sqstm1;Uba52*). These data are the first to suggest that vocalization dysfunction in *Pink1*-/- rats may be linked to nucleus ambiguus motoneuron inflammation.

Disclosures: E.H. Dao: None. **S.A. Lechner:** None. **J.M. Welsch:** None. **C.A. Kelm-Nelson:** None.

Poster

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Topic: F.01. Neuroethology

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1UL1TR002373

Title: Rescue of vocal deficits with exercise in a Pink1-/- rat model of Parkinson Disease

Authors: *J. D. HOFFMEISTER[1,2], C. K. BROADFOOT[4], N. SCHAEN-HEACOCK[2], S. A. LECHNER[3], M. KRASKO[2], A. NISBET[2], J. RUSSEL[2], J. SZOT[2], T. J. GLASS[6], N. P. CONNOR[7], C. A. KELM-NELSON[8], M. R. CIUCCI[5];
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Abstract: Vocal deficits are common in Parkinson Disease (PD) and because they do not respond to drugs, targeted vocal exercise is the primary treatment. While exercise improves symptoms for some individuals, many do not respond. To optimize treatment, it is essential to understand the mechanisms by which vocal exercise modifies behavioral and neural function. Previously, we have shown vocal exercises rescue vocal deficits at an early/prodromal timepoint (4 months) in the *Pink1-/-* rat model of PD, and that vocal exercise was associated with increased 5HT immunoreactivity in the hypoglossal nucleus. Here, we extend that work to later timepoints (6 and 10 months of age--midstage) as most patients with PD do not begin vocal exercises until later in the disease. We hypothesized vocal exercise would rescue ultrasonic vocalization intensity at both time points. (Note: as part of a larger study, these rats also underwent tongue training, not discussed here). For both the 6- and 10-month cohorts, *Pink1* -/- rats were randomly assigned to vocal exercise or non-exercise control conditions, and wild type (WT) rats were all assigned to non-exercise control condition. All rats were tested at baseline, and after 8 weeks of exercise or non-exercise training. Vocal exercise used food to reward increased USV intensity, frequency modulation, and complexity. Primary outcomes were USV intensity, average peak frequency, and call complexity. Average peak frequency of frequency-modulated calls was greater for the WT group than for *Pink1-/-* exercise (6-month $p<0.0001$, 10-month $p< 0.05$) and *Pink1*-/- non-exercise (6-month $p<0.0001$, 10-month $p<0.05$) groups. The 6-month *Pink1-/-* exercise group had a greater percentage of complex calls than the 6-month WT group ($p<0.05$). While vocal exercise resulted in greater vocalization complexity at 6-months, this was not observed at 10-months, and intensity was not rescued at 6 or 10 months, contrary to earlier timepoints in this model. Tissue analyses of monoaminergic immunoreactivity in brainstem nuclei are ongoing. These behavioral findings suggest benefits of vocal exercise in PD may be more-beneficial earlier in the disease, and will be considered relative to neural tissue analyses.

Disclosures: J.D. Hoffmeister: A. Employment/Salary (full or part-time):; University of Minnesota, University of Minnesota Physicians. **C.K. Broadfoot:** B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; NIH F31 NIDCD. **N. Schaen-Heacock:** A. Employment/Salary (full or part-time):; Stipend-University of Wisconsin-Madison. **S.A.**

Lechner: A. Employment/Salary (full or part-time):; University of Wisconsin-Madison. **M. Krasko:** A. Employment/Salary (full or part-time):; Stipend-University of Wisconsin-Madison. **A. Nisbet:** A. Employment/Salary (full or part-time):; University of Wisconsin-Madison. **J. Russel:** A. Employment/Salary (full or part-time):; University of Wisconsin-Madison. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; NIH NCI R01. **J. Szot:** A. Employment/Salary (full or part-time):; University of Wisconsin-Madison. **T.J. Glass:** A. Employment/Salary (full or part-time):; University of Wisconsin-Madison. **N.P. Connor:** A. Employment/Salary (full or part-time):; University of Wisconsin-Madison. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; NIH NIDCD, NCI R01. **C.A. Kelm-Nelson:** A. Employment/Salary (full or part-time):; University of Wisconsin-Madison. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; NIH NIDCD R01. **M.R. Ciucci:** A. Employment/Salary (full or part-time):; University of Wisconsin-Madison. B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; NIH NIDCD R01.

Poster

P591. Vocal/Social Communication: Non-Avian II

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P591.10

Topic: F.01. Neuroethology

Title: Parallel evolution of neuroepigenomic specializations in vocal learning mammals

Authors: M. E. WIRTHLIN[1], I. KAPLOW[1], S. ZHANG[1], T. A. SCHMID[2], M. M. YARTSEV[3], A. R. PFENNING[1];
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Abstract: Vocal behavior is a highly variable behavioral phenotype, ranging from simple grunts and hisses to the complex vocal learning exemplified by human speech. Although the neuroanatomical, electrophysiological, and molecular bases for this behavior are being explored in a handful of species, an overall picture of how genomic differences subserve this trait remain elusive. Here, we assessed the transcriptomic and epigenomic profile of several brain regions

involved in vocal control in a bat species with extensive vocal plasticity (Rousettus aegyptiacus) and compared them to datasets collected from comparable brain regions in rodents, rhesus macaque, and human. Despite the extensive nucleotide sequence turnover resulting from 150 million years of mammalian evolution, we were able to identify conserved sets of regulatory sequences associated with brain region-specific transcriptional specializations, as well as uniquely shared specializations in the vocal control circuits of humans and bats. These findings suggest that there may exist fundamental genetic mechanisms for the evolution of complex vocal behavior, as well as implicating genomic changes in gene regulatory sequences, rather than changes in genes themselves, as a hotbed for the evolution of mammalian vocal-motor behavior.

Disclosures: M.E. Wirthlin: None. **I. Kaplow:** None. **S. Zhang:** None. **T.A. Schmid:** None. **M.M. Yartsev:** None. **A.R. Pfenning:** None.

Poster

P605. Early-Life Stress: Adolescence

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P605.01

Topic: F.04. Stress and the Brain

Support: National Institute of Mental Health R01MH098348 (D.C.K. and S.M.)

Title: Discrimination, neural reactivity to stress, and psychological distress

Authors: *D. K. GREY[1], J. B. PURCELL[1], K. N. BUFORD[1], M. A. SCHUSTER[2,3], M. N. ELLIOTT[4], S. TORTOLERO EMERY[5], S. MRUG[1], D. C. KNIGHT[1];
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Abstract: Exposure to discrimination has been linked to disruptions in stress reactivity and emotion regulation processes. Chronic stress is a risk factor for emotional dysfunction, including depression, anxiety, and posttraumatic stress (Cano et al., 2016; Brooks Holliday et al., 2018). However, the neural mechanisms by which discrimination ultimately affects mental health remain unclear. Thus, it is important to determine the inter-relationships among discrimination, neural reactivity to stress, and mental health. The acute stress response is mediated by a neural network that consists of the prefrontal cortex (PFC), inferior parietal lobule (IPL), hippocampus, and amygdala. We hypothesized that stress-induced activity within these brain regions would vary with prior exposure to discrimination. Furthermore, changes in the activity of these regions may alter reactivity to future stressors, which may harm mental health. In the present study, adolescent exposure to discrimination was measured at four-time points between the ages of 11

and 19 years. Psychological distress (i.e., depression, anxiety, and posttraumatic stress symptoms) and neural reactivity to acute psychosocial stress were measured when participants were young adults (Mean Age: 20.03). Neural reactivity to stress was assessed in 301 participants using a variant of the Montreal Imaging Stress Task (MIST) during functional magnetic resonance imaging (fMRI). A linear mixed-effects model (LME) compared stress-evoked brain activity to discrimination as a dichotomous variable (i.e., Discrimination versus No Discrimination). Greater neural reactivity was observed within the dorsolateral PFC, inferior and superior parietal lobes, lingual gyrus, and middle temporal gyrus in participants who reported discrimination versus those who reported no discrimination. A secondary LME analysis compared stress-evoked brain activity to discrimination as a continuous variable. Hippocampal reactivity to stress varied negatively with the amount of discrimination experienced. In addition, participants that reported discrimination also reported higher levels of depression, anxiety, and posttraumatic stress. The present results suggest that exposure to discrimination may alter the stress reactivity of brain regions that support emotion regulation processes (e.g., PFC, hippocampus, and IPL). Taken together, the present findings suggest that discrimination may alter neural reactivity to stress, which may in turn have long-term implications for the psychological well-being of those exposed to discrimination.

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Poster

P605. Early-Life Stress: Adolescence

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P605.02

Topic: F.04. Stress and the Brain

Title: The relationship between threat sensitivity and anxiety symptoms in community violence exposed urban adolescents

Authors: *A. HEETER, N. LIPANSKI, B. BORG, H. MARUSAK;
Wayne State Univ. Sch. of Med., Detroit, MI

Abstract: Community violence (CV) exposure, defined as witnessing or experiencing acts of violence, is a chief public health concern. Prior research by our group and others demonstrates that adolescents living in urban environments are disproportionately exposed to CV than their non-urban peers, and CV exposure is associated with greater anxiety symptoms in urban youth. Given prior research demonstrating that threat sensitivity may mediate anxiety symptoms in adult and adolescent populations, threat sensitivity may serve as a psychobiological mechanism linking CV to the elevated risk of anxiety in urban youth. Our ongoing survey study examines the impact of CV on anxiety symptoms in urban adolescents exposed to high CV rates to test this

hypothesis. Here, we present a preliminary analysis from this ongoing study on 24 adolescents (*M*±*SD*=15.3±1.3 years; 58% female; 58% Black American) residing in Detroit, Michigan. Youth completed validated measures of CV exposure (Things I Have Seen and Heard scale) and anxiety symptoms (Screen for Child Anxiety Related Disorders; scores >25 indicate a clinically significant anxiety disorder). Threat sensitivity was measured using a novel questionnaire that captures sensitivity to ambiguous (i.e., potential) and unambiguous (i.e., acute) threats via CV-related vignettes. Correlation analyses were conducted on continuous variables. Twenty-three adolescents (96%) reported exposure to some type of CV during their lifetime and had on average six different exposure types. The most endorsed CV items were witnessing adults yelling (92%), an arrest (83%), a beating (75%), and hearing gunshots (75%). Our sample also reported high average anxiety symptoms (M=32±17), with most youth (58%) reporting scores indicative of an anxiety disorder. There was a relationship between reported symptoms of anxiety and threat sensitivity, such that more anxiety symptoms were associated with greater sensitivity to unambiguous but not ambiguous threat. Overall, our results suggest a relationship between sensitivity to acute (i.e., predictable, unambiguous) threat, particularly anxiety in urban adolescents who experience high rates of CV. Heightened sensitivity to unambiguous threats may be adaptive in the short term by allowing potential victims of CV to be on alert and avoid potential danger. However, long-term heightened threat sensitivity may be detrimental. Given that anxiety frequently begins during adolescence understanding the underlying psychobiological mechanisms may lead to better interventions that target threat sensitivity (e.g., cognitive bias modification) to prevent anxiety in at-risk adolescents.

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Poster

P605. Early-Life Stress: Adolescence

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Program #/Poster #: P605.03

Topic: F.04. Stress and the Brain

Support: NIH Grant P20GM103430
Rhode Island Foundation 5213_20200592

Title: Examination of sex specific behavioral and metabolic effects incurred by acute stress and adult dietary exposure

Authors: *K. SALHANY, Jr., J. GAULIN, J. MELO, Z. DJABAKATIE, A. TOORIE;
Biol., Rhode Island Col., Providence, RI

Abstract: Stress experienced throughout discrete phases of development (i.e., adolescent vs adult period) can confer long lasting effects on neuroendocrine physiology, psychological

function, and metabolism in a sex-specific manner. While the hypothalamus-pituitary-adrenal (HPA) axis mediates basal homeostatic aspects of metabolism, acute stress increases HPA axis activity, which leads to enhanced sympathetic nervous activity and elevated circulating glucocorticoids. Sustained elevations in circulating glucocorticoids are associated with an increased risk of developing metabolic syndrome (MetS) and its associated symptoms (i.e., glucose dyshomeostasis, dyslipidemia, and systemic insulin resistance). MetS can arise independently due to genetic makeup, social constraints, and/or environmental exposures. However, multifactorial interactions, such as gene by environmental interactions or stress by diet interactions, can result in diverse cellular and systemic outcomes serving to protect or predispose against pathologies, including MetS. A Sprague Dawley model of acute restraint stress was utilized to evaluate the sex-specific neuroendocrine, behavioral, and hepatic glucoregulatory metabolic effects occurring as a result of prior stress experience (adolescent vs adult) and adult diet interaction following long-term maintenance on a control or obesogenic (45% fat, 17% sucrose; fat-sucrose diet (FSD)) diet. Preliminary findings suggest that prior stress experience alters diet-induced metabolic and neurobehavioral effects in a sex-specific context.

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Poster

P605. Early-Life Stress: Adolescence

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Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P605.04

Topic: F.04. Stress and the Brain

Title: Early micronutrient supplementation modulates the early-life stress induced alteration in emotional behaviour and hypothalamic DNA methylation in adolescent male mice

Authors: *V. BEGNI[1], K. REEMST[2], J. KOTAH[2], D. CHEISHVILI[3,5], M. SZYF[4], M. RIVA[1], A. KOROSI[2];
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Abstract: Exposure to early life stress (ELS) is a well-known risk factor for psychiatric disorders. Emerging evidence suggests a possible role of nutrition in stress-induced enduring alterations in cognitive and emotional functions. In this regard, we have recently shown that ELS reduces the level of the amino acid methionine, a micronutrient essential for methylation. We showed that a short early nutritional supplementation with methyl donor micronutrients (including methionine) can rescue the ELS-induced cognitive impairments and hypothalamic-pituitary-adrenal (HPA) axis hyperactivity (Naninck et al., 2017). We thus set out to study how

such early micronutrient supplementation, with and without a previous ELS exposure, modulates the response to an acute restraint stress (AS) during adolescence. To this end we exposed C57BL/6J male mice to a ELS model consisting of limited bedding and nesting materials from postnatal day 2 to 9. Control (CT) animals were left undisturbed in their home cages (n=45 CT and 45 ELS). During the same period both groups were fed with either standard or supplemented diet. Half of both CT and ELS adolescent animals were then exposed to AS and next tested for anxiety and depressive-like behaviors. The hypothalamus was dissected and processed for genome wide methylation analyses using the Illumina Mouse Methylation BeadChip. At the neuroendocrine level, we observed that neither ELS nor the supplemented diet affects the dynamic of corticosterone release in response to AS. Concerning the behavioral aspect, while only few changes were observed at basal condition (i.e. impaired self-care behavior), following AS a more clear anxiogenic profile was present in ELS-exposed mice (i.e. reduced time spent in the open arms of elevated plus maze and in the light chamber of dark/light box) and the supplemented diet partly prevented these ELS-induced impairments. Finally, at the level of hypothalamic DNA methylation, we observed that AS exposure induced a differential response in DNA methylation profile depending on ELS background. Interestingly, several of the differentially methylated genes belong to the Y chromosome, and the supplemented diet modulated these effects, rescuing the ELS-induced alterations in this response. Altogether, the supplemented diet appears to (at least partially) prevent the observed behavioral alterations. Remarkably it also induces long-lasting effects on hypothalamic methylation levels, which is dependent on ELS history and AS exposure during adolescence. In conclusion, we observed that an early nutritional intervention might modulate the impact of ELS-exposure on coping with stressors later in life.

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Poster

P605. Early-Life Stress: Adolescence

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Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P605.05

Topic: F.04. Stress and the Brain

Support: COLCIENCIAS No. 646

Title: Lifelong enhancement of body mass from adolescent stress

Authors: *K. M. MORAN, L. F. GONZÁLEZ-MARTÍNEZ, Y. DELVILLE;
Psychology, Univ. of Texas, Austin, Austin, TX

Abstract: In hamsters, exposure to stress in adulthood causes increased body weight. We addressed how social stress during puberty would impact food intake and body weight. Stressed hamsters started gaining significantly more weight than controls after only two days of stress exposure. Over a two-week period, stressed subjects gained 10% more weight and consumed more food than controls. At the end of the stress period, stressed hamsters collected nearly twice as many palatable sugar pellets from an arena than controls. Stressed subjects presented 15-20% more body fat in mesenteric, inguinal, and retroperitoneal fat pads. In order to assess the duration of these effects, we analyzed data from previous studies keeping hamsters for over two months past the stress period in puberty. Our analysis shows that stressed hamsters stopped gaining more weight after the stress period, but their body weights remained elevated for over two months, consistently weighing 10% more than their non-stressed counterparts. We also analyzed conditioning training data collected after the period of stress in late puberty and early adulthood (P56 to P70) that was part of the original studies. Training consisted of lever pressing for palatable food rewards. At these times, previously stressed hamsters retrieved similar numbers of food pellets from the conditioning chambers, suggesting no difference in appetite after the stress period. These data showing a long-lasting effect of stress on body weight may be relevant to studies on the ontogeny of lifelong obesity.

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Poster

P605. Early-Life Stress: Adolescence

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P605.06

Topic: F.04. Stress and the Brain

Support: Texas A&M Triads for Transformation (T3) Grant

Title: Corticosterone administration in adolescence enhances habit learning in adulthood: Blockade by mifepristone

Authors: *T. M. GADBERRY, M. G. PACKARD;
Psychological and Brain Sci., Texas A&M Univ., College Station, TX

Abstract: Extensive evidence indicates a link between early life stress (ELS) in humans and a subsequent predisposition to psychopathologies that are characterized in part by maladaptive habitual behaviors. Stress and anxiety are factors that can influence the relative use of mammalian memory systems implicated in such disorders. Specifically, cognitive memory functions of the hippocampus are typically impaired by stress/anxiety, whereas habit memory functions of the dorsolateral striatum (DLS) are enhanced. However, these effects have been primarily observed in adult animals, and the effects of stress on these memory systems have not

been extensively investigated in the context of ELS. We have previously observed that administration of the stress hormone corticosterone (CORT) during adolescence enhances subsequent DLS-dependent habit formation in adulthood. The present study was designed to further elucidate the role of CORT in this phenomenon by examining the effects of concurrent administration of the broad steroidal antagonist Mifepristone (MIFP). Four groups of Long-Evans male adolescent rats (PND 37±4) received two peripheral injections daily over five days: first, either MIFP (30mg/kg) or its vehicle, followed by a 15-min interval, then either CORT (5mg/kg) or its vehicle. Subjects then matured into adulthood undisturbed (~3 weeks) before training in a DLS-dependent water plus maze task. In this task, rats were released from different starting positions (i.e. north or south) and were required to make a consistently reinforced body turn response (i.e. always turn right) at the maze choice point to mount a hidden escape platform. Consistent with our previous findings, administration of CORT during adolescence facilitated task acquisition in adulthood relative to controls. However, MIFP given prior to CORT in adolescence blocked subsequent enhancement of habit formation. Finally, rats given MIFP alone did not significantly differ from controls. Taken together, these findings provide further evidence that chronic administration of corticosterone during adolescence can facilitate subsequent DLS-dependent habit formation and highlights glucocorticoid function as a potential underlying mechanism by which ELS may influence habit learning in adulthood.

Disclosures: T.M. Gadberry: None. **M.G. Packard:** None.

Poster

P605. Early-Life Stress: Adolescence

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Program #/Poster #: P605.07

Topic: F.04. Stress and the Brain

Support: NIH Grant K01MH127306
NIH Grant R01MH118369

Title: Adolescent social isolation differentially perturbs adult value-based decision making in female and male mice

Authors: *E. N. HOLLY[1], P. PAREEK[2], M. V. FUCCILLO[1];
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Abstract: Adolescence is a critical period in social and cognitive development, characterized by rapid increases in exploration, social interaction, and neural connectivity. While social isolation at any developmental stage can have long-term consequences on physical and mental health, mounting evidence demonstrates a profound impact of adolescent social isolation (ASI) on a wide range of behavioral and physiological endpoints in adulthood. Goal-directed behaviors and

value-based decision making are integral in daily function, but the impacts of ASI on these behaviors remain largely unexplored. To model ASI, female and male C57bl6/J mice were bred in house and weaned at P21 into either single or group housing. After P60, mice were trained in a self-initiated operant task consisting of three phases: (1) initiation: entry in a central port initiated the trial, and two retractable levers were extended, (2) choice: mice must respond on one of the levers within 10s, (3) outcome valuation: levers retracted and the outcome was delivered. Responding on the active lever resulted in chocolate or vanilla liquid reward, while responding on the inactive lever resulted in 4s darkness. After 1s intertrial interval, a new trial began. To test initial learning, mice were first trained to respond on one lever. After acquisition, mice were run in a serial reversal task, where the active lever reversed after 8/10 correct responses for 10d. We then probed how mice use information about reward value and temporal costs to guide decision making. To test how ASI affected how mice valued rewards, mice were presented with rewards of varying contrasting volumes (15µl vs 0, 5, or 10µl). Next, to test how costs were integrated into decision making, varying temporal delays (1.5, 3, or 4.5s) were applied to the larger 15µl reward. Preliminary data suggest ASI differentially impacted how female and male mice learned and made decisions about values and costs in adulthood. Compared to socially housed females, ASI female mice acquired the operant task significantly faster, were more explorative in choice behavior throughout the serial reversal task. In contrast, compared to the socially housed males, ASI males took significantly more time to acquire the operant task, but showed increased sensitivity to reward value, more consistently choosing the larger reward even when the reward difference was small and when the temporal cost was high. Together, these data demonstrate long-lasting impacts of adolescent social isolation on adult value-based decision making in both males and females. Ongoing work is building neuroeconomic models to further dissect the impacts of isolation stress on choice behavior.

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Poster

P605. Early-Life Stress: Adolescence

Location: Virtual Only

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Program #/Poster #: P605.08

Topic: F.04. Stress and the Brain

Support: Graduiertenkolleg GRK2350/1
NSERC DG

Title: Acute and persistent sex-dependent effects of social instability stress on anxiety-like and social behaviour in Wistar rats.

Authors: *A. SURAKKA[1], A. ELTAHIR[2], S. MURRAY[2], S. PATEL[2], A. C. HANSSON[1], R. SPANAGEL[1], C. M. MCCORMICK[2];

[1]Central Inst. of Mental Hlth. Mannheim, Mannheim, Germany; [2]Brock Univ., St:Catharine's, ON, Canada

Abstract: In adolescence, the quantity and quality of social interactions shape adult social function. To better understand effects of adolescent social instability stress (SS: 1-hr daily isolation and change in cage mate between postnatal day [P] 30-45) in rats, female and male Wistar rats were tested in adolescence or in adulthood. In the elevated plus-maze (on P46 or P71), SS rats showed reduced anxiety compared to control (CTL) rats, as did females relative to males and adults relative to adolescents. When tested on P47 or P48, SS rats spent less time in social interaction than did CTL rats, and females spent less time in social interaction than did males. Females made more entries into arms (locomotion measure) than males. In the social novelty test (on P48 or P72), SS male rats spent more time in social approach toward a novel peer than toward their cagemate, but only when tested in adolescence. There was no effect of SS in females, although they spent more time near the cagemate than the novel peer. SS rats also made more rears in the test arena and explored the corners more than did CTL rats, In the social memory test (P50 or P74), the three-way interaction was significant, with lower memory performance in SS than CTL males in adulthood. Thus, social instability stress in adolescence in Wistar rats alters the behavioural repertoire, with enduring decrements in social behaviour, enhanced exploratory behaviour, and reduced anxiety-like behaviour.

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Poster

P605. Early-Life Stress: Adolescence

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Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P605.09

Topic: F.04. Stress and the Brain

Support: Hope for Depression Research Foundation
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Office of Naval Research (ONR) 00014-19-1-2149
Pritzker Neuropsychiatric Research Consortium
University of Michigan Undergraduate Research Opportunities Program (UROP)
Michigan Research and Discovery Scholars (MRADS) program

Title: Adolescent environmental enrichment and social stress modulate expression of the same gene families within both nucleus accumbens and hippocampus in a genetic rodent model of stress vulnerability

Authors: *A. M. O'CONNOR, M. H. HAGENAUER, L. C. T. FORRESTER, E. R. RICHARDSON, F. I. ROB, E. K. HEBDA-BAUER, S. J. WATSON, Jr., H. AKIL; Univ. of Michigan, Ann Arbor, MI

Abstract: Background: Adolescence is a critical period for the development of emotional circuitry and a time of increased vulnerability to environmental influences and stressors. This study examined the impact of adolescent environmental enrichment (EE) and social defeat (SD) stress on gene expression within two emotional brain regions (nucleus accumbens (NACC) and hippocampus (HC)) using RNA-sequencing. These interventions were studied in selectively bred Low Responder (bLR) rats that exhibit elevated anxiety-like behavior and stress vulnerability. **Methods:** From postnatal day 35 (P35) to P60, male bLR rats (generations F49-F56) were placed in a complex social and physical environment for 1 hr/day for 5 days/week (EE group) or kept in standard housing (NIL group). From P61-64, the rats experienced either social defeat within the resident-intruder paradigm (SD group) or were placed in a novel cage (NIL group) for 15 min/day. From P65-67, the rats underwent behavioral testing (P65: open field, P66: social interaction, P67: elevated plus maze), and were sacrificed on P68. Brains were flash-frozen and dissected at -20°C (NACC: hole punch, HC: whole dorsal HC). For each sample, a cDNA library was constructed and sequenced (Illumina NovaSeq S2), producing 50bp paired-end reads. Reads were aligned to genome assembly Rnor6 (STAR) and summarized into counts per transcript (featureCounts: Ensembl 96 annotation). Following quality control and normalization, the final datasets contained ~17,000 transcripts/region, with sample sizes of NACC: n=46 (n=10-14/subgroup) and HC: n=25 (n=5-8/subgroup). Differential expression was calculated using the limma/voom method and a model including technical co-variates (RNA concentration, batch). Results were compared to gene sets related to functional ontology (MSigDB:C5), cell type/anatomy (DropViz, BrainInABlender, HippoSeq), and manipulations in brain tissue (GeneWeaver, Gemma, literature) using a custom .GMT file and fast Gene Set Enrichment Analysis. **Results:** Within both brain regions there was a convergence of differential expression in association with both manipulations (EE & SD) on a set of genes dominated by the protocadherin family and major histocompatibility complex (RT1), as well as an enrichment of effects within gene sets previously linked to stress, social behavior, and cell type. **Conclusion:** Although often treated as opposing interventions, EE and SD share many common features, including social interaction, stress, and a novel environment. Our results indicate that these two interventions can produce both opposing and converging changes in gene expression in emotional circuitry during adolescence.

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Poster

P605. Early-Life Stress: Adolescence

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P605.10

Topic: F.04. Stress and the Brain

Support: NIH Grant R01DA037911
CIHR Grant MOP-74709
NSERC Grant 2982226
CONACYT 545327
FRQ-PBEEE 0000258419

Title: Molecular and behavioral consequences of social defeat stress in adolescent female mice

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Abstract: Social stress in adolescence increases risk to psychiatric disorders, including depression, anxiety, and substance abuse. During adolescence, the prefrontal cortex (PFC) is still maturing remaining sensitive to environmental influences. The Netrin-1/DCC guidance cue pathway controls dopamine (DA) axon growth to the PFC across adolescence, by promoting mesolimbic DA axon targeting in the nucleus accumbens (NAc). Using a modified version of the adult mouse accelerated social defeat stress paradigm (AcSD), we demonstrated that AcSD in adolescence dysregulates the Netrin-1/DCC system in mesolimbic DA regions, altering PFC DA development and inducing deficits in inhibitory control in adulthood (Vassilev, Pantoja-Urban, et al., 2021). Given that men and women show differential vulnerability to stress and to psychopathologies of adolescent onset, we adapted our adolescent AcSD model in males to adolescent female mice and assessed whether it (i) induces social avoidance 24 h later, (ii) regulates Netrin-1/DCC expression in pre- and postsynaptic components of mesolimbic DA circuitry one week later, and/or (iii) alters behavioral inhibition in adulthood. Adolescent C57BL/6J female mice were introduced, twice a day, to aggressive CD-1 mice for 4 days. A social interaction test was used to measure social avoidance behavior and to categorize them as "resilient" or "susceptible". Sample size for each experiment: control (n=10) and social defeat stress (n=15). Levels of *Dcc* mRNA were quantified in the ventral tegmental area (VTA) using qPCR. Behavioral inhibition was measured using the Go/No-Go task. As we found in adolescent male mice, the majority of adolescent females exposed to AcSD do not show social avoidance. This proportion was not determined by the CD-1 aggression, since the number of attacks did not differ between female resilient and susceptible groups across all stress sessions. In adulthood, female mice that showed a social avoidance resilient phenotype in adolescence, display deficits in cognitive inhibition compared to control groups. Instead, female mice that showed social avoidance in adolescence seem to be protected against cognitive dysfunction. Adult female mice exposed to AcSD in adolescence do not differ in their locomotor activity or motivation to work for a food reward. The expression of *Dcc* mRNA in the VTA, contrary to males, does not differ across female groups. We are currently examining Netrin-1 expression in the NAc and assessing DA connectivity in the PFC. These results suggest that the effects of social stress in adolescence

on cognitive and metabolic function may be sexually dimorphic and involve different molecular processes.

Disclosures: A. Pantoja Urban: None. **S. Richer:** None. **A. Mittermaier:** None. **M. Giroux:** None. **C. Flores:** None.

Poster

P605. Early-Life Stress: Adolescence

Location: Virtual Only

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Program #/Poster #: P605.11

Topic: F.04. Stress and the Brain

Support: SEP-CINVESTAV #141
CONACYT GRANT 485970

Title: Chronic social defeat during adolescence increases depression-like behaviors and induces long-lasting learning and memory impairments in male Swiss Webster mice participation of adrenal and gonadal axes

Authors: *H. MANCHA-GUTIÉRREZ[1], E. M. ESTRADA-CAMARENA[2], L. MAYAGOITIA-NOVALES[2], E. LOPEZ-PACHECO[1], C. LOPEZ-RUBALCAVA[1]; [1]CINVESTAV-IPN, Mexico City, Mexico; [2]Inst. Nacional de Psiquiatría, Mexico City, Mexico

Abstract: Chronic stress exposure during adolescence is a significant risk factor for the development of depression. Chronic social defeat (CSD) is an animal model of depression with excellent ethological, predictive, discriminative and face validity. Because the CSD model has not been thoroughly examined as a model of stress-induced depression within the adolescence stage, we evaluated the short and long-term behavioral and neuroendocrine effects of the CSD during early adolescence. To do this, male Swiss Webster (SW) mice at postnatal day (PND) 28 were exposed to ten consecutive days of CSD (PND28-37), control mice were handled daily. Twenty-four hours (mid-adolescence) or four weeks (early adulthood) after the CSD, independent groups of mice (n=7) were tested in the social interaction test (SIT), the forced swimming test (FST), or in the Barnes maze. After that, their brains (n=4) were obtained for serotonin transporter autoradiography. Independent groups of mice (n=3) were euthanized (at five days intervals) before, during, and after the CSD to obtain trunk blood for serum hormones (corticosterone and testosterone) immunoassays. The CSD during early adolescence induced 1) enduring depression-like behaviors as inferred from reduced social interaction ratio (social avoidance) in the SIT and increased immobility behavior (behavioral despair) in the FST, moreover, the CSD-induced depression-like behaviors correlated in an age-dependent manner with the SERT binding in the hippocampus, 2) long-lasting learning and memory impairments as deduced from increased primary latencies to find the escape box in the Barnes maze, and 3)

higher levels of serum corticosterone in both mid-adolescence and early adulthood and lower levels of serum testosterone from mid to late adolescence. Overall, our results 1) show that CSD during early adolescence induced long-lasting depression-like behaviors and reveal the critical and age-dependent role of SERT in the normal brain development, as well as in the development of pathological behaviors, 2) show that CSD during early adolescence induced enduring cognitive deficits, and 3) highlight the vulnerability of the interaction between adrenal and gonadal axes, evidencing the importance of adequate interaction between these axes during adolescence for the normal brain and behavioral development.

Disclosures: H. Mancha-Gutiérrez: None. **E.M. Estrada-Camarena:** None. **L. Mayagoitia-Novales:** None. **E. Lopez-Pacheco:** None. **C. Lopez-Rubalcava:** None.

Poster

P605. Early-Life Stress: Adolescence

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P605.12

Topic: F.04. Stress and the Brain

Support: The College of Health Sciences, University of KwaZulu-Natal

Title: Alteration of the α5 GABA receptor and 5HTT lead to cognitive deficits associated with major depressive-like behaviors in a 14-day combined stress rat model

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Abstract: Introduction: Current models used to study the pathophysiology of major depressive disorder (MDD) are laborious and time consuming. This study examined the effect of a 14-day combined stress model (CS; corticosterone injection (40mg/kg, s.c.) and chronic immobilization stress for 14 days) versus a 28-day corticosterone only model (daily injection of 40mg/kg corticosterone for 28 days) on 1) depressive-like behaviour and cognitive deficits, 2) serotonergic and GABAergic systems of the hippocampus and prefrontal cortex (PFC), regions implicated in the pathophysiology of MDD in adult male Sprague Dawley rats. **Materials and methods:** Depressive-like behaviors and cognitive deficits were assessed in the forced swim test (FST), sucrose preference (SPT), Morris water maze (MWM) and novel object recognition (NORT) tests. Effects of the stress model on the GABAergic and serotonergic systems were measured using real-time PCR and ELISA respectively to detect expression of , α5 $GABA_A$ receptor, the serotonin transporter (5-HTT), the serotonin 1A receptor ($5\text{-}HT_{1A}$), and the concentrations of corticosterone (plasma), GABA and acetylcholinesterase (AChE) in the hippocampus and Prefrontal cortex (PFC). **Results:** Findings revealed that the CS group showed

increased immobility time in the forced swim test, time to reach the Morris water maze (MWM) platform, higher corticosterone level, and increased expressions of hippocampal and PFC 5-HT_{1A} and α5 $GABA_A$ receptors, and AChE compared to their control groups. In contrast, reductions in sucrose preference ratio, discrimination index in NORT, time in MWM target quadrant, and expression of hippocampal 5-HTT was noted relative to their control group. Compared to the 28-day corticosterone only group, PFC 5-$HT_{1A,}$ Hippocampal 5-HTT were reduced, while PFC 5-HTT, Hippocampal α5 $GABA_A$ receptors, and Hippocampal α5 $GABA_A$ receptors, and AChE concentrations were higher in the CS group. **Conclusion:** Our CS model induced depressive-like behavior with early cognitive deficits in rats affecting both hippocampus and PFC. The CS model may be useful in investigating new and comprehensive treatment strategies for MDD. **Key word:** Combined stress, Depression, Cognitive deficit, , α5 GABA receptors, GABA, 5-HT_{1A}, 5-HTT.

Disclosures: G. Ngoupaye: None. **M. Mokgokong:** None. **T. Madlala:** None. **M.V. Mabandla:** None.

Poster

P605. Early-Life Stress: Adolescence

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P605.13

Topic: F.04. Stress and the Brain

Title: Early Life Stress Inoculation Effects on Adult Anxiety-Related Behaviors

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Abstract: Stress is frequently discussed in the context of its harmful psychological and physiological effects. However, mild stressors may also build resiliency when encountering intense stressors later. This stress inoculation creates a buffering effect to reduce the negative outcomes associated with later stress. Rodent models have been used to investigate the resilience-promoting effects of early life inoculation protocols, such as brief handling by an experimenter. Further, environmental enrichment (EE) that involves exposure to small toys and access to physical exercise, may promote stress resilience by decreasing anxiety and depressive-like behaviors. The socially monogamous prairie vole is an ideal model to investigate whether stress inoculation protects against social stress given their social structure of pair bonding, living in family groups, and demonstrating negative behavioral and physiological responses to social stressors. The current study investigated if anxiety-like behaviors would be reduced during adulthood after early life exposure to either (EE) or brief handling. Male (n=24) and female (n=24) prairie voles were randomly assigned to either EE continuously (n =8/sex) or 10 seconds

of cup handling daily (n=8/sex) for 7 days, or left unmanipulated (n=8/sex) during the early life period [postnatal (PND) day 29-35]. On PND36, animals underwent a 4-week social isolation period and subsequently a 5-minute open field test. Preliminary data analyses indicate that EE animals spent significantly more time grooming in the open field compared to both unmanipulated and handling groups, with no sex effect. This observation may support prior research that EE reduces anxiety-like behaviors in socially isolated prairie vole adults. However, center section exploration was slightly reduced in EE and handling groups vs. the unmanipulated group, which may indicate slightly increased anxiety. It is possible that the lengths of the early life manipulations may need to be increased to achieve stress-resilience effects on anxiety-related behaviors, as seen in 21-day manipulations (Pryce et al., 2001, 2003), or that the manipulations may be more harmful than helpful when presented at an early age. This study provides a foundation for investigating potential early life manipulations to promote stress inoculation against social stressors in the prairie vole model. Further, the clinical implications may provide deeper insight into the effectiveness of early life resiliency strategies as forms of protection against adulthood social stress.

Disclosures: O. Akinbo: None. **L. Matuszewich:** None. **A.J. Grippo:** None.

Poster

P605. Early-Life Stress: Adolescence

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Program #/Poster #: P605.14

Topic: F.04. Stress and the Brain

Support: NIH P20 GM103643
NIH R15 HD091841

Title: The effect of early life trauma on amygdalar corticotropin releasing factor (CRF) expression and later life pain sensitivity

Authors: J. ZUKE, *M. BURMAN;
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Abstract: Increases in neonatal intensive care unit (NICU) admissions over the past several decades are likely responsible for the concomitant drop in infant mortality. However, the experiences these infants endure in the NICU leave them vulnerable to later life psychological disorders such as anxiety and depression. Utilizing a rodent model, our lab explores how a common NICU procedure (maternal separation, handling, and needle pricks to the paw) 4 times daily for the first 7 days of life, alters behavior and CRF receptor expression in juvenile rats. We've previously demonstrated that neonatal pain increases amygdala CRF expression during our neonatal manipulations. Additionally, we've observed that neonatally manipulated rats

display reduced amygdala CRF expression as juveniles. Moreover, rats that receive neonatal pain demonstrate reduced anxiety or fear during the early juvenile period, but demonstrate a stress-induced tactile hypersensitivity following a foot-shock stessor. We consider this a two-hit process, in which the neonatal pain creates a vulnerability to a pain-like state that is activated by the occurrence of later-life stress.The current studies further this work in two regards. First, we examine whether neonatal pain alters amygdala CRF receptor expression, in addition to altering the expression of CRF itself. Second, we also investigated whether a physiological stressor (Hind paw incision) is sufficient to produce the juvenile hypersensitivity. In Experiment 1: juvenile rats that experienced neonatal pain (or not) were subject to a secondary psychological stressor (using electric shock stress) or left undisturbed (basal) during the juvenile stage. Brain tissue was collected to examine amygdalar CRF receptor (receptors 1 and 2) expression using RNAscope® in situ hybridization (ISH). These results were quantified (number of cells, luminance of expression and area expressing) and compared to our previous ISH work quantifying CRF expression in the basolateral and central nucleus of the amygdala. In Experiment 2: juvenile rats that experienced neonatal pain (or not) were subject to a secondary physiological stressor (hind-paw incision) or left undisturbed (basal) during the juvenile stage. Subjects were tested in both a 2-day thermal conditioning place preference assay and single day hind paw weight bearing assay during the juvenile period. Only rats that received neonatal pain and the hind paw incision demonstrated a significant avoidance of the previously hot side, consistent with enhanced sensitivity. Together, these studies add to the growing that amygdalar CRF mediates the effects of neonatal trauma on subsequent pain sensitivity.

Disclosures: J. Zuke: None. **M. Burman:** None.

Poster

P680. Neuropharmacology: Appetitive Behavior and Sex Differences

Location: Virtual Only

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Program #/Poster #: P680.01

Topic: G.02. Reward and Appetitive Learning and Memory

Support: Vanderbilt University SOM Department of Pharmacology startup funds (E.S.C)
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Edward J. Mallinckrodt Jr. Foundation (to E.S.C)
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Title: Sex differences in cholinergic regulation of dopamine release mediate sexually dimorphic behavior

Authors: *L. J. BRADY[1,2], J. TAT[3], K. C. THIBEAULT[4,2], S. O. NOLAN[1,2], C. A. SICILIANO[1,4,2], E. S. CALIPARI[1,2,4];
[1]Pharmacol., [2]Vanderbilt Ctr. for Addiction Res., [3]Neurosci. Educ., [4]Vanderbilt Brain Inst., Vanderbilt Univ., Nashville, TN

Abstract: The mesolimbic dopamine system is involved in the expression of sex-specific behaviors and is a critical mediator of many psychiatric disease states. While work has focused on sex differences in the anatomy of dopamine neurons and relative dopamine levels, an important characteristic of dopamine release from axon terminals in the nucleus accumbens (NAc) is that it is rapidly modulated by local regulatory mechanisms independent of somatic activity. One of the most potent regulators of dopamine terminal function is through nicotinic acetylcholine receptors (nAChRs). In the NAc dopamine is released in tonic (slow and regular) and phasic (short, burst/spikes) patterns that are subject to heavy modulation by cholinergic (ChAT) systems through α4β2* containing nAChRs located directly on dopamine terminals. However, our understanding of the interaction between the ChAT and dopamine systems has been completed almost entirely in male subjects and our preliminary data show that this interaction is fundamentally different in females. Using ex-vivo fast-scan cyclic voltammetry combined with pharmacological antagonism of α4β2*-nAChRs with DhβE in NAc slices from males, naturally cycling (intact) and ovariectomized females, we find that ChAT regulation of dopamine release through nAChRs is not present in intact female mice. Deficits in nAChR modulation of dopamine release in intact females are not affected by the estrous cycle but can be rescued by ovariectomy – indicating that ovarian hormones play a role in this process. Previous work has shown that estradiol (E2) has affinity for nAChRs; however, these studies have shown that E2 is capable of inducing nAChR-mediated currents in cell culture and the consequences of these effects on dopamine release remain to be explicitly defined. We find that E2 increases dopamine release acutely, an effect that is blocked by antagonism of α4β2-nAChRs. Finally, we observed sex-specific effects of ChAT interneuron activity on reward-seeking behavior using Gq-DREADDs injected into the NAc of ChAT-cre$^{+/-}$ mice to selectively activate ChAT interneurons during positive reinforcement operant conditioning with sucrose. Overall, we show that circulating ovarian hormones alter the ability of α4β2*-nAChRs on dopamine terminals to modulate dopamine release in the NAc suggesting that sex differences in ChAT regulation of dopamine neurotransmission underlies sex-dependent differentiation in reward learning. Moving forward it will be critical to directly link these sex-differences to reward processing and reinforcement learning for the development of sex-specific pharmacotherapies to treat a variety of psychiatric disorders.

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Poster

P680. Neuropharmacology: Appetitive Behavior and Sex Differences

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P680.02

Topic: G.02. Reward and Appetitive Learning and Memory

Support: Wake Forest University & the Department of Psychology

Title: The effects of homeostatic-related peptides in the nucleus accumbens shell on palatable feeding in the rat, with and without concurrent μ-opioid receptor stimulation

Authors: *W. E. PRATT, A. E. CARNEY, C. DO, A. M. GROOME, A. J. SMITH, C. J. CALAFIORE;
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Abstract: Amongst its many important motivational functions, the nucleus accumbens (NAc) is central for regulating consumption of food based upon palatability, promoting intake on sweet and fatty foodstuffs high in caloric density that are hedonically pleasing. Nonetheless, the medial NAc shell receives inputs from the hypothalamus that indicate current energy state via release of the homeostatic-related peptides NPY, AGRP, CART, αMSH, orexin/hypocretin, and MCH, although the function of these signals on palatable feeding is not well-studied. This project examined if these homeostatic-related peptides affect feeding through direct actions on the medial NAc, and if they may interact with regional μ-opioid receptor signaling (known to enhance both hedonic responses to sugar solutions and increase food intake). 23 gauge stainless steel guide cannulas were surgically implanted bilaterally above the anterior medial NAc shell of male Sprague-Dawley rats. After recovery, non-restricted animals were given access to a high fat/sucrose diet for daily 2-hr feeding sessions. Food intake, water intake, and locomotion were monitored. Once each group of animals (N= 8/group) was acclimated to the feeding chamber and injection procedures, rats received bilateral NAc injections of vehicle (0.5 μl/side) or one homeostatic-related peptide, counterbalanced across days. On separate treatment days (all separated by at least 48 hrs), each group also received peptide injections with or without co-administration of the μ-opioid receptor agonist DAMGO (0.025 μg/side). NAc injections of the orexigenic peptide NPY (0, 0.67, and 1.0 μg/side) increased intake of the sweetened fat diet during the 2-hr session, both alone and when co-administered with DAMGO. AGRP (up to .94 μg/side) had no effect on palatable feeding within the session, though AGRP treatments led to weight gain over the next 24 hrs. Orexin A injections (at 3.56 μg/side) increased sweetened fat diet intake, but did not enhance the pro-feeding effect of μ-opioid receptor stimulation of the region. MCH did not significantly increase feeding on its own, but caused a biphasic effect on DAMGO-elicited intake, significantly decreasing consumption at 0.5 μg/side and enhancing it at 1.0 μg/side. The anorectic peptide CART (at 1.0 μg/side) inhibited feeding with and without DAMGO coinfusion, and reduced body weight in the next 24 hrs. αMSH had no significant effects on consumption of the sweetened fat diet. These findings suggest that homeostatic-related signals from the hypothalamus to the NAc have various and distinct effects on the consumption of palatable diets, in some cases modulating μ-opioid receptor stimulation of feeding.

Disclosures: W.E. Pratt: None. **A.E. Carney:** None. **C. Do:** None. **A.M. Groome:** None. **A.J. Smith:** None. **C.J. Calafiore:** None.

Poster

P680. Neuropharmacology: Appetitive Behavior and Sex Differences

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Program #/Poster #: P680.03

Topic: G.02. Reward and Appetitive Learning and Memory

Support: UNAM-DGAPA IN307720

Title: Role of rD4 receptors on the consumption of a sucrose solution and the appetite process

Authors: S. HERNÁNDEZ-CORREA, V. E. LÓPEZ-ALONSO, F. CORTÉS-SALAZAR, *R. E. ESCARTÍN-PÉREZ, J. M. MANCILLA-DÍAZ, ***D. DÍAZ-URBINA**;
Lab. of Neurobio. of Eating, UNAM-FES Iztacala, Tlalnepantla de Baz, Mexico

Abstract: Ingestion of sugar solutions induces binge-eating behavior (BEB). Moreover, the dopamine D4 receptor (rD4) is implicated in the preference for palatable diets. However, whether the central rD4 modulate the consumption of a palatable solution, altering the behavioral process controlling feeding has not been explored yet. Here we explored the preference for a sucrose solution (5%, 10% and 20% sucrose) in rats and then we evaluate the effect of the central blocking of rD4 on the microstructure of drinking behavior in satiated rats with access to a sucrose solution. In the first experiment, Wistar male rats had 7 days of access to the 3 sucrose solutions at onset of the light phase for one hour. We measured the consumption (1h) and the microstructure of drinking behavior of each solution, and a clear preference for the 20% solution was observed. The microstructure of drinking analysis indicated the increase of frequency and duration of bouts, and increase of meals and time-inter meals, which suggest the affectation of the appetite process, delaying the satiety development. In the second experiment, Wistar male rats had one hour of access only to the 20 % sucrose solution. After 10 days, animals received an intracerebroventricular injection of vehicle or the rD4 selective antagonist L745870 (1µg/5µl or 2 µg/5µl). We observed that L745870 (1 and 2 µg) decreased the sucrose consumption (g/1h). L745870-2µg increased bouts frequency but decreased the duration of bouts, time-inter bouts, and meal size, suggesting the decrease of the appetite for the solution. Togheter this findings suggest that rD4 are involved in the regulation of palatable solutions, suggesting a potential use of the rD4 for the treatment of BEB or obesity

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Poster

P680. Neuropharmacology: Appetitive Behavior and Sex Differences

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Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P680.04

Topic: G.02. Reward and Appetitive Learning and Memory

Support: DA016285-05
State of Washington

Title: The mGluR2 agonist LY379268 reduces sucrose taking, seeking, and motivation in male and female rats

Authors: *J. W. GRIMM, F. SAUTER, D. MACDOUGALL, E. SPAULDING, K. STENSGAARD, M. HARDY, K. R. GRIFFIN, B. MARX;
Psychology and Program in Behavioral Neurosci., Western Washington Univ., Bellingham, WA

Abstract: There are sex differences in the prevalence and severity of substance use disorders and in disordered eating and obesity rates. However, preclinical assessment of pharmacotherapies for the addiction behaviors underlying drug and food addiction, such as craving, have been conducted primarily with male rodents. For example, Bossert et al. (PMID 16834996) found that the mGluR2 autoreceptor agonist LY379268 reduced sucrose-seeking, but not sucrose-taking, in male rats. The aim of the present study was to explore the generality of the anti-craving ("anti-seeking") effect of LY379268 across sexes. In addition, the effect of the drug on motivation to receive sucrose was assessed using a progressive ratio (PR) schedule of reinforcement. Different groups of adult male and female Long-Evans rats (N = 91) were challenged with LY379268 in three experiments: 1.) fixed ratio (FR) schedule of reinforcement (taking), 2.) extinction of responding previously reinforced on the FR (seeking), or 3.) responding reinforced on a progressive ratio (PR) schedule of reinforcement (motivation). For each experiment, rats first responded for 10% liquid sucrose on a FR in 10 daily 2-h sessions. For the PR study, this was followed with training on a PR for 7 daily 3-h sessions. Rats were then challenged in a counterbalanced order with LY379268 (0, 1.5, 3, 6 mg/kg; IP; 30 minute pretreatment) on test days followed by either three reacquisition days of FR (experiments 1 and 2) or PR (experiment 3) responding. Female rats responded at a higher rate for sucrose on both the FR and PR. LY379268 reduced responding in a dose-related manner in all three experiments. There were no clear sex-specific effects of the drug on responding. These findings replicate previous findings of sex differences in sucrose taking, seeking, and motivation. The generality of the anti-taking, -seeking, and -motivation effects of LY379268 across male and female rats support further evaluation of this and other compounds that modulate synaptic glutamate as potential anti-addiction pharmacotherapies.

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Poster

P680. Neuropharmacology: Appetitive Behavior and Sex Differences

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Program #/Poster #: P680.05

Topic: G.02. Reward and Appetitive Learning and Memory

Title: Exercise tendencies modulate changes to opioid or exercise-induced reward

Authors: *A. R. TITUS;
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Abstract: Reward changes were observed in rodents with different exercise tendencies by utilizing the conditioned place preference (CPP) paradigm. Colleagues at the University of Missouri - Columbia established three distinct genotypes with reliable phenotypes among Wistar rats: low volume runners (LVR), high volume runners (HVR), and wild-types (WT). Adult male Wistar rats from each of these groups were given a rewarding stimulus of either access to a running wheel or an injection of morphine. Results indicated that there was no difference in the strength of CPP between the rewarding stimuli. Extinction was significantly more effective in LVR than HVR and WT animals, as LVR animals spent a lower percentage of time in their assigned conditioning chambers after eight days of exposure to the CPP paradigm without a rewarding stimulus. It was confirmed that running wheel access and opioid administration were comparably rewarding in all groups. The conclusion is that, compared to HVR and WT, LVR animals with their different genotypes and phenotype also have a unique pattern of behavioral responses to an opioid and to running.

Disclosures: A.R. Titus: None.

Poster

P680. Neuropharmacology: Appetitive Behavior and Sex Differences

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P680.06

Topic: G.02. Reward and Appetitive Learning and Memory

Title: Impact of prenatal escitalopram on probabilistic reversal learning and social approach in male and female offspring

Authors: *J. BEZENAH[1], A. TEJADA[1], G. KELLY[1], M. ESPINOSA[1], L. R. AMODEO[2]; [1]Psychology, California State University, San Bernardino, San Bernardino, CA; [2]Psychology, California State Univ. San Bernardino, San Bernardino, CA

Abstract: Selective serotonin reuptake inhibitors (SSRIs) are frequently used during pregnancy and while SSRIs are considered safe during pregnancy, there is limited knowledge of the long-term consequences of perinatal SSRI exposure on cognitive and behavioral development. SSRIs increase extracellular serotonin (5-HT), which contributes to their clinical effectiveness in major depressive and anxiety-related disorders. However, this hyper-serotonergic state during pregnancy can be transferred from the mother to the fetus through the placenta, causing potential alterations in neurotransmission and neural functioning. Further, recent human studies have demonstrated prenatal exposure to SSRIs in humans may increase susceptibility to autism spectrum disorder (ASD) and developmental delays. In a recent systematic review and network meta-analysis comparing the efficacy and acceptability of antidepressant drugs, escitalopram was shown to be one of the most effective antidepressants. However, as one of the newer SSRIs on the market introduced in 2002, there is still a lack of information on its safety profile during pregnancy. This study focused on behavioral flexibility, repetitive behaviors, and social behavioral deficits seen in ASD individuals. To this end, we administered escitalopram (0 or 10 mg/kg, s.c.) to nulliparous female Long-Evans rats for the first (E0-10) or last half (E11-20) of the embryonic period. Young adult male and female offspring were subsequently tested on a battery of behavioral tasks including probabilistic reversal learning task, open field conflict, marble burying and socialapproach task. Preliminary results demonstrate that prenatal escitalopram sex-dependently alters open field conflict, marble burying, and reversal learning but has noimpact on social behaviors

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Poster

P680. Neuropharmacology: Appetitive Behavior and Sex Differences

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Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIH R00 MH106731

Title: Serotonin 1B receptor mediated impulsivity is associated with increased subjective reward valuation

Authors: *S. S. DESROCHERS, K. M. NAUTIYAL;
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Abstract: Dysregulation of impulsive behavior is an important characteristic of many different psychiatric disorders, including ADHD and conduct, gambling, and substance use disorders. However, disordered impulsivity may be caused by the dysregulation of several different behavioral and cognitive substrates. Our prior preclinical studies have demonstrated that adult mice lacking serotonin 1B receptor expression (5-HT_{1B}R; tetO1B model) have increased impulsive action (e.g. inability to withhold responding) in tests such as the Go/No-go, as well as enhanced preference for larger rewards, but no difference in impulsive choice (e.g. intolerance to delays), making it an ideal model for studying the underlying causes of one kind of impulsivity. In the present results, we first confirm prior impulsive action findings using a different paradigm: the 5-choice serial reaction time test (5CSRTT). Mice lacking the 5-HT_{1B}R have increased premature responding (a measure of impulsive action), but show no differences in measures of attention. We hypothesize that this impulsive action phenotype is the result of changes in reward processing, specifically causing increased valuation of palatable substances and a subsequent inability to withhold responding for reward. Therefore, to directly examine reward valuation, we next studied hedonic responding for different concentrations of a palatable reward in a brief access lickometer protocol. Mice lacking the 5-HT_{1B}R reach maximum licking at relatively lower concentrations of reward than controls, indicating increased 'liking'. Finally, we examined how reward-associated stimuli influence behavior with a modified Pavlovian-to-instrumental transfer (PIT) test, where classically conditioned reward paired cues were subsequently used as an instrumental outcome. There are no genotype differences in acquisition of Pavlovian responding, but interestingly 5-HT_{1B}R knockout mice show enhanced instrumental responding for reward paired cues, which could be due to a subjectively increased reward value associated with the cue. Future work using variable reward amounts in wild type mice will more generally address how reward value impacts behavior this modified PIT test. Together these data suggest that an absence of 5-HT_{1B}R expression not only increases impulsivity, but also causes more general changes in subjective reward valuation. To determine whether these behavioral results are due to developmental or on-line adult mechanisms, data from an adult rescue of 5-HT_{1B}R expression are forthcoming.

Disclosures: S.S. Desrochers: None. **K.M. Nautiyal:** None.

Poster

P680. Neuropharmacology: Appetitive Behavior and Sex Differences

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P680.08

Topic: G.02. Reward and Appetitive Learning and Memory

Support: NIH

Title: The role of lateral hypothalamic orexin-glucose inhibited neurons in reward-based feeding

Authors: *S. TEEGALA[1], K. D. BECK[2], V. H. ROUTH[3];
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Abstract: Weight loss maintenance rarely succeeds suggesting that lasting changes occur in brain circuitry controlling food intake. Lateral hypothalamic area (LHA) orexin neurons drive "reward-based" feeding by activating the ventral tegmental area (VTA) dopamine (DA) neurons. LHA orexin neurons are inhibited by glucose (GI neurons). Weight loss decreases levels of the satiety hormone leptin and increases the orexigenic hormone ghrelin. Ghrelin directly enhances, while leptin suppresses the activation of orexin-GI neurons in low glucose. Increasing LHA glucose after weight loss blunts motivation for food reward. Based on these data, we hypothesize that *preventing changes in the glucose sensitivity of LHA orexin-GI neurons following weight loss blunts reward-based feeding and aids in weight loss maintenance.* We will first determine if glucose and ghrelin converge on the same signaling pathway in orexin-GI neurons. We found that pertussis toxin (PTX; 100 ng/ml), a G-protein coupled receptor (GCPR) inhibitor, blocked the effect of low glucose ($p<0.001$) in mice. PTX predominantly blocks Gαi/o which is negatively coupled to adenylate cyclase (AC) and it's downstream effector, protein kinase A (PKA). In support of this, the PKA inhibitor, Rp-cAMP (10μM) blocked the effect of low glucose (N=5 mice, $p<0.05$) on orexin-GI neurons while inhibition of protein kinase C (PKC; coupled to Gαq) did not. These data suggest that the inhibitory effect of glucose is due to a Gαi/o-mediated reduction in AC activity and that reducing glucose relieves this inhibition, allowing AC to activate PKA. We found previously that a 24-hr fast (high ghrelin/low leptin) increased activation of LHA orexin-GI neurons in low glucose and induced glutamate plasticity on VTA DA neurons. Here, we determined if weight loss also enhances glutamate transmission on VTA DA neurons. Compared to control adlib fed mice, mice weight restricted to 85% of their original body weight increased the amplitude of glutamatergic α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) currents (N = 6 mice, $P<0.05$), and the AMPA/N-methyl-D-aspartate (NMDA) receptor current ratio ($p<0.010$) on VTA DA neurons. Dialyzing glucose into the LHA of weight restricted rats during a progressive ratio test significantly reduced the number of active lever presses (N=6 rats, $p<0.01$) and breakpoint ($p<0.001$) for sucrose. These data suggest that weight restriction leads to persistent changes on VTA DA neurons by decreasing the glucose sensitivity of orexin-GI neurons and inhibiting these neurons could prove beneficial in long-term weight loss maintenance.

Disclosures: S. Teegala: None. **K.D. Beck:** None. **V.H. Routh:** None.

Poster

P680. Neuropharmacology: Appetitive Behavior and Sex Differences

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Program #/Poster #: P680.09

Topic: G.02. Reward and Appetitive Learning and Memory

Support: Stillman Drake Funding to P. J. C
Esther Hyatt Wender Research Fellowship to O. D
Reed College Neuroscience Fellowship to A. M. B
Reed College Science Research Fellowship K. J. C

Title: Glucagon-like Peptide-1 (GLP-1) Signaling in the Ventral Tegmental Area (VTA) Mediates Neuropeptide-Y (NPY) and Ghrelin Stimulated Operant Responding for Food and Alcohol Reward

Authors: *K. J. COLVIN, A. M. BRIGANDE, O. DAO, J. G. DARWICH, P. J. CURRIE; Psychology, Reed Col., Portland, OR

Abstract: Glucagon-like peptide-1 (GLP-1) is a gastric and brain derived neuropeptide recently implicated in natural reward and drug-induced euphoria. GLP-1 receptors are expressed throughout the brain, including mesolimbic neurons of the reward circuit. Here, it is hypothesized that GLP-1 interacts with additional brain peptides to alter reward salience. In the present study, we investigated the impact of the GLP-1 agonist, exendin-4 (Ex-4), on the stimulatory effect of neuropeptide Y (NPY) and ghrelin in relation to food and alcohol reward. In order to probe the interaction effects of GLP-1, NPY, and ghrelin within the ventral tegmental area (VTA), we measured alcohol intake or operant responding for sucrose pellets after the administration of GLP-1 and NPY, GLP-1 and ghrelin, or GLP-1 with combined NPY-ghrelin treatment in male Spraque-Dawley rats. Rodents were stereotactically implanted with guide cannulae to permit direct VTA administration. After training on a progressive ratio 3 (PR3) schedule, animals were first injected with Ex-4 into the VTA at doses 0-0.1 µg. Rats were then immediately injected with NPY (0-300 pmol) and/or ghrelin (0-200 pmol). Separate rats were administered similar treatment and then tested in an alcohol two-bottle choice paradigm. Our findings indicated that while NPY and ghrelin stimulated operant responding for sucrose pellets and increased alcohol intake, Ex-4 suppressed intake, and more importantly, significantly reduced the NPY and ghrelin-stimulated sucrose pellet and alcohol consumption. Overall, our findings suggest that VTA NPY and ghrelin administration increase operant responding for food and alcohol reward and that these effects are attenuated by VTA Ex-4 pretreatment. These data provide compelling evidence that central GLP-1, NPY, and ghrelin systems interact within the brain to modulate reward salience.

Disclosures: K.J. Colvin: None. **A.M. Brigande:** None. **O. Dao:** None. **J.G. Darwich:** None. **P.J. Currie:** None.

Poster

P680. Neuropharmacology: Appetitive Behavior and Sex Differences

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Program #/Poster #: P680.10

Topic: G.02. Reward and Appetitive Learning and Memory

Support: DGAPA UNAM IN232120

Title: Effects of CB2 receptor activation in the anterior cingulate cortex on motivation for a sucrose solution

Authors: E. M. HERNANDEZ-MORALES[1], J. C. RODRIGUEZ-AGUILAR[2], N. F. ROMERO-OCHOA[1], F. CORTÉS SALAZAR[1], V. E. LOPEZ-ALONSO[1], ***R. E. ESCARTIN-PEREZ**[1];
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Abstract: Researchers in the area of Neurobiology of obesity have focused on the brain mechanisms that regulate eating behavior to understand the neuronal processes, which contribute to abnormal weight gain. The areas of the reward circuit and those related to emotional regulation are fundamental, and one of the anatomic sites where the activity of such circuits converges is the anterior cingulate cortex (ACC), where cannabinoid neurotransmission has important neuromodulatory activity. Although it is well established that CB1 receptors (CB1r) consistently stimulate the consumption of hypercaloric and palatable food, contributions of CB2 receptors (CB2r) in this phenomenon are not yet completely clarified. To our knowledge, there are only a few reports that CB2r activation produces opposite effects to those produced by CB1r activation and that these effects are observed several hours after CB2r activation. Consequently, to evaluate the contribution of the CB2r of the rat ACC on the motivation for palatable food, we propose the hypothesis that the activation of CB2r would decrease the motivation for a sucrose solution (10%). Male Wistar rats (190-230 g, n= 12) were trained with a sequence of operant conditioning programs, starting with a fixed ratio 1 (FR1, 30 min), then with a fixed ratio 5 (FR5, 30 min), and finally with a progressive ratio program (RP, response ratio= $[5e^{(0.2 \times \text{trial number})}] - 5$) to measure motivation for the sucrose solution. PR sessions finished when the subject failed to obtain at least one reward in 60 min or in absence of responses in a 4-min period. Microinjection cannulas were stereotaxically implanted unilaterally in the ACC, and subjects received intra-ACC injections of vehicle (0.9% saline, 5% DMSO) or the CB2r agonist (GW405833, 0.25 µg). The effects of CB2r activation were evaluated in independent subjects at two different periods, 15 minutes and 8 hours after intra-CCA injections. We measured the response rate (lever presses/min), the total number of responses, and the breakpoints (number of lever presses to obtain the last reward) under the PR program. Our results showed that activation of CB2r did not produce significant effects on the breakpoints, and we only observed a tendency to decrease the motivation for the sucrose solution. However, when we analyzed the lever presses rate, we found that 8 hours after intra-ACC administration of GW405833, the percentage of responses per minute significantly decreased. Our results are consistent with the hypothesis that the activation of cB2r in the ACC may affect the reinforcing value of the sucrose solution. However, these effects are not detectable in the hour after the administration of the agonist.

Disclosures: E.M. Hernandez-Morales: None. **J.C. Rodriguez-Aguilar:** None. **N.F. Romero-Ochoa:** None. **F. Cortés Salazar:** None. **V.E. Lopez-Alonso:** None. **R.E. Escartin-Perez:** None.

Poster

P762. Opioids: Circuitry and Neurophysiology of Addiction I

Location: Virtual Only

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Program #/Poster #: P762.01

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH R01 EY029227

Title: Morphine accumulates in the mouse retina following systemic injection

Authors: *N. BERGUM[1], M. LAMCZYK[2], J. VIGH[2];
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Abstract: Melanopsin-expressing intrinsically photosensitive retinal ganglion cells (ipRGCs) mediate the entrainment of mammalian sleep-wake rhythms to environmental light. Interestingly, recent mouse studies from our lab revealed that ipRGCs express functional μ-opioid receptors (MORs), the primary molecular target for opioid analgesics. Furthermore, MOR agonists can directly inhibit ipRGC firing, which could prevent ipRGCs from regulating sleep-wake rhythms in response to light. In humans, opioid metabolites can be detected within eye following opioid use. However, it remains unclear whether opioids accumulate in the mouse retina following systemic exposure. To confirm the fidelity of a mouse model for studying how opioids might affect sleep-wake behavior via ipRGCs *in vivo*, we set out to investigate whether morphine reaches the mouse retina following systemic delivery. We collected tissue (retina and serum) from the adult male mice (2-4 months) at 8 different time points over a 12-hour period (n=8/time point) following a single 20 mg/kg intraperitoneal morphine injection. Morphine levels in serum and retina were measured using liquid chromatography-tandem mass spectrometry. Thus, we were able to establish a pharmacokinetic profile of morphine in the retina relative to serum. While morphine levels were high in both tissues initially, morphine persisted in the retina whereas serum morphine levels decreased to near undetectable levels after three hours. Interestingly, results from this study not only show that systemically administered morphine is deposited in the retina, but also that morphine remains there long after the drug has been metabolized in the blood. This supports the idea that opioids could persistently activate MOR on ipRGCs, altering the ability of ipRGCs to transmit light information to the brain's sleep-wake circuitry.

Disclosures: N. Bergum: None. **M. Lamczyk:** None. **J. Vigh:** None.

Poster

P762. Opioids: Circuitry and Neurophysiology of Addiction I

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Program #/Poster #: P762.02

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant DA009815
NIH Grant UG3 DA030325
PA DOH TSF SAP #410007972

Title: Glucagon-like peptide-1 analog reduces heroin seeking and taking in rats

Authors: J. DOUTON, N. HORVATH, A. HAJNAL, ***P. S. GRIGSON**;
Neural and Behavioral Sci., Penn State Univ. Col. of Med., Hershey, PA

Abstract: Drug addiction is a chronic brain disease characterized by the uncontrolled use of a substance. Due to its relapsing nature, addiction is difficult to treat, as individuals can relapse following even long periods of abstinence and, it is during this time, that individuals are most vulnerable to overdose. In 2020, America reported the highest number of overdose deaths ever recorded. Opioid overdoses, in particular, have been increasing for decades, making it imperative that we find new treatments for opioid use disorder. We hypothesize that drugs of abuse generate a state of 'need' in the individuals that can only by corrected by more drug. This state is akin a state of physiological need such as thirst or hunger. Indeed, hormones that modulate hunger and satiety, like glucagon-like peptide-1 (GLP-1) and its analogs, have shown promise in reducing motivated behaviors for cocaine, amphetamines, nicotine, alcohol, and recently opioids. In this study, we test the effectiveness of the GLP-1 analog, liraglutide (LIR), in reducing heroin addiction-like behavior, and the potential side effects associated with the treatment. We show that daily treatment with LIR (0.1 mg/kg sc) increased the latency to take heroin, reduced heroin self-administration, prevented escalation of heroin self-administration, and reduced drug-induced reinstatement of heroin seeking behavior in rats tested 6 h later. A one-hour pretreatment time, however, was not effective in reducing cue-induced seeking. Moreover, we showed that, while LIR (0.1, 0.3, 0.6 and 1.0 mg/kg sc) supported conditioned taste avoidance of a LIR-paired saccharin cue, it did not elicit intake of the antiemetic kaolin. Further, while repeated administration of LIR did not reduce water intake, it did reduce food intake and body weight gain at all doses, except 0.1mg/kg. Overall, the data show that LIR is effective in reducing heroin taking and heroin seeking at doses that do not cause malaise and have a modest effect on food intake and body weight gain.

Disclosures: J. Douton: None. **N. Horvath:** None. **A. Hajnal:** None. **P.S. Grigson:** None.

Poster

P762. Opioids: Circuitry and Neurophysiology of Addiction I

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Program #/Poster #: P762.03

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Center on Compulsive Behaviors (CCB) Intramural Fellowship

Title: Regional and evolutionary conservation of opioid-mediated suppression of parvalbumin cell inhibition

Authors: *A. P. CACCAVANO, K. A. PELKEY, C. J. MCBAIN;
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Abstract: The opioid system in the central nervous system plays a crucial role in pain sensation, stress response and mood, and is a key target for drugs of abuse. With a high risk of dependency, mortality from respiratory depression, and epidemic levels of abuse, opiates pose significant public health challenges. Although there have been significant advances in understanding the neural substrates of endogenous and exogenous opiates, critical gaps remain in our understanding of how opiates modulate different neuronal subtypes and their activity within microcircuits. Within the hippocampus, opiates suppress the activity of inhibitory parvalbumin-expressing (PV) cells, and thus disinhibit excitatory pyramidal cells (PCs). This effect is primarily mediated through mu opioid receptors (MORs), which are associated with the reward-processing of both natural stimuli and drugs of abuse. While these insights from the rodent hippocampus have been critical for understanding the effect of opiates in the brain, it is unknown if opioid-mediated suppression of inhibition is conserved in other cortical regions, or in higher species. This study utilized an optogenetic approach, employing Cre-recombinase mice to selectively express a light-sensitive opsin in inhibitory PV cells. Light-evoked inhibitory synaptic currents were recorded in excitatory PCs via whole cell patch clamp, and the opioid sensitivity assessed with a MOR-specific agonist (DAMGO) and antagonist (CTAP). Excitatory PCs were recorded within hippocampal subregions, as well as visual, motor and prefrontal neocortex. Consistent with prior studies, light-evoked responses consistently exhibited robust DAMGO sensitivity in the CA1 region of hippocampus. However, across neocortical regions this DAMGO sensitivity was highly variable, indicating a potential specialization of hippocampal PV cells. To assess if opioid-mediated suppression of PV-cell activity is evolutionarily conserved, similar experiments will be conducted in non-human primates utilizing a viral strategy targeting the evolutionally-conserved and PV-specific E2 enhancer of the voltage-gated sodium channel gene Scn1a.

Disclosures: A.P. Caccavano: None. **K.A. Pelkey:** None. **C.J. McBain:** None.

Poster

P762. Opioids: Circuitry and Neurophysiology of Addiction I

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Topic: G.09. Drugs of Abuse and Addiction

Support: NIDA Grant UH3 DA048353
NIDA Grant R01 DA047855

Title: Dual agonism at mu-opioid and adrenergic-alpha$_2$ receptors are necessary to produce the discriminative-stimulus effects of the primary kratom alkaloid mitragynine in rats

Authors: *M. L. CROWLEY[1], A. PATEL[2], S. OBENG[2], L. F. RESTREPO[2], L. R. GAMEZ-JIMENEZ[2], N. P. HO[2], F. LEON[1], M. M. MOTTINELLI[1], C. R. MCCURDY[1], L. R. MCMAHON[2], T. HIRANITA[2];
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Abstract: The kratom alkaloid mitragynine may combine two critical pharmacological mechanisms in a single molecule, mu-opioid and adrenergic-alpha$_2$ agonism, that currently is achieved clinically with separate drugs (e.g., methadone and lofexidine, respectively, for opioid use disorder/withdrawal syndrome). We have demonstrated that the mu-opioid receptor agonist morphine (15 min prior to sessions) partially mimics the discriminative stimulus effects of the primary kratom alkaloid mitragynine (32 mg/kg, i.p.) in Sprague-Dawley rats. Moreover, our group has shown that the antiallodynic effects of mitragynine in rats are blocked by the adrenergic-α_2 receptor (Aα_2R) antagonist yohimbine. Using rats discriminating i.p. 32 mg/kg of mitragynine from its vehicle, here we compared time-effect functions (5, 15, 30, 60, 90, 120, 180, 240, 360, and 240 prior to sessions) of substitution of morphine, the Aα_2R agonists (clonidine and lofexidine) and antagonist (yohimbine). Greater than 80% drug-lever responding was produced by morphine at 3.2 mg/kg 60 min post-injection, clonidine at 0.1 mg/kg 60 min post-injection, and lofexidine at 0.56 mg/kg 180 min post-injection. At the dose producing full substitution, morphine did not significantly modify rates of lever-pressing, thermal antinociception in the hotplate assay at 52°C, or thermomodulation, i.e., rectal temperature. However, clonidine and lofexidine decreased rates of responding to 25% and 50% of control and rectal temperature by 3.8°C and 3.9°C, neither clonidine nor lofexidine significantly modified hotplate response latency. The dose-effect function of mitragynine discrimination was shifted rightward and downward by the opioid receptor antagonists naltrexone (0.032 mg/kg) and naloxone (0.1 mg/kg), as well as by the Aα_2R antagonists yohimbine and atipamezole (both at 3.2 mg/kg). These results suggest that mitragynine exerts its in vivo effects via dual agonism at μ-opioid and adrenergic-α_2 receptor.

Disclosures: M.L. Crowley: None. **A. Patel:** None. **S. Obeng:** None. **L.F. Restrepo:** None. **L.R. Gamez-Jimenez:** None. **N.P. Ho:** None. **F. Leon:** None. **M.M. Mottinelli:** None. **C.R. McCurdy:** None. **L.R. McMahon:** None. **T. Hiranita:** None.

Poster

P762. Opioids: Circuitry and Neurophysiology of Addiction I

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Program #/Poster #: P762.05

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant DA050325

Title: Acute glucagon-like peptide-1 receptor agonist liraglutide prevents cue-, stress-, and drug-induced heroin seeking in rats

Authors: *J. E. NYLAND[1], J. E. DOUTON[2], S. L. MILLS-HUFFNAGLE[1], N. K. ACHARYA[1], D. SUN[1], P. S. GRIGSON[1];
[1]Penn State Col. of Med., Hershey, PA; [2]Univ. of North Carolina, Chapel Hill, NC

Abstract: In the last decade, opioid use disorder (OUD) has been a threat to public health, being declared an epidemic by the Centers for Disease Control (CDC). The recent COVID-19 pandemic has intensified the crisis, with a national 29% increase in overdose deaths from October 2019 to September 2020, and preliminary data from the CDC suggesting overdose deaths in 2020 surpassed those during the peak of the opioid epidemic. For those who decide to seek medication for addiction treatment (MAT), initiating buprenorphine among patients who are not yet experiencing withdrawal may paradoxically precipitate withdrawal. For this reason, many MAT pathways recommend initiating treatment only among patients already experiencing symptoms of withdrawal and subsequently titrating the dose during an observation period. During this period, patients are particularly vulnerable for relapse. Recent research has shown that hormones that modulate hunger and satiety can also modulate reward behavior associated with drugs of abuse. For example, the short-acting analog of glucagon-like peptide-1 (GLP-1), an incretin hormone that regulates homeostatic feeding, has been shown to reduce responding for normally rewarding stimuli such as food, cocaine, heroin and nicotine in animal models. In this study, we tested the effects of a single, acute exposure to the long-acting GLP-1 analog, liraglutide, on a rat model of heroin reinstatement. We found that, in rats with heroin self-administration experience, an acute dose of 0.3 mg/kg liraglutide (sc) was effective in preventing relapse after exposure to the three major precipitators of relapse: drug-associated cues, stress, and the drug itself. Finally, we confirmed that the effects observed in drug seeking are not due to a locomotor depression or impairment, as liraglutide did not significantly alter performance in a rotarod test. These results provide an avenue to explore non-opioid treatments for OUD in

humans that can bridge the gap between cessation of opioid use and initiation of MAT to prevent relapse.

Disclosures: J.E. Nyland: None. **J.E. Douton:** None. **S.L. Mills-Huffnagle:** None. **N.K. Acharya:** None. **D. Sun:** None. **P.S. Grigson:** None.

Poster

P762. Opioids: Circuitry and Neurophysiology of Addiction I

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Program #/Poster #: P762.06

Topic: G.09. Drugs of Abuse and Addiction

Support: NIH Grant DA049504

Title: Reduced Dopamine Terminal Function in the Nucleus Accumbens following Heroin Self-Administration

Authors: *B. GEORGE, M. H. DAWES, E. G. PECK, S. R. JONES;
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Abstract: Recent evidence has illustrated that heroin produces its rewarding and addictive effects, at least partially, through activation of the mesolimbic dopamine system. Notably, it has been shown that heroin seeking can be attenuated by systemic administration of antagonists of negative regulators of the dopamine system, such as kappa opioid receptors (KOR) or presynaptic D3 receptors (D3R), thus increasing dopamine levels. Therefore, it is likely that withdrawal from chronic heroin exposure drives a state of hypodopaminergia in the nucleus accumbens (NAc), as previously observed following withdrawal from chronic stimulant and alcohol use. To this end, this study aimed to (1) investigate alterations in the dopamine terminal function following heroin self-administration in male and female rats and (2) identify a mechanism for hypodopaminergia following chronic heroin self-administration. Adult male and female Long Evans rats were trained to self-administer heroin (0.05 mg/kg/inf) and then placed on long access (LgA; FR1, 6-hr session, unlimited infusions, 0.05 mg/kg/inf) heroin self-administration paradigm to induce escalation of heroin intake. Following LgA, rats were utilized for neurochemical analyses - *in vivo* microdialysis and *ex vivo* fast-scan cyclic voltammetry (FSCV) in the NAc. The results support previous literature in that following LgA, male and female rats had decreased basal extracellular levels of dopamine as well as a reduced dopaminergic response to a heroin challenge (0.1 mg/kg/inf, IV) in the NAc during withdrawal from heroin self-administration. FSCV results revealed that heroin exposed rats have reduced dopamine release during single-pulse stimulations but increased phasic-like dopamine release during stimulation trains (5 pulses, 5-100Hz) when compared to their heroin naïve counterparts. In addition, we found that presynaptic D3R and KOR activity in the NAc was increased

following LgA in male and female rats. These results reveal a marked reduction in dopamine system function following heroin exposure and identify a potential mechanism in the NAc that may drive the hypodopaminergic state observed during heroin withdrawal.

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Poster

P762. Opioids: Circuitry and Neurophysiology of Addiction I

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Topic: G.09. Drugs of Abuse and Addiction

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NIH Grant GM008620
NIH Grant MH096889
NIH Grant DA044118

Title: Unexpected role of endogenous opioid receptors in adversity-induced addiction in females

Authors: *S. C. LEVIS, M. T. BIRNIE, N. KAMEI, J. L. BOLTON, J. S. MONTESINOS, C. R. PERRONE, T. Z. BARAM, S. V. MAHLER;
UC Irvine, Irvine, CA

Abstract: Background: Early life adversity (ELA) is associated with vulnerabilities to reward-related problems, such as addiction to pro-hedonic opioid drugs. Women may be particularly vulnerable, suggesting a sex-specific derangement of reward circuit maturation by ELA. However, the mechanisms by which this occurs are poorly understood. We employed a naturalistic model of ELA in rats to examine the impacts of ELA on functional maturation of the reward circuit, focusing on disruption of opioid drug seeking behaviors. **Methods:** Adult female ELA-experienced rats were tested for aspects of opioid addiction-like behaviors including free consumption and economic demand elasticity to measure motivation for opioid drugs. ELA-induced changes in reward circuitry were examined using c-Fos expression as a marker of neurons activated by heroin. To gain insight into ELA-induced molecular changes in reward-related regions, we employed RT-qPCR for a suite of molecular candidates. Finally, following on our intriguing molecular findings, we pharmacologically manipulated endogenous opioid signaling during opioid self-administration to test the mechanisms of ELA-enhanced opioid seeking. **Results:** ELA led to enhanced motivation for opioid drugs in female rats, in accord with our prior findings. Reward circuit activation by heroin distinguished ELA from control females, with differential activation in nucleus accumbens. Preliminary data suggest that these ELA-induced vulnerabilities and opioid-induced activity may involve altered opioid receptor expression and signaling in specific nodes of the in the reward circuit. **Conclusions:** ELA

produces a sex-specific pro-addiction phenotype in female rats, which may be caused by disrupted functional maturation of reward circuits, leading to aberrant neural activity and opioid signaling upon opioid exposure.

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Poster

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Topic: G.09. Drugs of Abuse and Addiction

Support: NIH/NIDA P50DA005010

Title: Bac-mediated overexpression of striatopallidal neuron specific gene gpr6 heightened aversive effects of opiate withdrawal

Authors: *J. LINDEN, C. S. PARK, H. LAM, N. T. MAIDMENT, X. W. YANG; Psychiatry and Biobehavioral Sci., UCLA, Los Angeles, CA

Abstract: The G protein-coupled receptor GPR6 is highly selectively expressed in the striatopallidal or D2 medium spiny neurons (MSNs) in the striatum. Our prior study mice deficient in *Gpr6* showed elevated reward-driven instrumental conditioning behavior. Here, we hypothesize that *Gpr6* function, in part through its constitutive signaling to increase cAMP production in D2-MSNs, could also modulate aversive behaviors such as learned avoidance tasks. Importantly, the D2-MSNs in the nucleus accumbens has been shown to be critical for aversive learning and, in the context of drug addiction, avoidance behavior associated with opiate withdrawal.

Here, we genetically test the in vivo function of GPR6 in modulating the aversive effects of opiate withdrawal. We developed a new conditional BAC transgenic mouse model (BAC-*Gpr6*) overexpressing murine *Gpr6* under its own genomic regulation. We evaluated these animals using a conditioned place aversion (CPA) assay using the μ opioid receptor antagonist naloxone, either alone or after chronic morphine exposure. We showed that in BAC-*Gpr6* mice, aversion to the drug-paired compartment was induced for a low dose of naloxone that failed to produce any avoidance in wildtype controls. Similarly, in naloxone-induced morphine withdrawal CPA, we found that low dose naloxone potentiates withdrawal avoidance in BAC-*Gpr6* but not in wildtype animals. To decipher the possible transcriptomic mechanism through which Gpr6 modulates withdrawal aversion, we subjected BAC-Gpr6 mice and wildtype controls to low-dose naloxone and performed RNA sequencing in tissue from dorsolateral and ventral striatum, two regions enriched in μ opioid receptor. We will present our findings on genes that are

differentially expressed based on Gpr6 gene dosage increase and hence could play a role in modifying aversive behaviors in opiate withdrawal.

Disclosures: J. Linden: None. **C.S. Park:** None. **H. Lam:** None. **N.T. Maidment:** None. **X.W. Yang:** None.

Poster

P833. Hippocampal Interactions II

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Program #/Poster #: P833.01

Topic: H.08. Learning and Memory

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Title: Modulation and Visualization of Hippocampus-driven Defensive Networks

Authors: *K. E. DORST[1], R. A. SENNE[2], S. RAMIREZ[2];
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Abstract: This research aims to provide a framework for the network interactions that govern differential defensive responses to aversive mnemonic stimuli. Specifically, both the brainwide and behavioral effects of artificially reactivating a particular fearful memory in a given region are largely unknown. Defensive behaviors, which are aberrantly expressed in PTSD and anxiety, include both active avoidance and passive freezing but manifest depending on the brain-state and the environment of the animal. Here, we alter environmental contingencies to test for the capacity of a defined set of hippocampal cells to differentially drive defensive behaviors when optogenetically activated. Our preliminary results show that artificial reactivation of the same set of cells processing fear manifests as anxiogenic responses, or freezing behavior, depending on whether these cells are stimulated in a large open field, or in a small chamber. These results suggest that a subset of cells, upon activation, recruit different neural substrates to generate diverging behavioral outputs. Our current work utilizes immunohistochemical and graph theory analyses to identify candidate regions mediating state-dependent defensive behavioral switches. Together, our work provides insight into the capacity of discrete sets of cells to produce various behavioral responses.

Disclosures: K.E. Dorst: None. **R.A. Senne:** None. **S. Ramirez:** None.

Poster

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Center of Systems Neuroscience and Neurophotonics Center at Boston University

Title: Erasable hippocampal neural signatures predict memory discrimination

Authors: N. KINSKY[1], D. ORLIN[2], ***E. RUESCH**[3], S. RAMIREZ[3];
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Abstract: Despite the ubiquitous use of contextual fear conditioning (CFC) to study episodic memory, there is a knowledge gap pertaining to how neural code evolves during the consolidation of short-term memory into long-term memory. Previous studies have demonstrated that CFC causes a reorganization, or remapping, of hippocampal place fields. This phenomenon suggests that the hippocampal neural code may provide a substrate for the retention and recall of CFC memories. Here, we leveraged the spatial and temporal resolution of single-photon calcium imaging in conjunction with CFC to further characterize the heterogeneity of hippocampal remapping over long-time scales in freely moving mice. We exposed mice to two different arenas throughout the experiment so we could test the specificity of the CFC memory. Moreover, we tracked behavior in the days before, during, and after shock to examine the long-term stability of the CFC memory. A cohort of mice received anisomycin, a protein synthesis inhibitor that disrupts long-lasting synaptic plasticity, immediately after shock to block consolidation. We hypothesized that anisomycin would temporarily arrest learning-related changes in the event rate of neurons and turnover of active neurons between sessions, and would prevent remapping of their place field locations. We found that freezing returned to pre-shock levels in the days following CFC for anisomycin mice but not for control mice corroborating previous findings. Surprisingly, we also observed that anisomycin temporarily accelerated the turnover of active neurons. This increased cell turnover was concomitant with a general shutdown of neural activity up to two days later. Thereby, we posit the amnestic effects of anisomycin could stem from the disruption of constitutive processes in conjunction with blocking learning-related plasticity. Additionally, the degree to which mice discriminated between arenas correlated with the amount of cell turnover between the arenas. This finding supports the idea that the hippocampal neural code provides a substrate for differentiating between arenas. Future analyses will investigate whether cell turnover between arenas relates to cell turnover across days and how freezing-related neural responses evolve through the course of consolidation and amnesia. Finally, we are

currently working to utilize single-photon calcium imaging to investigate the heterogeneity of CFC induced remapping in the dentate gyrus sub-region of the hippocampus. Together, our data provide physiological insight on how hippocampal cell populations are differentially contributing to the processes underlying memory consolidation and recall.

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Title: Dorsal versus Ventral Lateral Septum in the Regulation of Fear

Authors: *C. A. RIZZI-WISE, D. V. WANG;
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Abstract: Appropriate behavioral instincts such as fear or avoidance behaviors in response to threat are critical for survival. However, excessive or misplaced fear is maladaptive and can have a negative impact on daily living for people who suffer from fear-associated disorders such as PTSD. With this clinical implication, there are still many avenues regarding the neural circuitry underlying the regulation of fear that have yet to be comprehensively studied. Recently gaining interest, the Lateral Septum (LS) is positioned to have an important role in the regulation of affect, including fear, given its critical position in relaying spatial and contextual information from the hippocampus to downstream hypothalamic regions. Using optogenetics and a contextual fear conditioning procedure, our data reveals the presence of two functionally different yet parallel pathways that originate in the hippocampus and project to the LS: the dorsal (d) CA1-to-dLS pathway likely increases the expression of fear whereas the ventral (v) CA1-to-vLS pathway decreases the expression of fear. To further explore these differences, our use of in vivo electrophysiology has uncovered firing property differences between the dLS and vLS relating to how this region is able to regulate different affects, with a focus on fear. In line with our findings that the dLS versus vLS has opposing behavioral controls, this appears to manifest from opposing firing changes as well. In response to different affective or neutral environments, the dLS has a pattern of increasing its firing rate whereas the vLS has more neurons that decrease their firing rate.This in-depth research will provide a novel understanding of LS circuitry which is helpful for the development of advanced treatment options for those with dysregulated behaviors relating to negative emotions.

Disclosures: C.A. Rizzi-Wise: None. **D.V. Wang:** None.

Poster

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Title: Two populations of retrosplenial cortical neurons in memory consolidation

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Abstract: About one third of life is spent asleep, indicating that this offline state serves an important physiological relevance. During sleep, a process called memory consolidation occurs where newly acquired information is transformed into long-lasting memories. It is well-established that this process consists of hippocampal-cortical communication, where hippocampal ripple oscillations (150-250 Hz) coordinate with cortical slow-wave delta oscillations (1-4 Hz). These cortical delta oscillations have a cyclic pattern: the Up state of each cycle consists of increased neuronal activity, whereas the Down state of each cycle consists of almost complete neuronal silence. We recently have shown an interaction between the retrosplenial cortex (RSC) and hippocampus, such that clusters of RSC neurons fire at distinct time points in relation to the hippocampal ripple oscillation during sleep: RSC Up states precede the hippocampal ripple, while RSC Down states follow the hippocampal ripple. This suggests that RSC population activity may orchestrate memory consolidation. Here, we employed *in vivo* electrophysiology and advanced data analyses, including cell assembly analysis, to study RSC neuronal activity in freely behaving mice during consolidation. Interesting, our results revealed that ~25% of the RSC population started to fire at the Down state and reached maximal firing at the Down-to-Up transition. Therefore, we termed these RSC neurons the Down state assembly (DSA), and the remaining RSC neurons as non-DSA. We found that DSA activity preceded non-DSA activity, leading us to hypothesize that the DSA may initiate and maintain subsequent non-DSA network activity crucial to memory consolidation. In support of this, DSA activity increases during consolidation of a contextual fear experience. Although preliminary cross-correlation analyses reveal a low probability of DSA neurons directly interacting with non-DSA neurons, DSA neurons may project to another region for the initiation and maintenance of non-DSA network activity. Ongoing analyses will further investigate patterns within non-DSA activity, along with the interaction between these two RSC subpopulations. Results from this study may reveal that DSA activity coordinates activity of select non-DSA populations, and whether this activity drives the communication with other relevant brain regions to enable the

processing of distinct memories. Altogether, these findings will provide insight on the mechanisms of memory, which could shed light on how detailed information is integrated and organized into brain networks.

Disclosures: A.N. Opalka: None. **D.V. Wang:** None.

Poster

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Title: Emerging neural ensemble codes in the hippocampal-amygdala circuitry underlie memory formation

Authors: *J. LIU, D. V. WANG;
Drexel Univ., Philadelphia, PA

Abstract: Memory formation requires communication between the hippocampus and its functionally connected brain regions. Although it is well-established that the hippocampal-amygdala circuitry is essential for contextual fear memory, it remains unknown how the neural ensembles between these two brain regions cooperate to form a stable fear memory. We focused on studying memory acquisition and subsequent consolidation by employing a contextual fear conditioning procedure. First, we applied an independent component analysis for cell assembly detection of data recorded in the basolateral amygdala (BLA) during the post-training slow-wave sleep (SWS), a stage critical for memory consolidation. We found that only one BLA cell assembly (consisted of ~5% of the neurons) was strongly regulated by hippocampal CA1 ripples, indicating a specificity of hippocampal-BLA communication for memory consolidation. Moreover, all neurons from this assembly started to fire robustly after the mouse received footshocks during training, and were reactivated during a subsequent recall test. Our further analyses showed that this cell assembly was modulated by ripples only during the post-, but not pre-training sleep. A rapid hippocampal-BLA information flow around the times of sharp wave ripples was confirmed by generalized linear models (GLMs). Importantly, unlike the BLA, the vast majority of CA1 neurons were recruited in the memory acquisition and subsequent consolidation and retrieval processes, indicating two distinct strategies for information coding between the hippocampal-BLA dialogue. These results suggest that a BLA fear memory cell assembly is rapidly generated during training, subsequently consolidated with hippocampal CA1 during post-training sleep, and maintained for fear memory retrieval, whereas most of the CA1

neurons participate in the same processes. Our data provide new insights into the understanding of the mechanism of memory formation in the hippocampal-BLA network.

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Poster

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Program #/Poster #: P833.06

Topic: H.08. Learning and Memory

Title: Organization of oscillatory activity across layers and cell types of the granular retrosplenial cortex in awake mice

Authors: *K. ARNDT[1], L. M. F. KLAVER[1], E. T. GILBERT[2], S. A. MCKENZIE[4], D. F. ENGLISH[3];
[2]Sch. of Neurosci., [3]Neurosci., [1]Virginia Tech., Blacksburg, VA; [4]UNM, Albuquerque, NM

Abstract: The hippocampus (HPC) is the seat of learning and memory where theta and sharp wave ripples organize place cells to tile experience. Downstream regions must be able to read out this information for memory consolidation to occur. Recently, the gRSC has been identified to exhibit similar activity to CA1 (i.e. theta, sharp wave ripples, and place cells). However, how this activity is produced in gRSC and its role in learning and memory is far less understood than in HPC CA1.
By densely sampling (20um resolution) the LFP across all layers of the gRSC in awake head-fixed mice using linear 64 channel silicon electrodes we aim to understand how the gRSC circuitry supports oscillatory activity for learning and memory. We identify high frequency oscillations (HFOs) in the 150-250 Hz frequency band, sharp waves, theta oscillations, and theta-nested gamma oscillations occurring in different layers across the gRSC. Specifically, sharp waves and HFOs are predominantly found on layer (L) 1 and L2/3 respectively. Theta and gamma oscillations are distributed across the layers. We also show differences in putative pyramidal cell and interneuron (Cell Explorer) responses to these distributed LFP activities. Pyramidal cells and interneurons increase firing rate at times of sharp waves in L1 and HFOs in L2/3. As well as distributed phase and frequency locking to the different LFP oscillations found across the layers. These findings show important network dynamic differences between the layers of the gRSC. Functionally, this implies each layer has different responsibilities in information processing and provides the neuroscience community with a more detailed understanding of the layer specific differences in the gRSC.

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Poster

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Title: Oscillations in the amygdala and hippocampus during an affective object-context association task

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Abstract: Oscillations in the amygdala and hippocampus during an affective object-context association task
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Key words: rat, hippocampus, amygdala, affective salience, context, memory
Abstract
The amygdala and hippocampus are bidirectionally interconnected and support complementary functions. In particular, the hippocampus is essential for remembering associations between objects and spatial contexts, and the amygdala can in turn modulate those hippocampus-dependent memories. For example, optogenetic stimulation of the basolateral complex of the amygdala (BLA) in a previous study accelerated acquisition of hippocampus-dependent object-context associations. Under naturalistic conditions, the BLA is engaged by stimuli with affective or social salience. For instance, pharmacological inactivation of the BLA in a previous study altered female rats' spontaneous preferences for conspecific urine odorants relative to plant-based non-social odorants. The present study addressed how affective odorants influenced oscillations in the BLA and hippocampus as rats performed a hippocampus-dependent object-context association task. Female rats learned to associate objects, which were small pots filled with scented digging media, with spatial contexts, which were visually-distinct boxes. Specifically, two objects (A and B) were differentially rewarded in two boxes by burying a food reward in only one object in each context (i.e., Box 1: A+B-; Box 2: A-B+). Local field potentials were recorded from the BLA and ventral hippocampus as rats performed the task under four different conditions. All four conditions included one object scented with a plant-based odorant. The second object was scented with one of the following: conspecific male urine, conspecific female urine, fox urine, or a second plant-based odorant. The overall question of interest was whether oscillatory activity in the BLA and hippocampus would differ between

conditions. Preliminary results suggested that oscillatory dynamics in the BLA and hippocampus correlated with task performance. The results have implications for understanding bidirectional interactions between the amygdala and hippocampus.
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Poster

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Title: Hippocampal-amygdala memory circuits govern experience-dependent observational fear

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Abstract: The empathic ability to vicariously experience the other's fearful situation, called observational fear (OF), is critical for survival. In OF, an observer witnesses a demonstrator in an aversive situation and responds with fear. Rodent studies have established behavioral models of OF and identified several brain regions and molecules important for OF. In particular, neural processes in the anterior cingulate cortex (ACC) and basolateral amygdala (BLA) networks are considered to be the center for OF. It is well known that both prior similar fear experience in the observer and social familiarity with the demonstrator can facilitate OF. Such a unique feature allows individuals to increase their empathic ability. For example, if the demonstrator's reaction to aversive stimuli is robust, the observer expresses OF without prior experiences (we refer to as Naive OF), which has been generally used as a rodent OF model. However, in nature and our lives, the demonstrator's reaction is often ambiguous and difficult to understand. To fully understand the other's situation, the observer uses both prior similar fear experience and social familiarity with the demonstrator, which facilitates OF. Some studies suggest that ACC activity

might be modulated by prior similar fear experience in the observer and social familiarity with the demonstrator. However, the neural circuit mechanisms of experience-dependent OF (Exp OF) have not yet been systematically investigated, and it remains unknown whether ACC and BLA are essential for Exp OF. Moreover, the hippocampus (HPC), particularly dorsal HPC (dHPC), is crucial for formation of own contextual fear experience, whereas ventral HPC (vHPC) is crucial for the formation and recall of social recognition memory. Thus, pre-requisite understanding of others' aversive situations may recruit dorsoventral HPC function to integrate memory information about prior similar fear experience in the observer and social familiarity with the demonstrator to elicit Exp OF. Here, we demonstrate that dorsoventral HPC to BLA pathways, without involving ACC, are crucial for Exp OF, while the ACC to BLA pathway is essential for Naive OF. Second, we found that fear memory engram cells in BLA are encoded during prior similar own fear experience, are reactivated during Exp OF, and are necessary for Exp OF. dHPC neurons generate fear memory engram cells in BLA during prior fear experience, while vHPC neurons respond to the familiar demonstrator's fearful situation, which reactivates the engram cells in BLA to elicit Exp OF. Our study provides new insight for memory engram-dependent perception-action coupling that underlies empathic behaviors like Exp OF.

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Poster

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Title: Hippocampal cells multiplex positive and negative engrams in healthy and maladaptive states

Authors: *M. SHPOKAYTE[1], O. P. MCKISSICK[4], B. RAHSEPAR[2], F. R. FERNANDEZ[2], E. RUESCH[1], S. L. GRELLA[3], R. SUTHARD[1], S. LIU[5], S. RAMIREZ[1];
[2]Biomed. Engin., [3]Ctr. for Memory and Brain, [1]Boston Univ., Boston, MA; [4]Brown Univ., Providence, RI; [5]Whitehead Inst. For Biomed. Res. | MIT, Cambridge, MA

Abstract: The hippocampus is involved in processing a variety of mnemonic computations including the spatiotemporal components, as well as the corresponding emotional dimensions, of contextual memory. Recent studies have demonstrated vast structural and functional heterogeneity along the dorsal-ventral axis of the hippocampus, and while much is known about how the dorsal hippocampus processes spatial-temporal content, much less is known about whether or not the ventral hippocampus (vHPC) partitions positive and negative experiences into distinct sets of cells. Here, we combine transgenic and all-virus based activity-dependent tagging strategies to visualize multiple valence-specific engrams in the vHPC and demonstrate two partially segregated, albeit physiologically similar, populations that respond to positive and negative experiences. We then optogenetically manipulated the terminals of these engrams to drive preference or aversion and asses their capacity to switch. Next, using an RNA Sequencing approach, we find that vHPC positive and negative engram cells display distinct transcriptional programs in which -neuroprotective and neurodegenerative -related gene sets are enriched, respectively. Ongoing studies are investigating the role of negative engrams in the induction and cascade of Alzheimer's disease. Together, our findings provide a novel means by which to visualize multiple engrams within the same brain, point to their unique genetic signatures as putative avenues for future therapeutic interventions, and investigate their role in neurodegenerative disease.

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Poster

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Topic: H.08. Learning and Memory

Title: Engram transition from recent to remote memory recall is accompanied with connectivity-related changes

Authors: *R. REFAELI[1], T. KREIZEL[2], I. GOSHEN[2];
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Abstract: When memory is acquired, a group of neurons with high activity levels during learning are allocated to support it. Some of these neurons will later participate in the recall of this memory. Their activity during the *acquisition* of memory is necessary for recall of recent (days post-acquisition) and even remote (a month and more post-acquisition) memories. Recent memory involves relatively fast processes that take place during the first hours to days following learning. Remote memories, however, are consolidated over much longer time scales, in yet not

well-defined processes termed together 'systems consolidation'. This process involves changes in multiple brain regions that enable the persistence of the memory. In this research we targeted the transition of memory from recent to remote recall, in order to find missing links between these memory stages. For that purpose, we tagged a first cFos+ cells using genetic manipulation, and a second group of cFos+ cells using IHC, within the same mouse. Thus allowing comparison of the two populations in each animal. First, we show that neuronal activation throughout different stages in memory consolidation is consistent in the hippocampus, but not in other memory-related brain areas, compared to the acquisition. Secondly, during the transition from recent to remote recall, several brain areas maintain fixed ensembles, not necessarily similar to the acquisition. Furthermore, hyperactivation of hippocampal astrocytes during acquisition, known to enhance recent memory, increases the percentage of CA1 reactivated cells between recent and remote, strengthening the hypothesis that the strength of recall is bound to the engram reactivation. In order to define whether the ensembles supporting remote memory are selected based on their connectivity and if they differ from the ensembles supporting recent memory, CA1 cells were marked based on their projection target. We found that the transition from recent to remote does include recruitment of projection-based sub-population; CA1 to ACC projecting cells are more likely to be activated during remote than during recent recall or acquisition. Additionally, hyperactivation of CA1 astrocytes during acquisition of memory also increases the activation of the CA1 to ACC-pc during recent but not remote. Lastly, analyzing projections of active cells from the CA1 during different stages of memory, using full brain reconstruction, revealed that as time progresses, the CA1 to ACC projections ratio increases, compared to other areas monosynaptic connected to the CA1, such as the Supra Mammillary Bodies or Nucleus Accumbens.

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Poster

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Title: Ensemble activity patterns underlying spatial navigation and reversal learning in aged and young adult mice

Authors: *W. MAU[1], L. CHEN[2], Z. DONG[2], T. SHUMAN[3], D. J. CAI[4];
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Abstract: Normal aging is often accompanied by cognitive decline. The hippocampus is particularly afflicted during aging, resulting in impaired memory in rodents, non-human primates, and humans. Age-related deficits have been observed in spatial reversal learning, yet we do not know how hippocampal activity patterns might contribute to this deficit. To answer this question, we used calcium imaging with head-mounted Miniscopes to study the stability and transience of hippocampal activity patterns in aged and young adult mice performing a spatial reversal task. We trained mice to navigate to goal locations on a circular track for rewards. Then after multiple days of this training, mice are forced to ignore the old goal locations and navigate to new ones for reward (spatial reversal learning). Young adult mice (~7 months old) consistently learn the initial goals, then during spatial reversal learning, they also learn the new goal locations within 30 minutes. Aged mice (~17 months old) learn the initial goals just as quickly as young mice, but perform worse during spatial reversal learning, suggesting that they are more inflexible in learning new goal locations. To examine the neural correlates of this age-related reversal learning deficit, we turned to the calcium imaging data. Using independent component analysis (ICA), we identified neuronal ensembles comprised of co-activated neurons (Lopes-dos-Santos et al., 2013, *J. Neurosci Methods*). In young mice, these ensembles weaken during spatial reversal learning, suggesting that CA1 ensembles undergo reorganization in order to learn new goal locations. This reorganization is less prevalent in aged mice, which could explain their impairments in spatial reversal learning. We also observed specific deficits in learning-induced excitability in CA1 of aged mice. These excitability deficits could underlie differences in CA1 ensemble patterns that could ultimately contribute to spatial reversal learning deficits seen in aging.

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Poster

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Title: Network dynamics of dorsal hippocampal CA1 and CA3 during fear memory encoding and recall

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Abstract: Episodic memories—memories of experiences placed in time and space— form through associations made in the hippocampus between distinct stimuli within specific contexts. Recent studies have shown that in a contextual fear conditioning (CFC) paradigm, which causes the formation of an episodic-like memory, optogenetic activation of neurons in hippocampal CA1, CA3 or Dentate Gyrus that are "tagged" during induction of CFC can sufficiently trigger retrieval of the fear-memory. The existence of "engram-cells" in different hippocampal subnetworks suggests that the intact hippocampus is critical for episodic memory formation and recall. However, how the anatomically connected but functional distinct subnetworks in the hippocampus, such as CA3 and CA1, coordinate the information to support memory formation and recall remains unknown. This question can only be solved by studying the cross-network interactions during memory formation and recall. Here, we implement a novel behavioral paradigm for CFC where functional two-photon imaging of the hippocampal CA1 and CA3 will be performed in head-fixed mice navigating virtual contexts undergoing fear conditioning and during fear memory recall across multiple days. We observed CFC behaviors in head-fixed mice similar to freely moving animals. More importantly, we have now connected behavior with the dynamics and interactions of hippocampal subnetworks in CA1 and CA3 to help better understand the role of cross-network dynamics in episodic memory formation and recall.

Disclosures: C. Dong: None. **S. Krishnan:** None. **M. Sheffield:** None.

Poster

P836. Hippocampal Interactions IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P836.01

Topic: H.08. Learning and Memory

Support: NIH Grant R01MH115304

Title: Do engram cells discharge like memory cells during optogenetic activation?

Authors: *C. GARCIA JOU[1], E. PARK[2], D. DVORAK[3], E. R. LEVY[3], A. A. FENTON[3];
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Abstract: Engram cell (EC) experiments genetically tag neurons that are active during learning so that they can be optogenetically manipulated to test whether their reactivation is necessary and sufficient for the expression of a memory. Hippocampal ECs have been identified in mice using threat-conditioning paradigms, but the results challenge established notions of how the hippocampus encodes and processes information. Consider for example, that hippocampal ECs are place cells that discharge at specific locations of a conditioning environment to represent that environment. During conditioning, a pair of cells with coincident firing fields in the north of the environment may discharge together, but never when cells with firing fields in the south discharge. However, these three place/engram cells would be activated synchronously during optogenetic activation, making it difficult to imagine how the optogenetic activation could result in replaying or reactivating the representations that were stored during conditioning. We have begun to investigate this contradiction. We first established that optogenetic tagging and activation of CA1 was sufficient to express a conditioned-place avoidance known to require place cells and their dynamic temporal discharge. 5-Hz optogenetic activation of memory-tagged cells was sufficient to express the conditioned avoidance in both neutral familiar and entirely novel environments without either the CS or US. We recorded ensembles of CA1 cells in a subset of mice under anesthesia after the optogenetic test and behavioral demonstration of sufficiency. Firing rates increased ~77% during 7 mW, 473 nm, theta-frequency optogenetic activation, and increased their likelihood of discharging to 1% in response to each 15 ms light pulse. Remarkably, CA1 cell pairs maintained their pre-established co-firing relationships such that pairs of cells that cofired before stimulation continued to cofire during stimulation, and cell pairs that did not cofire continued not to cofire (7mW: $r = 0.36$, compared to control: $r = 0.51$). Ensemble CA1 recordings in awake, head-fixed mice also confirmed that 5 and 10 Hz optogenetic stimulation produces an evoked discharge pattern in single cells that can be distinguished from control discharge. These initial observations indicate the importance of monitoring patterns of neural discharge during EC experiments in order to assess whether or not memory-related temporal discharge patterns are maintained during optogenetic stimulation in EC experiments that elicit conditioned responses.

Disclosures: C. Garcia Jou: None. **E. Park:** None. **D. Dvorak:** None. **E.R. Levy:** None. **A.A. Fenton:** None.

Poster

P836. Hippocampal Interactions IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P836.02

Topic: H.08. Learning and Memory

Support: ANR Hippencode
EU Horizon 2020 MSCA ITN SyDAD

EUR Bordeaux Neurocampus

Title: Exploring the early properties of CA3 engram neurons with a novel viral tool

Authors: D. CUPOLLILO[1], C. MARNEFFE[1], C. REYNAUD[1], S. DEFORGES[1], N. GROSJEAN[1], ***C. MULLE**[2];
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Abstract: Forming new memories after a one-time experience requires initial encoding then consolidation over time. The hippocampal CA3 region is a key structure involved in multimodal information integration and initial memory storage. In this work, we describe the development and validation of a novel tool for fast-labeling of engram neurons (FLEN). FLEN is based on c-Fos activity-dependent transient expression of a destabilized fluorescent marker ZsGreen1 rapidly after one-trial learning (few hours). With FLEN, we explored the electrophysiological properties of c-Fos activated CA3 neurons following one-trial learning. In parallel, we have employed the Robust Activity Marker (RAM) system to label CA3 engram neurons 24 hours following a novel experience. Comparing FLEN positive and RAM positive neurons allows to show that both intrinsic excitability and synaptic properties of CA3 pyramidal neurons undergo progressive plastic changes over the first day following a one-trial memory task.

Disclosures: D. Cupollilo: None. **C. Marneffe:** None. **C. Reynaud:** None. **S. Deforges:** None. **N. Grosjean:** None. **C. Mulle:** None.

Poster

P836. Hippocampal Interactions IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P836.03

Topic: H.09. Spatial Navigation

Support: NIH grant GM118801

Title: Etomidate blocks the formation of stable hippocampal engrams in WT but not interneuronα5-GABA$_A$R-KO mice

Authors: *M. ZHU[1], M. PERKINS[2], R. LENNERTZ[2], R. A. PEARCE[2];
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Abstract: Studies of place cells have shown that the hippocampus encodes a cognitive map, and that stable place-specific firing patterns (engrams) represent memory traces. Here, we examined what an amnestic dose of etomidate does to engram formation and stability in WT mice and in mice lacking α5-GABA$_A$Rs in interneurons (α5-i-KO), which serve as its target (A Abdulzahir,

SfN 2021). We measured cellular activity in freely exploring mice using GCaMP6f and a mini-endoscope (Inscopix), and we captured the mouse's position using Noldus EthoVision. Mice underwent two 10-min recording sessions per day, separated by four hours (AM and PM), with unique sets of sensory cues used each day to create new contexts. Image processing and statistical analysis were performed using IDPS software (Inscopix) and custom-written MATLAB routines.
Under control conditions (saline IP), place-specific firing patterns, as determined by mutual information between spatial position and cellular activity, were observed in 10-20% of imaged cells (up to 1200 per field) in both WT and α5-i-KO mice. 50-60% of place cells observed during AM sessions exhibited similar firing patterns in PM sessions, demonstrating that mice had formed and recalled stable engrams. This similarity was quantified by Population Vector (PV) correlation between position-specific firing rate maps (WT-sal 0.52±0.12, α5-i-KO-sal 0.43±0.14, $p<0.0001$ vs. shuffle for both groups). In WT mice, etomidate (2-8mg/kg IP 30 minutes prior to the AM session) produced dose-dependent reductions in both place cell formation and stability, with significant loss of PV correlation at 7 mg/kg (WT-etom 0.23±0.16, $p<0.001$ vs. WT-sal). By contrast, the PV correlation was unchanged in α5-i-KO mice injected with 7 mg/kg etomidate (α5-i-KO-etom 0.42±0.12, $p=0.83$ vs. α5-i-KO-sal), indicating that mice had successfully formed stable engrams. To demonstrate that mice formed context-specific engrams, in a separate group of experiments with WT mice we altered the context between AM and PM sessions. This change resulted in significantly reduced PV correlations (WT-alt-context 0.35±0.11, $p<0.001$ vs. WT-sal). Additionally, 7mg/kg etomidate did not cause any changes in overall cellular activity (events per second) for either genotype (WT-sal 0.08±0.02, etom 0.07±0.02, $p=0.14$; α5-i-KO-sal 0.09±0.03, etom 0.09±0.03, $p=0.37$), or alter the probability that the any given cell would be active during both AM and PM sessions (WT-sal 0.57±0.11, etom 0.53±0.18, $p=0.39$; α5-i-KO-sal 0.66±0.08, etom 0.70±0.07, $p=0.56$). These findings provide additional evidence that etomidate targets interneuronal α5-$GABA_A$Rs to suppress contextual memory formation.

Disclosures: M. Zhu: None. **M. Perkins:** None. **R. Lennertz:** None. **R.A. Pearce:** None.

Poster

P836. Hippocampal Interactions IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P836.04

Topic: H.08. Learning and Memory

Support: GM118801
GM128183

Title: α5-$GABA_A$ receptors located on inhibitory interneurons mediate contextual memory suppression by etomidate in mice

Authors: *A. ABDULZAHIR[1], R. NAGARAJAN[2], U. RUDOLPH[3], R. A. PEARCE[4]; [1]Univ. of Wisconsin Madison, Madison, WI; [2]Dept. of Comparative Biosci., Univ. of Illinois at Urbana-Champaign, Urbana, IL; [3]Dept. of Comparative Biosci., Univ. of Illinois At Urbana-Champaign, Urbana, IL; [4]Anesthesiol., Univ. of Wisconsin, Madison, WI

Abstract: Suppression of memory is one of the primary goals of general anesthesia. Previous investigations have shown that etomidate blocks both long term potentiation of excitatory synapses (LTP – a cellular model of memory) and contextual memory (a surrogate for episodic memory) by targeting $GABA_A$Rs that incorporate α5 subunits (α5-$GABA_A$Rs).[1,2] We recently reported that selectively eliminating α5-$GABA_A$Rs from hippocampal pyramidal neurons did not interfere with etomidate's ability to suppress LTP *in vitro*,[3] suggesting that α5-$GABA_A$Rs on non-pyramidal cells might be instrumental in etomidate-induced amnesia. Here, we tested this hypothesis by selectively eliminating α5-$GABA_A$Rs from inhibitory interneurons and measuring etomidate suppression of contextual memory in the Context Pre-exposure Facilitation Effect (CPFE) paradigm.[4] Experiments were performed using α5-$GABA_A$R-interneuron knockout (α5-i-KO) mice, in which the α5-$GABA_A$Rs were selectively eliminated from interneurons by crossing mice carrying a 'floxed' Gabra5 gene with mice expressing Cre-recombinase under the control of the Gad2 promoter. Restriction of Cre expression to interneurons was confirmed by crossing Gad2-Cre mice with a TdTomato reporter mouse. Western blot analysis showed that α5 subunit expression was not measurably reduced in α5-i-KO mice compared to WT, consistent with the predominant expression of α5-$GABA_A$Rs in pyramidal neurons. Both WT and α5-i-KO mice injected with saline before pre-exposure (sal) displayed significantly higher freezing scores than no-pre-exposure (npe) controls (WT-npe 25 ± 5 %, α5-i-KO-npe 17 ± 5 %, n=5 each; WT-sal 41 ± 3 %, α5-i-KO-sal 41 ± 4 %, n=12 each; $p<0.01$ sal vs. npe), at levels not different from each other ($p=0.5$), demonstrating that the pre-exposure resulted in contextual memory formation, and that eliminating α5-$GABA_A$Rs from interneurons did not itself interfere with memory. Administration of etomidate significantly reduced freezing scores in WT mice (α5-i-KO-sal 17±2%, n=12, $p=0.01$ vs. WT-sal) but not in α5-i-KO mice (α5-i-KO-sal 37±4 %, n=12, $p=0.47$ vs. WT-sal). These findings support the hypothesis that α5-$GABA_A$Rs on interneurons mediate contextual memory suppression by etomidate.
References: 1. Cheng, V. Y., *J. Neurosci.* **26**, 3713-3720 (2006); 2. Martin, L. J., *Anesthesiology* **111**, 1025-1035 (2009); 3. Rodgers, F. C., *J. Neurosci.* **35**, 9707-9716, (2015);4. Matus-Amat, P., *J Neurosci.* **24**, 2431-2439 (2004).

Disclosures: A. Abdulzahir: None. **R. Nagarajan:** None. **U. Rudolph:** None. **R.A. Pearce:** None.

Poster

P836. Hippocampal Interactions IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P836.05

Topic: H.08. Learning and Memory

Support: NIH Grant R01MH115304

Title: Is the rat prefrontal cortex crucial for cognitive control during spatial cognition?

Authors: *E. PARK[1], G. GRUBS[1], D. TABORGA[1], K. NICHOLAS[1], A. S. AHMED[1], K. O'REILLY[2], A. A. FENTON[1];
[1]Ctr. for Neural Sci., New York Univ., New York, NY; [2]Child and Adolescent Psychiatry, New York State Psychiatric Inst., New York, NY

Abstract: Cognitive control tasks require using relevant information while ignoring competing irrelevant information. Prior work inactivating hippocampus during an active place avoidance task suggested hippocampus is crucial for cognitive control of spatial information. The task requires rodents on a rotating arena to avoid a stationary shock zone by using stationary cues while ignoring information from the rotating arena. During this task, the discharge of hippocampal neurons alternates judiciously between representing stationary and rotating locations according to the animal's proximity to shock and ability to avoid the shock zone, demonstrating cognitive control in hippocampal discharge. The central role of the medial prefrontal cortex (mPFC) in cognitive control is well established; mPFC damage causes deficits in cognitive control tasks. Here we evaluated whether rat mPFC lesion impairs active place avoidance to test a "central-computation" hypothesis that mPFC is essential for the computations required for cognitive control. An alternative "local-computation" hypothesis asserts that cognitive control can be accomplished via local computations by neural circuits, independent of mPFC. We compared active place avoidance after ibotenic acid lesion of mPFC (n = 10) and sham (n = 8) rats. After testing behavior, in a subset of the rats (lesion n = 8, sham n = 8), we evaluated the lesion and its consequences by measuring cytochrome oxidase (CO) activity in 14 functionally-related brain regions. CO activity correlations between the dorsal and ventral hippocampi, amygdala, retrosplenial cortex and nucleus reunions were weaker after mPFC lesion indicative of reduced functional coupling between related brain areas. Behaviorally, post-surgery, both groups learned to enter the shock zone less (Group: $F_{1,16} = 0.002$, $p = 0.9$; Day: $F_{1,16} = 26.3$, $p = 10^{-4}$; Trials: $F_{3.22,\ 51.50} = 36.8$, $p = 10^{-13}$), and increased the latency to first enter the shock zone across trials (Group: $F_{1,16} = 0.1$, $p = 0.8$; Day: $F_{1,16} = 69.6$, $p = 10^{-7}$; Trials: $F_{5.33,\ 85.33} = 12.2$, $p = 10^{-9}$). Memory retention at 24-h did not distinguish the groups (entrances: $t_{16} = 1.0$, $p = 0.4$; time to first enter: $t_{16} = 1.0$, $p = 0.3$). Both groups learned a new conflicting avoidance when the shock was relocated 180° to further assess cognitive flexibility (Group: $F_{1,16} = 0.21$, $p = 0.7$; Day: $F_{1,16} = 8.9$, $p = 0.01$; $F_{1.89,\ 30.12} = 18.55$). These data demonstrate that computations required for cognitive control can occur locally in brain networks independently of the mPFC, in support of the local-computation rather than the central-computation hypothesis.

Disclosures: E. Park: None. **G. Grubs:** None. **D. Taborga:** None. **K. Nicholas:** None. **A.S. Ahmed:** None. **K. O'Reilly:** None. **A.A. Fenton:** None.

Poster

P836. Hippocampal Interactions IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P836.06

Topic: H.08. Learning and Memory

Support: R01MH115304

Title: Photoactivated proteolysis targeting chimeras (PHOTACs) for light-activated degradation of specific synaptic proteins in hippocampal tissue

Authors: *A. FENTON[1], C. JOU[4], T. KO[2], A. B. GRAU-PERALES[3], M. REYNDERS[5], D. TRAUNER[2];
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Abstract: The degradation of proteins with Proteolysis Targeting Chimeras (PROTACs) is currently revolutionizing pharmacology. Our group has introduced photoswitchable versions, so called PHOTACs, that allow for optical degradation of specific proteins. Our initial targets are proteins that are hypothesized to be crucial for maintaining memory. The synaptic plasticity and memory hypothesis, a dogma of neuroscience, asserts that memory is maintained by sustaining neurotransmission across specific synaptic pathways, but definitive identification of the crucial molecular mechanisms requires tools to selectively degrade candidate proteins at specific sites at specific times of the learning/memory process. To test the feasibility of the PHOTACs approach, we developed a CaMK2-PHOTAC for neuronal CaMKIIα because this protein is crucial for maintained synaptic structure and function. Light activation (385 nm) of the CaMK2-PHOTAC ($n = 7$) decreased CaMKIIα immunolabeling 3-fold compared to controls ($p<0.001$), specifically, the non-activated CaMK2-PHOTAC (dark condition, $n = 7$, $p<0.001$), tissue incubated with ACSF with light ($n = 6$, $p<0.03$) or without illumination ($n = 3$, $p<0.01$). No differences were observed in parvalbumin immunolabeling ($p>0.9$), nor in the number of DAPI-stained nuclei ($p<0.9$), demonstrating specificity of the CaMK2-PHOTAC. Because CaMKIIα maintains synaptic structure, we next tested whether CaMK2-PHOTAC depresses synaptic transmission in the medial perforant pathway (MPP) that terminates in the suprapyramidal dentate gyrus of acute mouse hippocampus slices. While 100 ms 385 nm light pulses every 10 s did not change responses in the control slices, the illumination depressed baseline synaptic responses in CaMK2-PHOTAC-incubated slices ($n = 7$), as predicted for loss of CaMKIIα in maintaining baseline synaptic function. The effect required light activation because the synaptic responses persisted in the CaMK2-PHOTAC slices that were not illuminated ($n = 5$). These biochemical

and electrophysiological findings demonstrate that CaMK2-PHOTAC is effective in brain tissue, is cell permeable, and targets CaMKIIα with no evidence of toxicity. Finally, we will discuss the development of PHOTACs that target other proteins hypothesized to be crucial for maintaining long-term potentiation of synaptic function (LTP) and the persistence of long-term memory.

Disclosures: A. Fenton: None. **C. Jou:** None. **T. Ko:** None. **A.B. Grau-Perales:** None. **M. Reynders:** None. **D. Trauner:** None.

Poster

P836. Hippocampal Interactions IV

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P836.07

Topic: H.08. Learning and Memory

Support: Memory RO1MH115304

Title: Visualizing remote spatial memory in the hippocampus

Authors: *A. GRAU-PERALES[1], R. M. HARRIS[3], H.-Y. KAO[4], J. M. ALARCON[5], H. A. HOFMANN[6], A. A. FENTON[2];
[2]Ctr. for Neural Sci., [1]New York Univ., New York, NY; [3]Integrative Biol., The Univ. of Texas At Austin, Austin, TX; [4]Dept of Neurol., Univ. of Michigan, Ann Arbor, MI; [5]Dept. of Pathology, SUNY Downstate Med. Ctr., Brooklyn, NY; [6]Univ. of Texas at Austin, Austin, TX

Abstract: The input-specific populations of hippocampal synapses can undergo long-term synaptic changes, which are assumed to store memory. However, which specific constellations of hippocampal synaptic populations undergo these experience-dependent modifications is an open question. The atypical protein kinase C isoform, PKMζ is necessary and sufficient for the maintenance of LTP at hippocampal synapses and the persistence of long-term memory in a hippocampus-dependent active place avoidance task. Thus, PKMζ expression can be used to test hypotheses for which input-specific synaptic populations change long-term memory storage and cognitive information processing. To investigate the subcellular distribution of PKMζ in the hippocampus we used ArcCreER^{T2} x ChR2-EYFP mice to label memory-associated cells. Trained (n=4) and untrained (n=4) mice received 30-min trials on a rotating arena, to learn to avoid a stationary shock zone. Before the last training trial, all mice received 4-OH tamoxifen to induce Arc-driven expression of ChR2-EYFP. Trained mice expressed conditioned avoidance 1 month later. Immunohistochemical analysis of EYFP identified an increased number of memory-activated pyramidal cells in CA1 and CA3, and granule cells in the suprapyramidal blade of dentate gyrus (DG) compared to untrained mice. Memory-induced increases in PKMζ also followed this pattern, and identified input-specific patterns of memory-related synaptic populations. Memory training increased PKMζ-EYFP co-expression at CA3-CA1 Schaffer

collateral synapses but not where the TA entorhinal inputs terminate. To discover what other candidate molecules may be implicated in this memory, we used transcriptome profiling of hippocampus subregions after active place avoidance training (n's=17). A day after expressing the avoidance memory, 214 genes were differentially expressed in DG, 16 in CA1, and none in CA3 compared to controls. Most immediate early genes were upregulated. Gene Ontology analysis of the differentially expressed genes showed that memory was associated with activation of transcription and synaptic differentiation in DG but not CA3 or CA1. Hypothesis-driven candidate molecule analyses did not yield significant changes in the transcripts for PKMζ and other LTP-associated molecules, nor did they covary with memory expression, ruling out strong transcriptional regulation but not translational regulation. These observations confirm the importance of PKMζ activity in long-term memory maintenance and identify an input-specific distribution of memory-related hippocampal synaptic populations for long-term active place avoidance memory.

Disclosures: A. Grau-Perales: None. **R.M. Harris:** None. **H. Kao:** None. **J.M. Alarcon:** None. **H.A. Hofmann:** None. **A.A. Fenton:** None.

Poster

P836. Hippocampal Interactions IV

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Program #/Poster #: P836.08

Topic: H.08. Learning and Memory

Support: NIH grants R01MH115304

Title: A code for space in hippocampal coactivity dynamics independent of place fields

Authors: *E. R. J. LEVY[1], E. PARK[2], A. A. FENTON[1];
[1]Ctr. for Neural Sci., [2]New York Univ., New York, NY

Abstract: The hippocampus is crucial for spatial navigation as well as the acquisition and persistence of memories, especially episodic memories that contain environmental information. The standard cognitive map hypothesis for decoding information from neuronal discharge states that information is encoded in the activity of single neurons called place cells. Place cells are hippocampal principal cells with activity tuned to locations in a particular environment. It is not straightforward how such a code can represent the current environment without extensive spatial exploration and at the ms-to-s timescales of neural computations. Because hippocampal principal cells are governed by intrinsic and behaviorally modulated temporal dynamics, such as oscillations in the local field potential, we considered an alternative hypothesis. This coactivity hypothesis does not rely on single cell place tuning, rather it asserts that spatial information is encoded in the moment-to-moment cofiring relationships amongst an ensemble of neurons.

To compare the importance of these two coding schemes for encoding environments, we recorded CA1 activity across a multi-week experience of two physically distinct environments. Using a bright-field miniature microscope placed above a surgically implanted GRIN lens, we imaged mouse CA1 principal neurons expressing virally-induced GCaMP6f.
In addition to replicating the standard findings of CA1 remapping experiments, we find that at the 1-s time resolution, cell pair coactivity is stable within environments and distinct between environments. We show that this environment-discriminating information in cell pair coactivity is not a result of the cells' place tuning. Decoding environments from cell-pair coactivity is effective and even improves after removing cell-specific place tuning. Similarly, decoding environments directly from ensemble activity in CA1 is accurate and does not rely on place cells, high-rate cells, or low-rate cells. Instead, we find that discrimination relies on cells that have a propensity to anti-cofire with other cells at the 1s timescale.
Evaluating the 1-s low dimensional structure of CA1 activity with a non-linear algorithm, IsoMap, we find that it is organized on a 2D manifold that naturally discriminates the two environments. This non-linear projection discrimination relies on cell-pair coactivity, in particular, anti-coactive cell pairs.
We conclude that ensemble cofiring relationships are a neural code for environmental space, independent of place fields, and is advantageous for moment-to-moment identification of the current environment.

Disclosures: E.R.J. Levy: None. **E. Park:** None. **A.A. Fenton:** None.

Poster

P836. Hippocampal Interactions IV

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Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P836.09

Topic: H.08. Learning and Memory

Support: NS108765
NS108296

Title: Temporal Order of Increased Signal Coordination Within and Between PFC-Hippocampus After Learning of a Dynamic Spatial Task

Authors: *C. DICKSON[1], J. M. BARRY[2];
[1]Univ. of Vermont, BURLINGTON, VT; [2]Neurolog. Sci., Univ. of Vermont, Burlington, VT

Abstract: The mPFC is responsible for goal planning and decision making while the hippocampus is essential for spatial learning and memory. In a spatial active avoidance task requiring planning and spatial memory, we therefore predicted dynamically coherent neural activity at a theta timescale within (mPFC-mPFC, dHC-dHC, vHC-vHC) and between (mPFC-

dHC, mPFC-vHC) these two regions according to moment by moment cognitive and behavioral task demands. Moreover, we aimed to measure the development of this dynamic signal coordination as a function of task learning. Adult male rats were bilaterally implanted with EEG electrodes and local field potentials recorded in the mPFC, dorsal hippocampus (dHC), and ventral hippocampus (vHC) while subjects learned the active avoidance task. Whole training session mPFC-HC, mPFC-mPFC, and HC-HC theta coherence increased as a function of spatial learning. For mPFC-HC, the greatest coherence change within the theta bandwidth coincided with the unimodal coherence peak at ~7.5Hz. While rats were beginning to learn the task (Early session), the mPFC-dHC was more coherent than the mPFC-vHC, despite significantly lower synaptic connectivity. Prior to avoidance, theta coherence demonstrated a regional temporal sequence in which transitory peaks were observed between the mPFC-mPFC and mPFC-HC and dHC-dHC. Moreover, coherence within the theta bandwidth fluctuated between epochs for mPFC-HC and mPFC-mPFC, showing distinct frequency distributions in coherence. Theta coherence within and between the mPFC and hippocampal poles change as a function of learning and cognitive demand during active avoidance on a rotating arena. These temporally ordered coherence signatures reveal unique field potential coherence properties operating on a second-by-second timescale that coincide with transient cognitive states. Importantly, these results proved the importance of analytical timescale, demonstrating how information is lost when coherence is averaged across time or across the frequency band. It remains unclear what the content of these discrete frequency distributions within the theta bandwidth conveys and if they are functionally important. However, the results support the findings of other experiments indicating a causal role for 8-9 Hz mPFC-HC coherence for spatial avoidance. Future work will use optogenetic interventions to examine whether theta frequency specificity in signal coordination across the mPFC and hippocampal circuit provides qualitatively different types of information.

Disclosures: C. Dickson: None. **J.M. Barry:** None.

Poster

P911. Genetic Techniques I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P911.01

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Title: Deep-learning augmented single-neuron brain mapping

Authors: D. LU[1], *X. TIAN[2], E. KNOTT[3], M. REN[3], X. LU[2];
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[2]Louisiana State Univ. Hlth., Shreveport, LA; [3]LSU Hlth. Shreveport, Shreveport, LA

Abstract: Due to the complex anatomy, cell-type diversity, lack of simple, noninvasive delivery methods, and the challenge to evaluate the outcome, genetic engineering in the central nervous system (CNS) has been largely limited to costly, time-consuming, and laborious germline transgenesis. In order to overcome these hurdles, a neurotropic genome editing approach was developed for noninvasive generation of somatically engineered mice with a wide-scale and dose-dependable genome editing footprint in adult mouse brains. Our strategy facilitates sparse, stochastic, and dosage-dependent genetic labeling to characterize the morphology and neuropathology of single-neurons. To overcome the trade-offs in volume imaging for single-neuron analysis, an unsupervised deep learning network was used to augment the quality of volume imaging. we here describe the use of deep learning-based generative adversarial networks (GANs) to aid in the identification and visualization of subcellular neuronal structures within volume imaging data. Our empirical results demonstrate that CycleGAN can learn an unsupervised image model from high-quality images to identify relevant signals in low-quality images. In particular, we also show that CycleGAN can robustly adjust image noise to identify Purkinje neuron dendrites to enable rapid multi-neuron 3D reconstruction and segmentation using IMARIS. We developed a workflow and proposed the concept of "broad spectrum single-cell pathology analysis" to gain unprecedented temporal and spatial profiling of single-cell neurodegeneration and neuroinflammation after stroke. This approach can be used as a powerful genetic tool to generate loss- or gain- of function genetic modifications in CNS to probe structure, function/dysfunction and causal pathogenesis in Neurodegenerative/neurodevelopmental disorders and to develop disease-modifying therapy.

Disclosures: D. Lu: None. **X. Tian:** None. **E. Knott:** None. **M. Ren:** None. **X. Lu:** None.

Poster

P911. Genetic Techniques I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P911.02

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH R01 EY030169
NIH R01 EY018204
Whitehall Foundation Grant
Research to Prevent Blindness

Title: Intersectional mapping of multi-transmitter neurons in the brain

Authors: J. XU[1], A. JO[2], R. DEVRIES[3], S. DENIZ[2], S. CHERIAN[2], I. SUNMOLA[2], X. SONG[4], J. MARSHALL[2], T. L. DAIGLE[5], A. CONTRACTOR[2], T. N. LERNER[2], H. ZENG[5], ***Y. ZHU**[2];
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Med., Chicago, IL; [3]The Univ. of Chicago, Chicago, IL; [4]Shanghai Jiao Tong Univ., Shanghai, China; [5]Allen Inst. for Brain Sci., Seattle, WA

Abstract: Many neurons can produce, store, and release multiple neurotransmitters. The existence of such multi-transmitter neurons complicates our understanding of the brain and neural circuits. Intersectional strategies based on dual-recombinase system are particularly powerful in mapping brain-wide distributions of these multi-transmitter neurons based on expressions of genes needed to establish neurotransmitter identity. In this study, we performed a brain-wide mapping of neurons that are potentially capable of co-releasing glutamate and GABA. By creating intersections between VGLUT (VGLUT1, VGLUT2, or VGLUT3) and VGAT with Cre/Flp double recombinase system, we identified 30 brain regions that contain co-expression neurons in the adult, including 19 VGLUT2/VGAT regions and 11 VGLUT3/VGAT regions. While some of these regions have been previously identified, more than half of them have not been reported before. We also discovered another 32 brain regions co-expressing VGLUT and VGAT during the development but not in the adults. Using viral tracing with Intersect AAVs, we identified 14 brain regions that innervate the lateral habenula (LHb) with glutamate/GABA co-transmission, which doubles what is known. Furthermore, with co-injection of two AAVs that label VGLUT-expressing and VGAT-expressing neurons with different colors, we identified another 25 LHb-projecting regions exclusive of VGLUT2/VGAT projecting neurons. These regions provide glutamatergic innervations alone (6 regions), GABAergic innervations alone (11 regions), or mixed innervations originated from sole glutamatergic and sole GABAergic neurons (8 regions). These results reveal an unanticipated abundance of glutamate/GABA co-transmission in the brain and add to our understanding of the afferent circuitry to the LHb.

Disclosures: J. Xu: None. **A. Jo:** None. **R. DeVries:** None. **S. Deniz:** None. **S. Cherian:** None. **I. Sunmola:** None. **X. Song:** None. **J. Marshall:** None. **T.L. Daigle:** None. **A. Contractor:** None. **T.N. Lerner:** None. **H. Zeng:** None. **Y. Zhu:** None.

Poster

P911. Genetic Techniques I

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Program #/Poster #: P911.03

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: Lithuanian Research Council grant Nr. P-MIP-19-138
The European Social Fund under the Global Grant measure, grant number VP1-3.1- ŠMM-07-K-02-059

Title: Short promoters for directing AAV protein expression in specific classes of cortical interneurons

Authors: *G. BARANAUSKAS[1], K. RYSEVAITE-KYGUOLIENE[2], I. SABECKIS[2], D. H. PAUZA[2], T. TKATCH[3];
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Abstract: BACKGROUND: Gene delivery to neurons by nonpathogenic recombinant adeno-associated virus (AAV) is becoming a method of choice in many fields of neuroscience and is showing increasing promise for treatment of human diseases. However, there is still a limited choice of promoters suitable for neuron-type specific expression of AAV delivered genes. Therefore, we designed two promoters suitable for expression of AAV delivered genes in two types of cortical interneurons, somatostatin (SOM) and parvalbumin (PV) expressing cells.AIM: To identify the specificity of expression of AAV delivered genes controlled by two promoters designed by us.
METHODS: Regulatory sequences for two promoters were selected based on homology of gene surrounding regions of several mammalian species including humans. The specificity of gene expression was established by determining co-localization of the fluorescence of viral YFP and the fluorescence of antibodies to neuronal markers. AAV particles were injected in visual cortex area V1 of adult rats (2 - 4 months old). 10 - 14 days post-injection the rat brains were perfused with 4 % PFA phosphate buffer solution and then sliced to 100 um thickness. The slices were treated with antibodies to GAD65/67, CamKIIa, GABA, SOM, PV, vasoactive intestinal peptide (VIP) and cholecystokinin (CCK). Confocal images of the first 20 um from the slice surface were then taken and used for co-localization analysis.
RESULTS: The first promoter was designed to be specific to PV interneurons. Indeed, the analysis of confocal images taken from 3 rats showed that approximately 2/3 of viral YFP labelled neurons were also labelled by PV antibodies (383/605, 63 %). Meanwhile, only 16 % of neurons expressing viral YFP were also labelled by antibodies to SOM (37/228). In addition, co-localization tests with CamKIIa antibodies showed little overlap: only <7 % of neurons expressing viral YFP were also labelled by CamKIIa antibodies. Currently we are performing experiments to determine the specificity of a putative SOM promoter.
CONCLUSIONS: Our results show that we were able to design a promoter that restricts AAV delivered gene expression to interneurons with a strong preference to PV class of interneurons.

Disclosures: G. Baranauskas: None. **K. Rysevaite-Kyguoliene:** None. **I. Sabeckis:** None. **D.H. Pauza:** None. **T. Tkatch:** A. Employment/Salary (full or part-time):; Present address: Department of Physiology, Northwestern University, Chicago, IL, USA.

Poster

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Program #/Poster #: P911.05

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

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BRAIN-R01MH117069
Director's Pioneer Award Program (DP1OD025535)
Chan Zuckerberg Initiative
the Beckman Institute at Caltech
Vallee Foundation
Moore Foundation

Title: Efficient and Specific Sensor and Actuator Delivery to the Rodent Peripheral Nervous System through Systemic AAVs

Authors: *X. CHEN[1], S. RAVINDRA KUMAR[1], D. YANG[2], C. D. ADAMS[3], T. WANG[1], D. WOLFE[1], J. GRIFFITHS[1], R. NOSEDA[4], J. R. KEAST[3], I. M.-C. CHIU[2], V. GRADINARU[1]; [1]Biol. and Biol. Engin., Caltech, Pasadena, CA; [2]Microbiology and Immunobiology, Harvard Med. Sch., Boston, MA; [3]Univ. of Melbourne, Parkville, Australia; [4]Anesthesia, Beth Israel Med. Ctr/ Harvard Med. Sch., Boston, MA

Abstract: The peripheral nervous system (PNS) is critical in regulating end-organ function and feedback signal transmission to the central nervous system (CNS). To achieve precise genetic manipulation within the PNS, more efficient and targeted gene delivery vehicles are needed. Adeno-associated viruses (AAVs) have become the vector of choice for gene delivery in vivo but non-invasive administration of natural serotypes targets the PNS with mixed efficiency and specificity. Attempts to circumvent these limitations by direct administration of AAV are often either surgically challenging or have incomplete coverage. To further explore the potential of targeting PNS with engineered-capsid, we utilized the advanced selection platform Multiplexed-CREATE (Kumar et al, Nature Methods, 2020) to perform selection in vivo on PNS areas of interest. Preliminary Next Generation Sequencing (NGS) analysis of the recovered sequences revealed two novel AAV variants: AAV-PHP.PNS1 and AAV-PHP.PNS2. In vivo validation in C57BL/6J adult mice by i.v. delivery of the variants packaging EGFP under a strong ubiquitous CAG promoter showed enhanced EGFP expression in neurons of the nodose ganglia and Dorsal Root Ganglia (DRG). Consistent with NGS enrichment data, AAV-PHP.PNS1 showed greater specificity to the nodose ganglia and DRG by exhibiting lower overall transduction in peripheral organs including liver (an otherwise strong AAV9 target), while AAV-PNS2 showed higher overall efficiency in targeting ENS. Intraperitoneal delivery of the variants in P1 pups also exhibit strong transduction in DRG, labeling ~40% of the Tuj1+ cells. Among the neuron subtypes, ~53% of CGRP+ neurons were transduced. Intravenous delivery into adult Sprague Dawley rats also resulted in robust transduction in main sensory ganglia (DRG, Trigeminal Ganglia), sympathetic and parasympathetic ganglia, and in the ENS. To explore the vector's potential beyond neuron labeling and tracing, we demonstrate that we could record neuronal activity in nodose ganglia following I.V. delivery of GCamp with PNS1 in adult C57BL/6J mice. We next took a step forward to see whether we could achieve functional manipulation with the new PNS capsids, demonstrating that I.P. delivery of a Cre-dependent excitatory DREADD in Trpv1-Cre mice with PNS1 at P1 in adult mice can induce pain phenotypes. With improved efficiency and specificity towards the PNS in rats and mice for effective sensor and actuator

performance, the AAV-PHP.PNS1 and AAV-PHP.PNS2 capsids can help enhance anatomical and functional studies.

Disclosures: X. Chen: None. **S. Ravindra Kumar:** None. **D. Yang:** None. **C.D. Adams:** None. **T. Wang:** None. **D. Wolfe:** None. **J. Griffiths:** None. **R. Noseda:** None. **J.R. Keast:** None. **I.M. Chiu:** None. **V. Gradinaru:** None.

Poster

P911. Genetic Techniques I

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Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P911.06

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH NIBIB Trailblazer award (R21 EB031249)

Title: Evaluating the use of systemic AAV-PHP.S to transduce peripheral neurons in a rat model

Authors: *O. YANG, G. ROBILOTTO, A. MORRIS, A. D. MICKLE;
Univ. of Florida, Gainesville, FL

Abstract: Viral transduction is a commonly used tool to manipulate gene expression in rodents. Adeno-associated viruses (AAV) are the most widely used due to their relative safety and long-term efficacy without causing immunological complications. Many different serotypes have been discovered and developed to transduce different populations of neurons preferentially. Unfortunately, efficient targeting of peripheral neurons remains a challenge for many researchers. Recently, Chan et al. (2017) generated AAV-PHP.S, a capsid engineered from the AAV9 serotype, which produced higher transduction efficacy in the peripheral nervous system in c57/B6 mice compared to AAV9. This project aimed to evaluate the efficiency of systemic PHP.S to transduce peripheral neurons innervating the bladder and external urethral sphincter, compared to AAV9. We injected (IP) Wistar rat pups (P1) with either AAV9-CAG-tdTomato or AAV-PHP.S-CAG-tdTomato and after 6 weeks, collected bladder, external urethral sphincter, dorsal root ganglia, major pelvic ganglia, and spinal cord tissue. We then evaluated expression patterns in peripheral tissue and quantified expression in the dorsal root ganglion and major pelvic ganglion. Initial results indicate that systemic delivery of AAV-PHP.S-CAG-tdTomato was more efficient in the transduction of peripheral neurons in Wistar rats than AAV9-CAG-tdTomato. Our results indicate that AAV-PHP.S exhibits improved transduction of neurons in the peripheral nervous system in rats compared to the AAV9 serotype. These results suggest that despite the AAV-PHP.S serotype being optimized for targeting the mouse peripheral nervous system, it may also be a valuable tool in targeting the rat peripheral nervous system.

Disclosures: O. Yang: None. **G. Robilotto:** None. **A. Morris:** None. **A.D. Mickle:** None.

Poster

P911. Genetic Techniques I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P911.07

Topic: B.11. Glial Mechanisms

Title: Dual labeling Vesicular Stomatitis Virus Allows Identification of Virus-Host Dynamics in vivo in the CNS

Authors: *T. KRAUSE, X. MA, C. CEPKO;
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Abstract: Viral infection is often assayed by markers of ongoing viral gene expression. Such assays do not identify the cells that do not host ongoing viral replication, despite being exposed to virus, or those having a past history of viral gene expression. This leads to an inability to identify cells that are able to reduce or block virus replication. Using a dual labeling Vesicular Stomatitis Virus (VSV), we were able to overcome these limitations. The virus encodes Cre, expressed from a transcriptional unit that is active immediately after infection, which leads to stable marking of any cell with at least a low level of viral gene expression. It also encodes GFP, which marks cells with higher levels of viral gene expression/replication. Assay of the Cre and GFP readouts allows one to better delineate the dynamics of viral infection in tissue or in culture. By performing stereotactic injections of VSV into the murine CNS parenchyma, we found that the majority of productively infected cells (GFP positive) were neurons. In contrast, the cell types that were exposed to virus, but resistant to viral replication, were glia. Using this dual labeling system, we plan to further elucidate the mechanisms behind viral tropism and establishment of infection.

Disclosures: T. Krause: None. **X. Ma:** None. **C. Cepko:** None.

Poster

P911. Genetic Techniques I

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Program #/Poster #: P911.08

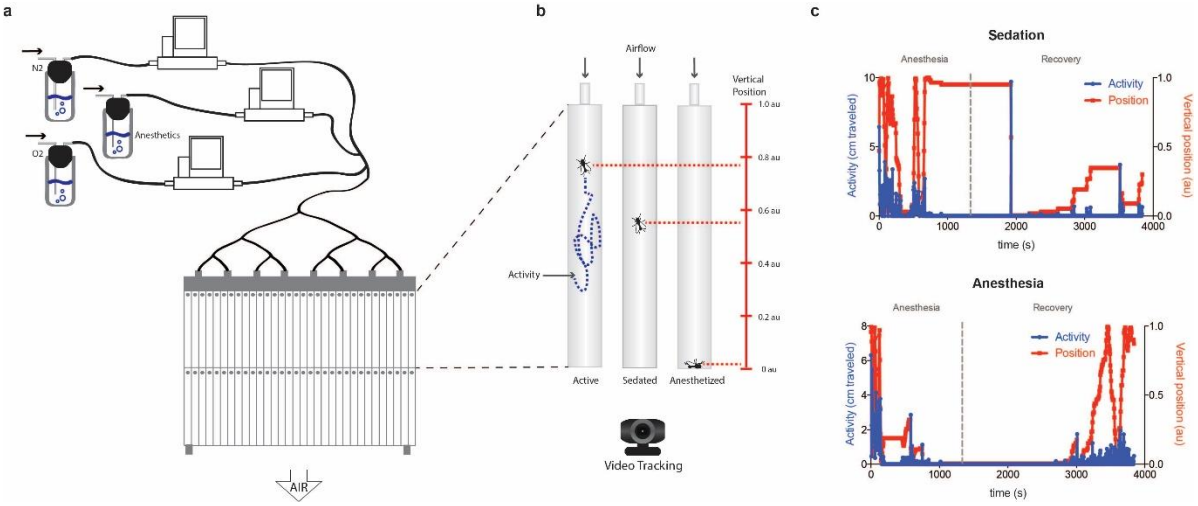
Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH Grant DP2NS087943
NIH Grant R01NS112534

Title: High-throughput testing of sedation and anesthesia in Drosophila by volatiles and aerosols with positional recording (VAAPR) system

Authors: *Z. YUAN, S. HANSEN;
The Scripps Res. Inst., Jupiter, FL

Abstract:



General anesthesia is a drug-induced reversible loss of consciousness (LOC) in animals. Anesthetics have been used clinically for more than 150 years in humans. Systems for accurately measuring anesthesia are complicated by sedation that precedes anesthesia and the very steep hill slopes encountered during dosing of anesthetics (i.e., anesthetics exert their effects over a very narrow concentration range). A high throughput method is in need for testing effects of various kinds and doses of anesthetics and screening potential anesthetic-sensitive genes with high sensitivity and temporal resolution. Previously we built a volatiles and aerosols with positional recording (VAAPR) system for testing anesthetics on Drosophila in 16-well chambers. Here we introduce a new version of VAAPR with 64-well chambers, automated multivalve electric flow meters, and data processing to distinguish sedation and anesthesia. To distinguish between sedation and anesthesia we record and process both activity and vertical position of animals. Flies that stop moving but do not fall, or flies that fall but move and can't regain a vertical position, are identified as sedated. Animals that fall and stop moving are identified as anesthetized. The key event for the latter is the event of falling, which is well-defined and allows for very precise temporal resolution. Furthermore, we introduce the use of survival curves for determining statistically significant anesthetic events in a large number of animals. The use of automated electric flow valves greatly increases the precision that volatile anesthetics can be delivered. Here we use three separate electric flow valves to control O_2 and N_2 separately. This allows us to maintain a constant amount of O_2 while dosing anesthetics in order to avoid potential effects from hypoxemia. We expect this technique to be used for identifying more genes involved in anesthetic sensitivity and other behaviors related to aerosolized drugs.

Disclosures: Z. Yuan: None. **S. Hansen:** None.

Poster

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Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH 5R01MH110932
NIH 1RF1MH120005
NSF 1707316

Title: Trans-synaptic labeling and circuit reconstruction with single neuron resolution by light microscopy in the Drosophila brain

Authors: *Y. LI, K. ATHUKORALA, M. CHEN, L. WALKER, D. CAI;
Univ. of Michigan, Ann Arbor, MI

Abstract: Proper performance of the nervous system relies on the precise function of individual neurons and their intricately-woven network topology. Identifying involved neurons in a circuit, and further resolving the connections between them are critical steps to understand specific brain functions in healthy and disease conditions. Progress in 3D electron microscopy-based circuit reconstruction sheds light on revealing the neural network structure with synapse resolution. However, EM's limitation of data throughput and difficulty of retrieving molecular identity of the reconstructed neurons makes light microscopy a crucial complementary modality.
Recently developed genetic tools, including GRASP, trans-Tango, and TRACT permit identifying anterograde mono-synaptic connections in the Drosophila nervous system. In particular, the latter two can label the full morphology of connected neurons, which promises the potential of revealing the second order neurons' identities and their other connections by the soma locations and projection patterns. That said, it would be ideal to obtain individual neuron's morphology so that the reconstructed network may be mapped to electron microscopy data for detailed connectomic analysis. Furthermore, identity of the second order neurons may be revealed by comparing their morphologies to numerous available light microscopy-based single neuron reconstructions using an informatics tool, e.g. NBLAST. However, current trans-synaptic tools only utilize a single reporter to label the second order neurons, which severely constrains the ability to reconstruct individual neuron's morphology.
To solve this problem, we created a new version of transgenic Bitbow fly to enable multispectral labeling with trans-Tango. Using sample expansion and a custom-built light microscope, we can achieve imaging the whole fly brain with sub-hundred nanometer resolution in a few hours. Combining with computer-aided neuronal tracing, we can now produce brain-scale data with single neuron annotations to build circuit consensus in a high-throughput manner. We envision that these new tools will greatly speed up our progress in brain circuit discovery.

Disclosures: Y. Li: None. **K. Athukorala:** None. **M. Chen:** None. **L. Walker:** None. **D. Cai:** None.

Poster

P916. Single-Cell Techniques I

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Program #/Poster #: P916.01

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIMH grant MH067842
The Saban Research Institute of Children's Hospital Los Angeles Pre-Doctoral Award

Title: SCAMPR: A pipeline for quantitative analysis and spatial mapping of neuronal mRNA expression

Authors: *R. ALI MARANDI GHODDOUSI[1,2], V. MAGALONG[2], A. KAMITAKAHARA[2,1], P. LEVITT[1,2,3];
[1]USC, Los Angeles, CA; [2]Children's Hosp. Los Angeles, Los Angeles, CA; [3]Keck Sch. of Med., Los Angeles, CA

Abstract: Unraveling the role of neuronal heterogeneity in the production of diverse behaviors requires detailed classification of neurons into related and/or distinct types. Cellular location and gene expression patterns are two of the most used characteristics for identifying neuron types, however, until recently it has been difficult to investigate both of these features in the same tissue. Hiplex RNAScope, a commercially available multiplexed *in situ* hybridization technique, combines high dimensional transcriptomics and topology in the same tissue section by allowing for visualization of over 48 different mRNAs at a single-cell resolution. Existing methods for quantitative analysis of spatial mRNA expression have some challenges, particularly the incomplete and/or inaccurate identification of cell boundaries that impact the correct assignment of transcripts to specific cells. Other factors include a requirement for high computing resources and the generation of exceptionally large datasets during analysis. We developed a dual immunohistochemistry-RNAscope protocol that allows for relatively rapid segmentation of neurons based on their cytosolic and nuclear boundaries. Single-Cell Automated Multiplex Pipeline for RNA (SCAMPR) was developed for semi-automated image processing, quantitative analysis, and data visualization of Hiplex RNAscope. SCAMPR requires modest computing power, generates datasets that are practical in size, and allows for sensitive and specific quantification of mRNA expression in both the nuclear and cytoplasmic compartments of the cells. We demonstrate that SCAMPR is compatible with both peripheral and central neurons. Comparisons between manual segmentation of cellular regions of interest (ROI) and SCAMPR demonstrate a minimum of a 3.5 fold increase in speed during ROI segmentation without a

decrease in accuracy. SCAMPR's utility is shown in pairwise and clustering-based comparisons of single-cell gene expression and the mapping of these relationships back onto the source tissue. Lastly, SCAMPR was applied in two different ways to measure, at cellular resolution, differences in gene expression between experimental groups. We used SCAMPR to compare gene expression in the developing nodose ganglion of mice that were raised under normal rearing conditions (care as usual) to those that were raised in the presence of early life stress. We also compared neurotrophin receptor mRNA expression in the embryonic brainstem of wildtype mice to the expression in mice containing conditional knockouts of the gene encoding the pleiotropic MET receptor tyrosine kinase (*Met)*.

Disclosures: R. Ali Marandi Ghoddousi: None. **V. Magalong:** None. **A. Kamitakahara:** None. **P. Levitt:** None.

Poster

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Program #/Poster #: P916.02

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Title: Every Cell Matters: Molecular Cartography™ of the mammalian brain

Authors: B. NILGES, F. REINECKE, A. BOGDOLL, ***N. KASHIKAR**;
Resolve Biosci. GmbH, North Rhine-Westphalia, Germany

Abstract: The brain is a complex organ and understanding its working is one of the holy grails of human scientific pursuits. Quantitative studies of the molecular inventory of the brain stimulate new hypotheses and experiments in basic and translational neuroscience. However, despite the recent advances in sequencing approaches, the spatial architecture of the mRNA expression at single-cell resolution has remained poorly understood. Only the addition of a spatial component to transcriptomic information can provide the context to understand the function of cell types, individual subtypes as well as connectivity within and across anatomically defined regions. Here, using Molecular Cartography (MC) on the mouse brain, we present a novel resource for the neuroscientific community, where we spatially map and quantify the transcript abundance of 100 genes that represent some of the key pathways and markers defining neural cells and CNS signaling. MC enables digital quantification of mRNA molecules, high sensitivity that parallels conventional single-molecule FISH, subcellular resolution in three-dimensions within a 10 µm thick tissue section. The genes were chosen to serve as combinatorial markers for specific cell types and subtypes in the different regions of the brain. We focus on the part of the hemisphere that contains the most relevant neuroanatomical areas such as the cortex, hippocampus, caudoputamen, thalamus, hypothalamus. We demonstrate that: a. MC data matches conventional *in situ* hybridization-based methods demonstrating a high degree of

specificity, b. MC is highly quantitative and correlates with an R^2 of >0.9 with bulk RNAseq data, c. identification and spatial clustering of more than 30 cell types and subtypes, and d. identification of all major cell layers and the heterogeneity of cells within.
Although we focus on the mouse brain as a resource case, MC is equally applicable to other species of neuroscientific interest, e.g., human samples from clinics or brain banks, cell culture models of major neural cell types, and reptilian or fish brains that enable to infer the functions of genes cells and structures based on their conservation throughput evolution. We trust that MC provides a novel and unique opportunity for neuroscience researchers to redefine the mapping of the brain in health, disease, and development.
The mouse brain dataset is available to download for registered users on resolve-biosciences.com.

Disclosures: B. Nilges: A. Employment/Salary (full or part-time):; full time. **F. Reinecke:** A. Employment/Salary (full or part-time):; full time. **A. Bogdoll:** A. Employment/Salary (full or part-time):; full time. **N. Kashikar:** A. Employment/Salary (full or part-time):; full time.

Poster

P916. Single-Cell Techniques I

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Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P916.03

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIMH U19MH114831

Title: Epigenomic Diversity of Cortical Projection Neurons in the Mouse Brain

Authors: *Z. ZHANG[1], J. ZHOU[1,6], P. TAN[1,8], Y. PANG[2], A. RIVKIN[1], M. A. KIRCHGESSNER[2], E. WILLIAMS[3], C.-T. LEE[4], H. LIU[1,6], A. D. FRANKLIN[2], P. A. MIYAZAKI[2], A. BARTLETT[1], A. ALDRIDGE[1], M. VU[2], C. FITZPATRICK[5], J. NERY[1], R. G. CASTANON[1], T. ITO[2], C. O'CONNOR[5], L. BOGGEMAN[5], K.-F. LEE[4], X. JIN[3], E. A. MUKAMEL[7], M. BEHRENS[9], J. R. ECKER[10,12], E. M. CALLAWAY[11];
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Abstract: Neuronal cell types are classically defined by their molecular properties, anatomy, and functions. While recent advances in single-cell genomics have led to high-resolution molecular characterization of cell type diversity in the brain, neuronal cell types are often studied out of the context of their anatomical properties. To better understand the relationship between molecular

and anatomical features defining cortical neurons, we combined retrograde labeling with single-nucleus DNA methylation sequencing to link neural epigenomic properties to projections. We examined 11,827 single neocortical neurons from 63 cortico-cortical (CC) and cortico-subcortical long-distance projections. Our results revealed unique epigenetic signatures of projection neurons that correspond to their laminar and regional location and projection patterns. Based on their epigenomes, intra-telencephalic (IT) cells projecting to different cortical targets could be further distinguished, and some layer 5 neurons projecting to extra-telencephalic targets (L5 ET) formed separate clusters that aligned with their axonal projections. Such separation varied between cortical areas, suggesting area-specific differences in L5 ET subtypes, which were further validated by anatomical studies. Interestingly, a population of CC projection neurons clustered with L5 ET rather than IT neurons, suggesting a population of L5 ET cortical neurons projecting to both targets (L5 ET+CC). We verified the existence of these neurons by dual retrograde labeling and by anterograde tracing of CC projection neurons, which revealed axon terminals in ET targets including thalamus, superior colliculus, and pons. These findings highlight the power of single-cell epigenomic approaches to connect the molecular properties of neurons with their anatomical and projection properties.

Disclosures: Z. Zhang: None. **J. Zhou:** None. **P. Tan:** None. **Y. Pang:** None. **A. Rivkin:** None. **M.A. Kirchgessner:** None. **E. Williams:** None. **C. Lee:** None. **H. Liu:** None. **A.D. Franklin:** None. **P.A. Miyazaki:** None. **A. Bartlett:** None. **A. Aldridge:** None. **M. Vu:** None. **C. Fitzpatrick:** None. **J. Nery:** None. **R.G. Castanon:** None. **T. Ito:** None. **C. O'Connor:** None. **L. Boggeman:** None. **K. Lee:** None. **X. Jin:** None. **E.A. Mukamel:** None. **M. Behrens:** None. **J.R. Ecker:** None. **E.M. Callaway:** None.

Poster

P916. Single-Cell Techniques I

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Program #/Poster #: P916.04

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: 1RF1MH114126-01
1UG3MH120095-01A1
P51OD010425
UL1TR000423

Title: A collection of enhancer-AAVs for targeting glial cell types across species

Authors: *B. LEVI[1], Y. BISHAW[4], J. K. MICH[1], R. MARTINEZ[4], B. GORE[4], J. T. MAHONEY[4], V. OMSTEAD[4], D. MACHEN[4], M. LEYTZE[4], N. TASKIN[4], N. WEED[4], X. OPITZ-ARAYA[4], N. DEE[4], N. SHAPOVALOVA[4], D. HIRSCHSTEIN[4], J. GOLDY[4], K. SMITH[4], S. YAO[4], Y. KOJIMA[7], S. GIBSON[8], G. D. HORWITZ[9], H. ZENG[5], T. L. DAIGLE[1],

B. TASIC[2], J. T. TING[3], E. LEIN[6];
[2]Cell and Circuit Genet., [3]Human Cell Types, [1]Allen Inst. For Brain Sci., Seattle, WA; [5]Structured Sci., [6]Human Cell Types, [4]Allen Inst. for Brain Sci., Seattle, WA; [7]Washington Natl. Primate Res. Ctr., Seattle, WA; [9]Physiol. and Biophysics, [8]Univ. of Washington, Seattle, WA

Abstract: Selective transgene expression is critical to the study of brain cell types including their roles in brain function and disease. However, few tools exist that can selectively target cell populations and can be applied across mammalian species. To enable specific brain cell population labeling across species, we are generating a collection of enhancer-AAV vectors that drive transgene expression in defined cell subclasses. We undertook a systematic screen to find enhancer elements capable of targeting gene expression from AAV vectors to major neocortical cell populations. In previous work, we have described enhancers for several different population of cortical neurons, and we demonstrated the maintenance of specificity in mouse and primate (Mich et al. Cell Reports 2021). Here, we show our progress generating a new collection of enhancer-AAV tools to target glial cell populations. We present a set of astrocyte- and oligodendrocyte-selective enhancers-AAV vectors, and present characterization of their activity across brain regions, and across species. Our growing toolbox of enhancer-AAVs for selective transgene expression will be important to dissect the roles of cell populations in brain function across species.

Disclosures: B. Levi: B. Contracted Research/Research Grant (principal investigator for a drug study, collaborator or consultant and pending and current grants). If you are a PI for a drug study, report that research relationship even if those funds come to an institution.; Receives research support from BioMarin. E. Ownership Interest (stock, stock options, royalty, receipt of intellectual property rights/patent holder, excluding diversified mutual funds); Inventor on licensed and unlicensed provisional patents. F. Consulting Fees (e.g., advisory boards); Consult with PatchBio. **Y. Bishaw:** None. **J.K. Mich:** None. **R. Martinez:** None. **B. Gore:** None. **J.T. Mahoney:** None. **V. Omstead:** None. **D. Machen:** None. **M. Leytze:** None. **N. Taskin:** None. **N. Weed:** None. **X. Opitz-Araya:** None. **N. Dee:** None. **N. Shapovalova:** None. **D. Hirschstein:** None. **J. Goldy:** None. **K. Smith:** None. **S. Yao:** None. **Y. Kojima:** None. **S. Gibson:** None. **G.D. Horwitz:** None. **H. Zeng:** None. **T.L. Daigle:** None. **B. Tasic:** None. **J.T. Ting:** None. **E. Lein:** None.

Poster

P916. Single-Cell Techniques I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P916.05

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Title: Spatial annotation of scRNA-seq data by 10x Visium

Authors: *A. C. MITCHELL, M. CHIANG, M. KUNST, K. BROUNER, A. GARY, A. TORKELSON, M. MCGRAW, K. SMITH, C. VAN VELTHOVEN, B. TASIC, H. ZENG; Allen Inst. for Brain Sci., Seattle, WA

Abstract: Over the past decade various single-cell genomics approaches (e.g., RNA-seq) applied to dissociated cells or nuclei have been used to define of cell types at the molecular level. These single-cell molecular profiles can also be used to predict spatial and anatomical location of cell types based on differential marker gene expression. However, to accurately and quantitatively define locations and distributions of cell types highly multiplexed molecular measurements *in situ* are needed. Here we use 10x Genomics Visium spatial gene expression platform (slides with 55 µm spots) to profile transcriptome-wide gene expression from 15 coronal and 8 sagittal sections spanning the mouse brain. The resulting data was integrated with the Allen Institute for Brain Science forebrain scRNA-seq-based taxonomy (https://portal.brain-map.org/atlases-and-data/rnaseq) and Bing Ren's lab BICCN snATAC mouse brain *cis*-element atlas (http://catlas.org/mousebrain). In conclusion, we find that despite lack of single cell resolution, 10x Visium spatial transcriptomics data are valuable for interpretation of scRNA-seq data to define spatial localization/distribution of cell types.

Disclosures: A.C. Mitchell: None. **M. Chiang:** None. **M. Kunst:** None. **K. Brouner:** None. **A. Gary:** None. **A. Torkelson:** None. **M. McGraw:** None. **K. Smith:** None. **C. van Velthoven:** None. **B. Tasic:** None. **H. Zeng:** None.

Poster

P916. Single-Cell Techniques I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P916.06

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH IRP Z01 MH001090

Title: Analyzing blood brain barrier involvement in major depressive disorder using neurovascular cell enrichment and single nucleus RNA sequencing

Authors: *M. LOTSTEIN, J. SAMUELS, M. LEHMANN, M. HERKENHAM; NIH, Natl. Inst. of Mental Hlth., Bethesda, MD

Abstract: Previous work in our laboratory showed that chronic psychosocial stress in mice induces vascular micro-damage in brain, including a decline in blood brain barrier (BBB) function. This damage activates a series of inflammatory pathways in brain-resident immune cells and precipitates depressive-like states. Not only relevant to psychosocial stress, BBB dysfunction underlies a subset of disorders, including Major Depressive Disorder (MDD). We

want to use transcriptomic analysis to examine how barrier cells respond to stress, specifically in MDD. We aim to identify the differences between postmortem samples from MDD and healthy control brains using single-nucleus RNA sequencing (snRNA-Seq). However, snRNA-Seq datasets have a low representation of neurovascular cells, typically limited to small endothelial and microglial clusters. These small sample sizes prevent any rigorous statistical analyses on neurovascular cells, which are frequently overlooked in favor of the more populous neurons and oligodendrocytes. We are using previously published snRNA-Seq datasets to identify signature markers of non-neurovascular nuclei that reside within the nuclear membrane. By staining our isolated nuclei samples with neuronal, oligodendrocyte, and astrocyte markers, we can sort out these dominating cell types. After performing snRNA-Seq experiments on sorted and non-sorted nuclei from frontal cortex of human postmortem brain, our sorted samples have a significant increase in neurovascular populations, including our main cell type of interest—endothelial cells. Further experiments are underway to examine the transcriptomic changes in barrier cells between MDD and healthy control brains after using the signature markers to enrich our samples for neurovascular cells.

Disclosures: M. Lotstein: None. **J. Samuels:** None. **M. Lehmann:** None. **M. Herkenham:** None.

Poster

P916. Single-Cell Techniques I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P916.07

Topic: I.01. Molecular, Biochemical, and Genetic Techniques

Support: NIH Grant 1R34NS111654-01

Title: Robotag-seq: robot assisted cellular barcoding for transcriptome-wide sequencing of anatomically identified cell populations in intact tissue

Authors: *J. O'BRIEN[1], B. AUCH[2], M. S. RIEDL[3], L. GENGELBACH[2], A. G. J. SKORPUT[3], D. M. GOHL[2,4], L. VULCHANOVA[3], S. B. KODANDARAMAIAH[1];
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Abstract: Single cell transcriptomics is a powerful tool for measuring the gene expression patterns within single cells, and it has revealed numerous insights that were previously hidden by the averaging of gene expression patterns across cell populations. The toolkit for single cell transcriptomics currently lacks the ability to integrate spatial and functional information with transcriptomic measurements at scale. Current techniques like Patch-Seq, which combine functional and transcriptomic profiling, are laborious, time consuming, and do not scale easily

for analysis on multiple cells in tissue. In contrast, spatial transcriptomics, which profiles transcriptomes in a spatially resolved manner, lacks the ability to conduct transcriptome-wide measurements at cellular resolution and is depth-limited to only analyzing cells near the tissue surface. Here, we present a robotic microinjector that is programmed to microinject 100's of pre-identified cells in intact neural tissue with unique oligonucleotide barcode-antibody conjugates (hashtags) which target the nuclear pore complex. After injection, nuclei are isolated from the surrounding tissue before transcriptomic profiling using emulsion droplet-based cell partitioning. During transcriptomic profiling, the injected hashtags are recovered and can be used to relate the profiled transcriptomes with the pre-identified cells thereby integrating spatial and transcriptomic information into a single approach. We have demonstrated successful automated microinjection of a fluorescently labeled dextran into neurons in mouse striatum and spinal cord dorsal horn slices with success rates of 96% and 68%, respectively. Additionally, we have demonstrated automated microinjection-mediated tagging of neuronal nuclei in mouse striatum slices with a fluorescently labeled antibody as a precursor step for fluorescence based nuclear enrichment and as a proof of concept for hashtag labelling of nuclei using the same monoclonal antibody. We are capable of performing high-throughput automated microinjections at a rate of approximately 100 neurons per hour which will facilitate transcriptome-wide profiling at scale. We are currently optimizing the processes for nuclear harvesting, enrichment, and subsequent transcriptomic profiling. When fully developed, we anticipate this to be a generalized strategy for barcode tagging specific populations of cells of interest for whole transcriptome analysis in a variety of tissues.

Disclosures: J. O'Brien: None. **B. Auch:** None. **M.S. Riedl:** None. **L. Gengelbach:** None. **A.G.J. Skorput:** None. **D.M. Gohl:** None. **L. Vulchanova:** None. **S.B. Kodandaramaiah:** None.

Poster

P916. Single-Cell Techniques I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P916.08

Topic: B.08. Intrinsic Membrane Properties

Support: DFG grant EXC 2049
DFG grant FOR 2143

Title: Intra-individual physiomic landscape of pyramidal neuron network in the human neocortex

Authors: H. PLANERT[1], F. X. MITTERMAIER[1], S. GROSSER[2], P. FIDZINSKI[3], U. SCHNEIDER[3], H. ALLE[1], I. VIDA[2], J. R. P. GEIGER[1], ***Y. PENG**[1];

[1]Inst. of Neurophysiol., [2]Inst. for integrative Neuroanatomy, [3]NeuroCure Cluster of Excellence, Charité-Universitätsmedizin Berlin, Berlin, Germany

Abstract: Computation within cortical microcircuits is defined by properties of the neurons and the synaptic interaction between them. While heterogeneity among different interneuron types is well established, excitatory pyramidal neurons of the cortex are considered to be largely homogeneous. To identify possible functional differentiations within this pyramidal neuron population and their recurrent synapses, we focused on the human layer 2/3 temporal cortex which is greatly expanded compared to the rodent cortex.
We performed multi-neuron patch-clamp recordings in acute brain slices obtained from patients (22 individuals) undergoing epilepsy resection surgery. In total, we recorded the electrophysiological properties of 1007 pyramidal neurons and 1242 monosynaptic connections. Our high-throughput approach allowed sampling of up to 90 neurons and 170 synaptic connections from single individuals enabling us to assess inter- and intra-individual variability. We observed substantial heterogeneity in cellular parameters with intra-individual variability severalfold larger than inter-individual variability. Hierarchical clustering of the high-dimensional parameter set yielded functionally distinct clusters of pyramidal neurons which were present in each patient. Synaptic properties of recurrent excitatory connections were highly diverse including preliminary evidence for some target-cell specificity.
Taken together, or results show that the human layer 2/3 pyramidal neuron population comprise distinct functional subtypes. This may represent a generic organizational principle that is shared across human individuals and relate to previously reported transcriptomic heterogeneity. The identified diversity in cellular and synaptic function has important implications for the computational capacity of the cortical microcircuit and highlights potential species-specific differences.

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Poster

P957. Experimental Computational Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P957.01

Topic: I.06. Computation, Modeling, and Simulation

Title: Markerless pose estimation programs use on 12 month old infants

Authors: *S. FINKBEINER[1], W. QUACKENBUSH[1], M. REIMERS[1], R. KNICKMEYER[1], C. BEST[2];
[1]Michigan State Univ., Lansing, MI; [2]Michigan State Univ., East Lansing, MI

Abstract: Neural network-based markerless motion tracking programs hold great promise for studying the behavior of both people and animals videotaped in freely moving conditions, by determining the movement of different body parts through space. However, little work has been done using these softwares to evaluate infant motion. The primary objective of the current study was to compare two such systems (DeepLabCut (DLC) and OpenPose (OP)) to each other, as well as to ground truth annotations done by our research team. DeepLabCut is a neural network that was originally trained on mouse models, it requires minimal training from the user before it's implemented; whereas OpenPose is a pre-trained neural network that was trained on humans and required no training before use. We used 22 videos of infants, about 12 months of age, acquired during a behavioral paradigm adapted from the Laboratory Temperament Assessment Battery (Lab-TAB) Mask episode. Videos were provided by researchers at University of North Carolina at Chapel Hill (UNC), who used them to assess infant fear reactivity. We determined that aside from the Neck point, OpenPose1.7.0 is providing more accurate annotations compared with the DeepLabCut2.0.7.2 neural network, which we trained on 2,809 frames. The videos used for annotation were captured from the infant's left side, creating lower confidence values for key points on the right side of the infant (eyes and ears). Aside from the right ear and right eye, the annotation that showed the lowest confidence when using OP was the neck annotation; 81% of infants analyzed showed consistent marginal drops in confidence when compared to other annotated key points. Our team is currently exploring whether we can supplement the OP neural network with data from DLC to attain more accurate neck annotations. Ultimately we will use these pose estimation programs to (1) determine the overall motion of the infants and compare it to parental ratings of infant activity level, (2) assess how infants direct their attention during the mask task and test whether this is associated with expert ratings of infant fear reactivity, and (3) determine if expert ratings of fear reactivity can be predicted from infant motion.

Disclosures: S. Finkbeiner: None. **W. Quackenbush:** None. **M. Reimers:** None. **R. Knickmeyer:** None. **C. Best:** None.

Poster

P957. Experimental Computational Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P957.02

Topic: I.06. Computation, Modeling, and Simulation

Support: Schweizerischer Nationalfonds zur Förderung der Wissenschaftlichen Forschung (Swiss National Science Foundation) - PP00P3_157539 [Mante]
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Eidgenössische Technische Hochschule Zürich (Federal Institute of Technology Zurich) - ETH-20 19-1 [Bohacek]
Fondation Botnar [Bohacek]
Swiss 3R Competence Centre (OC-2019-009, JB and MFY) , Directorate 3RCC c/o University of Bern, Hochschulstrasse 6, CH-3012 Bern, secretariat@swiss3rcc.org, +41 31 631 56 22 [Bohacek]

Title: Deep-learning based pose estimation, identification, and end-to-end behavior classification for interacting primates and mice in complex environments

Authors: *M. MARKS[1], J. QIUHAN[1], O. STURMAN[2], L. VON ZIEGLER[2], S. KOLLMORGEN[1], W. VON DER BEHRENS[1], V. MANTE[1], J. BOHACEK[2], M. F. YANIK[1]; [1]ETH Zurich and Univ. of Zurich, Zuerich, Switzerland; [2]ETH Zurich, Zurich, Switzerland

Abstract: The quantification of behaviors of interest from video data is commonly used to study the effects of genetic alterations, pharmacological interventions, and brain functionality. Existing approaches lack the capabilities to analyze the behavior of groups of animals in complex environments. We present a novel deep learning architecture for classifying individual and social animal behavior in complex environments from raw video frames, whilst requiring no intervention after minimal human supervision. Our behavioral classifier is embedded in a pipeline (SIPEC) to perform segmentation, identification, pose-estimation, and classification of complex behavior all automatically with state-of-the-art performance. SIPEC successfully recognizes multiple behaviors of freely moving individual mice as well as socially interacting non-human primates in 3D, using data only from simple mono-vision cameras in home-cage setups. We show how this pipeline can be easily deployed and used by researchers to analyze their behavioral data.

Disclosures: M. Marks: None. **J. Qiuhan:** None. **O. Sturman:** None. **L. von Ziegler:** None. **S. Kollmorgen:** None. **W. von der Behrens:** None. **V. Mante:** None. **J. Bohacek:** None. **M.F. Yanik:** None.

Poster

P957. Experimental Computational Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P957.03

Topic: I.06. Computation, Modeling, and Simulation

Support: National Eye Institute grant (R01-EY025330)
National Defense Science and Engineering Graduate fellowship

Title: A Unified Model of the Task-evoked Pupil Response

Authors: *S. MIRBAGHERI[1], C. S. BURLINGHAM[2], D. J. HEEGER[2];
[1]Grad. Program in Neurosci., Univ. of Washington, Seattle, WA; [2]Psychology, New York Univ., New York, NY

Abstract: Background: During task performance, pupil size entrains to trial timing, dilating and then constricting sluggishly following trial onsets. This so-called task-evoked pupil response is modulated in amplitude by task demands and behavioral performance. The task-evoked pupil response is commonly used to estimate arousal by assuming pupil size is a linear transformation of task events. We show that estimates of arousal based on this consensus model are not robust to small changes to the task that should not alter arousal level. We propose an alternative model linking the task-evoked pupil response and arousal, which leverages that a common neural input drives pupil size and saccades. **Method:** Human observers performed a forced-choice orientation-discrimination task on a briefly displayed peripheral grating. On easy trials, the grating was tilted far from vertical, and observers achieved ~93% accuracy on average. On hard trials, the grating's tilt was close to vertical, and observers achieved ~70% performance on average. The task difficulty was changed in alternate runs (75 trials/run, 4 s/trial). In a separate version of the task, the wait time between trials was reduced (2 s/trial). We fit a computational model to measurements of pupil size and saccades, generating estimates of arousal level. **Result:** Fitting our model to the data yields: 1) an estimate of the common input that drives both saccades and pupil size and 2) the gain of the input to the pupil, separately for each trial type. Gain scaled with task difficulty and behavioral performance and generalized across tasks with different inter-trial wait times, as expected of a measure of arousal. The estimated arousal from the consensus model, on the other hand, varied considerably with the wait time between trials, indicating that this model over-fits and that pupil size is not a linear transformation of task events. The estimated common input was modulated by the timing of the task and behavioral response, suggesting that it reflects a cognitive representation of task structure. Moreover, the model offers a unified explanation for other phenomena, including modulation of trial-to-trial pupil response variability with task difficulty and a constrictory pupil response time-locked to saccades that scales with task difficulty. **Conclusion:** We propose a novel model that predicts the shape and amplitude of the task-evoked pupil response from saccade timing, estimates arousal robustly, and explains many other phenomena. This model's success suggests strong constraints on the neural circuits that comprise the common driver of saccades and pupil size, as well as the arousal/gain modulator of pupil size.

Disclosures: S. Mirbagheri: None. **C.S. Burlingham:** None. **D.J. Heeger:** None.

Poster

P957. Experimental Computational Tools I

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Program #/Poster #: P957.04

Topic: I.06. Computation, Modeling, and Simulation

Support: NSF-1632259
DHHS NIDILRR Grant 90REGE0005-01-00 (COMET)
NIH/NIBIB Grant 1R01EB024058-01A1

Title: Nonlinear dimensionality reduction simplifies the control of dexterous prosthetic hands

Authors: *A. PORTNOVA-FAHREEVA[1], F. RIZZOGLIO[2], M. CASADIO[3], F. A. MUSSA-IVALDI[4], E. ROMBOKAS[5];
[1]Northwestern Univ., Evanston, IL; [2]Univ. Degli Studi Di Genova, Genova, Italy; [3]Univ. of Genova, Genova, Italy; [4]Arms And Hands, Shirley Ryan AbilityLab, Chicago, IL; [5]Univ. of Washington, Seattle, WA

Abstract: With the rise of technological advancements, prosthetic hands have seen many improvements in their designs, increasing the number of Degree of Freedoms (DOFs) from simple one-degree hooks to anatomically-correct testbed hand. Nonetheless, the control strategies have been limited, preventing prosthetic users from utilizing the entire plethora of finger dexterity in their devices and limiting them to control only a pre-selected number of grips, either via a simple finite state machine in an on-off pattern or more complex pattern recognition algorithms. In this study, we proposed the use of a nonlinear dimensionality-reduction method, the nonlinear Autoencoder (NAE), to explore the lower-dimensional, *latent*, space of complex human hand kinematics in order to facilitate prosthetic control. The NAE was compared to the more standard Principal Component Analysis (PCA), frequently used to explore the dimensionality of hand kinematics. Performance of the two methods was compared on (a) the ability to accurately reconstruct hand kinematic data from a latent manifold and (b) variance distribution across latent dimensions. The NAE exhibited high performance in its ability to reconstruct hand kinematic data with 94% accuracy from a two-dimensional latent space in comparison to just 78% accuracy in PCA. In addition, the latent dimensions of the NAE exhibited a more equal distribution of variance in comparison to PCA. To test the possibility for the NAE to offer an effective platform to control prosthetic hands, we developed a simple myoelectric controller, in which users were able to control a 15-DOF virtual hand using four forearm muscles. Muscle signals were combined in a vector summation algorithm, allowing the users to operate on a 2D space, which corresponded to the latent space of hand kinematics. As a result, we could decode every point on the control space into its corresponding 15 dimensions of virtual hand kinematics. Through this study, we also explored the complexity of learning a visually high-dimensional task (*i.e.*, controlling a 15D virtual hand) controlled by a low-dimensional space (*i.e.*, 2D latent space) and the effect of the users' training on the visually low-dimensional task (*i.e.*, cursor control) on their learning to perform the task that appeared high-dimensional. Two groups were created, in which one was trained on a 2D cursor-controlling task prior to learning to control the 15D virtual hand, the other one was only exposed to learning to manipulate the virtual hand, without any feedback about the true dimensionality of the control space.

Disclosures: A. Portnova-Fahreeva: None. **F. Rizzoglio:** None. **M. Casadio:** None. **F.A. Mussa-Ivaldi:** None. **E. Rombokas:** None.

Poster

P957. Experimental Computational Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P957.05

Topic: I.06. Computation, Modeling, and Simulation

Support: Canada First Research Excellence Fund

Title: A Simple Device for Real-Time Detection of Head and Body Movements in a Mock Scanner, for Screening and Training Human Subjects and Alert Animals

Authors: *F. AYAD[1], P. HUMANE[2], T. DAVID[3], A. BORTEL[4], A. SHMUEL[3];
[1]Biomed. Engin., McGill Univ., montreal, QC, Canada; [2]McGill Univ., Monteal, QC, Canada; [3]McGill Univ., Montreal, QC, Canada; [4]Montreal Neurolog. Institute, McGill Univ., Montréal, QC, Canada

Abstract: Introduction
With the advent of imaging at ultra-high field, head and body movements are one of the factors that limit our capacity to obtain high resolution/quality data. We have developed a small wearable device with the purpose of acquiring wireless data on head and body movements for real-time analysis while a subject is trained in a mock MRI scanner. This device works in parallel to an in-bore camera used for movement detection, to support subject screening and training to remain still in preparation for MRI.
Methods
Our setup consists of a wireless motion sensor system and an in-bore video camera. The sensor system consists of two highly sensitive, three-axis sensor nodes. The sensor nodes can be placed on the forehead and the hip of the participating human subjects by means of elastic bands, to measure head and body movement, respectively. For alert primates and mice, the sensor is sufficiently sensitive to detect platform vibrations caused by the animal's movement.
The software plots and analyzes the movement data in real-time and alerts the user and the subject when a movement beyond the accepted thresholds occurs.
Results
We have tested the system for training human subjects, alert primates, and alert mice.
Conclusions The proposed system can be used to screen and train human subjects or alert animals to stay still in the MRI. It can also be used for gradually accommodating special subject populations - such as subjects with neurodevelopmental disorders - to staying still in the scanner. The device can contribute to improving imaging quality and reducing the cost of imaging human subjects that are prone to moving and thus producing unusable data. Given the increase in ultra-

high-field (7 Tesla and higher) MRI systems, the implementation of this device is even more important, since movement artifacts are accentuated and especially detrimental in high-field imaging.

VIDEO Demonstration Link

https://mcgill-my.sharepoint.com/:v:/g/personal/fadi_ayad_mail_mcgill_ca/ESasA4OQc59KjlwIzO0vvC0B9zLAvEp3g0xNMGol7iYWXw?e=euncwd



Figure 1: The head movement Sensor attached to a subject's forehead.

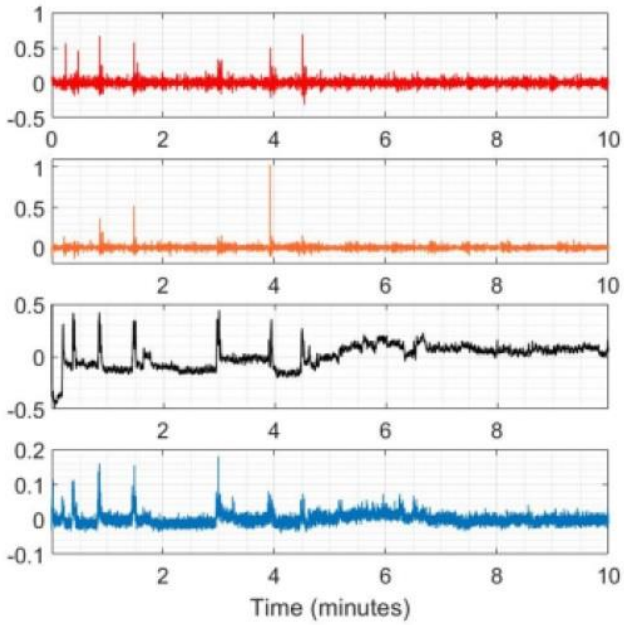


Figure 2: Sample data from Subject 1

Disclosures: F. Ayad: None. **P. Humane:** None. **T. David:** None. **A. Bortel:** None. **A. Shmuel:** None.

Poster

P957. Experimental Computational Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P957.06

Topic: I.06. Computation, Modeling, and Simulation

Support: NSF Graduate Research Fellowship (K.A. Malaga)
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UM Rackham Merit Fellowship (K.A. Malaga)
Wallace H. Coulter Foundation
A. Alfred Taubman Medical Research Institute

Title: Patient-specific diffusion tensor-based thalamic segmentation in deep brain stimulation for essential tremor

Authors: *K. A. MALAGA[1,2], L. HOUSHMAND[2], J. T. COSTELLO[3], J. CHANDRASEKARAN[2], K. L. CHOU[4,5], P. G. PATIL[2,4,5];
[1]Biomed. Engin., Bucknell Univ., Lewisburg, PA; [2]Biomed. Engin., [3]Electrical Engin., [4]Neurol., [5]Neurosurg., Univ. of Michigan, Ann Arbor, MI

Abstract: Thalamic deep brain stimulation (DBS) is an established surgical therapy for essential tremor (ET). Traditional DBS targeting and electric field modeling employ atlas-based approaches, which perform well on average but obscure patient-specific anatomical variability. The objective of this retrospective study was to assess an individualized modeling methodology (N-of-1 approach) for segmenting thalami in ET patients receiving unilateral DBS. The N-of-1 approach uses patient-specific diffusion tensor imaging data to perform thalamic segmentation and volume of tissue activation (VTA) modeling. For each patient (n = 22), the thalamus was segmented into 13 nuclei using k-means clustering. VTAs associated with therapeutic and side effects were computed using finite element analysis and then used to evaluate the segmentation. Segmentation and VTA results from N-of-1 and atlas-based modeling were compared. VTA size and shape varied considerably across patients. Using N-of-1 segmentation, 91% of tremor suppression VTAs overlapped with motor thalamus (100% using atlas-based). Therapeutic VTAs had significantly more motor thalamus overlap than sensory thalamus for both approaches. For sustained paresthesia, 94% of VTAs overlapped with sensory thalamus using N-of-1 segmentation (74% using atlas-based). Sustained paresthesia VTAs had significantly more sensory thalamus overlap than therapeutic VTAs for both approaches. For dysarthria/motor contraction, 68% of VTAs spread outside thalamus (in all directions) using the N-of-1 approach (79% using atlas-based). Dysarthria/motor contraction VTAs had significantly more external

spread than therapeutic VTAs for only the N-of-1 approach. The N-of-1 approach explained DBS side effects better, specifically when patient neuroanatomy differed from the atlas. Although atlas-based results were similarly accurate across patients, the N-of-1 approach may provide greater precision regarding individual outcomes. N-of-1 thalamic segmentation, in particular, enables optimized targeting based on patient-specific anatomical variability.

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Poster

P957. Experimental Computational Tools I

Location: Virtual Only

Time: Tuesday, November 9, 2021, 11:00 AM - 12:00 PM

Program #/Poster #: P957.07

Topic: I.06. Computation, Modeling, and Simulation

Title: Logistic regression in machine learning with application to face recognition and unknown rejection

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Abstract: Face recognition in machine learning is generally about recognizing known faces. As science attempts to close the gap between man and machine by building systems capable of learning, we must embrace the importance of the unknown. The ability to differentiate between known and unknown can be considered a critical element of any intelligent self-learning system. (Boult, Cruz, 2019, p. 1). This poster investigates the use of logistic regression in Machine Learning, using an example of how this can be applied to face recognition and unknown face rejection. Machine learning in face recognition, faces of a selected group of people are classified, and all other faces and non-faces are rejected. The classic approach is to estimate the class densities, assigning the face to the class with the highest posterior probability. Performance of the face recognition classifier is counted as good if the known test face is classified correctly. If the known test face is misclassified by the face recognition classifier, then the misclassification is counted as a classification error. If an unknown face is presented to the face recognition classifier, the face recognition classifier could still classify the unknown face erroneously as a known face. Distance thresholds as used in k-nearest neighbor or euclidean based classifiers could classify an unknown face as unknown should the distance threshold not be high enough to declare as a known face, but may not be consistent to reject the unknown. In contrast, logistic regression is a way to use a squashing function or a logistic function to map a real-valued input to the range 0 to 1. Since the logistic or sigmoid output values are between 0 and 1, it would be reasonable to view the output as a probability. Logistic regression predicts positive when the

probability is greater than 0.5 and negative when the probability is less than 0.5. Unknown face rejection is investigated in this poster using the Olivetti face dataset engaging logistic regression with sigmoid output probabilities. A whole approach uses all of the pixels in the Olivetti face image as a feature vector processed in a logistic regression binary classification scheme of one vs all where a face image is either in the Known Class or not in the Known Class (i.e. unknown). Unknown rejection performance is computed and compared using single-look processing with one input unknown face, and multi-look processing with two input unknown faces.

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Poster

P957. Experimental Computational Tools I

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Title: Large-scale acquisition of hidden fluorescent labels for training versatile markerless motion capture networks

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Abstract: Recent advances in machine learning-based markerless pose estimation have dramatically improved the scale and ease with which body landmarks can be tracked in studies of animal behavior. However, pose estimation for animals in a laboratory setting still faces some unique challenges: researchers typically manually generate new labels (~500-1000 labels) for each experimental setup (e.g., for differing behavioral assays, camera angles, lighting conditions, etc.), limiting the generalizability of this approach. Furthermore, each lab trains their own landmark trackers from scratch - the lack of a ‘one-size-fits-all’ kinematic tracker means that different labs track different anatomical landmarks and analyze the resulting kinematic data in idiosyncratic ways. To address these issues, we developed an approach, which we term GlowTrack, for markerless tracking that can generalize across behaviors, experimental setups, and labs. The key innovations are: a) an automated, high-throughput approach for generating

hidden labels free of human error using fluorescent markers at a scale that would not be feasible for human annotation (100's of thousands to millions); b) a multi-camera, multi-light setup for generating large amounts of training data diversity; and c) a technique for massively-parallel tracking of hundreds of landmarks simultaneously, which can provide dense coverage for kinematic analysis. These advances, together with modern approaches for data augmentation, anomaly detection, and temporal smoothing, yield neural network models that generalize across experimental setups, laying the foundation for standardized behavioral pipelines and more complete access to how animals move.

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